

Dear Dr. Ibrom,

Thank you for your comments and for your discussion with the reviewers. We understand that you and Referee #2 both feel the manuscript would be strengthened by including vegetation data to support the hypothesis that plant uptake might contribute to patterns of post-fire N availability in this system. We are concerned, however, that there has been an unintended miscommunication regarding the nature of these data. The data were not collected in any way in which they can be leveraged to empirically test this hypothesis. Even without vegetation data, however, we feel strongly that the manuscript remains an interesting and important contribution to the scientific literature. To date, the primary mechanisms put forward in the literature to explain patterns of post-fire nitrogen pulses are mineral inputs from ash and increases in microbial activity, although neither of these have been rigorously tested. Our study makes two clear advances – first we introduce a Bayesian analysis that better incorporates spatial nutrient heterogeneity into field measures of net mineralization rates. This allows us to demonstrate increases in N availability in our system. Secondly, we are able to refute both of the previously proposed mechanisms to which nitrogen pulses have been attributed. This implies that some unaccounted-for biogeochemical mechanism must be involved in regulating nutrient availability following fire disturbance. We view changes in plant uptake as the most parsimonious explanation for this pattern, but our primary goal was to propose this mechanism to the scientific community, encouraging studies that directly test alternative mechanisms of nutrient dynamics following disturbances and particularly those which examine nutrient sinks in vegetation.

We fear that including the vegetation data will weaken the manuscript by drawing attention to the methodological incompatibility of the vegetation and N availability data. Put simply, the unpublished data we cite in this manuscript come from a single time point collected two years prior to the data presented in this study. Because of this, we believe the vegetation data are incompatible with the data we present here; we cited them in the discussion to draw attention to the field-level implications of vegetation nutrient uptake and provide evidence that this proposed mechanism could be plausible. However, our intention was not to imply that our data provide direct support for this hypothesis. Of course it remains entirely possible that plant-uptake does *not* help to resolve patterns of post-fire nutrient availability, or that a different mechanism plays a larger role, as you suggest, but we were unable to identify other plausible mechanisms for this ecosystem.

We propose some organizational changes to that, we hope, clarify the distinction between which hypotheses we tested versus what we encourage future research to target. In particular, we would like to change the title of our manuscript to *The contributions of microbial activity and ash deposition to post-fire nitrogen availability in a pine savanna*. We re-organized the listed hypotheses in the abstract to differentiate the two we tested from the third untested hypotheses that we propose. Finally, in the Conclusions, we explicitly caution against drawing conclusions from our proposed plant uptake hypothesis because it remains untested.

Again, we very much appreciate the time you have invested in this manuscript. Below we attach a line-by-line responses to the reviewer comments as well as a marked up version of the manuscript tracking the changes we have made.

Sincerely,

Cari Ficken

Line Edits for RC1

The authors could improve model description for pool sizes and cycling rates by including more details. The paper is well written with a logic structure and concisely summarised in the abstract.

The model description will be improved based on your specific comments. See below for details.

However, in my opinion, the results do not sufficiently support the interpretations in the discussion, as the chosen setup of study sites does not seem adequate. Firstly, the aim of this study was to present data about the effects of prescribed fire on soil N dynamics; yet, one of the three treatment sites (B2) was affected by wildfire and had a shortened fire return interval compared to the other two sites. Secondly, the two sites affected by prescribed fire had very different responses to fire in terms of vegetation re- sprouting and different standing biomass stocks prior to fire. While the authors related the differences in magnitude of the mineral N pulse to these site differences the number of independent sites (N=2, with three replicate soil cores per site and week) seems too small to support the overall conclusion proposed in the paper - that plant uptake regulates post-fire N availability; especially given the high variance within site pre- and post-burn data and between sites.

We believe there are two important points to consider here. The first relates to the sample size and the effects of prescribed vs wildfire. In this system, prescribed fires are employed to mimic historically naturally occurring wildfires, which occurred on a 1-3 fire return interval (Frost 1998; mean fire return interval = 2.2 years; Stambaugh et al 2011). As such, the wildfire that unexpectedly occurred, did so within the range of both wild- and prescribed-fire intervals observed in this system. After 40 years of either 2- or 3-year fire intervals, there was no statistically significant difference reported in the % cover of any understory plant group (tree seedlings, shrubs, vines, graminoids, forbs, or ferns and mosses; Brockway & Lewis 1997). Consequently, we do

not anticipate substantial differences in biomass accumulation after one year of a shortened fire return interval, and biomass is a strong determinant of fire intensity, and we will amend the next draft of the manuscript to reflect this information as follows (page 5 line 28 – p6 lines 1-4):

The site that burned prematurely due to a wildfire was grouped with other burned sites, despite its shortened fire return interval (one year) relative to the other burned sites (three years). Previous work has found no significant difference in vegetation cover after 40 years of management with either a 2- or 3-year burn interval (Brockway and Lewis, 1997); because biomass is a strong determinant of fire intensity, we did not anticipate that a site experiencing a shortened burn regime for one year would have substantial effects on fire dynamics.

Nevertheless, N=3 sites remains a small sample size and certainly limits our ability to draw conclusions across ecosystems. We will be cautious about the strength of our claims in the next draft of this manuscript. If there are particular sentences in which you feel we have stretched the applicability of these findings, we would appreciate your direction to them.

Secondly, we agree that it is premature to draw conclusions that plant uptake is solely responsible for post-fire N availability dynamics. Rather than claiming that our data conclude this, we believe our data disprove two alternative hypotheses- an increase in microbial processing and ash deposition- that could independently account for the observed patterns. Instead, we hypothesise that the role of plant N uptake is another factor that should be explicitly considered in future studies. We believe we have been appropriately cautious as to the strength of our arguments and conclusions. For example, we acknowledge limitations in our ¹⁵N analyses on page 19 lines 3-7:

Given the uncertainties surrounding the redistribution of surface inputs down the soil profile, we cannot conclusively rule out the potential to surface additions to contribute to the observed NH₄⁺ pulse. Nevertheless, considering the unrealistic mass of ash-N needed to be deposited onto surface soils to account for our measured shifts in δ¹⁵N, we conclude that ash inputs are unlikely to fully account for the increase in measured soil inorganic N availability.

And on page 19, lines 15-20, we acknowledge that it would be inappropriate to conclude that post-fire increases in NH₄⁺ is solely driven by changes in plant sink strength:

If fire damage temporarily halted or slowed the plant uptake of inorganic N, we would expect to see an accumulation of soil N if microbial immobilization did not increase

sufficiently to deplete the pool. However, N accumulating in excess of demand can only partly explain observed increases in inorganic N availability, since the pulse of N we detected following fire was many times greater than what was produced by net mineralization and net nitrification. Nevertheless, a change in plant sink strength may have contributed to post-fire NH_4^+ pulse.

Brockway, DG and CE Lewis. 1997. *Long-term effects of dormant-season prescribed fire on plant community diversity, structure and productivity in a longleaf pine wiregrass ecosystem.* Forest Ecology & Management (96): 167-183.

Frost, C.C. (1998). Presettlement fire frequency regimes of the United States: A first approximation. Proceedings 20th Tall Timbers Fire Ecology Conference: Fire in Ecosystem Management: Shifting the Paradigm from Suppression to Prescription, Boise, ID (eds T.L. Pruden & L.A. Brennan), pp. 70–81. Tall Timbers Research, Inc., Tallahassee, FL.

Stambaugh, MC, RP Guyette, and JM Marschall. 2011. *Longleaf pine (Pinus palustris Mill.) fire scars reveal new details of frequent fire regime.* J. Veg. Science (22): 1094-1104.

Specific comments

The authors may consider revising Figure 1 as the schematic illustration of the paired- core sampling design is not readily understood. For example, it is unclear what the single circle below week 9 represents, is it the last sample for the measurement of pool size? It might be better to depict paired-soil cores for all nine weeks or omit the figure altogether as the sampling design is sufficiently explained in section 2.2.

The circle (core) below week 9 was meant to indicate that the incubating core from the final week's set was collected one week 10, since it was installed on week 9 and incubated for a week. This is consistent with how the other core sets were treated (the dashed circle depicts the core collection timing). We have received comments both that this figure is helpful and unhelpful and have opted to keep the figure, but expand its text to increase clarity.

Authorities for plant species names should be included when species are mentioned for the first time.

Authorities for plant species have been added.

In the methods section, the description of the Bayesian hierarchical models would benefit from including more details, specifically: -

Site effects (intercepts for B1-B3) should be reported –

We have included significant site effects in the text (see below for example). For visual clarity (because site effects were substantially greater than environmental effects), we have omitted them from figures. This omission note is also added to figure legends.

Page 12 lines 9-13: *Site effects (θ_0) had the strongest overall effect on NH_4^+ pool sizes, although this effect was not significant at C1. In burned sites B1-3, θ_0 was -26.9 (95% credible interval (CI) = -46.11– -6.57), -22.03 (95% CI = -38.91– -4.78), and -24.16 (95% CI = -41.78– -6.12), respectively. At C2, θ_0 was -23.23 (95% CI = -40.38– -5.69).*

Page 12 lines 22-23: *Site effects on NO_3^- pool sizes much weaker than for NH_4^+ and were only significant at B1 ($\theta_0 = -4.81$, 95% CI = -9.46 – -0.08).*

Did the authors standardise the coefficients? –

Coefficients were not standardised prior to analysis.

What is the underlying distribution for $\beta_{0i,j}$? –

$\beta_{0i,j}$ has a normal underlying distribution. This information has been added to the text on line 22-23 (page 10): *All predictors, including random site site effects, were modelled with normally distributed, uninformative priors.*

Should the formula in 3b have a minus before $\beta_{6iy0i,j}$ as the initial concentration is subtracted from the incubated concentration? –

The effects of initial concentrations are reflected in the negative posterior estimates of S_0 (Fig 8).

Using the rjags package, how many chains and iterations were run?

Three chains were run with 200,000 iterations after a 100,000-iteration burn-in period. This information will be included in the Methods section (page 10, lines 23-24).

How was convergence tested? –

Convergence was tested by examining chain density and trace plots to ensure proper chain mixing, and by calculating the Gelman-Rubin diagnostics using the `gelman.diag()` function in the coda package to ensure the scale reduction factors for each predictor was <1.05 . This information has been added to the methods section (p 10 line 25 – p11

line 1.

Does $\sigma \sim \text{unif}(0,100)$ relate to both models or just the cycling rates model?

This relates to both models and has been included in the set of equations describing the pool size model as well (p9 line 23).

On page 15 (line 19), please state how soon following fire vegetation re-sprouted in sites B1 and B2.

The information was amended as follow: Page 16 (line 20) *"We noticed that B1 and B2 exhibited rapid vegetation resprouting following fire, while regrowth in B3 was patchy. Vegetation began resprouting in B1 and B2 six days after fire, but not until 18 days after fire in B3 (C. Ficken, unpublished data)."*

On page 18 (line 4), should it read "...and sharp increases in soil temperature with depth. . ." instead of decline?

Yes, it should read "increases", and we changed the word. Thank you for catching this.

In the discussion on page 18 (line 27) authors refer to the preference of plants in pine savanna for uptake of ammonium. It would be good to include a reference confirming this statement about uptake preference in this ecosystem as the authors argue that plant preference for ammonium uptake could explain the relatively large nitrate pool sizes relative to ammonium.

As far as we know, there are no studies explicitly documenting the nitrate vs ammonium preference for species inhabiting pine savannas, but preference for one N form is likely the result of multiple drivers, including enhanced uptake of the dominant N source (e.g. Kronzucker et al 1997; Houlton et al 2007; Wang and Mack 2011). Because nitrification rates are low at low pH, acidic soils often have greater ammonium availability than nitrate. If the availability of each nitrogen form is one component of preference, we expected that plants inhabiting the acidic soils of our study system to take up relatively more ammonium than nitrate. Here, we draw an analogy between seasonal patterns of N availability in longleaf pine savanna (high soil ammonium and low nitrate during the winter, but low ammonium and high nitrate during the growing season; Christensen 1977), and seasonal patterns of N concentrations in northeastern US streams (winter maxima when terrestrial plant N uptake is low; Vitousek 1977). To clarify this, we added references explaining the drivers of plant N uptake patterns and resulting environmental availability, and amended the manuscript as follows (Page 20 lines 5:9):

Preference for NH_4^+ by plants inhabiting acidic soils, where nitrification is limited by low pH and NO_3^- availability is consequently low (Ste-Marie and Paré, 1999; Houlton et al., 2007; Wang and Macko, 2011; Kronzucker et al., 1997), could help to explain the relatively large pool sizes of NO_3^- relative to NH_4^+ during the growing season (Vitousek, 1977), and this pattern is consistent with previous seasonal trends in a longleaf pine savannas (Christensen, 1977).

Temperature is an important influential factor on N transformation processes (MIN, NIT) and soil temperatures might change after fire due to the blackened surface promoting increased heat absorption. The authors could discuss whether they consider soil temperature to have an effect on N transformation processes in the context of their study.

Indeed, temperature is an influential factor for N transformations. To reflect this, we added the following sentence on page 17 (line 10), and included two citations for readers interested in learning more. We limited the discussion of this driver, however, since we did not observe increases in cycling rates following fire.

Soil surface blackening after fire may increase soil temperature and stimulate immediate and prolonged N transformations after fire (Booth et al., 2005; Ojima et al., 1994).

Technical corrections Page 5, line 17: delete 'in'. Page 9, line 1: correct the word 'through'. Page 10, line 23: correct reference to figures to '3-4' instead of '2-3'. Page 11, line 7: correct figure number in brackets to Figure 5. Page 17, line 9: delete first 'ash' in sentence.

These technical corrections have been made, and we appreciate your careful review of our manuscript.

Line Edits for RC2

Specific comments. Pg 2, Line 24: This phrase is confusing and needs clarification: "localized studies with ecosystem-specific temporal data resolution"

This has been changed to “This suggests a need for localized studies with temporal data resolution appropriate for that ecosystem to evaluate the mechanism behind changes in soil N availability following fire.”

Pg 5, line 6: Please provide examples of the Ericaceous species

This has been amended to read “...dominated by Ericaceous species, including *Vaccinium formosum* Andrews, *V. fuscatum* Aiton, *V. tenellum* Aiton, *Lyonia lucida* (Lam.) K. Koch, *L. mariana* (L.) D. Don, and *Gaylussacia frondosa* (L.) Torr. and Gray ex. Torr. ...”

Pg 5, lines 10-15: It would be useful to know more about the burn regime of the study sites, especially since one of the fires was a wildfire. When are prescribed fires normally set? At the same time as the wild-fire? Was the wildfire similar in severity and timing as the prescribed fires?

We did not measure burn temperature in the wildfire site (B2, see p6 lines 14-19), but observationally, the burn in B2 was comparable to prescribed burns in B1 and B3. See p11 lines 8-11.

On P5 lines 12-16 we added “Prescribed burns are set primarily during the growing seasons, when wildfires also occur. To maintain control of the prescribed burns, they are performed as low-intensity backing fires (ignited along a road or other fire break and allowed to burn into the wind). This fire return interval mimics the historical fire return interval of 1 to 3 years (Frost, 1998), with burns occurring on average every 2.2 years (Stambaugh et al., 2011).”

Pg 5, lines 15-19: It’s unclear what is meant by “historical burn characteristics.” The information in parentheses makes it seem like this means that all the sites burned under similar conditions, but the last part of the sentence makes it seem like only the return interval was the same.

Prescribed fires of Sites on a 3-year burn rotation are staggered such that not every site burns on the same year (for example, some sites burn on years 1,4,7,10; other sites burn on years 2,5,8,11). We wanted to target sites that were on the same burn rotation and that burned in the same years. To clarify this, we added “the same” on page 5 line 25 as follow: “...we limited the number of burned sites to those with similar burn histories (i.e. all on the same 3-year burn rotation). ...”

Pg 5, line 20: The grouping of the wildfire with other fire sites needs more justification. The authors go to great length to explain in previous sentences why the three ‘burned’ sites were chosen based on their similarities and then seem to gloss over the grouping of this wildfire with other fire sites despite the fact that it burned at a totally different burn interval and likely under very different conditions.

Indeed the wildfire burned out of rotation, however the climatic and weather conditions at the time of all burns was similar (ie all fires occurred in a 5 day period in July). In this system, prescribed fires are employed to mimic historically naturally occurring wildfires, which occurred on a 1-3 fire return interval (Frost 1998; mean fire return interval = 2.2 years; Stambaugh et al 2011). As such, the wildfire that unexpectedly occurred, did so within the range of both wild- and prescribed-fire intervals observed in this system. Additionally, after 40 years of either 2- or 3-year fire intervals, there was no statistically significant difference reported in the % cover of any understory plant group (tree seedlings, shrubs, vines, graminoids, forbs, or ferns and mosses; Brockway & Lewis 1997). Consequently, we do not anticipate substantial differences in biomass accumulation after one year of a shortened fire return interval, and biomass is a strong determinant of fire intensity. We will amend the next draft of the manuscript to reflect this information (page 5 line 28 to page 6 lines 1-4).

Pg 5, line 22-24: The removal of the third control site from further analyses is questionable. Were these differences among control sites unknown prior to sampling, or were they only discovered after sampling? Is there nothing that can be gleaned from information on this site? How representative of the area is this site?

There was some indication prior to sampling that the third control site differed from the other sites. For example, it had cinnamon fern (*Osmunda cinnamomea* L.), a species that generally grows in wetter areas, growing near our sampling area, while neither the other control nor treatment sites had this species. However, it was not until we examined data on soil nitrogen content that we realized how different this site was from the others. For example, its nitrogen content was at least one order of magnitude greater than all other sites, and it had substantially greater soil moisture than the other sites. This site is not necessarily anomalous of longleaf pine forests- the area is underlain by an undulating water table which gives rise to drastic micro-topographical gradients and high floristic diversity. However, because it had such substantially different initial soil conditions than the remainder of our sites, we excluded it from analyses. We suspect differences in the depth of the water table (specifically, it may be closer to the surface) between the site we removed from analysis and our remaining sites may be a cause of such differences in vegetation cover.

Pg 5, line 23: Five sites is pretty small sample size, especially since the wildfire site might not represent the prescribed fire sites. If the sites are not considered replicates, and instead the cores within a site are the replicates, this should be clarified.

Sites are considered replicates.

Pg 5, line 26: How big was the sampling area?

The sampling area was 1 m². This information will be added to the next draft of the manuscript (p6 line 9)

Pg 5, line 27-28: The description of sampling above the 'ecotone' needs clarification. Was this just to avoid being in the 'extremes' of either upland or lowland?

Indeed this was to avoid sampling either in the riparian wetland areas or the very dry upland areas. We state (p6 line 10) "This topographic location was chosen to minimize the effects of extremely well-drained, hydrologically disconnected (as found in the uplands) or saturated, anoxic (as found in the wetlands) soils on microbial processing."

Pg 6, lines 1-2: The vegetation sampling needs to be described in more detail. Exactly how was this done? Given that the vegetation link to N availability is a big part of this study's conclusion, why aren't these data included or at least described in more detail?

We collected vegetation cover data at the onset of the experiment, prior to any fires, to help ensure that our sites were appropriate replicates. We clarify this as follows (p6 lines 12-13): "At the onset of the experiment, all vegetation within sampling plots was identified to species and the percent cover was estimated." However, we did not anticipate that vegetation regrowth could play a role in structuring post-fire nitrogen availability. Instead, this hypothesis arose as our data did not support other explanatory hypotheses. As a consequence, we did not track vegetation recovery following fire for this study.

Pg 6, line 3: This sentence should be moved to where the fire regimes are being described.

Because the complexities of the fire regime at each site necessitated substantial discussion in advance of actually listing the sites identifiers along with their burn dates, we found it difficult to relocate this sentence earlier in the manuscript. We chose not to move this information so that we could concisely provide the burn dates with the individual sites for all the burned sites we analyzed.

Pg 6, lines 4-7: What temperatures were recorded? Where in the burns were these pyrometers installed? How far apart from each other? How high off the ground?

Burn temperatures are reported in the results (page 11 lines 10-12). We amended the methods section to include to include information on the tag layout within the site.

Pg 6, lines 6-7: Are there other surrogates of fire intensity that could be used to assess this wildfire site? Canopy mortality? Depth of residual organic layer? Char cover?

These prescribed burns are low intensity and do not reach into the canopy. We described observational metrics of fire intensity in the results on page 11 lines 8-10.

Pg 6, line 13: How far apart were cores? "Adjacent to each other" is vague.

We collected cores within approximately 10 cm from each other, but we avoid sampling large roots. We will add this information to the manuscript (p 6 line 25).

Pg 6, line 19: "throughout the growing season" makes it seem like samples were collected over a much longer period. Longleaf probably grow for many weeks (much more than 9 weeks) in North Carolina.

We replaced "throughout" with "during".

Pg 6, line 21: "until they were analyzed" is vague. How long were soils typically stored frozen before analysis?

Soil cores were stored on ice while in the field and were moved to 4C within the same day. Cores were never frozen. We analyzed soils for nitrogen content within 48 hours of collection, as stated on page 7 line 10. Other analyses which are not time sensitive were analyzed in the fall.

Pg 7, line 21: What is the date referring to?

Thank you for catching this. This was an error on the part of our citation managing software. The correct citation for the monthly precipitation values has been added.

Pg 8, line 24: What is SOM? Soil organic matter? From the description in previous sections, no organic layer develops in this system. Please clarify.

An organic litter layer (ie an O horizon) does not develop, but the soil still has some (albeit very little) organic matter. We added a definition for SOM (soil organic matter) on p9 line 19).

Pg 10, line 11-12: The vegetation description should be expanded to show how the communities varied across sites. What functional types are the three species listed? Grasses, forbs, shrubs?

We added the functional types of the listed species.

Pg 10, line 17-18: This sentence seems out of place since no description of fuel load or moisture across the sites is given in this manuscript.

This information will be omitted.

Pg 14, line 16-17: The link to plant communities would be strengthened if there were more detailed plant data included in the study. As written, there's no way to assess whether N

availability co-varied with plant abundance. Apparently these data exist (Pg 15, lines 18 and 21), so why aren't they included?

Unfortunately, we do not have data on vegetation regrowth patterns following fire that can be incorporated in this manuscript. The data that are cited as unpublished corresponded to data collected for other studies with different temporal resolutions. Consequently, the data were not collected in any way in which they can be leveraged to empirically test this hypothesis. That plant uptake might help to explain post-fire patterns of N availability arose because our data refuted two primary mechanisms that are commonly used to explain post-fire increases in soil N availability. This implies that there is some un-accounted for biogeochemical mechanisms that influences post-fire soil N availability; we view plant uptake as a parsimonious alternative hypothesis. We stress, however, that this study was not designed as a test of the hypothesis (p22 lines 2-4). Instead, our goal was to propose it to encourage future explicit tests of this hypothesis (p1 lines 21-23 and p22 lines 4-5). We hope that we have been appropriately cautious about the strength of our arguments and conclusions (for example, we acknowledge limitations in our ¹⁵N analyses on page 19 lines 3-7); our aim was not to claim that our data conclude that plant uptake is solely responsible for post-fire N patterns, but, based on our results, to hypothesize that plant uptake may be another factor that should be explicitly considered in future studies. We have modified the title to reflect the initial goals the experiment. We have also replaced "propose" with "speculate" (e.g. p 15 line 16; p22 line 13) to emphasize that the plant uptake hypothesis has only been described, not explicitly tested, in this study.

Pg 17, this paragraph on ¹⁵N is way too long and hard to follow. Please simplify and condense, or break into a couple of paragraphs.

We have broken this into 3 separate paragraphs to improve the flow of this section.

Pg 18, lines 25-28: Inclusion of the vegetation data would substantially strengthen this statement.

We are unable to provide vegetation regrowth data because this experiment was designed to test how microbial cycling and ash deposition contributed to N availability.

Figure 1 is a bit confusing. The text description could use more detail for clarification.

Comments we received on earlier drafts of this manuscript have found this figure both helpful and unhelpful. Reviewer 1 here commented that the figure was redundant to

the information provided in the text and may not be necessary, but also indicated some uncertainty about illustrations in the figure. As a consequence, we are hesitant to remove the figure entirely and prefer instead to clarify it. We are attaching a revised version of Figure 1 along with this reply. We have substantially increased the text in the figure, and would appreciate your feedback on the revised version.

Technical Corrections: Throughout: “Southeastern” is sometimes one word and some- times two words.

We have replaced “south eastern” with “southeastern” in all cases. Thank you for catching this.

Contributions of microbial activity and ash deposition to post-fire nitrogen availability in a pine savanna

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Key words: nitrogen, mineralization, nitrification, fire, longleaf pine, ammonium, nitrate, plant resource uptake, $\delta^{15}\text{N}$

Abstract. Many ecosystems experience drastic changes to soil nutrient availability associated with fire, but the magnitude and duration of these changes are highly variable among vegetation and fire types. In pyrogenic pine savannas across the southeastern United States, pulses of soil inorganic nitrogen (N) occur in tandem with ecosystem-scale nutrient losses from prescribed burns. Despite the importance of this management tool for restoring and maintaining fire-dependent plant communities, the contributions of different mechanisms underlying fire-associated changes to soil N availability remain unclear. Pulses of N availability following fire have been hypothesized to occur through (1) changes to microbial cycling rates and (2) direct ash deposition. Here, we document fire-associated changes to N availability across the growing season in a longleaf pine savanna in North Carolina. To differentiate between possible mechanisms driving soil N pulses, we measured net microbial cycling rates and changes to soil $\delta^{15}\text{N}$ before and after a burn. Our findings refute both proposed mechanisms: we found no evidence for changes in microbial activity, and limited evidence that ash deposition could account for the increase in ammonium availability to more than 5-25 times background levels. Consequently, we propose a third mechanism to explain post-fire patterns of soil N availability, namely that (3) changes to plant sink strength may contribute to ephemeral increases in soil N availability, and encourage future studies to explicitly test this mechanism.

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Moved down [1]: we further hypothesize that (3) changes to plant sink strength may contribute to ephemeral increases in soil N availability.

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1 Introduction

Temporal heterogeneity in resource supply is ubiquitous across ecosystems (Schimel and Bennett, 2004; James and Richards, 2006; Archer et al., 2014) and such resource pulses can be important if they contribute disproportionately to the overall resource budget of an ecosystem (McClain et al., 2003).

5 Because they vary in magnitude and frequency, nutrient pulses across ecosystems differ in their potential to influence community and ecosystem dynamics. Despite compelling modelling-based evidence suggesting that nutrient pulses can influence ecological dynamics including species richness (Tilman and Pacala, 1993), physiological nutrient uptake constraints (Bonachela et al., 2011), and stoichiometric coupling (Appling and Heffernan, 2014), it can be difficult to predict when and where
10 temporal nutrient heterogeneity will occur. This uncertainty makes it difficult to assess the conditions under which temporal heterogeneity in nutrient supply might influence community- or ecosystem-level functioning.

Nutrient dynamics in pyrogenic systems may be especially variable in time because fire is a major disturbance that influences nitrogen (N) availability (Wan et al., 2001). There is a consensus that, across
15 ecosystems, pulses of soil N, in particular ammonium (NH_4^+), occur in response to fire (Huber et al., 2013; Wan et al., 2001). However the duration and magnitude of these pulses vary strongly by fuel composition, and consequently among forest and fire types (Wan et al., 2001). In northern conifer forests, stand-replacing fires can result in increased soil NH_4^+ concentrations detectable more than one year following the burn (Smithwick et al., 2005; Turner et al., 2007). In contrast, in pine forests of the
20 southeastern US (e.g. longleaf pine savannas), where prescribed fires only consume the understory vegetation, NH_4^+ concentrations following fires are more variable; some studies have documented no change in soil N pools following fire (Christensen, 1977; Richter et al., 1982), while others have documented immediate increases (2 days) that quickly dissipate (Lavoie et al., 1992). This suggests a need for localized studies with temporal data resolution appropriate for that ecosystem to evaluate the
25 mechanism behind changes in soil N availability following fire.

Pine savannas in the southeastern US are often managed with prescribed fires in the absence of recurring natural fires to maintain habitat for endangered species (Sorrie et al., 2006) and nutrient losses or pool redistributions from these fires can be substantial (Boring et al., 2004; Wilson et al.,

Deleted: ecosystem-specific

2002;Wilson et al., 1999). In addition to large quantities of carbon (C) released through fuel consumption (Boring et al., 2004), prescribed fires can release up to 50% of the phosphorus (P) and up to 75% of the N that was stored in the understory biomass (Carter and Foster, 2004;Wan et al., 2001). These nutrients can be lost through volatilization, or redistributed as ash in low intensity fires. Despite
5 the ecosystem-level nutrient losses associated with fires, short-term pulses of increased N availability in the soil are also observed following prescribed forest fires across vegetation types (Certini, 2005;Schafer and Mack, 2010;Smithwick et al., 2005;Wan et al., 2001) including in longleaf pine (*Pinus palustris* Mill.) savannas (Boring et al., 2004). In longleaf pine savannas, because these nutrient pulses occur during a period of rapid post-fire plant regrowth, they may influence successional patterns
10 (Shenoy et al., 2013), plant diversity, and ecosystem productivity.

The mechanisms driving these ephemeral increases in N availability following fire remain poorly resolved, and so it remains difficult to predict how a specific fire will influence local N availability and turnover. Fire can decrease N availability if N is volatilized and lost from the system in high intensity fires (Lavoie et al., 2010;Certini, 2005). On the other hand, fire can increase N availability if it spurs
15 microbial turnover of organic matter (Wilson et al., 2002;Certini, 2005), returns nutrient-rich ash to the system (Boring et al., 2004), or decreases the vegetation demand for N. Short term increases in soil N availability may not translate to longer-term ecosystem retention if N is lost through leaching or as gaseous products during turnover.

In addition to difficulties associated with assessing the relative importance of each mechanism
20 influencing post-fire N availability, logistical challenges remain to accurately measure N availability. First, changes to soil N availability are likely to occur rapidly following fire. Since microbial turnover occurs on a span of hours to days, and plants in fire-adapted systems begin re-sprouting within days to weeks, changes to N availability in pyrogenic systems are also likely to be ephemeral. Previous studies of post-fire N dynamics in longleaf pine savannas have relied on monthly or less-frequent soil samples
25 (Wilson et al., 2002;Lavoie et al., 2010), but this sampling resolution may be too coarse if changes in N dynamics occur rapidly following fire, or are transient. Secondly, net N cycling rates are often calculated as the difference in pool size between two time points. When measured in the field, repeated sampling of the same soil core would control for spatial heterogeneity in starting conditions, but would

likely distort estimates of N dynamics because soil disturbance can increase rates of C mineralization and microbial respiration. Instead, to avoid disturbance associated with repeated sampling of the same core, nutrients are assumed to be distributed homogeneously in a small sampling area. Thus, it is assumed that cores collected in close proximity to each other are comparable, and can be considered replicates. However, nutrient pool sizes can vary by orders of magnitude within a meter (Jackson and Caldwell, 1993), and so these assumptions, while practical, are problematic and often invalid. As such, field estimates of net cycling rates calculated from the difference between two nearby cores may be influenced by the idiosyncrasies of N spatial heterogeneity, and may not accurately represent local or larger-scale N dynamics. Without using expensive tracers, field-based sampling protocols to estimate net nutrient cycling remain imperfect and researchers must collect extensive soil replicates to overcome the issues associated with environmental heterogeneity.

In this study, our broad aim was to evaluate alternative mechanisms driving post-fire changes in N availability while addressing the above mentioned methodological and analytical challenges to estimating net cycling rates. We measured soil N status every week for nine weeks during the 2014 growing season in five longleaf pine savannas sites in North Carolina. Our study is the first that we know of to provide high-resolution temporal (i.e. weekly) data on the effects of prescribed fire on soil N dynamics in longleaf pine savannas. We then used a Bayesian hierarchical linear model to account for heterogeneous *in situ* N availability. The goals of our study were (1) to evaluate the short-term effects of fire on soil inorganic N availability and (2) evaluate whether changes in N pool sizes following fire could be attributed to changes in net microbial cycling rates or ash deposition.

2 Methods

2.1 Study Site and Fire Characteristics

Our study was carried out in a longleaf pine savanna on Fort Bragg Military Reservation (35.1391°N 78.9991°W) near Fayetteville, NC, USA. This area is characterized by deep, sandy and sandy loam soils from the Candor and Blaney series, which lack an O horizon. Mean monthly temperature ranges from 6.9 - 26.0 °C, and mean annual precipitation is 127.5 cm. This area includes numerous

microtopographical gradients represented by numerous low riparian wetlands in an upland matrix. The uplands are well-drained and savanna-like, with an open canopy of longleaf pine (*Pinus palustris* Mill.) and an understory dominated by wiregrass (*Aristida stricta* Michx.; Sorrie et al., 2006). Several hardwood species and *Pinus serotina* Michx. replace *P. palustris* in the wetlands lining streambeds; in these areas, the soil is often saturated and the ground covered with *Sphagnum* L. sp. Separating the uplands from the wetlands, the ecotones have dense, shrubby vegetation dominated by Ericaceous species, including *Vaccinium formosum* Andrews, *V. fuscatum* Aiton, *V. tenellum* Aiton, *Lyonia lucida* (Lam.) K. Koch, *L. mariana* (L.) D. Don, and *Gaylussacia frondosa* (L.) Torr. and Gray ex. Torr.

Since the 1980s, prescribed burns have been used as a management tool to maintain the longleaf pine savannas on the reservation; since the mid-1990s, these burns have occurred on 3-year rotations to promote longleaf pine regeneration and maintain habitat for rare and endangered species (Sorrie et al., 2006). Prescribed burns are set primarily during the growing seasons, when wildfires also occur. To maintain control of the prescribed burns, they are performed as low-intensity backing fires (ignited along a road or other fire break and allowed to burn into the wind). This fire return interval mimics the historical fire return interval of 1 to 3 years (Frost, 1998), with burns occurring on average every 2.2 years (Stambaugh et al., 2011). Fort Bragg is composed of burn parcels (hereafter “sites”) with independent burn histories. Permanent vegetation sampling transects spanning the topographic gradient have been maintained in 32 sites since 2011; the burn regime in the majority of these sites has been experimentally altered, with a subset of sites being maintained in 3-year burn intervals (Ames et al., 2015).

From these sites, we selected three sites scheduled to burn in 2014, and three not schedule to burn in 2014 for use in a before-after-control-impact experiment. To avoid any artefacts associated with different historical burn characteristics (e.g. historical burns occurring in wetter or dryer years; historical burn intensity and frequency), we limited the number of burned sites to those with similar burn histories (i.e. all on the same 3-year burn rotation). However, one site not scheduled to burn until 2016 experienced a wildfire in July 2014, and another site scheduled to burn did not. The site that burned prematurely due to a wildfire was grouped with other burned sites, despite its shortened fire return interval (one year) relative to the other burned sites (three years). Previous work has found no

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significant difference in vegetation cover after 40 years of management with either a 2- or 3-year burn interval (Brockway and Lewis, 1997); because biomass is a strong determinant of fire intensity, we did not anticipate that a site experiencing a shortened burn regime for one year would have substantial effects on fire dynamics. The site that failed to burn was considerably different from the remaining sites

5 (% soil moisture and % soil organic matter were both more than double that of the other sites), and as such was dropped from further analyses. Thus, we were left with 5 study sites: B1, B2, B3 experienced burns in 2014; C1 and C2 were control sites that remained unburned. Our study sites, renamed here for clarity, correspond to sites 3, 11 (wildfire site), and 15b (B1-3), and 9 and 32 (C1 and C2) described in Ames et al. (2015). At each site, we established a 1 m² sampling area approximately 5 m upslope of the
10 ecotone. This topographic location was chosen to minimize the effects of extremely well-drained, hydrologically disconnected (as found in the uplands) or saturated, anoxic (as found in the wetlands) soils on microbial processing. At the onset of the experiment, all vegetation within sampling plots was identified to species and the percent cover was estimated.

Burns occurred in treatment sites B1, B2 (wildfire), B3 on 4 July, 9 July, and 7 July 2014, respectively
15 (Julian days 185, 190, and 188). Using metal tags marked with Tempilaq temperature-sensitive paint (Air Liquide America 296 Corporation, South Plainfield, NJ, USA), we collected data on aboveground fire temperature at B1 (6 tags) and B3 (8 tags). Tags were installed on metal polls approximately 50 cm above the soil surface. They were installed surrounding our plots approximately 3 m apart from each other. We did not collect fire temperature data at B2 because it was not initially scheduled to burn.

20 2.2 Soil Analyses

From May 30 through July 25, 2014, soil cores were collected weekly (nine weeks) from each site for pool size measurements (Figure 1). We collected pool size cores for a minimum of three weeks after a prescribed burn. As such, our site-specific sampling allowed us to collect data before and after burns and detect any immediate changes in N concentration in response to the burn. Each week, three cores
25 (each 5 cm diameter x 12 cm deep and adjacent (~10 cm) to each other) were randomly collected from each site, for a total of 27 cores collected for pool size measurements at each site over the nine-week sampling period. (In burned sites, the number of cores collected prior to versus following prescribed

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fires differed between sites depending on when the burns occurred. In B1, N=15 cores were collected prior to burns and N=12 cores were collected following prescribed burns. In B2 and B3, N=18 cores were collected prior to burns and N=9 cores were collected following prescribed burns.) These cores were used to compare the pool sizes of nitrate (NO_3^-) and NH_4^+ during the growing season (Figure 1).

5 After collection, all cores were stored on ice, immediately transported back to the laboratory, and kept at 4 °C until they were analysed for inorganic N, soil moisture, soil organic matter, pH, and $\delta^{15}\text{N}$. All cores were homogenized by passing through a 2 mm sieve. Frequent fires in this ecosystem consume aboveground vegetation and litter, preventing the development of an O horizon in these soils (Boring et al., 2004). As such, sieving removed coarse root fragments, rather than partially decomposed organic

10 matter. Subsamples (~10 g) from each core were extracted within 48 hours of collection with 2 M KCl for inorganic N concentrations. The samples were shaken for 30 minutes, centrifuged, and the extract was then filtered out and stored frozen until analysis on a Lachat QuikChem 8500. Additional soil subsamples were oven dried for gravimetric soil moisture analysis, and combusted at 450 °C to measure organic matter content. Finally, we measured soil pH in 2:1 dH_2O :soil ratios with a bench top pH probe.

15 To measure net cycling rates, we also installed three PVC collars (5 cm internal diameter x 12 cm deep) in each site every week (N=27 cycling rate cores collected at each site over nine-week sampling period; the number of cores collected prior to versus following the prescribed burns differed between sites depending on when burns occurred, as above). These collars were installed adjacent to the soil cores collected for pool size measures (Figure 1), and were incubated *in situ* for one week, after which time

20 they were collected and analysed as above for inorganic N pool sizes, soil moisture, soil organic matter, and pH. After allowing for error in the initial pool size of the incubated soil core (see below for model details), net nitrogen cycling rates were calculated based on the difference in extractable NO_3^- and NH_4^+ in the incubated cores and the un-incubated cores. That is, while traditional methods assume that the N pool sizes in the initial and incubated soil cores are equivalent, our analyses calculated net cycling rates

25 allowing for differences in initial conditions between the two cores.

2.2 $\delta^{15}\text{N}$ Analysis and Mixing Models

To assess plant-derived ash inputs to soil N pools after fire, we analysed soils from a subsample of time points for $\delta^{15}\text{N}$. Plants generally discriminate against ^{15}N in favour of ^{14}N uptake (Craine et al., 2015; Hogberg, 1997), and, as such, vegetation tends to be depleted in ^{15}N relative to soil. If appreciable plant-derived N was deposited on burned sites as ash, we expected to see a decrease in $\delta^{15}\text{N}$ while observing an increase in N pool size. Although ash is deposited on surficial soils, heavy rains occur frequently during June-August in this region of North Carolina (11.5, 14.8, and 11.5 cm, respectively; State Climate Office of NC), and the well-drained, sandy soils could leach nutrients through the soil profile. Moreover, because plants begin resprouting within days of a fire (personal observation), we did not want to discount the potential for nutrients to be redistributed by roots. As a consequence of our uncertainty regarding the vertical distribution of deposited ^{15}N , we subsampled the full soil cores (0-12 cm) for ^{15}N analyses. The enrichment of the sample in ^{15}N is reported on a per mille basis (‰) and was calculated as follows:

$$\delta^{15}\text{N} (\text{‰}) = \frac{R_{\text{sample}} - R_{\text{standard}}}{R_{\text{standard}}} \times 1000 \quad (1)$$

where R_{sample} is the ratio of $^{15}\text{N}:^{14}\text{N}$ in the sample, and $R_{\text{standard}} = 0.0036$, the ratio in atmospheric N_2 .

We subsampled cores collected at each site for pool size estimates from the two sampling weeks pre-burn and the two sampling weeks post-burn, for a total of four consecutive sampling weeks surrounding each burn (for B1-3, N=6 unburned and N=6 burned samples; for C1 and C2, N=12 samples). For unburned sites, we subsampled $\delta^{15}\text{N}$ for four weeks corresponding to the same consecutive weeks surrounding the burn dates in burned sites (hereafter *burn season*). For example, site B1 burned on July 3, 2014 corresponding to our sixth sampling week. For this site, and for the unburned site C1, we therefore measured $\delta^{15}\text{N}$ from soil subsamples in the 4th-7th sampling weeks. Because our sites were not truly paired, we chose time points for $\delta^{15}\text{N}$ analyses in the unburned sites based on the burn dates of the closest burn site. In this way, we allowed ourselves to detect any ash drifting between sites. Soils were oven dried at 40°C until a constant weight, then ground finely, encapsulated in tin capsules, and combusted on a Carlo-Erba Elemental Analyzer coupled to a mass spectrometer at the Duke Environment Isotope Laboratory.

We used two end-member mixing models to estimate the mass of ash-N deposited onto the system. We used $\delta^{15}\text{N}$ and N concentrations from pre-burn soil, and published $\delta^{15}\text{N}$ signatures of ash (-0.81 ‰; Huber et al., 2013) as the end-members, and post-burn soil $\delta^{15}\text{N}$ and N concentrations as the resulting mixture from the two end-members. We solved for the mass of ash-N needed to be deposited in order to account for the observed shift in soil $\delta^{15}\text{N}$ signature.

To assess whether ash inputs could be detected in post-burn soil cores using the natural abundance of ^{15}N , we also calculated the mass N needed to be deposited to observe a shift in soil isotopic signature of the minimum external precision (0.1‰ $\delta^{15}\text{N}$ at 1 standard deviation).

2.3 Model Development and Statistical Analyses

2.3.1 Pool Sizes

To assess the spatial distribution of nutrient (NO_3^- and NH_4^+) and soil organic matter availability, we calculated the coefficient of variation (CV) for each site prior to prescribed burns. To understand how fire and soil variables affect N pool sizes, we fitted a Bayesian hierarchical linear model; this is akin to a multiple regression that also allows for variability in the relationship between true soil N pool sizes (μ) and measured pool sizes (y) which might occur, for example, through analytical error. Any effect of soil environmental conditions on N pool sizes would occur on μ , not y . Each core was modelled independently, and we accounted for site blocking effects by including random intercepts for each site. For core $i = 1 \dots n$ at site $j = 1 \dots 5$, observed N pool size (NH_4^+ or NO_3^-) was modelled as a function of random site effects, percent soil moisture (SM), percent soil organic matter (SOM), soil pH, and the number of days since the previous burn (days since fire; DSF) as follows

$$y_{0ij} \sim N(\mu_{0ij}, \sigma^2) \quad (2a)$$

$$\mu_{0ij} = \beta_{0ij} + \beta_{1i}\text{SM}_i + \beta_{2i}\text{SOM}_i + \beta_{3i}\text{pH}_i + \beta_{4i}\text{DSF}^{-1}_i \quad (2b)$$

$$\sigma \sim \text{unif}(0,100) \quad (2c)$$

For NO_3^- , we added an additional predictive parameter, $\beta_{5i}\text{NH}_4^+$ to allow for NO_3^- concentrations to additionally vary with nitrification substrate (NH_4^+) availability. We expected the effects of a burn to

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diminish with time, so we transformed DSF to DSF^{-1} , so that as DSF increased, $DSF^{-1} \rightarrow 0$. Full models included all main effects and no interactions.

2.3.2 Cycling Rates

We built a hierarchical state-space model within a Bayesian framework to understand how fire and soil variables affected N cycling rates. As in the models of N pool sizes above, our cycling rate models allowed for variation in the relationship between true (μ) and measured (z) cycling rates, modeled below as τ . In addition, we also allowed for error associated with the assumption that the N concentrations in initial cores (y_0 ; as from equation 2a) were equal to the initial concentrations of the incubating cores (y_1). By including these errors into our model, we essentially relaxed the assumption that paired cores (un-incubated and incubated cores) were true replicates and had equal initial N concentration and edaphic conditions (SOM, pH, SM). We removed four core pairs (of 135) that exhibited NH_4^+ or NO_3^- concentrations below the detection limit in the initial concentration. For core $i = 1 \dots n$ at each site $j = 1 \dots 5$, cycling rate was modelled as

$$z_{i,j} \sim N(\mu_{i,j}, \tau) \quad (3a)$$

$$\mu_{i,j} = y_{1,i,j} - y_{0,i,j} = \beta_{1,i}SM_i + \beta_{2,i}SOM_i + \beta_{3,i}pH_i + \beta_{4,i}DSF^{-1}_i + \beta_{5,i}D_i + \beta_{6,i}y_{0,i,j} \quad (3b)$$

$$\tau = \sigma^{-2} \quad (3c)$$

$$\sigma \sim \text{unif}(0,100) \quad (3d)$$

To model cycling rates, we included incubation length (D , in days) and initial NH_4^+ or NO_3^- concentration as additional predictors. Again, full models included all main effects and no interactions.

Prior to all analyses, we removed two cores from B1 (burned) and two from C2 that had pool size values below the analytical detection limit (four of 135 cores). All models were built with the rjags package (version 3.15) in R version 3.2.1 (R Development Core Team, 2011). All predictors, including random site site effects, were modelled with uninformative normally distributed priors. Models were run with three chains and run with 200,000 iterations after a 100,000-iteration burn in period. Convergence was tested by examining chain density and trace plots to ensure proper chain mixing, and by calculating the Gelman-Rubin diagnostics using the gelman.diag() in the coda package (version

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0.18.1) to ensure scale reduction factors for each predictor was <1.05. All values are reported with 95% credible intervals (CI).

3 Results

3.2 Site Conditions and Fire Characteristics

5 Although plant community composition varied, shrubs *Gaylussacia frondosa* (L.) Torr. & A. Gray ex
Torr., *Clethra alnifolia* L., and the native bamboo *Arudinaria tecta* (Walter) Muh. were dominant in all
of our study sites. Study sites were dry, low in organic matter, and acidic (Figure 2). Prescribed burns in
all three sites thoroughly consumed all or most of the aboveground biomass. Aboveground understory
vegetation in B1 and B3 was completely consumed. In B2 some scorched leaves remained on the
10 woody vegetation, but the herbaceous understory species were completely consumed. Fire temperatures
were similar between B1 and B3: average maximum fire temperature at B1 was 612 °C ±18 and at B3
was 635 °C ±18. Fire temperature was not measured in B2.

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3.3 Pool Sizes

To assess the fine-scale spatial variability in NH_4^+ and NO_3^- concentrations, we compared the CV of
15 NO_3^- and NH_4^+ pool sizes within each site prior to prescribed burns. Both NO_3^- and NH_4^+ pool sizes
were highly spatially heterogeneous, despite similar mean concentrations across sites prior to prescribed
burns (Figures 3-4). Spatial variability in NO_3^- pool size was high within each site, but each site
exhibited similar variation in NO_3^- pool size. CV- NO_3^- ranged from 42.3 in B1 to 57.6 in C2. NH_4^+ pool
sizes were also highly variable across sites, and there was a considerable range of the spatial
20 heterogeneity across sites. CV- NH_4^+ ranged from 57.0 in B2 to 114.2 in C2.

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In the first week of sampling, initial pool sizes of inorganic N were similar between sites. Over the pre-
burn season, sites had greater NO_3^- than NH_4^+ availability (3.06 ±0.16 µg NO_3^- per gram of dry soil
[gds⁻¹] and 0.86 ±0.07 µg NH_4^+ gds⁻¹). However, the ratio of NH_4^+ : NO_3^- increased following
prescribed burns and sites B2 and B3 both experienced a shift in the dominant inorganic N form to
25 NH_4^+ immediately after a burn.

There were observable increases in NH_4^+ pool sizes immediately after a burn relative to the same time points in unburned control sites and time points in burned sites immediately prior to the burn (Figure 5). Three days post-burn in Site B1, NH_4^+ had increased from $0.83 \pm 0.15 \mu\text{g NH}_4^+ \text{gds}^{-1}$ in the pre-burn season to $6.10 \pm 1.08 \mu\text{g NH}_4^+ \text{gds}^{-1}$. One day post-burn in Site B2, NH_4^+ had increased from 0.91 ± 0.12
5 $\mu\text{g NH}_4^+ \text{gds}^{-1}$ in the pre-burn season to $7.79 \pm 2.09 \mu\text{g NH}_4^+ \text{gds}^{-1}$. The pattern of NH_4^+ pool size change in Site B3 was qualitatively different than changes observed in Sites B1 and B2. In Site B3, there was an approximate exponential increase in NH_4^+ pool size that plateaued, but did not diminish, by the end of our sampling, more than 3 weeks post-burn. Three days following a burn in Site B3, NH_4^+ pool size had increased from $0.84 \pm 0.16 \mu\text{g NH}_4^+ \text{gds}^{-1}$ in the pre-burn season to $21.9 \pm 6.31 \mu\text{g NH}_4^+ \text{gds}^{-1}$. Site effects (β_0) had the strongest overall effect on NH_4^+ pool sizes, although this effect was not significant at C1. In burned sites B1-3, β_0 was -26.9 (95% credible interval (CI) = -46.11 – -6.57), -22.03 (95% CI = -38.91 – -4.78), and -24.16 (95% CI = -41.78 – -6.12), respectively. At C2, β_0 was -23.23 (95% CI = -40.38 – -5.69). After random site effects, pool sizes of NH_4^+ were most strongly correlated with days since fire (DSF; $\beta_{\text{DSF}} = 12.50$, 95% credible interval (CI) = 5.34 – 19.66 ; Figure 6) and pH ($\beta_{\text{pH}} = 6.16$,
15 95% CI = 1.79 – 10.46 ; Figure 6). Because we fit our pool size models to the inverse of DSF (i.e. DSF^{-1} ; see *Methods*), the positive correlation between NH_4^+ and DSF indicates decreasing pool sizes as time since fire lengthens. Pool sizes of NH_4^+ were larger for less acidic soils and in recently burned soils. Pool sizes of NH_4^+ were slightly greater in soils with more organic matter ($\beta_{\text{SOM}} = 0.2$, 95% CI = 0.01 – 0.30), but did not vary with soil moisture ($\beta_{\text{SM}} = -0.11$, 95% CI = -0.30 – 0.07 ; Figure 6).

20 In contrast to observed pulses of NH_4^+ availability following fire, we did not find strong fire-associated increases in NO_3^- pool size (Figure 5). NO_3^- availability ranged from 0.1 to $8.56 \mu\text{g NO}_3^- \text{gds}^{-1}$, and on average was $2.85 \pm 0.15 \mu\text{g NO}_3^- \text{gds}^{-1}$. Site effects on NO_3^- pool sizes much weaker than for NH_4^+ and were only significant at B1 ($\beta_0 = -4.81$, 95% CI = -9.46 – -0.08). DSF was not related to NO_3^- pool sizes ($\beta_{\text{DSF}} = 0.44$, 95% CI = -1.22 – 2.10 ; Figure 6). Pool sizes of NO_3^- varied with soil organic matter content
25 ($\beta_{\text{SOM}} = 0.03$, 95% CI = 0.01 – 0.07), NH_4^+ pool size ($\beta_{\text{NH}_4^+} = 0.06$, 95% CI = 0.02 – 0.10), and pH ($\beta_{\text{pH}} = 1.46$, 95% CI = 0.39 – 2.49 ; Figure 6).

We also examined whether there was a long-term legacy of fire detectable in sites that had not experienced prescribed burns in that growing season (i.e. across pre-burn time points in burned sites, and all time points from unburned sites). In these areas that had not experienced recent fires, DSF had no effect on NH_4^+ ($\beta_{\text{DSF}} = 0.70$, 95% CI = -60.54-61.46) or NO_3^- availability ($\beta_{\text{DSF}} = 1.79$, 95% CI = -60.31-63.75). Soil moisture ($\beta_{\text{SM}} = 1.55$, 95% CI = 0.65-2.45) and, to lesser extents, NH_4^+ availability ($\beta_{\text{NH}_4^+} = 0.19$, 95% CI = 0.45-0.72) and soil organic matter ($\beta_{\text{SOM}} = 0.03$, 95% CI = 0.01-0.07) were positively correlated with NO_3^- pool sizes.

3.3 Cycling Rates

Net N cycling rates were generally low and temporally heterogeneous across the growing season (Figure 7). They varied between net production and net consumption between sampling points (Figure 7). Across the whole growing season, net mineralization in unburned sites was $0.19 (\pm 0.16) \mu\text{g gds}^{-1} \text{day}^{-1}$; in burned sites, it was $0.26 (\pm 0.05) \mu\text{g gds}^{-1} \text{day}^{-1}$. Net mineralization rates were appreciably more variable the week following a burn, but patterns between sites were inconsistent (Figure 7). For the first week following burns in sites B1-3, net mineralization rates were $-1.72 (\pm 0.32)$, $0.31 (\pm 0.10)$, and $2.54 (\pm 0.46) \mu\text{g gds}^{-1} \text{day}^{-1}$ respectively. Despite the change in net mineralization pattern following a burn, there was no consistent effect of DSF on net mineralization rate (Figure 8; $\beta_{\text{DSF}} = 0.34$, 95% CI = -0.50-1.19). Of the measured edaphic variables, net mineralization rates were correlated only slightly with soil moisture (Figure 8; $\beta_{\text{SM}} = 0.31$, 95% CI = 0.14-0.48).

Net nitrification rates were temporally heterogeneous throughout the full growing season, but were not appreciably more variable immediately following burns. Net nitrification rates were very low across the growing season in burned ($-0.08 \pm 0.05 \mu\text{g gds}^{-1} \text{day}^{-1}$) and unburned sites ($-0.04 \pm 0.04 \mu\text{g gds}^{-1} \text{day}^{-1}$; Figure 7). Measured edaphic parameters were poorly correlated with observed net nitrification rates, although there was a slight positive relationship between net nitrification rates and soil moisture (Figure 8; $\beta_{\text{SM}} = 0.07$, 95% CI = 0.03-0.11).

3.4 Total Soil $\delta^{15}\text{N}$ and Ash Deposition

Soil N concentration was relatively stable throughout the burn season, and was similar between burned (0.35%N \pm 0.09) and unburned sites (0.38%N \pm 0.07; Table 1). Mean total soil N varied between sites (Table 1; Figure 9). On average, B2 had the lowest total soil N content (0.18%N \pm 0.02), and total soil N ranged from 0.08% at C1, to 0.98% at C2. Across all burned and unburned time points, confidence intervals overlapped between burned and unburned conditions at each site, indicating no persistent change in total N over the full growing season (Table 1).

Across the full growing season, soil $\delta^{15}\text{N}$ in burned sites was 2.76‰ (\pm 0.36) and in unburned sites was 2.00‰ (\pm 0.36). The response of $\delta^{15}\text{N}$ to burning varied between sites. Soil $\delta^{15}\text{N}$ in unburned Sites C1 and C2 was on average 1.22‰ (\pm 0.52) and 1.79‰ (\pm 0.36; Table 1), respectively. In burned sites, there were shifts in $\delta^{15}\text{N}$, although the direction of shift varied between sites. Soils in B1 were depleted in ^{15}N after a prescribed burn relative to before the burn; $\delta^{15}\text{N}$ shifted from 3.98‰ (\pm 0.64) to 3.22‰ (\pm 0.50). In Site B2 the soil $\delta^{15}\text{N}$ decreased from 2.67‰ (\pm 0.57) before the prescribed burn to 2.45‰ (\pm 0.24) after the burn. Finally, there was a slight enrichment in soil ^{15}N in Site B3 following fire; $\delta^{15}\text{N}$ shifted from 1.37‰ (\pm 0.36; Table 1) to 2.59‰ (\pm 0.93; Table 1). The 95% CIs surrounding the mean $\delta^{15}\text{N}$ (and %N) overlapped for all burned sites, indicating the soil $\delta^{15}\text{N}$ at each site was statistically indistinguishable pre- and post-burn.

We used pre-burn soil $\delta^{15}\text{N}$ isotopic signature in mixing models to calculate the mass of ash-N needed to be deposited on our sites to achieve both the minimum and the empirically measured shift in soil $\delta^{15}\text{N}$. To achieve a shift in soil $\delta^{15}\text{N}$ of the minimum external precision, sites B1, B2, and B3 would need 11, 5, and 20 g N m⁻² ash-N, respectively, deposited following fire. To achieve the measured shift in soil $\delta^{15}\text{N}$, sites B1 and B2 would need 100 and 11 g N m⁻² added through ash deposition; B3 would need 175 g N m⁻² to be removed from fire (Table 1). We also calculated the same values using fresh leaf $\delta^{15}\text{N}$ from leaves collected from our sample site (-2.9‰ \pm 0.1; J. Wright, unpublished data), rather than published ash $\delta^{15}\text{N}$ values (Supplementary Table S1).

4 Discussion

In this study, we collected weekly measurements of soil inorganic N availability to document short-lived changes in N dynamics following fire and throughout the growing season of a pyrogenic forest in the southeastern US. As far as we know, this is the first study to pair estimates of N pool sizes and cycling rates at high temporal resolution in a longleaf pine savanna. Prior to prescribed burns, there was high variability in N availability, particularly for NH_4^+ pool sizes. This heterogeneity reinforces the need for a methodological approach that considers initial edaphic conditions when carrying out *in situ* experiments on paired soil cores. To address this, we relaxed the assumption that initial and incubating cores were true edaphic replicates; we used a Bayesian statistical framework to allow for variability in the relationship between true versus measured inorganic N concentrations in our soils.

Immediately following prescribed burns, we found sharp increases in NH_4^+ pool sizes in all of our study sites. However, the magnitude and duration of this increase varied between sites. Unlike studies in southeastern US pine savannas with monthly or less-frequent temporal sampling protocols, our weekly sampling allowed us to capture highly ephemeral changes in soil inorganic N pools. Furthermore, we found no changes in cycling rates and no evidence that ash deposition could account for the large increases in N availability following fire. Instead, we speculate that an ephemeral dampening of plant uptake could contribute to the observed increases in inorganic N following fire.

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4.1 Changes in N dynamics across the growing season

Throughout the growing season, inorganic N availability and net cycling rates were low, as is common in longleaf pine savannas (Binkley et al., 1992). In unburned conditions over the growing season, there was greater NO_3^- availability than NH_4^+ . This pattern is consistent with previous work, which documented relatively high NH_4^+ availability in the winter, followed by decreasing NH_4^+ availability throughout the growing season (Christensen, 1977). Net nitrification rates were low across the growing season, may have been inhibited by the low soil pH (Ste-Marie and Paré, 1999). Net mineralization in our study was higher than measured over the summer months in previous studies (Wilson et al., 1999), so rather than low mineralization rates, our low soil $\text{NH}_4^+ : \text{NO}_3^-$ ratios may be a result of preferential plant or microbial uptake of NH_4^+ over NO_3^- .

We observed sharp increases in soil inorganic NH_4^+ , but not NO_3^- , immediately following fire across three longleaf pine savanna sites in North Carolina (Figure 5). Although a global meta-analysis found that post-fire soil NO_3^- concentrations peak ten months after NH_4^+ concentrations (Wan et al., 2001), studies in southeastern US ecosystems found no change in soil NO_3^- up to 30 days (pine savanna; Boring et al., 2004), 320 days (pine forest; Lavoie et al., 2010) and 500 days (shrubland; Schafer and Mack, 2010) following fire. Across the full growing season, we measured NH_4^+ pool sizes of burned sites that were nearly 5x that of unburned sites. The direction of the effect of fire was consistent across our study sites, however the magnitude of increase was highly site-specific. Within a site, increases in NH_4^+ availability immediately following fire ranged from 5x to more than 25x the pre-burn levels. This NH_4^+ pulse was short-lived, and only in B3 was the increased NH_4^+ pool size sustained for longer than one week. As a consequence, a decreased sampling frequency would not have detected the ephemeral changes in soil NH_4^+ pool size in B1 and B2.

It remains unclear why, however, sites experience such variability in the magnitude of NH_4^+ response following fire. Although we cannot rule out the possibility that our high-intensity sampling influenced nitrogen cycling and pool sizes, we saw no evidence of increasing inorganic N availability, or increasing variability in N availability, in our control sites, which experienced the same levels of sampling disturbance without fire. Our findings imply that an un-accounted for biogeochemical mechanism may be involved in regulating nutrient availability following fire disturbance. We suggest that plant nutrient uptake, heretofore overlooked as a possible mechanism, may be another mechanism driving post-fire nutrient availability. We noticed that B1 and B2 exhibited rapid vegetation resprouting following fire, while regrowth in B3 was patchy. Vegetation began resprouting in B1 and B2 six days after fire, but not until 18 days after fire in B3 (C. Ficken, unpublished data). Moreover, in 2012, the last year all sites were sampled prior to the 2014 burns, B3 had the smallest standing biomass stocks of all three burned sites (J. Wright unpublished data). If plant N uptake remained low following fire in B3, this might explain the persistent increase in N availability in this site. However, biomass stocks after three months of regrowth in 2014 were also substantially different between B1 and B2, despite these sites exhibiting similar patterns of NH_4^+ availability over time. In unburned years, Mitchell et al. (1999) found that annual net primary productivity (ANPP) in a longleaf pine savanna was positively correlated

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with local moisture availability and biomass estimates in this heterogeneous system are highly dependent on local woody versus herbaceous cover, as well as annual variability in environmental conditions. Factors controlling unburned ANPP may differ from those controlling biomass regeneration, and given the spatiotemporal heterogeneity of this system, teasing apart these drivers may require a large-scale manipulative experiment.

4.2 Assessing Mechanisms of N Pulses Following Fire

A nitrogen pulse may occur following fire through (1) an increase in microbial mineralization, (2) ash inputs, or, as we propose, (3) a decrease in plant uptake. Fire may stimulate microbial turnover of organic matter if additions of C or N from ash deposition or root exudation (southern shrubland; Schafer and Mack, 2010) enhance microbial activity. Soil surface blackening after fire may increase soil temperature and stimulate immediate and prolonged N transformations after fire (Booth et al., 2005; Ojima et al., 1994). Wilson et al (2002) found significant increases in microbial biomass following fire in a longleaf pine savanna. Although we did not directly measure microbial biomass, we found no changes in net microbial mineralization associated with the observed increase in pool size, although cycling rates were increasingly variable following fire in burned sites (but not in unburned sites). Indeed, the increase in mass of NH_4^+ following fire was much greater than the mass of NH_4^+ produced on a daily basis by net microbial mineralization, reinforcing the conclusion that changes in microbial cycling rates could not fully account for the observed increase in pool sizes.

We also found no indication that the newly available NH_4^+ substrate led to a delayed increase in net nitrification rate. This, along with high soil C:N (unburned sites- 47:1; burned sites- 53:1) relative to other longleaf pine soils (Lucash et al., 2007), might suggest that autotrophic nitrifying microbes are competitively inferior to heterotrophic microbes under post-fire conditions in our study sites. Alternatively, an unmeasured increase in gross nitrification might have allowed for a commensurate increase in microbial immobilization of NO_3^- following fire. In general, however, the lack of change in microbial N cycling rates suggests that changes in microbial activity fail to account for the observed increases in NH_4^+ availability following fire.

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Direct additions of N into the soil from ash may provide an alternative mechanism for the observed increase in mineral N availability. To test this, we examined changes in both total N and $\delta^{15}\text{N}$ immediately before and after burns. N from ash additions is primarily organic (Christensen, 1977; Huber et al., 2013; Raison, 1979), and is thought to increase N pools by stimulating microbial activity. We

5 found no change in total nitrogen (i.e. %N) or SOM before and after prescribed burns. These results support the findings of a global meta-analysis of fires in forested systems, which found no effect of fire on total N (Wan et al., 2001).

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We used the natural abundance of ^{15}N as an isotopic tracer of ash additions. However, fractionation during volatilization preferentially releases ^{14}N , resulting in ash material that is enriched in ^{15}N relative to

10 fresh plant matter and an increase in $\delta^{15}\text{N}$ signature in ash relative to fresh plant material (Saito et al., 2007; Stephan et al., 2015). A study in a subalpine grassland reported foliar $\delta^{15}\text{N}$ values (-2.9‰) and N concentrations in ash (11.63 ± 0.80 mg N g⁻¹ ash; Huber et al., 2013) comparable to foliar $\delta^{15}\text{N}$ values (-2.9‰; ± 0.1 ; J. Wright, unpublished data) and ash-N concentrations of our system (8.75 ± 0.90 mg N g⁻¹ ash; Christensen, 1977). Huber et al. (2013) also reported $\delta^{15}\text{N}$ values in ash of -0.81‰, which we

15 used in mixing models.

Using this isotopic signature of ash, we found that 5 to 20 g ash-N m⁻² would need to be deposited in our burned sites in order to observe a detectable shift in soil isotopic signature. These values are greater than the mass of ash-N deposition reported in a longleaf pine system (1.15 g m⁻²; Christensen, 1977), suggesting that this method may not be ideal for detecting ash inputs in systems with low aboveground

20 vegetation stocks. Nevertheless, we estimated that 100, 11, and 175 g ash-N m⁻² would need to be deposited on sites B1-3 to account for our measured shifts in soil $\delta^{15}\text{N}$. These deposition levels are highly unlikely to have occurred at our sites, since they would require substantial aboveground vegetation accumulation and our system is burned every three years. However, it is unclear how quickly surface inputs can be expected to distribute throughout the soil profile. In the sandy soils of our system,

25 frequent heavy summer rains or active root growth may quickly redistribute surface inputs. While one study of longleaf pine savannas found that changes (losses) in total soil N following fire were concentrated at the soil surface (Binkley et al., 1992), another study of subalpine woodlands and grasslands detected no changes in total N in surface soils following burning (Huber et al., 2013). Our

work is in agreement with that of (Christensen, 1977), who found significant differences in $\delta^{15}\text{N}$ with depth, but no change following fire.

Given the uncertainties surrounding the redistribution of surface inputs down the soil profile, we cannot conclusively rule out the potential to surface additions to contribute to the observed NH_4^+ pulse.

5 Nevertheless, considering the unrealistic mass of ash-N needed to be deposited onto surface soils to account for our measured shifts in $\delta^{15}\text{N}$, we conclude that ash inputs are unlikely to fully account for the increase in measured soil inorganic N availability.

Finally, we propose that changes in plant and microbial immobilization could cause an increase in soil inorganic pool sizes. Prescribed fires in longleaf pine savannas are low intensity, and sharp increases in

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10 soil temperature with depth, particularly in dry soils, are unlikely to substantially damage the soil microbial community below 5 cm (Hartford and Frandsen, 1992). In fact, previous work in longleaf pine savannas has documented increases in microbial biomass following fire (Wilson et al 2002). In contrast to the microbial community, prescribed burns in our study system generally top-kill a majority of the aboveground herbaceous and woody biomass with stem diameters less than 10 cm (Just et al.,
15 2015). If fire damage temporarily halted or slowed the plant uptake of inorganic N, we would expect to see an accumulation of soil N if microbial immobilization did not increase sufficiently to deplete the pool. However, N accumulating in excess of demand can only partly explain observed increases in inorganic N availability, since the pulse of N we detected following fire was many times greater than what was produced by net mineralization and net nitrification. Nevertheless, a change in plant sink
20 strength may have contributed to post-fire NH_4^+ pulse.

Previous work found no evidence that plant species in an African savanna re-translocated nutrients from root biomass to resprouting shoot biomass following a fire (Vijver et al., 1999), indicating that soil pools can be important sources of N for regenerating biomass. Indeed, the biomass of resprouting vegetation following fire has been shown to be highly enriched in ^{15}N relative to pre-burn biomass

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25 (Huber et al., 2013; Schafer and Mack, 2014), an indication that the source of N in resprouting biomass is also enriched (Evans, 2001). Root biomass is an important component of short-term N retention in grassland ecosystems (De Vries and Bardgett, 2016). In fire-prone systems, fire-tolerant plants could

play an important role in preventing N leaching losses if they are able to resume N uptake quickly following fire.

We propose that plant-demand for inorganic N may have a strong influence on soil N pool sizes in this system, and a temporary decrease in this demand may have contributed to the observed post-fire NH_4^+

5 pulse. Preference for NH_4^+ by plants inhabiting acidic soils, where nitrification is limited by low pH and NO_3^- availability is consequently low (Ste-Marie and Paré, 1999;Houlton et al., 2007;Wang and Macko, 2011;Kronzucker et al., 1997), could help to explain the relatively large pool sizes of NO_3^- relative to NH_4^+ during the growing season (Vitousek, 1977), and this pattern is consistent with previous seasonal trends in a longleaf pine savannas (Christensen, 1977). Furthermore, an increase in soil NH_4^+ pool size

10 after fire without a stable increase in net microbial mineralization rates could occur if there is a decrease in plant uptake. Similarly, a muted effect of fire on NO_3^- pool size may occur if plants have diminished uptake of this inorganic N form, and plant uptake exerts a relatively weaker control on soil NO_3^- pools. Plant control on ecosystem N status has been well documented in northeastern US hardwood forests, where a defoliation event resulted in substantial N losses from the ecosystem (Aber et al., 2002;Likens
15 et al., 1969).

If post-fire patterns in N availability were related to plant uptake, we would expect differences in the magnitude and duration of soil N change following fire to be related to plant N-demand and regrowth following fire. In stand-replacing fires in temperate forests, where vegetation is killed, relatively persistent increases in N pools should occur following fire. We similarly would expect smaller and
20 more ephemeral changes in N pools in systems in which plants are only top-killed. Indeed, stand-replacing fires have been shown to result in changes to soil N pools that persist more than one year following the fires (Smithwick et al., 2005;Turner et al., 2007). In contrast, elevated inorganic N immediately following fires in grassland decreases throughout the growing season (Augustine et al., 2014). Low-intensity fires in grass-dominated glades adjacent to oak-hickory forest sites in a Kentucky
25 study resulted in increases in post-burn soil NO_3^- pool sizes, but no increase in lysimeter-detected NO_3^- leaching losses below 10 cm (Trammell et al., 2004). Although the study did not examine microbial biomass, they found no effect of fire on net N mineralization, suggesting role of plant uptake in patterns of N loss and retention post-fire. Such instances of plant control of N availability provide an important

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setting in which to examine the role of nutrient availability– and nutrient pulses in particular– on plant community composition and ecosystem productivity. Differences in the ability of species to capture this ephemeral resource may help explain differences in post-fire resprouting patterns and biomass regeneration following fire.

5 4.3 Ecological Implications of Fire-Associated N Pulse

To put the fire-associated pulse of inorganic N that we observed into context, we compared its mass to N inputs in the longleaf pine savannas ecosystem. Although the pulse of soil N following fire is most likely a redistribution of N from other ecosystem pools, it is conceptually helpful to understand the magnitude of this pulse relative to other components of the N cycle in longleaf pine savannas. The increase in soil inorganic N following fire ($0.98 \text{ g N m}^{-2} 10 \text{ cm}^{-1}$) was approximately 10x the daily total net inorganic N production (i.e. net mineralization + net nitrification; $0.11 \text{ g N m}^{-2} 10 \text{ cm}^{-1} \text{ day}^{-1}$).

10 These ephemeral increases in soil inorganic N availability occur during an important ontological stage of plant development as longleaf pine understory species begin resprouting within a few days following fire (C. Ficken, personal observation). The mass of mineral N released following fire was more than
15 twice the mass of N longleaf pine understory species assimilate into their biomass over a full growing season (0.45 g N m^{-2} ; unpublished data). However, the extent to which plants or microbes have access to this ephemeral nutrient pulse remains unclear. Previous studies have documented rapid plant uptake of N tracers by intact plants (Aber et al., 2002; Likens et al., 1969), suggesting that plants may have access to this N pulse during regrowth.

20 4.4 Conclusions

We have documented large pulses of mineral N following prescribed burns in a longleaf pine savanna in North Carolina. Our weekly sampling revealed that while one site experienced a persistent increase in NH_4^+ pool size, other sites experienced only very short-lived pulses that would not have been detected with monthly sampling. The marked differences in the duration of the NH_4^+ pulse that we observed may
25 explain why previous studies with less-frequent soil sampling showed no change in mineral N following fire. However, the factors that influence the magnitude of the system's response to fire are still

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unresolved, because neither an increase in microbial mineralization nor ash-N deposition could fully explain our observed patterns of post-fire N availability. We propose here a role for plant uptake in regulating post-fire N availability, but caution against drawing conclusions from this as-yet untested hypothesis. Instead, we propose this hypothesis to encourage future work to explore the relationships

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5 between N availability and plant biomass dynamics immediately following fire. Due to the rapid changes in N availability following fire, as well as the fast resprouting of understory species, we recommend that responses of the the local plant community be considered when determining an appropriate sampling regime for biogeochemical responses to disturbance. In systems in which the plant community responds rapidly, soil samples should be collected quickly and frequently to capture post-
10 disturbance plant-nutrient and biogeochemical dynamics.

The pulses documented here were not associated with increases in microbial activity, and, although inconclusive, our data also do not support the hypothesis that the observed inorganic N pulse could be attributed to ash inputs to the system. We speculate that a temporary slowing of plant uptake may contribute to the observed mineral N pulse. Although plants begin resprouting a few days after a fire,

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15 the extent to which plants access the newly available NH_4^+ remains unclear. Given the magnitude of this pulse relative to other N transformations in the system, and particularly relative to the mass of N estimated to be assimilated into the understory biomass annually, these fire-associated N pulses may be important sources of plant-available N as the vegetation regrows. As such, they may also play an important role in structuring plant recovery from disturbance and shaping community diversity in this
20 system.

Author Contributions

CDF and JPW conceived of the project; CDF collected samples, performed laboratory and statistical analyses; CDF and JPW wrote the manuscript.

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		Burned Sites			Unburned Sites	
		B1	B2	B3	C1	C2
NH ₄ ⁺ (μg gds ⁻¹)	Preburn	0.83 (0.29)	0.91 (0.24)	0.84 (0.31)	1.25 (0.28)	0.80 (0.14)
	Postburn	2.78 (1.62)	5.18 (2.21)	29.68 (6.03)		
NO ₃ ⁻ (μg gds ⁻¹)	Preburn	3.63 (0.78)	2.61 (0.63)	2.74 (0.63)	2.34 (0.26)	2.44 (0.22)
	Postburn	4.18 (1.76)	1.69 (0.65)	4.66 (1.35)		
δ ¹⁵ N (‰)	Preburn	3.98 (0.64)	2.67 (0.57)	1.37 (0.36)	1.22 (0.52)	1.79 (0.36)
	Postburn	3.22 (0.50)	2.45 (0.24)	2.60 (0.93)		
Total N (%)	Preburn	0.61 (0.18)	0.19 (0.04)	0.54 (0.20)	0.29 (0.10)	0.39 (0.14)
	Postburn	0.56 (0.12)	0.18 (0.02)	0.30 (0.11)		
Ash-N (g N m ⁻²)		104	11	-175		

5 Table 1. Mean soil inorganic N pool sizes reported per gram of dry soil (gds), δ¹⁵N values, and total N for each study site. Values are reported ±95% CI. Sample sizes for inorganic N pool sizes differ between sites depending on when the prescribed burns occurred; see *Methods* for details on sample sizes. Soil %N and δ¹⁵N values were collected on a subset of time points; sample sizes for these variables are N=6 for burned site means, and N=12 for control (unburned) site means. Ash-derived N values are the estimated masses of N needed to be deposited onto each site to result in the observed post-burn δ¹⁵N. Note that Christensen (1977) measured 1.15 ±0.49 g N m⁻² deposited in ash fall in fire in a longleaf pine savannas.

	Burned Sites		
	B1	B2	B3
Minimum shift	8	3	11
Empirical shift	316	8	-108

5 Supplementary Table S1. The mass of N needed to be deposited at each site in order to achieve a shift in soil $\delta^{15}\text{N}$ of the minimum external precision, and the mass of N needed to account for the observed empirical shift in soil $\delta^{15}\text{N}$. These values were calculated from mixing models with fresh leaf $\delta^{15}\text{N}$ as one end member. Across 1,827 samples from 67 plant species, the mean $\delta^{15}\text{N}$ was -2.9‰ (± 0.1 ; J. Wright, unpublished data). Units are in g N m^{-2} . Minimum external precision is 0.1‰ $\delta^{15}\text{N}$ at 1 standard deviation.

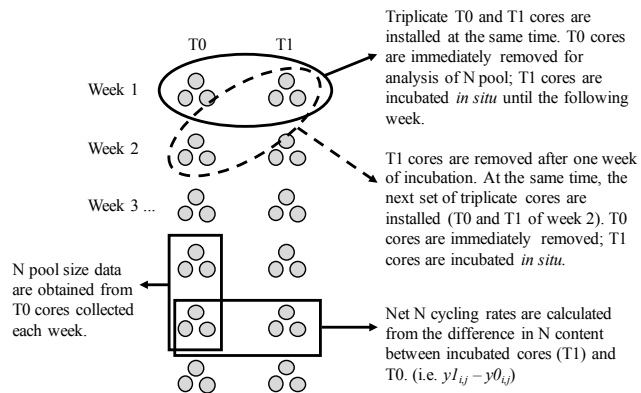
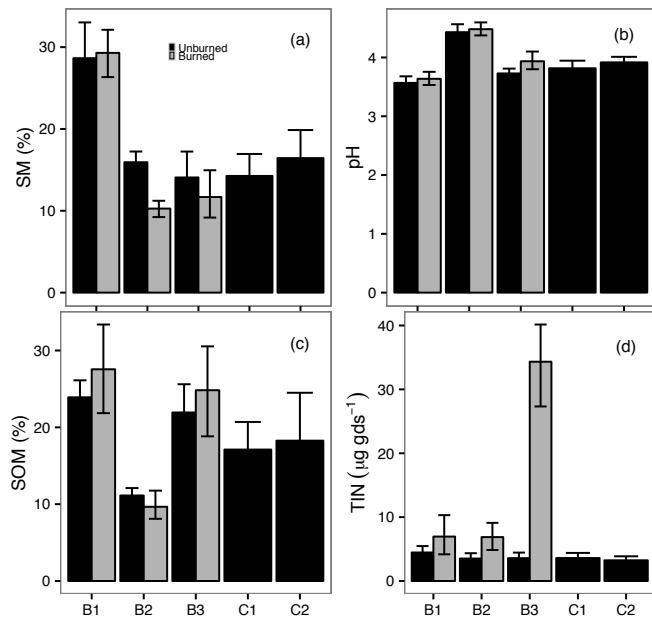


Figure 1. Schematic illustrating the sampling design across nine weeks during the growing season. Each week, two sets of three replicate cores were installed at each study site. One set was immediately removed for pool size estimates; the second set incubated *in situ* for one week to estimate net cycling rates.

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Figure 2. Mean soil moisture (SM; a), pH (b), soil organic matter (SOM; c) and total inorganic N (TIN, i.e. NH_4^+ and NO_3^- ; d) prior to and following burns at each site. TIN is reported per gram of dry soil (gds). Values are reported $\pm 95\%$ CI.

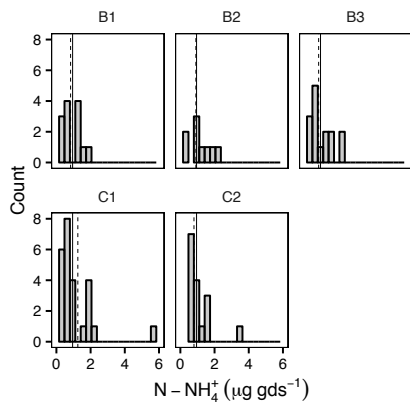
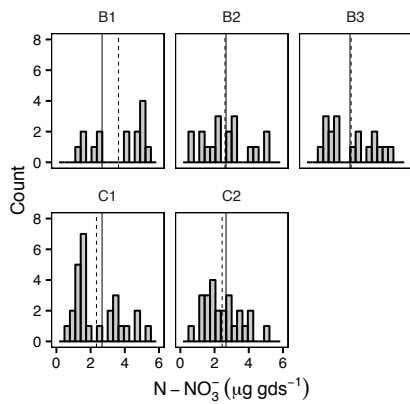
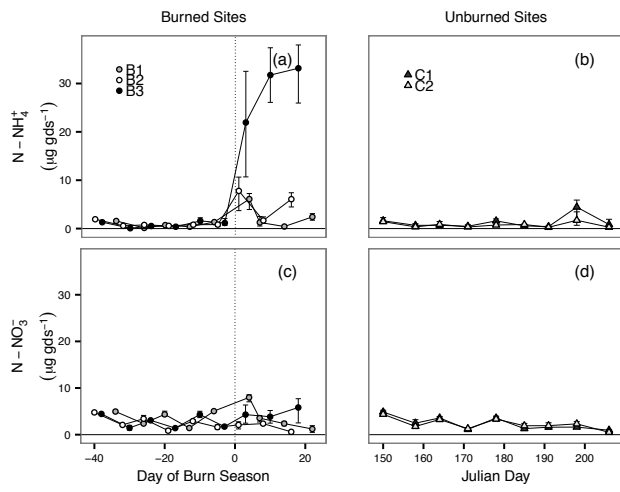


Figure 3. Histogram of soil NH_4^+ concentrations (reported per gram of dry soil, gds) prior to prescribed burns. Solid vertical lines indicate the median concentration across all sites; dashed vertical lines indicate the site-specific median concentration.



5 Figure 4. Histogram of soil NO_3^- concentrations (reported per gram of dry soil, gds) prior to prescribed burns. Solid vertical lines indicate the median concentration across all sites; dashed vertical lines indicate the site-specific median concentration.



5 Figure 5. Changes in pool sizes of (a) NH_4^+ in burned sites, (b) NH_4^+ in control sites, (c) NO_3^- in burned sites, and (d) NO_3^- in control sites. The x-axis for A and C depicts time (in days) centred on the date of burn; days immediately before the burn are negative x-values while days immediately following a burn are positive; the burn date is at 0 and is demarcated with a vertical dotted line. The x-axis for sites B and D depicts time in Julian Days. Prescribed burns in B1, B2, and B3 occurred on Julian Days 185, 190, and 188, respectively. Pool sizes are reported in in $\mu\text{g N per gram dry soil (gds)}$. Error bars ($\pm 95\%$ CI) for individual time points may be obscured by the point.

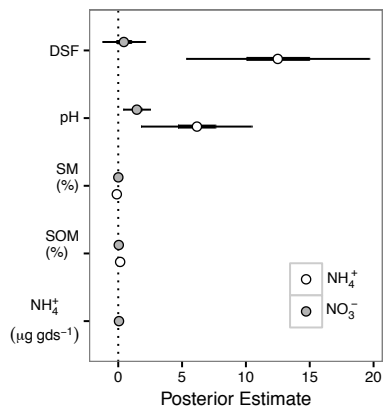
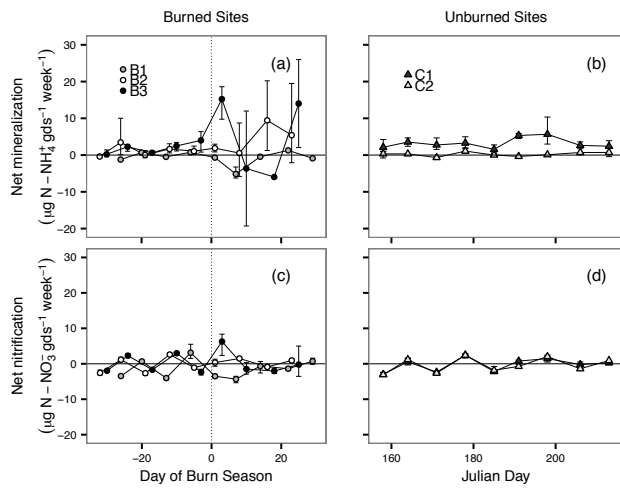


Figure 6. Mean posterior estimates for parameters predicting pool sizes of NH_4^+ and NO_3^- . Parameters are days since fire (DSF), soil pH, soil moisture (SM), soil organic matter (SOM), and substrate availability (i.e. NH_4^+ reported per gram of dry soil, gds). Thin black lines show 95% CI and thick lines show 50% CI. CI may be obscured by the mean point. **For visual clarity, posterior distributions of site effects are not included here. See text for details.**

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5 Figure 7. Changes in (a) net mineralization in burned sites, (b) net mineralization in unburned sites, (c) net nitrification in burned sites and (d) net nitrification in unburned sites. The x-axis for A and C depicts time (in days) centred on the date of burn; days immediately before the burn are negative x-values while days immediately following a burn are positive; the burn date is at 0 and is demarcated with a vertical dotted line. The x-axis for sites B and D depicts time in Julian Days. Prescribed burns in B1, B2, and B3 occurred on Julian Days 185, 190, and 188, respectively. Cycling rates are reported in $\mu\text{g N}$ per gram dry soil (gds) per week. Error bars ($\pm 95\%$ CI) for individual time points may be obscured by the point.

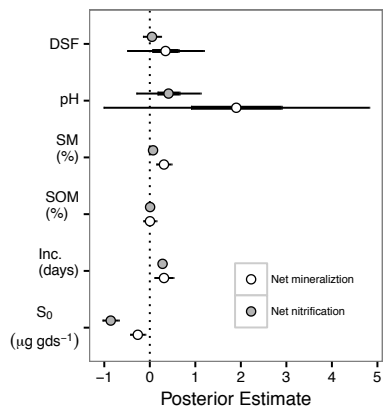
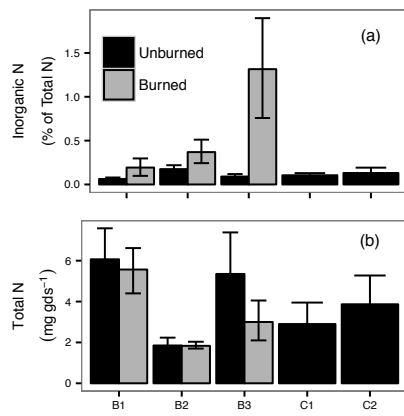


Figure 8. Estimates of parameter posterior mean effects with bars showing 95% credible intervals (CI) for net mineralization and net nitrification rates. Parameters are days since fire (DSF), soil pH, soil moisture (SM), soil organic matter (SOM), the incubation length (Inc.), and initial substrate availability (S_0) per gram of dry soil (gds). [For visual clarity, posterior distributions of site effects are not included here. See text for details.](#)

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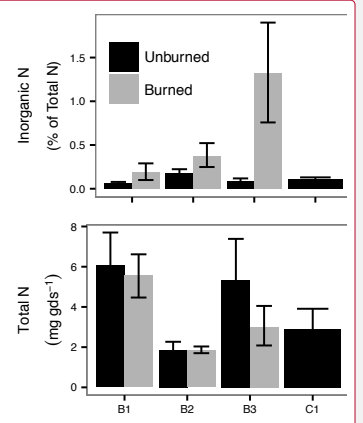


Figure 9. Bar chart showing the pulse of inorganic N relative to the total N pool size. Panel (a) shows the mean percent of inorganic N (i.e. NH_4^+ and NO_3^-) in soil under in unburned and burned periods. Panel (b) shows the mean total N soil content in burned and unburned periods, reported per gram of dry soil (gds). Error bars are $\pm 95\%$ CI.