

## ***Interactive comment on* “Free atmospheric CO<sub>2</sub> enrichment did not affect symbiotic N<sub>2</sub>-fixation and soil carbon dynamics in a mixed deciduous stand in Wales” by M. R. Hoosbeek et al.**

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Reply to referee #1

We would like to thank referee #1 for providing useful comments.

Indeed, next to symbiotic N<sub>2</sub>-fixation we should, in a revised version of our manuscript, also discuss heterotrophic N<sub>2</sub>-fixation (free-living N<sub>2</sub>-fixers). Hofmockel and Schlesinger (2007) hypothesized that heterotrophic N<sub>2</sub>-fixation would be enhanced due to increased litter production under elevated CO<sub>2</sub>. Increased N availability to plants would, in turn, meet the additional N required to sustain increased NPP under elevated CO<sub>2</sub>. They conducted series of experiments in which nitrogenase activity was

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measured in slurries and intact soil cores in response to different levels of substrate, moisture and nutrients. Forest floor and mineral soil samples were obtained from ambient and elevated CO<sub>2</sub> plots at the Duke Forest FACE site. Hofmockel and Schlesinger (2007) did not detect a FACE effect on potential nitrogenase activity and concluded that heterotrophic N<sub>2</sub>-fixation was not enhanced in temperate pine forests under elevated CO<sub>2</sub>. Based on these results we assume that heterotrophic N<sub>2</sub>-fixation at the BangorFACE experiment was not affected by elevated CO<sub>2</sub> and did not affect the outcome of our experiment.

The decline of NH<sub>4</sub><sup>+</sup> under elevated CO<sub>2</sub> was observed in 2008 (Fig 2; Hoosbeek et al., 2010). During this year total soil N did not change as compared to 2007. As mentioned, the decline of NH<sub>4</sub><sup>+</sup> may have been due to a combination of increased nitrification, microbial activity (N-fixation in SOM) or increased plant uptake under FACE. Indeed, based on the  $\delta^{15}\text{N}$  values of *Betula* we may infer that N sources have not changed due to FACE. Moreover, we agree that it would have been interesting to know the  $\delta^{15}\text{N}$  values of NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup> in order to compare these with plant  $\delta^{15}\text{N}$  values. Unfortunately, these data are not available.

The classical cascade model in which fresh SOM first enters the ‘fast’ or ‘labile’ pool after which it flows into the ‘intermediate’ pool and eventually end up in the ‘slow’ or ‘stable’ pool has been discussed and criticized for a while. The data presented by Lichter et al. (2008) support the conceptual model in which a part of OM entering the soil may be relatively quickly stabilized by, for instance, occlusion in aggregates or adsorption onto mineral surfaces (e.g. Golchin et al., 1994; Jastrow, 1996; Von Lützwow et al., 2006; Von Lützwow et al., 2008). It would have been interesting to apply this conceptual model to the BangorFACE experiment, but isotopic data of soil fractions are not available.

The Duke Forest is a N-limited system in which adding labile substrate due to FACE will not result in a noticeable priming effect. However, priming may occur in forest FACE experiments established on former agricultural soils. For instance, Hoosbeek

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et al. (2004) observed a priming effect during the second and third year of a FACE experiment with poplar trees established on former agricultural soils in central Italy. This temporal priming effect and the associated loss of older SOM was driven by the experiment, i.e. higher labile C availability due to FACE, and by the change of land use, i.e. going from a high fertility agricultural soil to a forest soil with a declining soil fertility status (Hoosbeek and Scarascia-Mugnozza, 2009). At BangorFACE we also observed that total soil C increased less and total N decreased more under FACE during the second year (Fig 1; Hoosbeek et al., 2010). We infer that soil microbial populations increased under FACE (higher labile C availability) and available N was taken up from this former agricultural soil (lower  $\text{NH}_4^+$  concentration under FACE in 2006). In a next step, or in addition, the extended microbial population decomposed N-rich older SOM making N available to the microbial population and plants. Since this source of N requires more energy, the population will decrease and adjust itself to the new availability of substrate and nutrients. After the increased decomposition of SOM diminished, concentrations of  $\text{NH}_4^+$  and  $\text{NO}_3^-$  will also go down again and adjust to levels that fit the new nutrient status of a young forest soil. This is in line with our observations, i.e.  $\text{NH}_4^+$  decreased after 2007 under FACE, while in the ambient  $\text{CO}_2$  plots  $\text{NH}_4^+$  did not decrease yet. While  $\text{NO}_3^-$  decreased both under FACE and ambient  $\text{CO}_2$  with lower concentrations under FACE. We postulate that the priming effect and subsequent lowering of soil  $\text{NH}_4^+$  and  $\text{NO}_3^-$  concentrations (not observed yet for  $\text{NH}_4^+$  in the ambient plots) are largely due to the change of land use and that these transitional processes were enhanced under elevated  $\text{CO}_2$ .

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