The Effects of Clear-cutting on Soil and Groundwater Nitrate Pools on Martha's Vineyard

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Abstract: This study examined the mechanisms responsible for high nitrate losses to groundwater from clear-cut forest sites on Martha's Vineyard, Massachusetts. The clear-cut sites are experimentally managed as part of an effort to restore native grassland. My goal was to determine whether or not a zone of nitrification existed below the surface of the mineral soil of the disturbed sites where excess nitrate was being produced. I also explored whether or not the nitrate in the groundwater could be accounted for by nitrate in precipitation leaching unprocessed through the soil profile. I collected samples throughout the soil profile to a depth of 2 m, and determined the pool sizes of nitrate and ammonium, rates of mineralization, nitrification, and potential nitrification, and carbon to nitrogen ratios. I also compared the $\delta^{15}N$ isotopic values of precipitation and groundwater samples, and calculated a nitrate budget for one of the clear-cut sites. I found that the nitrate in the groundwater is produced within the soil profile and cannot be accounted for by nitrate deposited in precipitation. In clear-cut sites, the extent of plant re-growth and plant uptake of nitrate are the primary factors controlling nitrate losses. However, low levels of nitrification do occur below the surface of the mineral soil in the clear-cut sites, and these low rates can account for the total nitrate appearing in the aquifer.

Keywords: disturbance, clear-cutting, soil, groundwater, precipitation, nitrate, nitrification, potential nitrification, nitrogen mineralization, ^{15}N

Introduction

Understanding how land-use change affects nitrogen transformations in the soil is important because the nitrogen cycle has been drastically modified by human activity. Today, 60% of the annual nitrogen deposition onto terrestrial surfaces comes from anthropogenic sources (Schlesinger 1997). Human activities that affect the nitrogen cycle include cultivation of nitrogen-fixing crop species, fertilization of crops, fossil fuel combustion (Schlesinger 1997), and alteration of landscapes which in turn affects the movement of nitrogen through ecosystems (Neill 2004). Deforestation is one example of anthropogenic land-use that modifies nitrogen cycling in the soil. Leaching of excess nitrate to the groundwater under clear-cut sites can have detrimental effects on the environment that include the depletion of minerals from the soil, acidification of the soil, eutrophication of downstream freshwater and marine coastal ecosystems (Vitouseq 1997) and increased rates of production and volatilization of nitrous oxide (Vitouseq 1982).

Soil Nitrogen Dynamics

Soil nitrogen availability is primarily determined by cycling of nitrogen from organic matter through decomposition. Mineralization is the process whereby inorganic nutrients and carbon dioxide are released from organic matter by the activity of degradative enzymes released by soil microbes (Schlesinger 1997). The ammonium released during mineralization is either taken up by plants, immobilized by microbes, adsorbed onto clay minerals, leached through the soil, or nitrified by bacteria in the genera, *Nitrosomonas* and *Nitrobacter*. The nitrate produced by nitrification can also be taken up by plants and microbes. Otherwise, it is leached through the soil profile or lost from the system through denitrification (Schlesinger 1997).

Mineralization rates are determined by the total organic nitrogen content in the soil, as well as the availability of carbon. High carbon availability, or organic matter with a high carbon to nitrogen (C:N) ratio, decreases the rate of mineralization because of nitrogen immobilization by microbes (Schlesinger 1997). Rates of nitrification are also lower with higher C:N ratios because of the high demand of heterotrophic microbes for nitrogen, leaving little ammonium left over for nitrification. Lovett et al (2002) found a strong negative correlation between soil C:N and nitrate export. Additionally, nitrification requires oxygen availability and therefore nitrification rates are low at low oxygen levels. Nitrification rates tend to also be lower at low pH and low levels of soil moisture. Release of terpenoid and tannin compounds by some types of vegetation might also inhibit nitrification (Schlesinger 1997).

Nitrogen transformations in the soil can also be greatly influenced by water movement through the soil profile. Flow rates and water pathways affect soil surface properties (and therefore adsorption and desorption dynamics), and determine which microbial communities have access to the dissolved nitrogen in the soil water as well as how long these communities have to transform the nitrogen (Neill 2004).

The relative amounts of the isotopes, ^{15}N and ^{14}N, can be used to gain insight into nitrogen cycling dynamics in forest ecosystems. Soils are typically enriched in ^{15}N because of fractionation during a variety of processes. Soil microbes mineralize and assimilate ^{14}N in favor of ^{15}N (Nadelhoffer 1994). The lighter $^{14}NH_4^+$ is volatilized to NH_3 gas before the heavier $^{15}NH_4^+$ (Schlesinger 1997). Nitrifying bacteria preferentially nitrify $^{14}NH_4^+$, and fractionation also occurs during denitrification (Schlesinger 1997). The two isotopes are also adsorbed differently onto soil exchange surfaces. Plants preferentially take up ^{15}N-depleted nitrogen, but isotopic fractionation during plant uptake is unlikely if systems are nitrogen limited (Nadelhoffer 1994).

Soils deeper in the soil profile are more enriched in ^{15}N than surface soils, and tend to have values around +8 $^{+}$/- 2‰ (Nadelhoffer 1994). Koopmans *et al* (1997) measured $\delta^{15}N$ values of -5.7‰ and -1.2‰ in the organic layers of two Douglas fir stands, and +4.1‰ and +4.7‰ at 70 cm depth. They found that soil water beneath the organic layer was enriched in $^{15}NH_4^+$ and depleted in $^{15}NO_3^-$, and attributed these results to isotopic fractionation in the organic layers associated with high nitrification rates. At 90 cm, they measured soil water $\delta\ ^{15}NO_3^-$ values of -6.7‰ and -6.1‰.

Nitrogen inputs to soils have variable $\delta^{15}N$ values, although they typically range from -10‰ to 0‰ (Nadelhoffer 1994). Koopmans *et al* (1997) measured precipitation $^{15}NO_3^-$ values of between -13‰ and -3‰. In contrast, Durka *et al* (1994) measured precipitation $^{15}NO_3^-$ values between 2.6-6.3‰. The natural abundances of ^{15}N in precipitation varies seasonally, and is influenced by the process of ammonium and nitrate formation in precipitation as well as the emission source of pollutant nitrogen gases (Koopmans 1997).

Disturbance and soil nitrogen dynamics

Various studies have examined the dynamics involved in nutrient loss from disturbed ecosystems. Nitrogen mineralization may increase post-disturbance because removal of the forest canopy increases the temperature and moisture of the soil as well as the intensity and frequency of cycling between wet and dry conditions in the forest floor. Additionally, clear-cutting may increase the availability of substrate for mineralization and decrease competition between heterotrophs and mycorrhizae (Vitouseq 1982). The increased availability of ammonium may lead to higher rates of nitrification, increasing soil nitrate levels to such an extent that re-growing vegetation and microbes are unable to take up the excess nitrate. Leaching of large amounts of nitrate to the groundwater ensues (Schlesinger 1997). Likens et al (1970) found that nitrate concentrations in stream-water increased dramatically after clear-cutting. They attributed this increase to the lack of plant uptake of nitrate produced by microbial nitrification. The increase in nitrate and hydrogen ions produced during nitrification also caused increased leaching of cations from the disturbed site (Likens 1970).

Various characteristics of a disturbed site may regulate the amount of nitrate lost from the system after disturbance. In general, forests with greater nutrient availability prior to disturbance exhibit higher rates of nitrification and nitrate leaching to groundwater (Schlesinger 1997). Processes that prevent nitrogen losses from disturbed systems include ammonium immobilization (Vitouseq 1985), clay fixation, ammonia volatilization, low gross mineralization, lags in nitrification, nitrate reduction to ammonium, nitrate reduction to nitrous oxide or nitrogen gas, nitrate immobilization, nitrate absorption, nitrate reduction at depth, or lack of water flow through the soil (Vitouseq 1982). In general, the intra-system nitrogen cycle stabilizes when vegetation begins to re-grow on the disturbed site (Vitouseq 1982).

Disturbance may also alter both the natural abundance of ^{15}N in the soil and in nitrogen compounds lost from the system. As mentioned above, disturbance enhances soil biological activity by increasing ammonium and nitrate pool sizes. Higher rates of biological activity, and therefore isotopic fractionation, tend to enrich the soil in ^{15}N and increase losses of ^{15}N-depleted compounds (Nadelhoffer 1994). Using $\delta^{15}N$ values and $\delta^{18}O$ values to trace leaching of nitrate through the soil profile, Durka *et al* (1994) found that the percentage of atmospherically deposited nitrate reaching the groundwater from two spruce forests differed depending on the extent of damage to the forests from acidification. In the declining forest, 59% and 114% of the deposited atmospheric nitrate leached through to spring water whereas in the healthy forests only 16-30% of the deposited nitrate leached through the soil profile unprocessed. The authors speculated that acidification prevented plants and soil microbes from consuming the deposited nitrate.

This study builds on a previous one conducted by Neill et al (2004) that also investigated the effects of clear-cutting on nitrogen transformations in the soil on Martha’s Vineyard. They found that immediately after cutting, soil ammonium pools increased dramatically but nitrate pools did not (figures 1 and 2). However, nitrate groundwater concentrations did increase by almost two orders of magnitude one year after cutting (figure 3). There was also no increase in net nitrification or mineralization in surface soils after cutting. The concentration of NH_4^+ in groundwater remained constant during the time when NO_3^- increased dramatically. The questions that my study attempts to answer, then, are: 1.) Is there a zone of nitrification deeper in the mineral soil from which nitrate is leaching down to the groundwater? and 2.) Is the nitrate in the groundwater simply a consequence of nitrate in precipitation leaching through the soil profile, unprocessed due to the absence of plant uptake of both water and nitrate?

Understanding the effects of clear-cutting on nitrogen soil transformations on Martha's Vineyard will provide a site-specific understanding of how disturbance and various management practices affect nitrate leaching to the groundwater. This understanding is particularly important given the current management goal to restore grassland on Martha's Vineyard, and the necessary clear-cutting involved in the restoration process. Understanding the affects of clear-cutting on nitrogen dynamics in turn allows us to predict the implications of this disturbance for downstream freshwater and marine coastal ecosystem productivity, as well as for the future productivity of the clear-cut site itself.

Methods

Study sites

I carried out my study on plots of land that are being experimentally managed as part of a project to restore native grassland on Martha's Vineyard. The study included two groups of sites: 1.) the Kohlberg sites, and 2.) the Mazar sites. The Kohlberg sites included a forested control plot, a plot that was clear-cut in 2001 (MUA-2001) and a plot that was recently cleared in the fall of 2004 (S3-2004). The Mazar group included a site that was cleared in 2002 (Mazar-2002) and an adjacent forested control plot. The vegetation of the control plots is predominantly characterized by black oak (*Quercus velutina*), white oak (*Q. alba*) and black huckleberry (*Galussacia baccata*) (Neill 2004) and the clear-cut sites were also dominated by these species prior to disturbance. In the MUA-2001 clear-cut, the trees were cut at the base of the trunk and removed, but since the clear-cut the stumps have been allowed to re-grow. In the Mazar 2002 clear-cut site the trees were also cut and removed. In contrast to the MUA-2001 site, however, the stumps were also removed by bulldozing. Grasses have re-colonized the plot, but periodic mowing of the site is used to prevent excessive re-growth. Like the Mazar-2002 clear-cut site, the trees and stumps in the S3-2004 clear-cut site have also been completely removed.

The soils of all the sites are sandy with low cation exchange capacity and low adsorption capabilities. The soil organic horizon has a pH of approximately 4, while the surface mineral soil has a pH of about 4.5. The bulk density values for the organic layer and the upper mineral soil, respectively, are 0.3 and 1.78 g dry soil / cm^3. The thickness of the vadose zone ranges from 3-5 m (Neill 2004).

Field techniques

Soils

In each of the 5 field sites (3 clear-cuts and 2 controls), I collected soil samples from three randomly chosen holes at 7 depths: the organic layer, 0-5 cm, 5-10 cm, 30 cm, 60 cm, 120 cm and 200 cm. I homogenized the samples in the lab and removed large roots and rocks. I kept the samples in the fridge and removed portions of soil for various analyses. I collected samples from the organic layer and the upper mineral soil (0-5 cm, 5-10 cm) with a corer, and used an augur 8 cm in diameter to collect soil at lower depths. In one hole at each site, I collected bulk density samples at 60 cm, 120 cm and 200 cm by marking the depth traveled by the augur, weighing the sample in the lab, and calculating the total volume of soil sampled using the volume of the augur cylinder (see Table 1 for bulk density results).

Precipitation and Groundwater

I collected 2-3 liters of groundwater from pre-installed wells in the Control North and MUA-2001 sites. In the S3-2004 site, we installed a well and collected a groundwater sample from there as well. I collected precipitation using buckets from the roof of the Marine Biological Laboratory Ecosystems Center. I froze all water samples, and defrosted them immediately prior to analysis.

Laboratory techniques and calculations

Soil moisture and C:N ratio

I determined the soil moisture content of the samples in order to report my results for nitrogen pools, mineralization and nitrification in ug N per g of dry soil. I weighed out 15-30 g of organic layer samples or 40-90 g of mineral soil samples and dried the samples at 60°C in a drying oven for more than 24 hours. I then combined the dried samples from the three holes at each site (1 g of each mineral soil sample, 0.5 g of organic layer samples), and ground the combined samples using a mortar and pestle. Using the Perkins-Elmer 2400 series 2 CHNS/O analyzer, I analyzed each sample for carbon and nitrogen content and calculated the C:N mass ratios.

Nitrogen pool sizes, net mineralization and net nitrification

To assess net mineralization and nitrification, I incubated 100-300 g organic soil or 200-500 g mineral soil in the dark at 25-30°C for 14 days. I extracted an additional 5 g of organic soil or 15 g of mineral soil with 50 mL of 1 *M* KCl to determine nitrate and ammonium pool sizes, as well as to acquire initial values for calculating net nitrification and net mineralization rates (Shaver 2004). The extracted samples shook for 1 hour. After allowing the shaken slurry to settle, I pressure-filtered 20mL of the supernatant using ashed GF/C filters. I then analyzed post-incubation soil samples for ammonium and nitrate content. I used the Lachat Flow Injection Analyzer to measure nitrate concentrations of the filtered samples and the Shimadzu 1601 Spectrophotometer (set at a wavelength of 640nm) to calorimetrically determine ammonium concentrations. I calculated net nitrification as the difference between the initial and final nitrate concentrations of the incubated soil samples, and net mineralization as the difference between the initial and final nitrate plus ammonium concentrations.

Potential Nitrification

I used the shaken soil-slurry method (Hart 1994) to determine the nitrification potential of soil at 5-10 cm, 30 cm and 120 cm in all three holes of the control sites, the MUA-2001 site and the S3-2004 clear-cut site. I weighed out 15 g of soil in a 250 mL plastic cup and added 100 mL of a 1.5 mM NH_4^+ and 1 mM PO_4^{3-} solution with a pH adjusted to 7.2. I shook the soil slurries for 24 hours on an orbital shaker at 150rpm. At 2, 4, 12 and 24 hours, I extracted 10 mL of the soil slurry, being careful to shake the sample before extraction to prevent settling of soil particles. I then centrifuged the extracted slurry for 8 minutes, and pipetted ~10 mL of the clear supernatant into scintillation vials which were then frozen until analysis. After thawing the vials, I determined the nitrate concentration of the samples using the Lachat Flow Injection Analyzer. To calculate potential nitrification rates, I graphed the sampling time points against ug nitrate-N g^{-1} dry soil and found the slope of the regression line.

$\delta^{15}N$ of groundwater and precipitation

I used an adaptation of the ammonia diffusion method to determine the δ^{15}Nitrogen isotopic values of nitrate in the precipitation and groundwater samples (Holmes 1998; Sigman 1997; LINX II Protocol). First, I determined the nitrate concentration in the groundwater and precipitation samples using the Lachat Flow Injection Analyzer. Next, I calculated the appropriate volume of sample that would yield >14 ug of nitrate for isotopic mass spectrophotometry analysis. I constructed filter packs by pipetting 25 uL of 2.5 M $KHSO_4$ onto 1cm diameter GF/D filters, and sealing the filters with the edge of a glass scintillation vial between two Teflon filters. I added 5 g of ashed NaCl and 3 g of ashed MgO per liter of sample. The NaCl prevents the filter packs from bursting, and the MgO buffers the sample at a pH of 9.7, enabling the conversion of NH_4^+ to $NH_3(g)$ (Holmes 1998). I then boiled down the samples to ~100 mL and poured them into 250 mL plastic bottles. To convert the nitrate to ammonia, I also added 0.5 g Devarda's alloy to each sample (Sigman, 1997). To samples with low nitrate concentrations (all three wells from the Control North site), I added 1 mL of NO_3^- stock solution (or 30ug of NO_3^-). I also assembled two standards by adding 2 mL of NO_3^- stock solution, 5 g of NaCl, 3 g MgO and 0.5 g Devarda's alloy to 100 mL of distilled water. I then incubated the samples and standards at 60°C for 27 hours, retightening the caps after 12 hours. After shaking the samples and standards for 4 days, I placed the filters in a dessicator with an open vial of 2.5 M $KHSO_4$ for over 36 hours. I then sent the filters to the mass spectrophotometer isotope lab for analysis.

I corrected the resulting sample δ^{15}N isotopic values for the N-content and δ^{15}N isotopic value of the Devarda's alloy using values determined in 2003 (Thomas 2003). Given some error in my standard results, I also used a pre-determined δ^{15}N isotopic value to correct the δ^{15}N isotopic values of the spiked samples (Thomas 2003). Finally, I corrected the standard value for < 100% recovery using a linear regression equation relating % NH_4^+ recovery to isotopic fractionation: $\delta^{15}NH_4^+$ (observed – actual) = 0.20 (% N recovered) - 19.95 (Holmes, 1998).

Nitrogen Budget for MUA-2001 Clear-cut

For the MUA-2001 clear-cut site, I used the nitrate concentrations I measured in precipitation and groundwater, the average net nitrification rates of the three soil profiles, and soil bulk density values to assess whether the amount of nitrate I measured in the groundwater could be accounted for by the amount of nitrate deposited by precipitation annually on the site. I assumed that the daily net nitrification rates I measured could reasonably occur 50 during the year. I also assumed that the aquifer was located at a depth of 3 m in the soil profile, that it was 3.5 m thick and that pore space made up 50% of the total aquifer volume. Additionally, I assumed that the total amount of annual precipitation amounted to 1236 mm (SES 2004). In addition to calculating the total wet deposition from the nitrate concentration I measured in my precipitation sample, I also calculated a second estimate using the following regression equation for annual wet nitrate deposition on Cape Cod, formulated by Valiela and Bowen (2001): NO_3^- (kg ha^{-1} yr^{-1}) = 0.025 (year) – 45.6 ($R^2 = 0.47$).

Results

Precipitation and groundwater nitrate concentrations

Consistent with concentrations measured by Neill and colleagues (Neill 2004), I found relatively high concentrations of nitrate in the groundwater beneath the MUA-2001 clear-cut site (Figure 4). Groundwater nitrate concentrations below the control plots were zero, and very low for the S3-2004 clear-cut site (1.84 uM). The nitrate concentration in precipitation was also low compared to that measured beneath MUA-2001, but significantly higher than concentrations measured beneath the S3-2004 and the Control North plot (Precipitation = 4.45 uM).

Nitrogen Pools:

The nitrate pools for the MUA-2001 and S3-2004 clear-cut sites were very similar to those found in the Control North site (figure 5) throughout the soil profile, with the organic layer having a significantly greater nitrate content that samples further down in the profile. However, the S3-2004 site did have slightly more nitrate in the upper mineral layer (5-10 cm and 30 cm) than the other two Kohlberg sites. The nitrate content profile of the Mazar Control site looked very similar to that observed in the Kohlberg sites. In contrast, the Mazar 2002 clear-cut treatment had the highest nitrate content of all the sites throughout the profile with the greatest amount of nitrate found in the organic layer and at 5-10 cm.

The Control North, MUA-2001 and Mazar Control sites had very low ammonium content throughout the soil profile compared to that seen in the S3-2004 and Mazar 2002 clear-cut treatments (figure 6). The organic layer of all the sites had the largest ammonium pool, with ammonium content approaching zero below 30cm. The organic layers of the Mazar 2002 site and the S3-2004 site had drastically larger ammonium pools than the other sites, with the Mazar 2002 (215.5 ug NH_4^+-N g^{-1} dry soil) organic layer having a pool almost seven times larger than the S3-2004 organic layer (32.03 ug NH_4^+-N g-1 dry soil). The 0-5 cm and 5-10 cm samples from S3-2004 and Mazar 2002 also contained significantly more ammonium than the other sites, although the amount (<10ug) was very small compared to the pools in the organic layer.

Net nitrification, net mineralization, and potential nitrification:

Net nitrification rates were zero throughout the soil profile for the control sites, and negative in the organic layers (figure 7). In the Kohlberg sites, net nitrification rates for the two clear-cut treatments were also close to zero below 60cm. However, for the MUA-2001 and S3-2004 clear-cut, nitrate was produced at 5-10 cm, and in the MUA-2001 treatment, at 0-5 cm and 30 cm as well. In the Mazar 2002 clear-cut, rates of nitrification in the organic layer and at 0-5 cm were very high compared to rates further down in the profile and rates for other sites. Nevertheless, a significant amount of nitrate was produced at 30 cm (0.099 ug NO_3^- - N g^{-1} dry soil) and smaller amounts were also produced at 5-10 cm and at 60 cm.

Rates of mineralization did not vary significantly between clear-cut treatments and control sites, except for in the organic layer of the MUA-2001 clear-cut site where the rate of mineralization was much lower than that in the organic layers of the other four sites (figure 8). Mineralization rates for all sites were highest in the organic layers, decreased drastically at 0-5 cm and approached zero at 30 cm.

Potential nitrification rates were comparable between sites at all three depths tested (figure 9). Samples from 5-10cm and 30cm appeared to have very similar rates, while the rates for the 120 cm soil were significantly lower. The 5-10 cm Control North sample, however, did have a slightly lower value than the 5-10 cm samples from the other three sites.

C:N ratios

The mass ratio of carbon to nitrogen did not vary significantly between sites (Figure 10). The soil composites from all sites exhibited higher C:N ratios in the organic layers and 0-5 cm mineral soil. Below 0-5 cm, the C:N ratio decreased with depth. Below 60 cm, the CHNS/O analyzer was unable to detect any nitrogen for most of the sites. Graphing C:N ratio against the nitrate content of the soils did not reveal any correlation between the two variables (Figure 11 A). Similarly, I did not observe any correlation between C:N ratio and net nitrification rates (Figure 11 B). The only site for which there did appear to be a positive correlation between C:N ratio and net nitrification was the Mazar 2002 clear-cut ($r^2 = 0.7576$).

$\delta^{15}N$ of groundwater and precipitation:

The $\delta^{15}N$ isotopic values of the precipitation and the groundwater samples differed tremendously (figure 12). Values for both precipitation samples were very negative (precipitation 1 = -9.04, precipitation 2 = -8.82) while groundwater samples ranged from 1.05 (S3-2004 clear-cut) to 13.08 (Control North 1). Of the groundwater samples, the S3-2004 clear-cut sample had the lowest $\delta^{15}N$ value while the Control North wells samples had the largest values.

Nitrogen Budget for MUA-2001 Clear-cut:

The total annual deposition of nitrate in precipitation cannot account for the high concentration of nitrate observed in the groundwater of MUA-2001 clear-cut (Table 2). My two estimates of annual wet deposition differed greatly, probably because I sampled precipitation during a heavy rain which diluted the nitrate content. Nevertheless, neither my measured approximation nor the calculated estimate (Bowen 2001) could account for the amount of nitrate in the groundwater. In contrast, although rates of net nitrification were very low throughout the soil profile of MUA-2001, these rates (including negative values) could account for the amount of nitrate I measured in the groundwater. Thus, low rates of nitrification can account for high nitrate concentrations in groundwater if all of the nitrate produced leaches completely through the soil profile.

Discussion

Groundwater and Precipitation:

The groundwater nitrate concentrations from the three MUA-2001 wells were variable; however, the well with the highest nitrate concentration (MUA-2001 E4 = 40.3 uM) was downhill relative to the other two wells, suggesting that the pulse of nitrate produced by the clear-cutting in 2001 is moving toward Edgartown Great Pond. I would expect that in approximately one year, the nitrate concentration of the groundwater below the S3-2004 clear-cut site will increase dramatically, given the high ammonium content in the organic layer. Additionally, the treatment is very similar to that of the Mazar 2002 clear-cut, and we see very high relative nitrate pools and rates of net nitrification throughout the soil profile at that site. I would also expect the concentration of nitrate in the groundwater below the Mazar 2002-clearcut site to be much higher than that observed in MUA-2001.

The large difference between the $\delta^{15}N$ values of my precipitation samples and those of the groundwater samples confirms that the nitrate in precipitation is not passing through the soil profile unprocessed to the aquifer. The high positive values of the groundwater samples suggest that the nitrate appearing there originates from organic matter, and has undergone several processes involving fractionation in the soil profile.

The lower values $\delta^{15}N$ values measured beneath the clear-cut treatment compared to the control plot may provide evidence of increased nitrification in the overlying soil profile. Although I did not measure high ammonium content in the MUA-2001 clear-cut organic layer, in 2001 Neill et al (2004) did. Nitrifying bacteria fractionate more when the pool of ammonium substrate is larger (Nadelhoffer 1994). In the control plot, the amount of ammonium available for nitrification is very limited; therefore, nitrifying bacteria would probably nitrify all available ammonium and would fractionate very little. In contrast, below the clear-cut site nitrifying bacteria could fractionate more given the larger pool of ammonium. However, this is simply a speculation and differences in the $\delta^{15}N$ values between the groundwater samples could have been produced by a number of other fractionating soil processes (see introduction).

Soil nitrogen dynamics

The low ammonium content in the organic layers of the MUA-2001 clear-cut and both control plots suggest that the oak forest in the control plots as well as the re-growing oak stumps in the MUA-2001 clear-cut site are extremely efficient at ammonium uptake. The ammonium pools and mineralization rates for the two control sites are very similar, ruling out the possibility that differences in nitrate losses from the Kohlberg and Mazar clear-cut treatments are controlled by pre-disturbance nitrogen availability (Vitouseq 1982). Net mineralization rates also did not differ significantly between control and clear-cut sites, suggesting that mineralization rates are not affected by clear-cutting.

The high ammonium content in the organic layers of the S3-2004 clear-cut and the Mazar 2002 clear-cut is probably produced by: 1.) increased availability of substrate for mineralization, and 2.) decreased uptake of ammonium by plants. This result is consistent with the management practices employed at the different sites. Whereas in the MUA-2001 clear-cut site, the oak stumps have been allowed to re-grow and are therefore taking up much of the ammonium mineralized in the surface soil, at the Mazar-2002 clear-cut site the stumps were removed completely. As a result, only periodically mowed grasses are currently taking up the mineralized ammonium. Additionally, the bull-dozing used to remove stumps probably churned a large amount of organic matter into the soil and left behind many dead and decaying roots, both which provide substrate for mineralization. Although the ammonium content of the S3-2004 clear-cut organic layer is much higher than the control sites, it is still significantly lower than that observed in the Mazar 2002 clear-cut. This smaller ammonium pool is probably a factor of the short time period between the clear-cut and the point at which I collected the soil sample.

The high ammonium content of the organic layers in the S3-2004 and Mazar 2002 clear-cut provides a potential substrate for nitrification. However, the low ammonium content further down in the soil profile in all of the clear-cut treatments suggests that if nitrification were occurring in these soils, it would probably be taking place in the surface soils where there is an available pool of ammonium. Thus, I suspect that the high nitrate content throughout the soil profile of the Mazar-2002 clear-cut site results from leaching of nitrate from the organic layer.

The negative net nitrification rates in the organic layers of the control plots are perhaps a result of immobilization of nitrate by heterotrophic bacteria. These negative values, in addition to the low rates of net nitrification observed throughout the soil profile in the control plots suggests that in the undisturbed forest, populations of nitrifying bacteria are very low and that plants and heterotrophic bacteria rapidly take up nitrate. Intense competition for ammonium between roots, mycorrhizae, heterotrophs and nitrifying bacteria, as well as allelochemic suppression (Rice and Pancholy 1972, Vitouseq 1982) may be inhibiting nitrifier population growth.

Despite the large pool of ammonium in the organic layer of the S3-2004 clear-cut site, net nitrification rates in the surface soils are also low. This is probably because nitrifying bacteria have not yet had sufficient time to establish themselves in the absence of competition, especially given the current cooler fall temperatures. The pool of nitrate at 30 cm is mysterious, given the low levels of nitrate in the soil layers immediately above it. However, given the relatively high rate of net nitrification and potential nitrification in the 5-10 cm soil sample, it may result from nitrate leaching down from the 5-10 cm layer. On the other hand, one of the 30 cm soil samples had a significantly higher nitrate content than the other two, so this sample (or laboratory tubes used for analysis) may have been contaminated.

Negative surface soil net nitrification rates in the MUA-2001 clear-cut again suggest that low populations of nitrifying bacteria are being inhibited by stump re-growth. The fact that Neill and colleagues (2004) found no nitrate in surface soils immediate post-clear-cut of the MUA-2001 site also implies that vegetation has to be completely removed for the nitrifying population to be able to establish itself. On the other hand, there is nitrification occurring at 0-5cm, 5-10cm, and 30 cm in the MUA-2001 clear-cut and, as demonstrated by the nitrate budget calculations for this site, these nitrification rates could account for the total amount of nitrate found in the groundwater. Nitrifying bacteria thus may be able to establish slightly deeper in the soil profile where competition from roots and heterotrophic bacteria is less intense.

My hypothesis concerning the inhibition of an existing nitrifier population by competition or allelochemic suppression is supported by the results of the potential nitrification analysis. Under optimum conditions of temperature and ammonium availability, we see nitrification in the 5-10 cm samples and the 30 cm samples of the control plots and the MUA-2001 and S3-2004 clear-cuts, signifying the presence of nitrifying bacteria (albeit, a small population) in those soil samples. Additionally, net nitrification rates are relatively much higher in the organic layer and 0-5 cm soil sample of the Mazar 2002 clear-cut site where re-growth of vegetation is inhibited. This suggests that the presence of vegetation in the control plots and in the MUA-2001 clear-cut site is responsible for the suppression of nitrification. The low rates of net nitrification measured in the control plots may also be affected by the acidity of the soil, as nitrification tends to occur in soils that have pH values greater than 4.0 (Nadelhoffer 1994).

In contrast to the study by Lovett (2002), I did not find any significant relation between nitrification and the C:N ratio of the soil samples (figure 11). Other factors, such as availability of ammonium and competition between nitrifying bacteria, roots and heterotrophic bacteria appear to be more important in determining nitrification rates.

In conclusion, nitrate losses from the studied sites appear to be controlled primarily by the presence of vegetation. Vegetation takes up excess ammonium, decreasing the pool size of ammonium available to nitrifying bacteria. Plant uptake of nitrate also prevents leaching of nitrate into the groundwater. The high level of nitrate observed in the groundwater beneath the MUA-2001 clear-cut treatment was probably a result of nitrification occurring at the top of the mineral layer, given the low rates of nitrification in the surface soils and the uptake of nitrate by re-growing stump roots. Compared to the Mazar-2002 clear-cut site, however, nitrate pools and net nitrification rates in the MUA-2001 are relatively small and I expect groundwater nitrate concentrations to be very large beneath the Mazar-2002 clear-cut site. If the concentration of nitrate is large beneath the Mazar 2002 site, the primary factor responsible for this is probably the lack of plant uptake at this site. In one year, patterns of nitrification and nitrate pool sizes throughout the soil profile in the S3-2004 clear-cut site will most likely mirror those observed in the Mazar 2002 site, given the similarity in management practice between these two sites.

Management implications

According to my results, nitrate loss from a disturbed site cannot be predicted by studying the C:N ratio of the soil, and low rates of net nitrification deeper in the soil profile can produce high nitrate concentrations in groundwater. Thus, it is difficult to predict nitrate losses from any one site. However, post-disturbance management practices do seem to greatly affect nitrate pools and rates of nitrification in surface soils. Bull-dozing and removal of stumps greatly increases the availability of organic matter for heterotrophic microbes and the availability of ammonium for nitrifying bacteria. This in turn leads to higher nitrate pools and rates of nitrification throughout the soil profile and, presumably, higher nitrate losses from the soil. Restoration projects should focus on minimizing disturbance to the soil and removing excess organic matter. Managers should plant native grasses as soon as possible after clear-cutting to take up excess mineralized ammonium. It would be very difficult to eliminate nitrate leaching from the disturbed site completely; therefore, managers must carefully assess the susceptibility of downstream freshwater and coastal ecosystems to eutrophication, and refrain from disturbing forested sites in close proximity to delicate aquatic ecosystems.

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Graphs and Figures

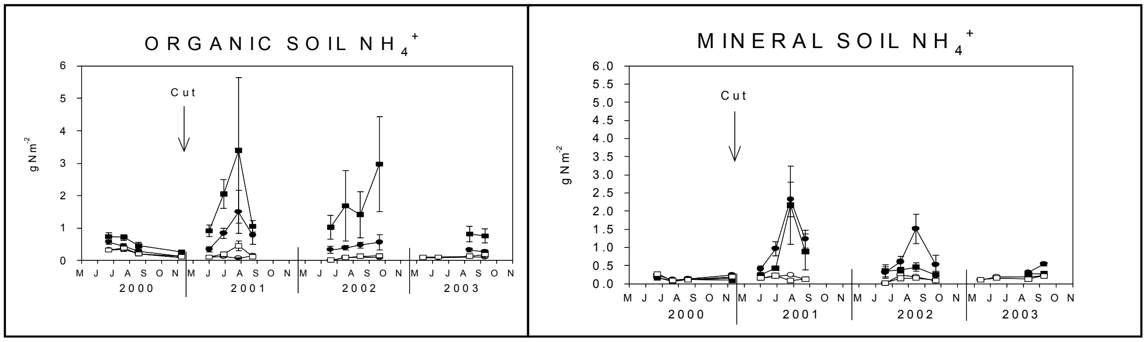

Figure 1: Neill et al (2004). Ammonium pools observed in organic soil and mineral soil (5-10 cm) pre- and post-clearcut in the MUA-2001 clear-cut site.

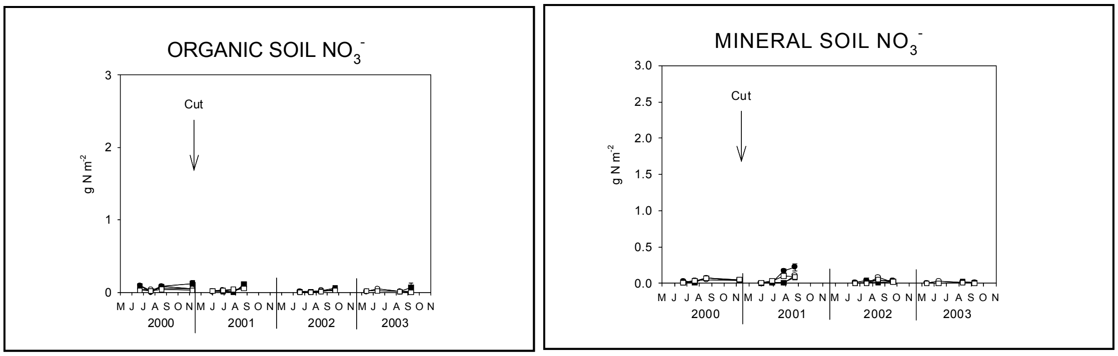

Figure 2: Neill et al (2004). Nitrate pools observed in the organic soil and mineral soil (5-10cm) pre- and post-clearcut in the MUA-2001 clear-cut site.

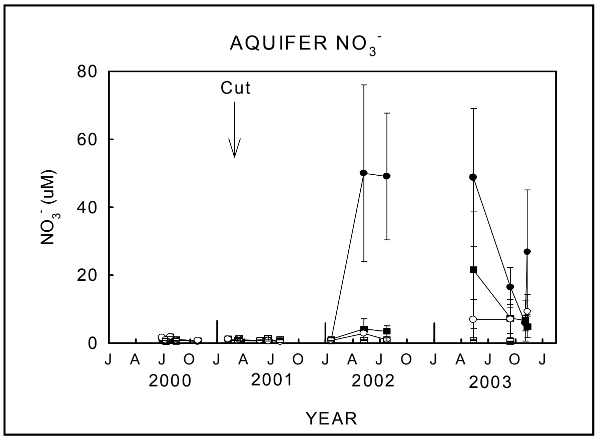

Figure 3: Neill et al (2004). Nitrate concentrations measured in the aquifer below the MUA-2001 clearcut site pre- and post-clearcut.

Soil Depth	*Control North*	*MUA-2001*	*S3-2004*	*Mazar Control*	*Mazar 2002*
60 cm	2.29	2.11	2.01	2.20	1.90
120 cm	2.28	2.33	2.47	2.42	2.26
200 cm	2.54	2.12	2.46	3.80	2.75

Table 1: Bulk density values (g / cm^3).

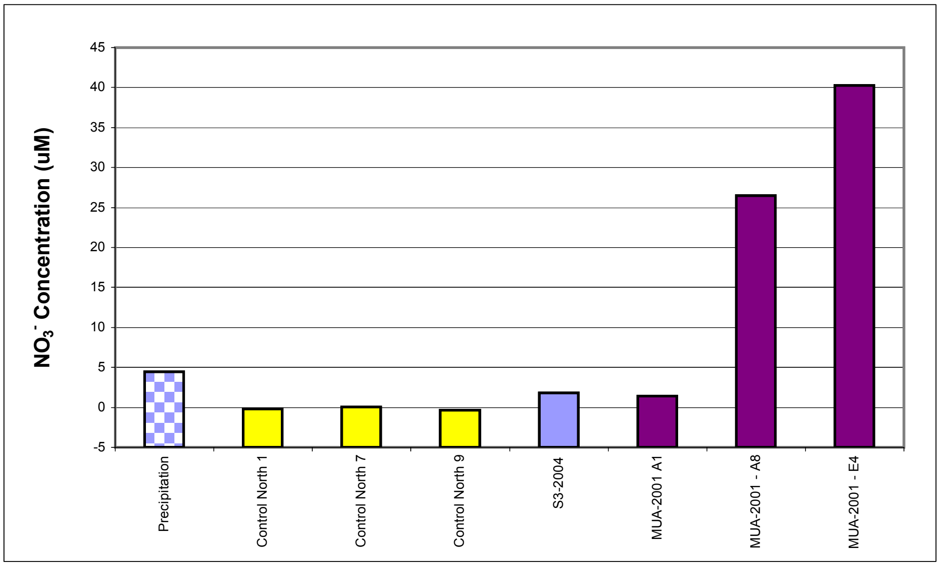

Figure 4: Nitrate concentrations in precipitation and groundwater.

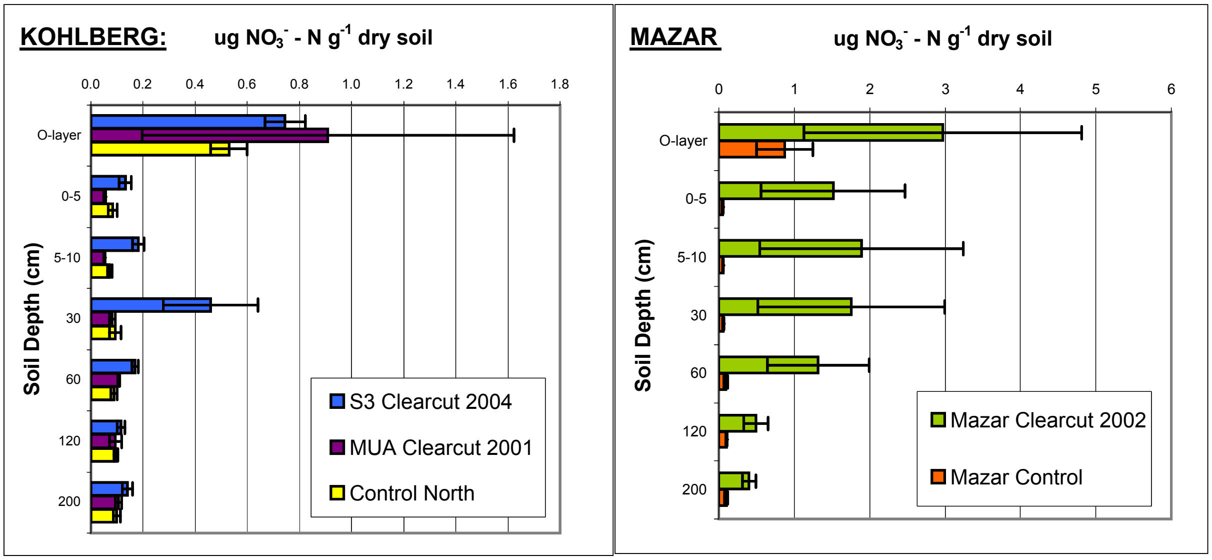

Figure 5: Soil nitrate pools. Error bars represent standard error (Mazar Control 60cm and Mazar 2002: n=4, all others n=3)

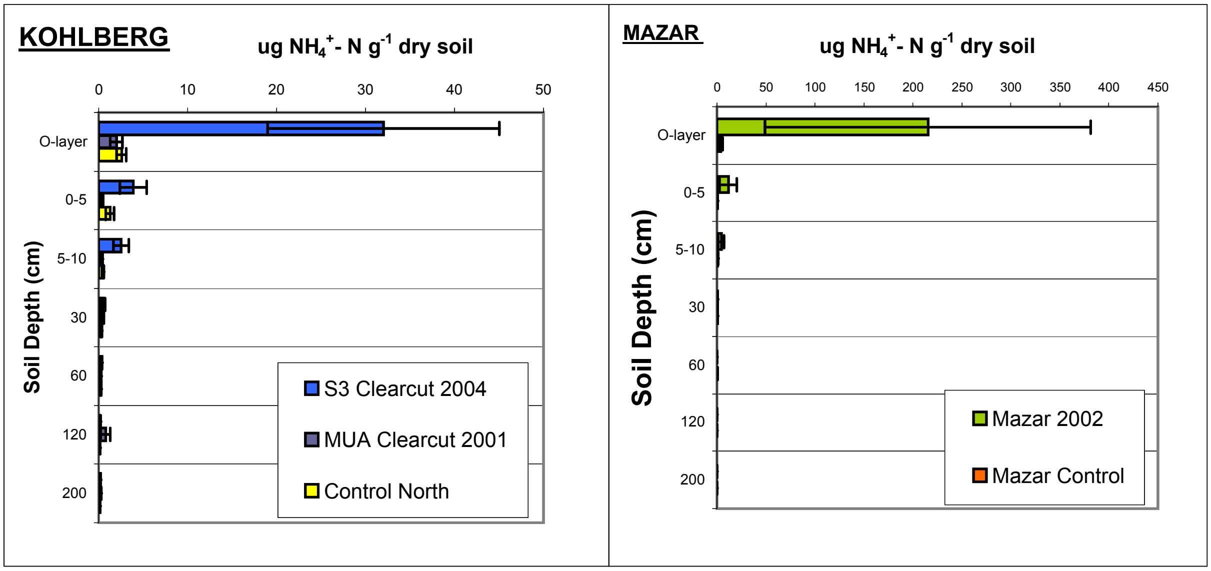

Figure 6: Initial ammonium pools. Error bars represent standard error (Mazar Control 60cm and Mazar 2002: n=4, all others n=3).

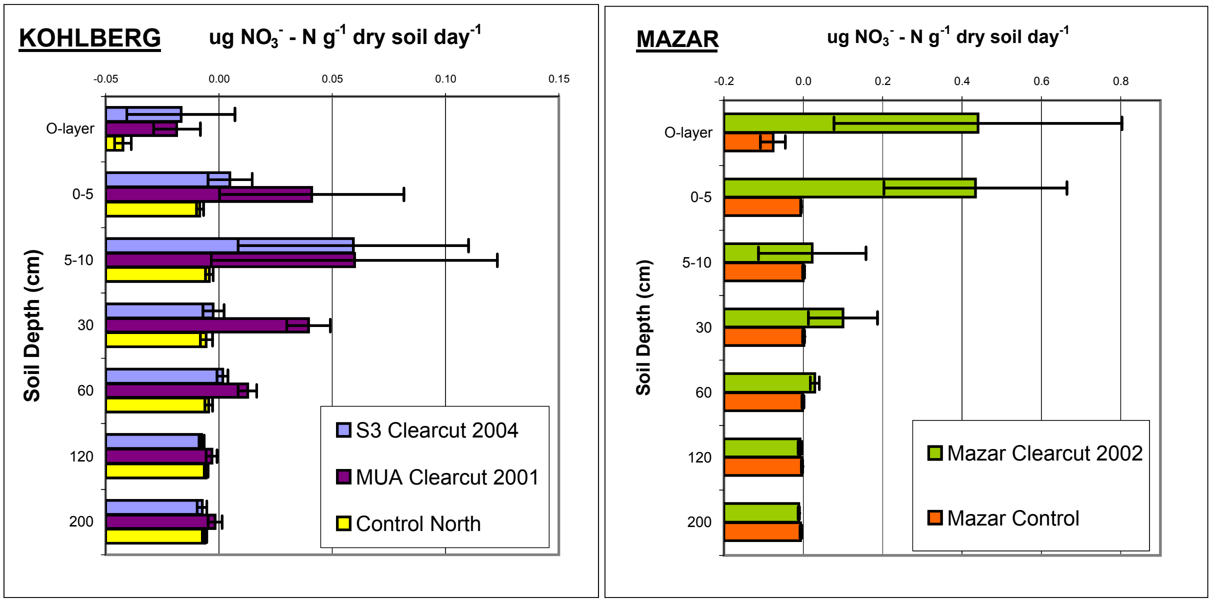

Figure 7: Net nitrification rates. Error bars represent standard error (n=3)

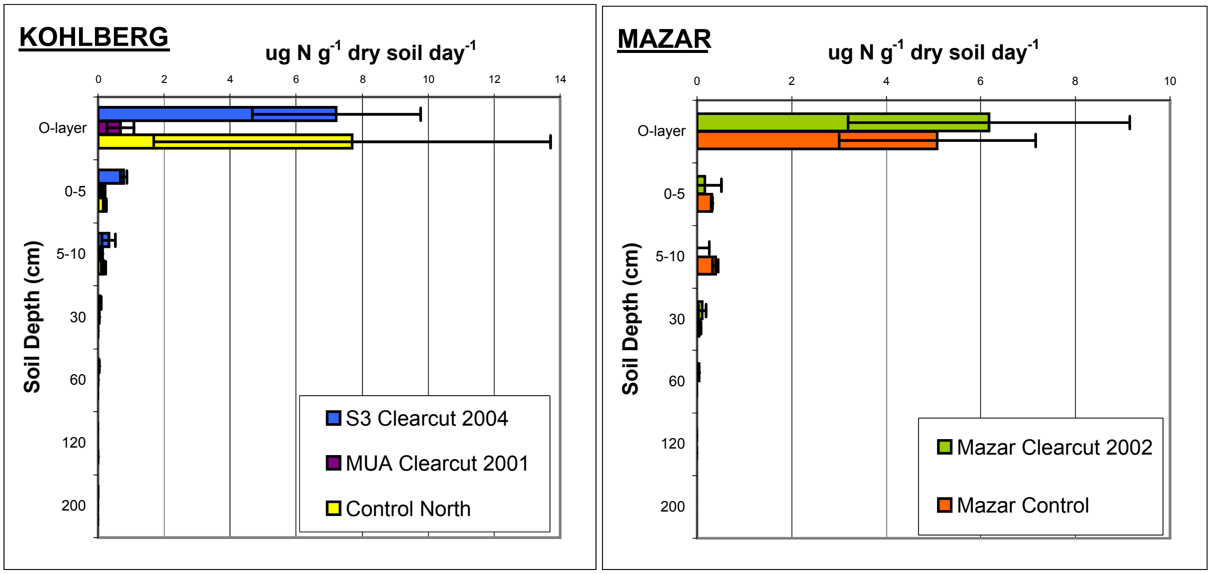

Figure 8: Net mineralization rates. Error bars represent standard error (n=3).

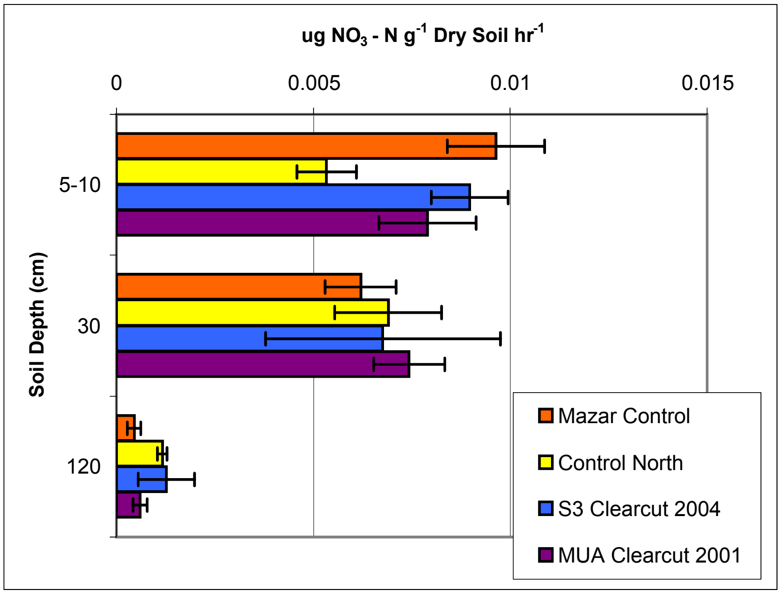

Figure 9: Potential nitrification rates. Error bars represent standard error (n = 3).

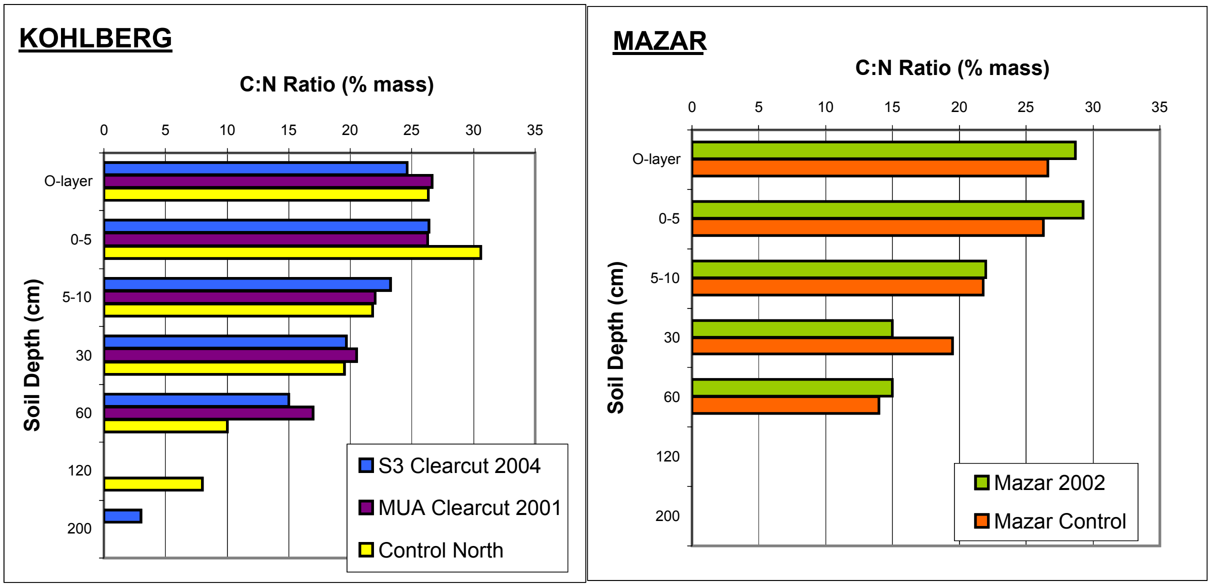

Figure 10: Ratio of carbon mass to nitrogen mass. Each bar represents the ratio for a composite soil sample comprised of depth-specific samples from the three pits at each site.

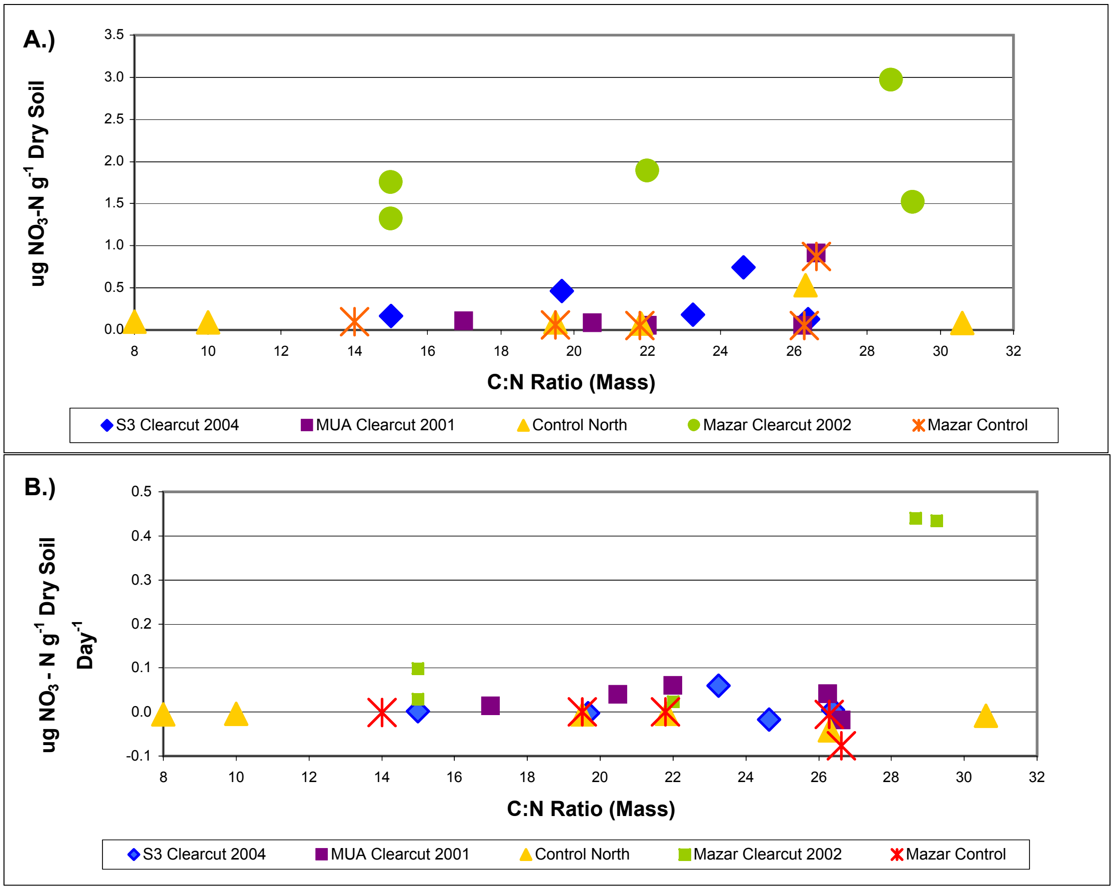

Figure 11: Carbon to Nitrogen mass ratios graphed against A.) initial nitrate pool and B.) net nitrification rates. Samples with infinite C:N ratios due to the inability of the CHNS/O analyzer to detect very small amounts of nitrogen are not graphed.

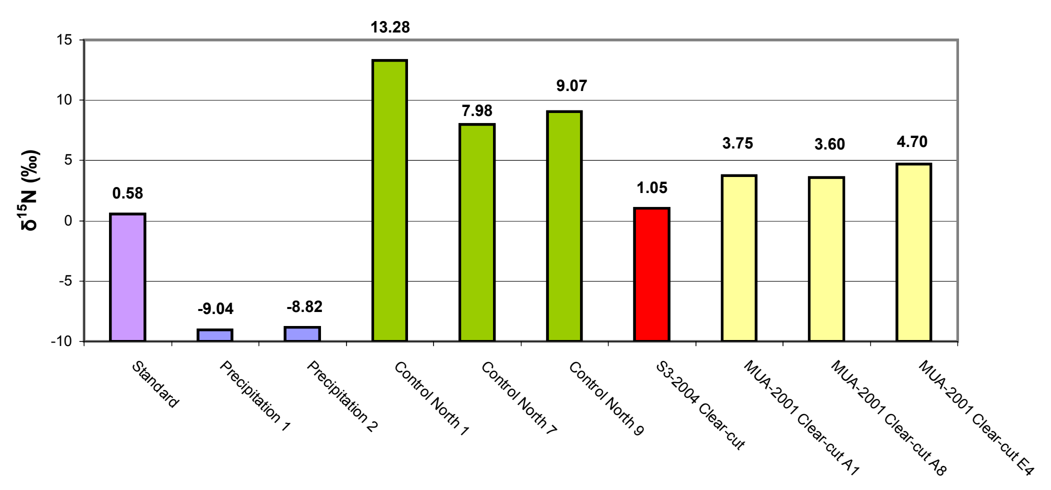

Figure 12: $\delta^{15}N$ isotopic values for precipitation and groundwater samples.

Total nitrate in aquifer (g NO_3^- - N m^{-2}):	Annual nitrate deposited (g NO_3^- - N m^{-2}) (measured):	Annual nitrate deposited in 2004 (g NO_3^- - N m^{-2}) (Bowen 2001):	Total nitrate produced in soil profile, assuming a 50-day growing season (g NO_3^- - N m^{-2}):
0.988	0.077	0.4425	1.743

Table 2: Nitrate budget values for nitrate deposition and production in the MUA-2001 clear-cut.