LUND UNIVERSITY

Department of Geology
Dr. Johanna Stadmark

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To the Chair of the Committee,

Thank you for appointing me as a reviewer of Michal Růžek's doctoral thesis. Already at the start I want to emphasize the high quality of the work presented in this thesis and its contribution to the understanding of carbon and nitrogen processes in stands of beech and spruce. The thesis adds to existing knowledge in this field by the results from conducted experiments and monitoring at Nacetin site, located in the Ore Mts, an area recovering from acidification.

The thesis builds on work outlined in five publications (four already published and one in review). In the two first-authored publications (I and V) M. Růžek has designed and performed field- and laboratory work and led the writing of the manuscript (V) and performed data processing and statistical analysis and prepared the manuscript (I). M. Růžek has also performed experiments and fieldwork as well as analysed data and contributed to the manuscript writing in Publications II and III and to a lesser extent in Publication IV. The published research is contributing to the international understanding of the biogeochemical processes in forests subject to acidification and/or nitrogen (N) fertilization.

The thesis itself firstly reviews existing knowledge on carbon and nitrogen cycles in forests soils, anthropogenic sulphur (S) and N deposition and their effects on forest ecosystems (Norway spruce and European beech) and presents the aims and objectives. In the following sections the sites and the methodological approaches are described and the major findings of the individual publications presented. Finally all results are discussed and synthesized under the following headlines: 1) Soil carbon (C) and N dynamics of spruce and beech stands under ambient condition, 2) Experimental manipulations and acidity constraints on soil organic matter (SOM) dynamics, and 3) Experimental manipulation and N availability constrains SOM dynamics.

An advantage of this thesis is the wide approach used to disentangle the questions addressed. In Publication I long-term monitoring data of concentrations of Na, K, Ca, Mg, Al, SO_4, NH_4, NO_3, DON, DOC and Cl in bulk deposition, throughfall, forest floor and mineral soils at three depths (30, 60, 90 cm) are presented for the two forest stands. Development over time and through the soil profile for the different compounds and their combination are discussed. Both sites are recovering from acidification, which is seen in the increasing pH and decreasing Al concentrations. In Publication II carbon and nitrogen stocks in vegetation biomass and in soils are estimated for the two forest stands and carbon and nitrogen fluxes presented based on three years of measurements. The tree species effects on C and N cycling and proportion of allocation above and below ground are discussed. In Publications III-V experimental sites with additions of S, N or their combination as well as control sites were sampled for the impact of treatments on carbon fluxes and fungi to bacteria ratio (III), soil microbial communities and hydrolytic enzyme production (IV) and degradation of litter (V).

The main goal of the thesis was to test whether soil processes that are altered by experimental treatments respond consistently with monitoring observations, i.e. acidification or N availability are the principal drivers of C accumulation in acidified soils. In this thesis M. Růžek outlined that he, in collaboration with others, wanted to:

1) *describe and compare current soil C and N fluxes and pools, in each of the forest stands (Publication I, II), with the hypothesis that there would be higher biomass nutrient pools in the beech stand and comparable soil C and N pools in both stands.* – These results are presented in Publications I and II. The stock of C in biomass was higher in the beech stand, while the N stocks in biomass were similar among stands. In soils both C and N stocks were higher in the beech stand than in the spruce stand, in contrast to earlier reported findings from other locations.

2) *quantify the effect of acidity/N availability changes on the amount of C and N lost from the forest soil via organic (DOC, DON) and inorganic (NO_3, CO_2) pathways, in two major forest types (Publication III), with the hypothesis that SO_4 addition, by altering soil acidity, causes a shift from organic C and N loss pathways (DOC, DON) at higher pH, to inorganic (CO_2, NO_3) loss pathways at lower pH.* – These results are partly presented in Publication III. Addition of S (and S+N) decreased pH and induced suppression of DOC concentrations in both forest stands. In the spruce stand also soil respiration was reduced. Addition of only N did neither affect DOC concentrations nor soil CO_2 efflux. The impact of increased acidification on DON loss is not the topic of Publication III. There was no clear impact of S addition on soil NO_3 concentration in either of the stands, but a decrease of DON in the soil solution was found in the beech stand in Publication IV.

3) *measure how acidity/N availability affects a range of key internal N and C transformations, the microbial community and "in situ" SOM decomposition (Publication IV, Manuscript V), with the hypothesis that microbial activity would be suppressed under acid treatments in both stands, due to the decrease of organic C-availability for microorganisms. A shift towards a higher production of C-mining enzymes was also expected. Further, it was hypothesized that N availability would reduce litter decomposition by the alleviation of N limitation. Increasing N availability would decrease the fungi/bacteria ratio and qualitatively transform the microbial community structure towards N-tolerant taxa.* – These results are presented in Publication IV and Manuscript V. Acidity induced changes in the bacterial communities in both forest stands and led to changes in enzymatic activity. Overall the enzymatic activity increased in the spruce +S treatment (Table 1. Publication IV). C-mining enzymes (Table 1. Publication IV) did not increase significantly, but it was hypothesized that production of such enzymes could counteract the impact of lower concentrations of SOM (DOC) in the more acidic treatments. Fungi were not influenced by any treatment in either of the stands. N additions did not in a consistent way impact the decomposition of any of the four litter types (Manuscript V) or decrease the fungi/bacteria ratio (Publications III and IV). Changes in microbial community structure were coupled to acidity, maybe because the location has already for a long time experienced high nitrogen loads and therefore no further changes in the community structure could be found in the +N-treatment.

One of the main achievements of this thesis is the combination of different aspects of soil science with the palette of methods used (long-term monitoring data, addition (S, N, S+N) experiments and manipulation experiments (material moved from one stand to another) and the results of analyses of soils, vegetation, soil waters, microbes, inorganic and organic species of C and N. The results derived from this thesis can provide data on C sequestration to models and to managing authorities, which further emphasize the importance of understanding of the ecosystems. Although upscaling from sites to regional or global scales should be done with caution the stands, as pointed out in the thesis, are not isolated occurrences but parts of larger forest areas in Europe with similar bedrock and soil conditions as well as deposition history. A large number of samples have been collected and analysed according to standard methods and I find the statistical handling sound.

Regarding the formal aspects of the thesis I found it well structured with chapters divided into introduction, background, motivation, paper by paper, synthesis and conclusion. The figures are illustrative, but sometimes they do not contain the information that is referred to in text, e.g., Figures 1 (to gain energy?) and 7 (melting snow?). I have found some typos, but no major flaws. There are also a few typos in the published papers, which just shows that one can never be too thorough with the proof-reading (e.g., TBCF in Publication II, three places, which made the sections on carbon allocation unnecessary complicated to understand).

I haven't found any major drawbacks, although there are aspects that can be elaborated on in the discussion, see some of my questions listed below. One thing that I lack in the thesis is the size of the forest stands and the possible interference between treatments in neighbouring plots. I assume the water transport during fertilizer/acidity applications mainly are into the ground without lateral transport in the forest floor or along roots. I would also appreciate a justification for the addition of 50 kg S ha^{-1} yr^{-1} in the experiments, which is currently not available in the thesis.

In my opinion this is a well-written thesis, covering a number of biogeochemical aspects in forest stands recovering from acidification, which definitely can be accepted as a basis for obtaining a doctoral degree.

Yours sincerely,

Dr. Johanna Stadmark

Below I list a selection of the questions I would like to ask, and discuss with, M. Růžek during the defence.

General questions

What do you view as the main outcome from you doctoral work?

If you in 1980 were asked to design a monitoring program what would you have included given what you/we know today? (Are there parameters that you are lacking in the long-term data?) Or if you could go back to the 1850's and collect samples – what would you have investigated?

If the current Norway spruce stands were harvested and replaced by European beech, how long time would you expect it to take before the carbon sequestration is as in your studies?

What type of forest would you advice forest owners/the government to plant to increase carbon sequestration?

What was your most unexpected result? Research always leads to new questions. Which ones would you like to investigate further?

Methodological questions

What are the main advantages and disadvantages by your experimental design?
(e.g. regarding the size of plots (3*3 m), are there edge effects?, transfer of water in the soil – compare disturbances in beech stand, how far away was the harvest, could it impact the NO_3 concentrations? or were there other disturbances?)

Regarding carbon sequestration in stands dominated by different tree species. How much of the differences between stands, in your results, are due to the different stand ages?

How old are the stands (spruce/beech)? (Thesis: 80 years/150 years, Paper I: 80/140, referring to Paper II, Paper II: around 80/already in 1842 a mixed beech-spruce-forest, could the beech be >170 years?, Paper III: around 80/referring to Paper II, Paper IV: 80/140, Manuscript V: 80/120)

What does the addition of S tell us in sites recovering from acidification? Could these results be used for predictions?

How are the CO_2 fluxes integrated over the year? Measurements at night/during winter?

How would you expect your results to differ if you conducted your experiments in areas with lower historical S- and N-deposition?

How much N-deposition would you expect would be needed to saturate the ecosystem, and initiate leaching?

Specific questions

Thesis

Page 29 ”Lower N demands in the spruce stand may partly explain the rapid decrease of inorganic N leaching following acid deposition declines.” 1) Why lower demands? 2) Why does a lower demand result in a decrease in leaching?

Page 31 How did you conclude that S-additions affected only the heterotrophic part of respiration? (Trees largely unaffected by treatments.)

Paper 2

Soil stocks of C and N are calculated to the depth of 40 cm. How much of the stocks are in deeper layers (i.e. not covered by these estimates). (Table 3)

Table 7 Could you please walk me through this table and explain if the soil stock is increasing ($GPP-R_{total}$ = NPP, but there are also Fs, which are larger than NPP -> is the soil stock decreasing?)

Paper 3

Figure 4 Are there significant relationships if the two forest stands are analysed separately?

Manuscript 5

How do you define high-quality litter? p. 14

Why did you use different kinds of bags (regarding mesh size) in the litterbag experiment?

It was a lot of bags, how did you know where to find them? How much disturbance did you make?

Are there really differences between stands in months 6 and 12 for the needles? (Table 3)