

# ***Interactive comment on* “The effect of drought and interspecific interactions on the depth of water uptake in deep- and shallow-rooting grassland species as determined by $\delta^{18}\text{O}$ natural abundance” by N. J. Hoekstra et al.**

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Main remarks. The paper deals with an essential topic, which refers to the ability of multispecific crops, and in this case, grasslands to sustain more severe droughts compared to pure stands. The choice of species is relevant for all Europe and more. The issue is also about methodology for studying resource sharing, and especially water, between species of a community. The choice of natural isotopic abundance of  $^{18}\text{O}$  is relevant and the experimental design is sound, using a comparison of control plots (i.e. rainfed) and plots protected by permanent rain out shelters. The duration of the

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drought studied is long enough to mimic a significant water deficit. The measurements made on biomass produced during the drought period itself are relevant for at least one important issue in drought resistance studies. The replicates number each year and the two site-year experiment provide a significant number of data for sound conclusions. The text is very clear, figures are mostly clear too. All of them if not more are necessary in the main text. Some conclusions are new and important.

I have however two serious concerns with the data itself on the one hand and with the treatment of the data on the other hand. Given the importance of the topic, the quality of the data and the novelty of the science, I really hope that the authors have the resources to work on these points and I therefore suggest that the paper should go through major modifications.

Firstable, the soil water isotopic composition is not clear. Fig B1 should be included as figure in main paper, not as appendix. The difference in soil profiles between the rain fed and rain out shelter is puzzling. No clear explanation is given for a difference as large as 2 o/oo at Tänikon (Fig B1). If the regional waters are close to -8 o/oo, how is it possible that we have -11 o/oo at the end of the drought period in Tänikon ? Are there any measurements of rainfall isotopic signature ? This would be a very useful measurement here. Futhermore, the gradient results from the soil surface evaporation and from the net subsequent diffusion of heavy isotopes downward. Soil evaporation could have been higher under the rainout shelters due to higher temperature but indeed, the first and main impact of such superstructure is a reduction of incident radiation. As a consequence and given the small difference in air temperature,  $ET^{\circ}$  could have been likely 10- 20 % less under rain out shelters. Were there any estimate of such reduction in incident radiation and at least, could the energy interception by the shelter be measured? This is critical to discuss several aspects of the responses (biomass production, water consumption, depth of water extraction, which depends on transpiration (see Boujamlaoui et al 2005) Finally, the soil water profiles clearly indicate a quite important water consumption below 40 cm which is not much addressed in the paper.

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Secondly, I strongly recommend to drop all reference to the first direct inference of water extraction from the comparison between the soil delta gradient and the so called “xylem water” signature. (incidentally, only a small fraction of the water extracted from the plant samples is truly xylem water.) It has been shown that such use of comparison is wrong (Durand et al. 2007). Furthermore, it is useless here and therefore unnecessarily weakens the paper a lot. If the question was about the ranking between treatments and species, then there was no need to infer any actual depth from the delta 18O data. The second estimate is clearly much more rigorous, providing an estimate of the average depth. By the way, why was direct comparison of the average depth of water extraction using the Philipps and Gregg methodology with direct inference not made? This would be much more convincing than a simple correlation. But again, even with the IsoSource computation, such statistical approach is not real evidence for actual depth. Isosource is certainly an important step forward and provides very interesting insights for interpreting the delta and I find that the use made of that software here is really relevant. What is worrying is that for the red clover however, there seems to be a contradiction between the conclusions obtained comparing the soil water profiles (drought and control) and the comparisons using the PCWU0-10. This raises the question of the accuracy of the methodology used when there are more sources than markers in this case at least.

Additionally, some more detailed remarks on the manuscript.

1. The species are given relative depth of rooting and water extraction a priori. However, it is very much related to the conditions of the experiments. For instance, Durand et al (1997, 2009) demonstrated that *L. perenne* could extract water from very similar depth as *F. arundinacea*, a well-known potentially deep rooted species. The qualification of shallow rooted for *L. Perenne* is therefore questionable (a mighty reason for doing this experiment indeed). Ascribing any depth of water extraction in the introduction or as a reputation should be made more reluctantly.

2. That local water extraction depends on local root density is very well established

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under well watered conditions (both in trees and crops) and is difficult to be introduced as a question.

3. Similarly, that water is extracted from deeper & wetter horizons when water is scarce near the surface is all but surprising. The water potential distribution in the plant – soil system inevitably leads to that and this has been documented (see literature cited like Sainclair , Garwood, but more recently modeled by Jarrige , Doussan or measured under various conditions by Gonzalez Dugo et al. . .)

4. The radiation below the shelters was not measured, which is an issue. The irradiative energy balance is likely more important for potential evapotranspiration<sup>o</sup> than the air temperature or humidity in that situation. How much could have been ET<sup>o</sup> been modified in these conditions ?

5. The apoplastic water in tiller's base may well not be more than 40 %, out of which, xylem water is even much less.

6. The outer sheath of grass tillers may transpire and therefore enrich the tissues water in heavy isotopes of water. This is not so much related to photosynthesis.

7. The direct inference of depth of water extraction using the soil profile delta 18O is flawed and misleading. The first paragraph of data analysis in material and methods and all paragraphs later on referring to it should be dropped (with absolutely no harm to the strength of the paper in the contrary !).

8. The use of water from deeper than 40 cm is not discussed. Could that have had some impact on computations of the PCWU0-10 ? It should be discussed somewhere anyway because we have no data on the delta 18O below 40cm.

9. The differences observed between the delta 18O profiles in the deep horizons at the same place are difficult to understand. What could have caused this ? Were the soil sampling conditions similar ?

10. P12 L 337: no agreement between the two estimates is presented but a correlation.

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11. Why no estimate of the average depth of water extraction using IsoSource is shown ?

12. Drought resistanceshould be defined here. In this case, drought resistance is mend as production during dry condition relatively to control conditions. The control is always an issue in drought response analysis. All that is needed here is a clear definition.

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