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Interactive comment on "Carbon flux to woody tissues in a beech/spruce forest during summer and in response to chronic elevated O₃ exposure" *by* W. Ritter et al.

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Author reply to comments of Reviewer #1

We thank Professor Howard S. Neufeld (Reviewer #1) for his constructive review that helped to improve our paper and present our findings more clearly. We considered the individual comments as follows (please find the revised version of our ms as a supplement):

Reviewer #1: Given the current discussion in the literature about technical limitations for determining the fate of recently assimilated carbon, I might ask if the conclusions expressed by Mencuccini and Hölttä regarding the suitability of pulse labeling for studying C2862

soil respiration are of any concern here? Certainly the conclusions reached by Ritter et al. in this paper concerning CO2 efflux from trees and the fraction of newly assimilated 13C in respired CO2 are bolstered by the detailed and thorough description of the ISOFACE system provided in Grams et al. (2011) as well as the discussion of the potential sources of error in this paper, and the data are consistent with that reported in other papers coming from this research group (i.e. Kuptz et al. 2011a, 2011b, Nikolova et al. 2009).

Author reply: Mencuccini and Hölttä (2010) call the suitability of isotopic approaches into question for relating sudden changes in canopy photosynthesis of (particularly tall) trees to the substrate availability and thus rate of soil respiration. In general, we agree with the referee that this criticism could be extended to the link between canopy photosynthesis and stem and coarse root CO2 efflux. The authors distinguish between the transfer time of individual isotopically labeled molecules and the faster propagation time of waves of turgor and osmotic pressure (i.e. sucrose concentration wave propagation time). However, they clearly pointed out that labeling studies are well suitable to track individually labeled molecules and to study mean residence time or half-life of C in systems such as tall trees. This is exactly what we were aiming at in our manuscript as our isotopic labeling focused on the carbon turn-over and translocation of recent photosynthates (i.e. isotopically labeled sugar molecules) at various positions along the canopy-stem-root-soil continuum during summer and in response to 2xO3. This is why we talk about "the flux of recent photosynthates" and "labeled C" throughout the manuscript. Therefore, we do not see our manuscript to be affected by the criticisms of Mencuccini and Hölttä (2010). Nevertheless, we adjusted the conclusive section of our manuscript to stress once more the fact that our approach is focused on tracking of labeled sugar molecules and should not be confused with the faster propagation of phloem pressure-concentration waves.

Reviewer #1: In the methods section of previous papers from this group, where repeated measures analyses of variance were used, those authors made mention of that fact, but it is omitted here. I would suggest that a sentence or two be inserted in the Statistical Methods section (2.11) regarding these analyses (see Figures 2 and 3). Also, according to Table 3, paired t-tests were used to detect differences among stem positions and coarse roots. However, it is not clear what was paired. Was upper stem compared to coarse roots, and then also to lower stem? Such comparisons run the risk of elevating the experimental wide error rate. Of course, the differences are so large that this will not change the conclusions, but I thought the authors might justify their statistical approach here perhaps. A similar mention of paired t-tests in Figure 4 leaves the reader confused as to which two items are being compared, so maybe the authors could clear that up.

Author reply: As suggested by the reviewer we now mention where repeated measure analyses of variance were used. Please note the corresponding improvements in section 2.11 Statistical Analyses. We are sorry about the apparent confusion on the use of the t-test and improved the clarity in this point: In Table 3, the CO2 efflux from the lower stem was compared to the upper stem and then to the coarse roots. In woody plants, CO2 efflux is known to vary between different stem heights/positions (Bowman et al., 2005; Hölttä and Kolari, 2009). For each species, we wanted to show whether the CO2 efflux differs between the stem tissue at the crown base and at breast height. Due to its proximity, CO2 efflux assessed from the lower stem position was compared to coarse-root CO2 efflux. In Figure 4, the t-test for paired comparisons was used to detect differences in delta13C shift between O3 regimes within gaseous samples regarding CO2 and solid samples of labeled beech and spruce trees. Please note our corresponding amendments in the legends of Table 3 and Figure 4.

Reviewer #1: There is no mention made of the potential impacts (or lack thereof) on photosynthesis from elevating the CO2 concentration during the stable isotope labeling period. The elevation amounts to almost a 30% increase in concentration over ambient levels prior to labeling, which should certainly affect instantaneous rates, even if briefly. The authors do note that stomatal conductance was most likely not affected, and that

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Ci to Ca ratios were altered only slightly, but rates of net photosynthesis must have increased somewhat. Perhaps a sentence or two regarding this could be made in the Discussion section. High CO2 can ameliorate O3 effects in some species, so would the conclusions drawn from this study be affected if the actual O3 effect was slightly attenuated during the labeling period?

Author reply: The referee is correct that we may expect some increases of the CO2uptake rate during the labelling period as CO2 concentrations were increased in the photosynthetically most active sun crowns by 18 and 27 % (i.e. 71 and 103 μ l L-1) in spruce and beech, respectively. The methodological aspects of the isoFACE labelling infrastructure are dealt with in great detail in Grams et al. (2011). Therefore, we decided to be brief on these aspects in this present ms but nevertheless now mention the potential increase in CO2 uptake rate in the M&M section. The observed O3 effects are likely to result from long-term exposure (7 years) to 2x O3. Such long-term effects are unlikely to be counteracted by short-term increases of CO2-concentration. In addition, and most importantly, in most cases amelioration of O3 effects by elevated CO2 concentrations are typically observed at much higher CO2 concentrations and are related to reductions in stomatal conductance (for e.g. beech see Grams et al. 1999). Since stomatal aperture was not affected during the operation of the isoFACE infrastructure (Grams et al. 2011), amelioration effects are unlikely. This aspect has now been added to the discussion of the revised paper.

Reviewer #1: One item that has not been addressed is the fact that the high O3 treatment was 2x ambient, which is fairly high relative to current O3 levels. Perhaps some statement could be made about the relevance of the treatment effects found at 2x ambient, given the known current and projected ambient O3 conditions for that region.

Author reply: C allocation was assessed in response to long-term, 7-year long exposure of adult beech and spruce trees to a twice-ambient O3 (2x O3) treatment. During the exposure to 2x O3 maximum concentrations were restricted to < 150 nL L-1 to prevent risk of acute O3 injury (cf. Matyssek and Sandermann, 2003). This exposure strategy resulted in a chronically enhanced O3 regime simulating the widely observed trend of currently increasing O3 background concentrations within a range, in which O3 levels that sporadically occur at the site were provided at higher frequency (Fowler et al. 2008; Sitch et al. 2007; Vingarzan 2004). In this way, the experimentally enhanced 2x O3 regime was realistic in view of projected scenarios as also applying to Central Europe. This argumentation has now been added to the M&M section of the revised version of the ms.

Reviewer #1: Finally, I think the paper would benefit from some brief speculation on why beech and spruce differ in their carbon allocation responses after O3 exposure, perhaps by using the model proposed by Kuptz et al. (2011a) in their paper on seasonal respiratory carbon allocation patterns in these trees.

Author reply: As suggested by the reviewer we added a brief speculation on the differences in O3 sensitivity of beech and spruce to the Discussion section.

Reviewer #1: I have made a few minor suggestions to correct or improve the English and they are given after the references cited below.

Author reply: All suggestions have been incorporated accordingly. Please see our improvements in the revised paper.

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Please also note the supplement to this comment: http://www.biogeosciences-discuss.net/8/C2862/2011/bgd-8-C2862-2011supplement.pdf

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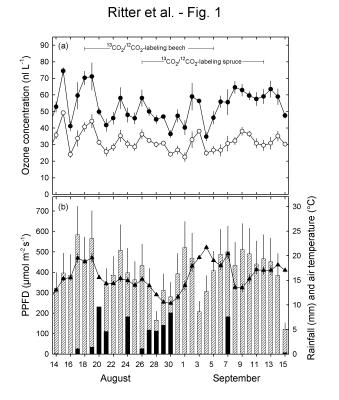


Fig. 1.

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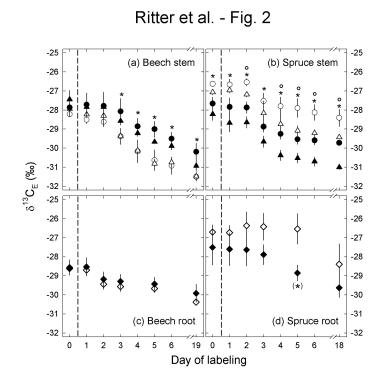
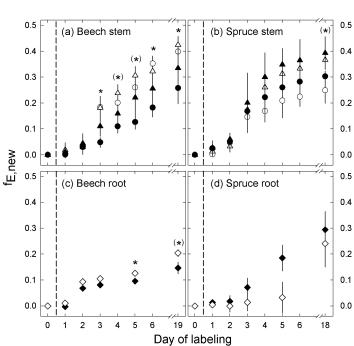


Fig. 2.

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Ritter et al. - Fig. 3

Fig. 3.

Ritter et al. - Fig. 4

	Unlabeled control		Labeled trees 1xO ₃		Labeled trees 2xO ₃	
	CO ₂ samples	Solid samples	CO ₂ samples	Solid samples	CO ₂ samples	Solid samples
0 -1 -2 -3	— 」 <i>— 四 </i> — 甲 単 					0 -1 -2 -3 -3 -3 -3 -3 -3 -3 -3 -3 -3 -3 -3 -3
after labeling (‰) 6 & 2 9 5 5	Canopy air CZZ Upper stem CO ₂ efflux ZZZ Lower stem CO ₂ efflux ZZZ Lower stem CO ₂ efflux EZZ Coarse root CO ₂ efflux EXX Soli respired CO ₂ at 8 cm EXX Soli respired CO ₂ at 15 cm	Phloem sugars CCCC Sun leaves Shade leaves Fine roots (a) Beech		(b) Beech		-4 -5 -6 -7 -8 (c) Beech
8-7-2 8-1-5 8-2-9-5-7 8-2-0 8-2-10 8-2-10 8-2-10 8-2-10 8-2-10						0 -1 -2 -3 -4 -5 -6 -7 -7 -8
-9	<u></u>	(d) Spruce	-	(e) Spruce	<u> </u>	(f) Spruce _9

Fig. 4.

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