Part 1 RCs

RC1: 'Comment on egusphere-2023-1522', Anonymous Referee #1, 29 Aug 2023 reply

Quick et al. present a study of tree water use under elevated CO2 in a FACE experiment. BIFoR is a unique FACE experiment in that it is located in an old growth forest and could potentially offer better insight into the future of global forests than previous FACE experiments in young plantation forests. The question of plant-water relations under high CO2 is a critical one and includes processes that are often uncertain so data such as those presented in this paper are extremely valuable.

While it is obvious that a lot of effort has gone into data collection, the paper is very poorly written, with little focus or in depth analysis of what was measured. While two hypotheses are raised in the introduction, these neither follow from the rest of the introduction nor are they followed through in the rest of the paper. The statistical analysis is lacking and it is hard to understand what the conclusions of this study are. I hope that my comments can help improve the manuscript so that this very valuable data can be published.

RC1 Major comments:

RC1 Major Comment 1:

- Abstract - this is very detailed and it is unclear what the focus of the paper is. Details of measurements do not generally belong in an abstract and distract from the main point of the paper.

RC1 Major Comment 2:

- Introduction - this starts very abruptly and the different sections feel very disjointed, without a clear storyline. In general, paper introductions do not need subsections and removing the headers and joining the paragraphs logically might help the reader.

RC1 Major Comment 3:

- Methods - there are a lot of details on the measurements at the site, perhaps too many for the main body of the paper, but little to no details on the statistical analysis of the data. From the results section it is apparent that some linear regressions have been performed, although these are not documented in the methods and it is unclear how and why these particular analyses were chosen. It is unclear if the data have been tested for normality or any other assumptions of the methods employed.

RC1 Major Comment 4:

- Statistical analyses - the study contains only very basic stats. There is no attempt made to test whether any of the observed differences are significant or not. Most importantly, since ecological processes are complex and many factors contribute to water use, as highlighted by the authors themselves, it is unlikely that a simple linear regression is the best model to use. Some sort of mixed effect or general linear model that can take into account the multiple factors involved might give a better picture of the response of tree water use to CO2.

RC1 Major Comment 5:

- Initial conditions - this is the case with any manipulative experiment, but one always has to wonder about the effect of initial conditions in each treatment on the actual treatment effect. As a minimum, initial conditions should be shown and discussed. Initial conditions could also be included in any more complex statistical model involved.

RC1 Major Comment 6:

- Discussion & interpretation - this is almost entirely lacking. The last two sections are in some part a discussion, but they are very superficial, do not discuss the implications of the findings or any limitations of the study.

RC1 Minor comments:

RC1 Minor Comment 1:

L 14 “diurnal (i.e. daylight)” maybe use daytime to avoid confusion?

RC1 Minor Comment 2:

L 47 This is a rather abrupt start to the introduction - why large trees?

RC1 Minor Comment 3:

L 50 unclear what the relaxed xylem is

RC1 Minor Comment 4:

L 60 wet deposition of?

RC1 Minor Comment 5:

L 71 photosynthesis is the name of the process even when the pigments are not green.

RC1 Minor Comment 6:

L 71 slightly odd references to model predictions, especially Guerreri et al. there is a lot of literature out there on model predictions of CO2 fertilization effect.

RC1 Minor Comment 7:

L 110 Do you mean drought?

RC1 Minor Comment 8:

L114 What is meant here by location - geographical coordinates, altitude, slope?

RC1 Minor Comment 9:

L 141 This sentence is very convoluted and it is unclear what separation refers to

RC1 Minor Comment 10:

L 156 monthly distributions of what? Significant differences between what and what?

RC1 Minor Comment 11:

L 182 is 1840 the plantation year?

RC1 Minor Comment 12:

L 318 It is unclear how these characteristics were used

RC1 Minor Comment 13:

L 335 I'm not sure this section and level of detail are necessary

RC1 Minor Comment 14:

L 477 It is unclear why there is a model fitted to each day rather than across days. It would also be good to see R2 values

RC1 Minor Comment 15:

L 482 'smaller', 'similar' Were there any statistical tests done to see if these differences were significant?

RC1 Minor Comment 16:

Figure 7 It is hard to tell what the difference between a) and b) and c) and d) is and why these could not just be one plot.

RC1 Minor Comment 17:

Figures 7 and 8 missing statistical values such as R2 or p

RC1 Minor Comment 18:

Figure 9 Would be good to show where the statistical differences are

RC1 Minor Comment 19:

L 549 So what does this figure show us?

RC1 Minor Comment 20:

L 564 Since you have precipitation data, could you test this deduction?

RC1 Minor Comment 21:

L 591 this information belongs in the methods

RC1 Minor Comment 22:

Sections 3.4 and 3.5 are these meant to be a discussion?

RC2: 'Comment on egusphere-2023-1522', Anonymous Referee #2, 12 Sep 2023 reply

This study evaluates the possible effects of enhanced CO2 (eCO2) on tree-water use, the measurements were conducted at one of the FACE experiments on mature forests. Here, the authors defined tree-water use as the daytime accumulated sapflow (TWU) and proposed two hypotheses: H1 A detectable eCO2 treatment effect on TWU is present and H2 TWU is greater in the presence of FACE infrastructure. In my opinion the hypotheses are relevant and could provide valuable information that could improve our understanding on ecosystem responses to environmental change and on eCO2 experimental design.

Unfortunately, the sections of the manuscript feel unconnected due to inclusion of irrelevant information, which could easily be moved to the supplementary material. The hypotheses are not properly tested, so the interpretation of the results and the conclusions are mostly based on visual interpretation of the plots.

The data used in this study is incredibly rare and valuable, so I really hope that the authors can make use of the following comments to better leverage these data.

RC2 Major comments:

RC2 Major Comment 1:

Abstract:

There is an unnecessary amount of detail here, it distracts the reader.

It is not clear to me how this info is going to contribute to the development of more “realistic dynamic vegetation models”

RC2 Major Comment 2:

Introduction:

The sections inside the introduction feel disconnected and instead of leading the reader to the research questions they feel like tangents.

Not a single mention to the Huber value or water use efficiency, which are key concepts to connect CO2 and water in trees.

RC2 Major Comment 3:

Methods:

Almost half of this manuscript is used to describe the methods. It is too much in my opinion. If the authors feel like the figures in this section are really necessary, they could be moved to the appendix section.

RC2 Major Comment 4:

There is no mention to any statistical test to evaluate the hypotheses.

<u>RC2 Major Comment 5:</u>

I don’t really think TWU is a good metric to detect effects of eCO2 in only 5 years of experiment if the trees are 180 years old, the change of sapwood area of trees under aCO2 and eCO2 would be negligible. However, if measurements of Huber value and Leaf area index are available, TWU can be converted to transpiration, enhancing the differences between treatments if any.

There is no connection on how soil moisture and throughfall are going to be used complement the analysis.

<u>RC2 Major Comment 6:</u>

Results and Discussion

There is almost no discussion at all. The results are not put in context, it is hard to tell how the reported values compare to other studies. A relative change between aCO2 and eCO2 would be very useful here.

<u>6a:</u>It is well known that sap flow escalates with sapwood area (proportional to Rb), and Rb to Ac, I don’t really see novelty on reporting linear models on these relationships. How these regressions help with the evaluation of the hypotheses?

<u>6b:</u>All the interpretations to the results rely on visual inspection, I believe this is not enough to evaluate if your hypotheses are true.

<u>6c:</u>It is very interesting to see here interception, throughfall, and soil moisture measurements, however these results appear disconnected from the other results.

<u>6d:</u>The word “significant” is used to described key results of the study, however there is no statistical test associated.

<u>6e:</u>You state that hypothesis 2 is true, however this is hard to tell without proper stats. It is unlikely that the effect of the infrastructure alone is bigger than the effect of the eCO2. If that were true, we could see the combined effect of infrastructure + eCO2 effect in the eCO2 treatment as well.

RC2 Minor comments:

<u>RC2 Minor Comment 1:</u>

L 70 “…could be beneficial for individual tree Productivity” hard to tell without considering respiration and temperature increase. These ideas would be helpful to contextualize better.

<u>RC2 Minor Comment 2:</u>

L 155 how did you test for significant differences?

RC2 Minor Comment 3:

L 335 Is this relevant?

RC2 Minor Comment 4:

Figure 9. unreadable

RC2 Minor Comment 5:

L 570 how can you tell there are" significant differences" without any test? Why do you attribute similar general responses to daylength? What about temperature or VPD?

RC2 Minor Comment 6:

L 585 "Influence of … and herbivory on water usage dynamics" is misleading, you just point out that an attack of herbivory happened.

Reply

Citation: https://doi.org/10.5194/egusphere-2023-1522-RC2

Part 2 AC

General (RC1, RC2):

AC re General (RC1, RC2): *Thank you for highlighting the importance of this topic and suggesting general and specific improvements. We recognise there are some shortcomings in the submitted paper. We are very grateful for the efforts to improve, and the points made, by all referees/ community to bring this paper to the required standard, both in terms of the quality and flow of the paper, explaining the statistical analysis and in respect of testing the hypotheses. Obtaining the data reported has occupied 6 years of intensive field work and we are very pleased and relieved that referee RC1 agrees that the data are of great value and that RC2 agrees that the hypotheses are relevant and will aid understanding of ecosystem responses to environmental change and improvement of eCO2 experimental design. The two hypotheses and objectives are reviewed in section 3.5.*

Abstract:

<u>RC1 Major Comment 1 and RC2 Major Comment 1:</u>

AC lines 13-41 re <u>Major Comment 1 and RC2 Major Comment 1</u>: *The abstract is being refocused on the main methodology, objectives and findings. Its redraft will be finalised in the marked up revision.*

We also make reference to where dynamic vegetation models can be improved in our Introduction. (old LL123-127)

Introduction:

<u>RC1 Major Comment 2 and RC2 Major Comment 2:</u>

AC re* <u>RC1 Major Comment 2 and RC2 Major Comment 2</u>*: *We have improved the starting sentences and clarified the story line by reordering some of the sentences to improve the flow and logic and moving previous section 1.2 nearer the beginning. The aims of the paper have been clarified and sharpened. Some headers are retained as we believe they help the reader frame the sections.*

See also AC on RC2 Major Comment 5 in respect of Huber value and water use efficiency.

Materials and Methods:

<u>RC1 Major Comment 3 and RC2 Major Comment 3:</u>

AC re <u>Major Comment 3 and RC2 Major Comment 3</u>: *We recognise that this may be too detailed for the reader and have moved some material to the SI (e.g. re non sap flow instrumentation) and some re sap flow to a new Appendix.*

In respect of the Figures in this Section of the paper, some of them add knowledge to the characteristics of water flow within the xylem. We have reviewed all these figures and explained their purpose within captions as appropriate. We moved any remaining figures to the new Appendix C which covers detail of the sap flux methodology. (e.g.

Fig.3 is now Fig.C1). We show the remaining text for Sections 2.7 to 2.8 in part 3 of our response as an example.

A summary of the analysis tools used is contained in Section 2.9.

Regression analysis in respect of tree size was progressed following visualisation, facilitating normalisation of the water usage data in respect of tree parameters and consequent comparison of data from different treatments; the linear model provided a simple best fit for this range of data – e.g. See old LL 473&474.

There is reference to the minimum detectable sap flux using this method (old LL425-426) showing that the effects are tree-size dependent and referring to Appendix A2, Stage 5.The limitations and truncation effect of the sap flow transducer method is also outlined in Appendix B. The first paragraph of this has been expanded to explain the influence of truncation on the normality of the data. Some text has also been moved here from the Results section.

Further response on statistical methods is outlined below.

Statistical analysis

RC1 Major Comment 4:

AC re RC1 Major Comment 4: *Timely sharing of sap flux data from this plant-water experiment is important to enable its use in land surface models such as JULES and vegetation models. As a result we have restricted our statistical approach to show the validity of the data and pointed to where this fills gaps in existing knowledge.*

RC2 Major Comment 4:

There is no mention to any statistical test to evaluate the hypotheses.

AC re RC2 Major Comments 4:

We recognise there were some limitations in our pre-print in respect of statistical testing of the hypotheses. We have now undertaken additional analyses to underpin our findings in respect of the hypotheses, focusing on 2019-2021. A brief insertion of new figure content and p values tables (draft below) has been added to the Results. The Discussion will be modified to reflect the new findings.

A summary of the analysis tools used is contained in Section 2.9 – see revised section below. Regression against tree size allowed normalisation of the water usage data in respect of tree parameters and consequent comparison of treatments; the linear model provided a simple best fit for this range of data – e.g. See old LL 473&474.

RC1 Major Comment 5:

AC re RC1 Major Comment 5: *With the exception of reporting of precipitation and throughfall data where this is available pre-treatment (sections 2.4, 2.5 and 3.4), reporting of the initial conditions is not possible. To assist the reader some data has been added to the SI document clarifying this point.*

RC2 Major Comment 5:

AC re RC2 Major Comment 5: Our *normalised daylight TWU is a whole-tree integrative measure of transpiration and, as such, a measure suitable for determining the effects of the CO_2 treatment. No other FACE studies of mature trees have had sufficient periods of sap flux data across treatment and control replicates to investigate seasonal CO_2 treatment effects over successive years. Estimates of water usage as a function of land area across each of the 9 arrays for all oaks in the experiment is outside the scope of the present study.*

Huber value and leaf area index for the years reported cannot be calculated directly on a per tree basis.

Results and Discussion

RC1 Major Comment 6 and RC2 Major Comment 6:

AC re RC1 Major Comment 6 and RC2 Major Comment 6: *Refinement of the Results and Discussion section is being carried out and will be presented in the marked up revision as an improvement to the pre-print.*

AC re RC2 Major Comment 6 sub comments

6a: *There is a paucity of results relating sap flow with sapwood area for mature Q. robur of the range of DBH considered here. The relationship in this study enables normalisation of the TWU data. This is not the only option for normalisation but, as we include all sap flux data across all days of our treatment season April to October, including dark and/ or rainy days, we do not use the tree-size normalisation method adopted in a similar study (i.e. Web-FACE, reported Leuzinger and Körner, (2007)) which compared species water usage. They normalised each species by the highest tree specific accumulated sap flux for a given period of monitoring (22 days).*

6b: *We agree that the analyses should not rely on visual qualitative observations. We have added statistical tests to underpin our findings in respect of the hypotheses. These are reported in a new section of the results.*

6c: *Please see response to* **RC2 Major Comment 5.**

6d: *We report the significance of our normalisation linear models in old LL500-507and LL522-530 including reference to p-values in the supporting tables. Old lines 571-572 and 643-651use the term significant incorrectly and these paragraphs have been reviewed and rephrased.*

and 6e: *Please see response to* **RC2 Major Comments 4**

Minor comments RC1 and RC2 in line order:

RC1 Minor Comment 1:

L 14 **AC:** *Noted. We have selected the terms most used in supporting plant-water and assimilation literature i.e. diurnal as the antonym to nocturnal. We recognise that the term can also be used in place of circadian, hence the clarification of definition and usage for this paper.*

RC1 Minor Comment 2:

L 47 **AC:** *Noted. We have now added couple of sentences forming a better start to the Introduction. See below.*

RC1 Minor Comment 3:

L 50 **AC:** *Whilst we previously referred to relaxed xylem to reflect changes in diameter due to lack of plant water tension (i.e. due to lack of or reduced transpiration), this sentence lacked clarity and the references had been incorrectly grouped. Above ground both ambient air temperature and lack of transpiration affects the stem DBH and related physical traits such as stem circumference. We intended to point to the tree anatomy response to a variety of stressors. Sentences in pre-print (old LL49-51) have been regrouped and redrafted. See below.*

RC1 Minor Comment 4:

L 60 **AC*:*** *We have deleted this sentence as part of refocusing content of the Introduction.*

RC2 Minor Comment 1:

L 70 ***AC:*** *old LL71-74 have been rephrased to clarify.*

RC1 Minor Comment 5:

L 71 **AC:** *Noted. Changed to refer to chlorophyll.*

RC1 Minor Comment 6:

L 71 **AC:** *We are not discussing the CO2 fertilization effect here, but rather the implications of CO2 fertilisation for water usage. Guerrieri et al. (2016) is quoted as a recent example paper as we are concerned here with the interface of forest carbon uptake and water use. We have changed the references to include some additions. (e.g. Norby et al. 2016)*

RC1 Minor Comment 7:

L 110 **AC:** *Yes, water availability and drought are relevant. We have modified the sentence.*

RC1 Minor Comment 8:

L114 **AC***: yes this is ambiguous, we have redrafted the phrase to be more specific.*

RC1 Minor Comment 9:

L 141 **AC:** *Dendrometer sentences have been removed as not relevant to this paper.*

RC2 Minor Comment 2:

L 155 **AC:** *A summary of the analysis tools used is contained in Section 2.9.*

RC1 Minor Comment 10:

L 156 **AC:** *monthly distributions are of '…seasonal and inter-year patterns of daily water usage by old growth oak trees…' as stated. Significant differences sentence has been rephrased to clarify.*

RC1 Minor Comment 11:

L 182 **AC:** *See also L796-798. Dendrochronology was used to date the age range of oaks in the FACE experimental forest. It is unclear what age the trees were when they were planted hence the reference to circa 1840.*

RC1 Minor Comment 12:

L 318 **AC*:*** *See stage 5 in Appendix Table A2 also Figure 2. The tree radius at insertion point derived from the circumference minus the bark thickness gives R (m).*

RC1 Minor Comment 13 and RC2 Minor Comment 3:

L 335 **AC:** *Moved to Appendix C*

RC1 Minor Comment 14:

L 477 **AC:** *the model is fitted across all the days of July. See also comments on Figs 7 & 8. Seasonal water usage modelling would need to account for day length and this modelling has not been undertaken.*

RC1 Minor Comment 15:

L 482 **AC:** *See L501 and Table S5, also comments on Figs 7 & 8. It would be possible to do more extensive treatment comparisons, but these would not result in significant differences, as discussed.*

RC1 Minor Comment 16:

Figure 7 **AC:** *putting all the treatments, separated by year, on one plot makes the results difficult to visualise. b) and d) combine the regression line results for all the years available for a particular treatment.*

RC1 Minor Comment 17:

Figures 7 and 8 **AC:** *p is stated in the related tables (Fig. 7 relates to Table 2 and Table S5, Fig.8 relates to Table 3 as stated in the text).*

RC1 Minor Comment 18:

Figure 9 **AC:** *Noted. Please see additional ANOVA analyses, see also response to* **RC2 Major Comments 4**

RC2 Minor Comment 4:

Figure 9 (L543). **AC** *We shall improve readability by increasing the size of this Figure and its labels in the final manuscript. The Figure can then be opened in a Figure viewer.*

RC1 Minor Comment 19:

L 549 **AC:** *We describe and discuss the effect of TWU normalisation here (Fig. 9). The caption and related paragraphs show the seasonal and inter-year patterns. These are also now explored statistically in the new ANOVA models.*

RC1 Minor Comment 20:

L 564 **AC:** *We have deleted this reference to precipitation as it is known that there are multiple environmental and soil drivers affecting transpiration which we will not be testing statistically within this paper.*

RC2 Minor Comment 5:

L 570 **AC** *We have rephrased this sentence. We previously described Figure 11 rather than statistically tested the conditions in each month. The assumption in this paper is that all trees experience the same or very similar temperatures and VPDs on a given day, so that comparison of treatments will be evident visually and give pointers for our statistical analysis of the hypotheses (see comments on ANOVA) and exploration in our future research.*

RC2 Minor Comment 6:

L 585 **AC** *We describe the effects of herbivory in respect of decreasing leaf area and canopy closure timing. By removal of leaf tissue herbivory reduces the whole tree transpiration (less leaf so less water usage).The supporting reference describes this in more detail. LL622-623 also explain the effects of the herbivory attack on canopy closure and hence throughfall.*

RC1 Minor Comment 21:

L 591 **AC:** *Noted. We have moved this paragraph to the end of Section 2.2 Measurements overview .*

RC1 Minor Comment 22:

AC: *Section 3.4 is a combined results and discussion as are all subsections in section 3. The two hypotheses and objectives are reviewed in section 3.5. If a separate Discussion section would be clearer we will incorporate this when we redraft the manuscript.*

Part 3 AC – changed text.

Abstract: (RC1, RC2)

Revised Abstract, 2nd, 3rd & 4th sentences (old LL14-22).

The present study investigates diurnal (i.e. daylight) water usage of old growth oaks within an experimental treatment season from April to October inclusive. Over five years, from 2017 to 2022, we collected individual tree data from eighteen oaks (*Quercus robur L.*) within a large-scale manipulative experiment at the Birmingham Institute of Forest Research (BIFoR) Free-Air CO_2 Enrichment (FACE) temperate forest in central England, UK. Diurnal tree water usage per day (TWU) across the leaf-on seasons was derived from these data….

Introduction: (RC1, RC2)

Revised old LL70-71

Primary producers may respond to elevated CO_2 (eCO_2) levels by assimilating and storing more carbon, which for plants containing chlorophyl happens during photosynthesis.

Revised old LL71-74

Global carbon and water cycle models (Guerrieri et al., 2016; De Kauwe et al., 2013; Medlyn et al., 2015; Norby et al., 2016) predict that, at least until the middle of the 21st century, trees and plants could potentially photosynthesise more efficiently, which may induce increased carbon storage. This could be beneficial for individual tree productivity.

Revised old LL109-111

Within the UK maritime temperate climate, only a few ecohydrological studies (e.g. Herbst et al., 2007; Renner et al., 2016) have previously considered the sap flow responses to water availability and drought for old growth Quercus species.

Revised old LL113-

The response of woody plants to drought varies considerably by species (Leuzinger et al., 2005; Vitasse et al., 2019), location (e.g. north versus south in Europe Stagge et al., 2017)), soil characteristics such as soil texture (Lavergne et al., 2020) and combinations thereof (Fan et al., 2017; Salomón et al., 2022; Sulman et al., 2016; Venturas et al., 2017).

Revised L156

We test for significant differences between treatments within these water usage distributions and patterns.

Revised complete sections(old LL 46-159)

Long-term manipulation experiments enable prediction of how, under climate change, increased atmospheric carbon dioxide levels and climate environmental extremes might affect plants and ecosystems. Plant hydraulics are adapted to expected ranges of environmental parameters, with larger plants exhibiting greater resilience to wider parameter variation due to their ability to maintain water and food reserves. Large trees can maintain their transpiration rates even during water stress but remain vulnerable (Süßel and Brüggemann, 2021). To maintain transpiration demands, trees accommodate to: diel variation in solar radiation; respiration fluctuation; high temperatures; and seasonal soil water deficits. Short-term mechanisms include stomatal regulation, stem diameter variations (Sánchez-Costa et al., 2015), use of stored plant water and use of available water at variable soil depth (David et al., 2013; Flo et al., 2021; Gao and Tian, 2019; Nehemy et al., 2021). Longer term strategies include development of resilient root structures, (David et al., 2013; Flo et al., 2021) and minimisation of embolism mitigated by different xylem structures (Gao and Tian, 2019). The ability of mature trees to withstand climate extremes may rely in part on using these buffering traits which act to prevent permanent damage and maintain viability (Moene, 2014; Iqbal et al., 2021). This prompts the further question of how increasing atmospheric carbon dioxide levels will affect the hydraulic resilience of trees. The response of woody plants to drought varies considerably by species (Leuzinger et al., 2005; Vitasse et al., 2019), location (e.g. north versus south in Europe Stagge et al., 2017)), soil characteristics such as soil texture (Lavergne et al., 2020) and combinations thereof (Fan et al., 2017; Salomón et al., 2022; Sulman et al., 2016; Venturas et al., 2017). Trees require water/ water vapour at all stages of life experiencing insufficient water at times (e.g. under elevated temperatures and drought) so tree species have evolved different root traits (Montagnoli, 2022) and hydraulic characteristics (Sperry, 2003) to maintain their fitness to their environment. Volkmann et al., (2016) used rainwater isotopes to track soil water sources for sessile oak (*Quercus petrea*) and beech (*Fagus sylvatica*). Sánchez-Pérez et al., (2008) studied oak (*Quercus robur*), ash (*Fraxinus excelsior)* and poplar (*Populus alba*). Both studies found that use of soil water at different depths varied between species and seasonal variation of climatic conditions. Trees therefore exhibit variable resilience to water shortage/ excess and other environmental stressors (Brodribb et al., 2016; Choat et al., 2018; Grossiord et al., 2020; Landsberg et al., 2017; Martínez-Sancho et al., 2022; Niinemets and Valladares, 2006; Schäfer, 2011; Süßel and Brüggemann, 2021) with a broad spectrum of sometimes species-specific strategies and coping mechanisms (Schreel et al., 2019).

Future-forest atmospheric carbon dioxide and water usage

Primary producers may respond to elevated CO_2 (eCO_2) levels by assimilating and storing more carbon, which for plants containing chlorophyl happens during photosynthesis. Global carbon and water cycle models (Guerrieri et al., 2016; De Kauwe et al., 2013; Medlyn et al., 2015; Norby et al., 2016) predict that, at least until the middle of the 21st century, trees and plants could potentially photosynthesise more efficiently, which may induce increased carbon storage. This could be beneficial for individual tree productivity. Stomatal regulation determines the trade-offs between carbon assimilation and water loss and determines the rate and quantity of water usage seen in the stems of woody plants. Water usage at tree level is, therefore, a strongly integrative measure of the whole plant response to environmental drivers (such as temperature and precipitation) and experimental treatments (such as eCO_2).

Untangling the canopy water exchange and soil moisture hydraulic recharge dynamics within forest Free-Air CO_2 Enrichment (FACE) experiments can be complex, but responses to eCO_2 manipulations (including stepwise increases (Drake et al., 2016)) inform our understanding of plant responses to climate change scenarios. Specific studies concerning transpiration and water savings of eCO_2 responses (Ellsworth, 1999; Li et al., 2003) have already improved the model predictions (De Kauwe et al., 2013; Donohue et al., 2017; Warren et al., 2011) and here we seek to enable further prediction improvements.

Experimental research into ecohydrological responses of old growth deciduous forest to changing atmospheric CO_2 levels has been limited. The Web-FACE study (Leuzinger and Körner, 2007) reported on temperate old growth species and found that *eCO_2* reduced water usage in *Fagus sylvatica* L. (dominant) and *Carpinus Betula* L. (subdominant) by about 14% but had no significant effect on the water usage of *Quercus petraea* (Matt.) Liebl*.,* the other dominant species present. There were a small number of trees (six) of a Quercus species included in Leuzinger and Körner's (2007) study, with water savings monitored by accumulated sap flux (normalised against peak values in each tree) over two 21-day periods. Changes in water usage by old growth oak trees at *eCO_2* when measuring for longer periods (greater than a month) across the leaf-on season have not previously been reported.

The paucity of studies of the water usage of mature temperate trees under *eCO_2* significantly weakens model-data comparisons at FACE sites (De Kauwe et al., 2013). Warren et al., (2011) reviewed the forest FACE experiments which, apart from Web-FACE, all constituted younger deciduous and mixed plantations less than thirty years old (Schäfer et al., 2002; Tricker et al., 2009; Uddling et al., 2008; Wullschleger & Norby, 2001; Wullschleger et al., 2002). Some of these studies are long-term (> ten years) but all are limited in their period of monitoring sap flow, maximum continuous data periods being covered by Schäfer et al., (2002) at Duke forest USA (1997-2000) and lesser periods by Oak Ridge National Environmental Research Park (ORNL) USA and POP/ EuroFACE (Wullschleger & Norby, 2001; Tricker et al., 2009). Larger numbers of young trees (252 aspen–birch) were monitored for sap flux by Uddling et al., (2008), whereas most recent sap flow studies of oak have either been single trees of different species (e.g. Steppe et al., 2016) or short-term proof-of-concept studies using experimental instrumentation (Asgharinia et al., 2022).

There are further (2010 onwards) sap studies of deciduous oak which do not manipulate CO_2 but which offer helpful data for comparison, for example within Europe (Aszalós et al., 2017; Hassler et al., 2018; Perkins et al., 2018; Schoppach et al., 2021; Süßel and Brüggemann, 2021; Wiedemann et al., 2016) and North America (Fontes and Cavender-Bares, 2019). Robert et al. (2017) have reviewed the characteristics of these old growth species from multiple studies which help us to place our results in context. Within the UK maritime temperate climate, only a few ecohydrological studies (e.g. Herbst et al., 2007; Renner et al., 2016) have previously considered the sap flow responses to water availability and drought for old growth Quercus species.

Improving global vegetation models and questions of scale.

Global vegetation models have been developed based on leaf-level plant knowledge alongside that of soil-tree-atmosphere exchange (e.g. Medlyn et al., 2015). These models have predicted reduced canopy conductance, *Gs* and increased run-off in future climate scenarios, but an important gap has been identified between estimated and observed water fluxes (De Kauwe et al., 2013).

Canopy/ leaf transpiration estimates from stem xylem sap flux (Granier et al., 2000; Wullschleger and Norby, 2001; Wullschleger et al., 2002), use the parameter canopy conductance (*Gs*) to reflect how the whole canopy transpires rather than concentrating on individual leaf stomatal conductance to water. Measurements of *Gs* and transpiration and partitioning of evapotranspiration in deciduous forests (Tor-ngern et al., 2015; Wehr et al., 2017) have now clarified relationships between canopy parameters and environmental variables PAR, VPD and precipitation. Long-term carbon and water flux data from flux towers in forest ecosystems (e.g. Ameriflux (Baldocchi et al., 2001), Euroflux (Valentini, 2003), FluxNet (Baldocchi et al., 2005)) and satellite datasets such as EOS/Modis worldwide (Huete et al., 1994), have provided canopy level and landscape wide data. Plant focused environmental manipulation studies, such as FACE, can provide data on individual parameters and processes to inform and challenge the models.

At the forest scale, studies of the effects of the European drought (2018-2019) on forested landscapes have shown that recovery time for surviving trees may be several years, affecting both plant growth, stem shrinkage (Dietrich et al., 2018) and branch mortality during that time, especially for old growth deciduous species (Salomón et al., 2022). At this forest scale (Keenan et al., 2013; Renner et al., 2016), there is also a more complex impact on ecosystem and atmospheric demands, as planetary-scale CO_2 levels increase affecting boundary layer feedbacks.

In contrast to forest- and leaf-scale studies, the present study is tree-focused and bridges the data gaps identified previously (Medlyn et al., 2015; De Kauwe et al., 2013) in respect of model-data scale mismatch. Tree-scale studies have provided essential data for calibration and validation of tree-water models (De Kauwe et al., 2013; Wang et al., 2016;), identified key parameters driving responses to expected water shortages (Aranda et al., 2012) and compared species differences in mature tree responses to ambient (Catovsky et al., 2002) or eCO_2 (Catoni et al., 2015; Tor-ngern et al., 2015). Xu and Trugman, (2021) have updated the previous empirical parameter approach to global vegetation modelling, reinforcing the need to use measured tree parameters (such as sapwood area) to improve model predictions of climate change response.

Here we focus on whole tree species traits and link these parameters to diurnal (i.e. daylight) tree water usage per day (TWU, litres d-1) from stem xylem sap measurements, affirming the influence of leaf-on season precipitation and solar radiation/ air temperature.

Measurements of xylem sap flux are marginally intrusive, providing highly time-resolved plant water usage data for several years with minimal maintenance. Heat-based measurement techniques (Forster, 2017; Granier et al., 1996; Green & Clothier, 1988) have been used over the past 40 years in measurement of plant xylem hydraulic function (Landsberg et al., 2017; Steppe and Lemeur, 2007) with automated data capture enabling increasingly realistic models of whole tree xylem function.

Objectives, research questions and hypotheses

This study provides new gap filling data to characterise seasonal and inter-year patterns of daily water usage by old growth oak trees using monthly distributions. We test for significant differences between treatments within these water usage distributions and patterns. The paper examines the limitations of water usage measurement by compensation heat pulse (HPC) sap transducers. It also relates diurnal tree water usage per day (TWU, litres d-1) to measurable tree traits (bark radius and canopy area) and examines variation of TWU with environmental drivers and soil moisture.

Materials and Methods: (RC1, RC2)

(modified text old LL 211)

(MacKenzie et al., 2021). Table S2 shows instrumentation types and related parameters used for analysis within this paper.

((modified old LL212-256)

We experienced early leaf-on herbivory attacks in oak by Winter moth larvae, especially in 2018 and 2019 (Roberts et al., 2022) decreasing leaf area by 20-30% and affecting canopy closure timing. A longer dry period occurred in meteorological summer 2018 (Rabbai et al., 2023) with wide variation in summer monthly precipitation across the study years.

Seasonal definitions

The seven months of CO_2 treatment per year (with six months of leaf-on photosynthesis) do not easily divide into standardised meteorological seasons (Spring, Summer), so we define our months of interest, including non-treatment months as shown in Table 1. The table includes two months, March to April of pre-leaf growth when oak sap starts to rise.

Soil and throughfall precipitation data collection

Pre-treatment (2015-2017 for infrastructure arrays) and pre-project (i.e. 2017 onwards data for all *Ghost* arrays) on-site soil and throughfall data were used to characterise the site. Supplementary (2018 onwards) throughfall/ soil monitoring sites were added (see Mackenzie et al. (2021)). Shallow soil moisture and soil temperature data were captured at least half-hourly by the same CR1000 datalogger as the sap flow data.

For plants, incident precipitation affects their function in several ways during the leaf-on season. Firstly water droplets incident on the leaves which when combined with lack of sun prevent full photosynthesis. The canopy water mostly evaporates or may drip to ground. Secondly throughfall (P_{fs}, mm) reaching ground level may either runoff the surface being lost to the soil, infiltrate (providing some necessary support for root rehydration and plant water intake or evaporate. Lastly the soil water percolates through the soil layers to replenish the water table.

Water inputs of throughfall precipitation under the oak canopy (within 2 to 3 metres of a stem and situated near a soil moisture monitoring position) were measured in all arrays, with Fig. S2 showing a typical installation set-up.

Calendar months	**FACE Treatment season label**	**Note**	**Oak phenology at BIFoR FACE**				
			2017	**2018**	**2019**	**2020**	**2021**
March – April (*eCO_2* starts beginning April)	**Budburst & first leaf**	March is pre-treatment. **First leaf dates for** oak shown	**6 April** *	**25 April** *	**29 Mar** *	No data* (c. 6th April)	**27 April** *
May – June	**Early leaf-on**	Includes **canopy closure early leaf** of oak	-	-	-	-	-

July – August	**Mid leaf-on**		-	-	-	-	-
September – October (eCO_2 until end October)	**Late leaf-on**	Includes **start** of senescence i.e. **first tint**	**6 Sept**	**12 Sept**	**1 Oct**	**15 Sept**	**28 Sept**
November - Feb	**Dormant**	All remaining non-treatment months	-	(after 21 Nov)**	**26 Nov****	(after 03 Nov)**	**07 Dec ****
		Assumed leaf-fall season	6 Sept 2017 to 25 April 2018	12 Sept 2018 to 29 Mar 2019	1 Oct 2019 to c. 6 April 2020	15 Sept 2020 to 27 April 2021	28 Sept 2021 to 24th April 2022

Table 1: **Definition of treatment season periods and dates for oak phenology at BIFoR FACE according to Nature's Calendar criteria for years 2017–2021 (note this excludes canopy closure data - not recorded). First tint is also recorded for year 2016 as 4th Oct. * On-site first leaf data (not obtained in 2020 due to the Covid-19 pandemic; 6th April 2020 was noted as budburst, unverified first leaf is recorded as 24th April 2020). Note: Separate records of leaf-fall season are recorded for LAI calculation purposes as Nature's Calendar data does not discriminate first leaf fall by leaf colour. ** First bare tree date recorded. Nature's Calendar link: (https://naturescalendar.woodlandtrust.org.uk/). Phenocam additionally available for all years (https://phenocam.nau.edu/webcam/roi/millhaft/DB_1000/).**

FACE and meteorological measurements

Local precipitation (from a mixture of sources including Met. towers, see MacKenzie et al., (2021)) was recorded. Treatment levels of eCO_2, diurnal CO_2 treatment period, top canopy air temperature (T_a , ^{o}C) and total solar radiation (TG, Watt m^{-2}) were available from the FACE control system (Hart et al 2020; MacKenzie et al 2021). Data were averaged across the six infrastructure arrays for TG and T_a as the *Ghost* arrays have no FACE measurements (see supporting information Fig. S9).

Tree selection

There is variation inherent in biological individuals, in the same or different treatment types (Chave, 2013), which may not behave typically in space or time. This individual-tree experiment design aims to minimise untypical variation. Accordingly the following criteria were used to select trees for sap flow monitoring:

- canopy cover completely within the array (eCO_2 & aCO_2 arrays)
- central within the plot near logger and adjacent to access facilities at height (eCO_2 & aCO_2 arrays – for sampling and porometry access)
- straight stem, preferably with little epicormic growth
- no large dead branches within the canopy which might affect the comparative biomass of the tree
- unlikely to experience seasonal standing or stream water at the base

Target oak trees for monitoring were also chosen to suit the physical limitations of the transducer to logger constraints rather than randomly.

(modified Section 2.7-2.8 old LL257-401)

Tree characteristics

The tree size measurement approach is shown in Figure S3 and the range and mean-per-treatment values of bark circumference (metres) for all target trees are tabulated (Table S3). All oak trees were of similar height (circa 25 m). Tree stem circumference at insertion height of probes was measured at installation (from 2017 onwards), and in subsequent winters (Jan 2020-Feb 2022). We note that tree size will affect TWU (Bütikofer et al., 2020; Lavergne et al., 2020; Verstraeten et al., 2008).

Canopy spread of all target oak trees was measured around installation date (2017-2018) and repeated for all oaks in early 2022 (Fig. S3). We assume that the two-dimensional canopy area, derived from the mean canopy diameter plus stem diameter, is a good approximation to actual canopy spread and hence the whole canopy surface experiencing leaf transpiration. For trees of similar height we assume that allometric shape to estimate whole canopy volume will be similar. On the second occasion in 2022 we measured the asymmetry of each tree stem across the probeset cardinal positions (East-West) and right-angles to this (North-South) as a check of mean bark radius value for sap flux calculations.

Short incremental wood cores (circa 100mm long, 4mm diameter) were taken from two old growth oaks outside of the experimental arrays. Microcores were also taken near all 36 target oak probeset installation positions. These cores were used to determine wood hydraulic properties (Edwards and Warwick, 1984; Marshall, 1958) for sap flux calculations (see also stage 4, Table A2 and definitions Table A1). In summer 2021 woodcores taken from some of the target oaks were further analysed to check the conversion (xylem woody matrix**)** factors from heat velocity to sap velocity and to verify the active xylem radial width. The visibly active xylem (sapwood) is typically between seven and 50 mm when viewed in wet cores but can more easily be measured in dried cores or disks. The uncertainty in heartwood boundary *H* (m), as described in Appendix B, could be resolved in future similar studies by taking short cores prior to installing instrumentation.

Xylem sap flux

Details of the xylem sap flux measurement method and associated calculations are in Table A2 and Appendix C. Each target oak tree had two probesets, East and West facing and contained a central heat pulse probe and two measurement probes (each containing four thermocouples upstream and downstream of the heater (Fig. 2). Appendix B discusses the limitations which the time-out characteristic places on this set of HPC data and consequent results. Validity of high outlier values is considered within our analysis.

To determine whole tree sap flux several tree characteristics were used: (a) tree stem circumference at insertion point, (b) bark thickness, from which we derived tree stem cambium radius at insertion point *R* (m) and subsequently heartwood radius *H* (m) from sensor spacing. *H* (m) could not be determined from 10 cm cores as these were not taken for all sap trees monitored.

The xylem sap flow installations in target trees commenced in Jan. 2017. All no-treatment-no-infrastructure control (*Ghost*) oak trees provided data from August 2017 and commissioning of all 18 oak trees was completed by autumn 2018. All oak sap flow installations were successful and a total of 12,259 days of individual tree data (770,667 diurnal sap flux measurements across all months) were processed for the 2017–2021 TWU analysis. Resulting data gaps in the earliest installations affected four of the 36 probesets installed in four trees August 2017 until September 2019.

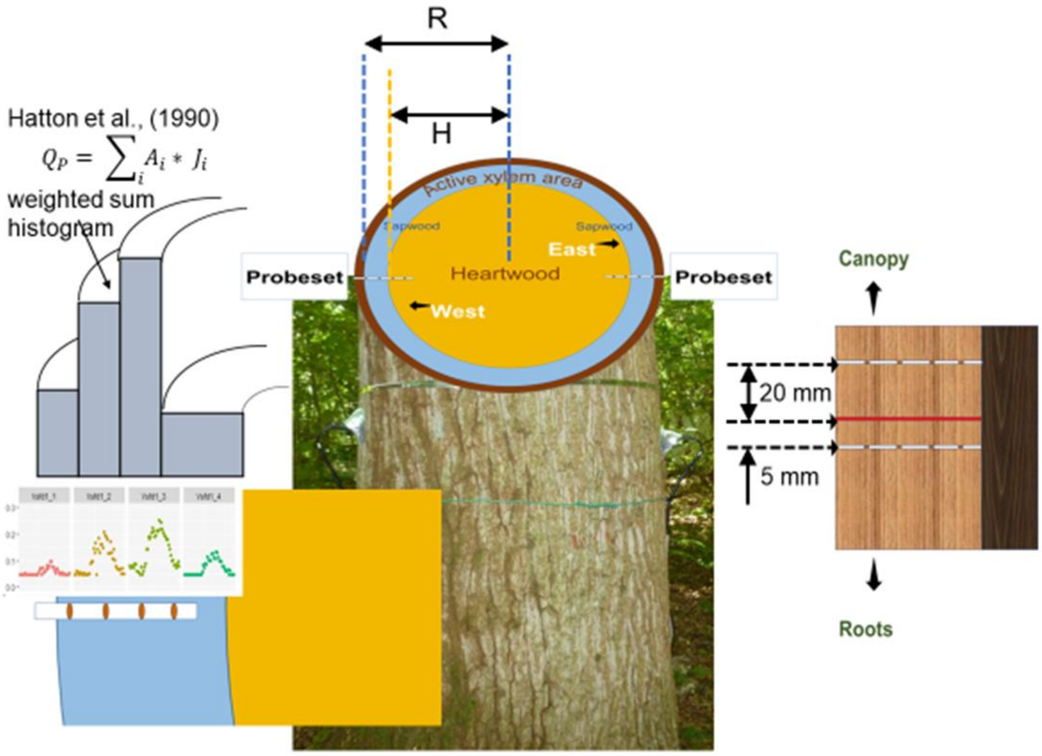

Figure 1: Showing sap probeset layout, spacing dimensions between probes and indicative illustration of Hatton et al., (1990) weighted sum histogram, where R (m) is the radius to the cambium and H (m) is the heartwood estimated radius, both at the probeset insertion height. All equations and variables also defined in Tables A1 and A2. Graphical insert is Figure C1(b).

Quality Assurance of raw HPC data

Commissioning and failure data were recorded for each probeset. This enabled a combination of data file amendment (especially for the earliest installations on separate loggers) and post capture filtering to eliminate periods of invalid data for each probeset.

Heat pulse to xylem sap flux calculations.

Figure C3 shows example positional (i.e. thermocouple-specific), point sap flux density data from four probesets in two trees. Data from the thermocouple radial position giving the highest diurnal values (one thermocouple position for each probeset) are selected from the four-position data and shown across a 24-hour period (Fig. C1(a)). Note the increase in sap flux density towards the centre of the sapwood, decreasing again towards the heartwood.

Converting point xylem sap flux data to whole tree water usage.

Using tree cambium radius (*R*) data, estimated heartwood radius (*H*) (0.05 m smaller than the inner sensor radial position), along with transducer radius positions (r_z), point sap flux density is converted to volumetric (half tree) total sap flux by using the integration of the point sap fluxes over the active sapwood conducting area. Output from this stage (Stage 5, Appendix Table A2 and Appendix C) gives a combined sap flux for each probeset (Fig. 3).

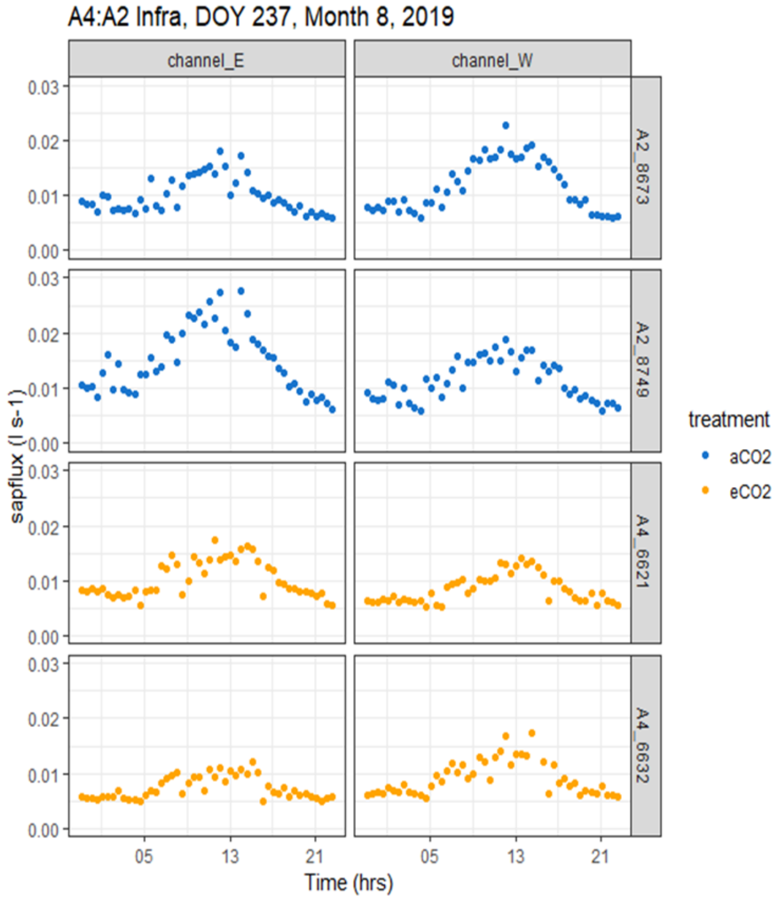

Figure 2: An example data visualisation from a sunny August day in 2019 showing output of Stage 5 combined point sap flux (litres s^{-1}) for four infrastructure trees with E facing (left) and W facing (right) probesets working: Array 4 (*eCO$_2$*), at top and A2 (*aCO$_2$*), at bottom. Time is in UTC.

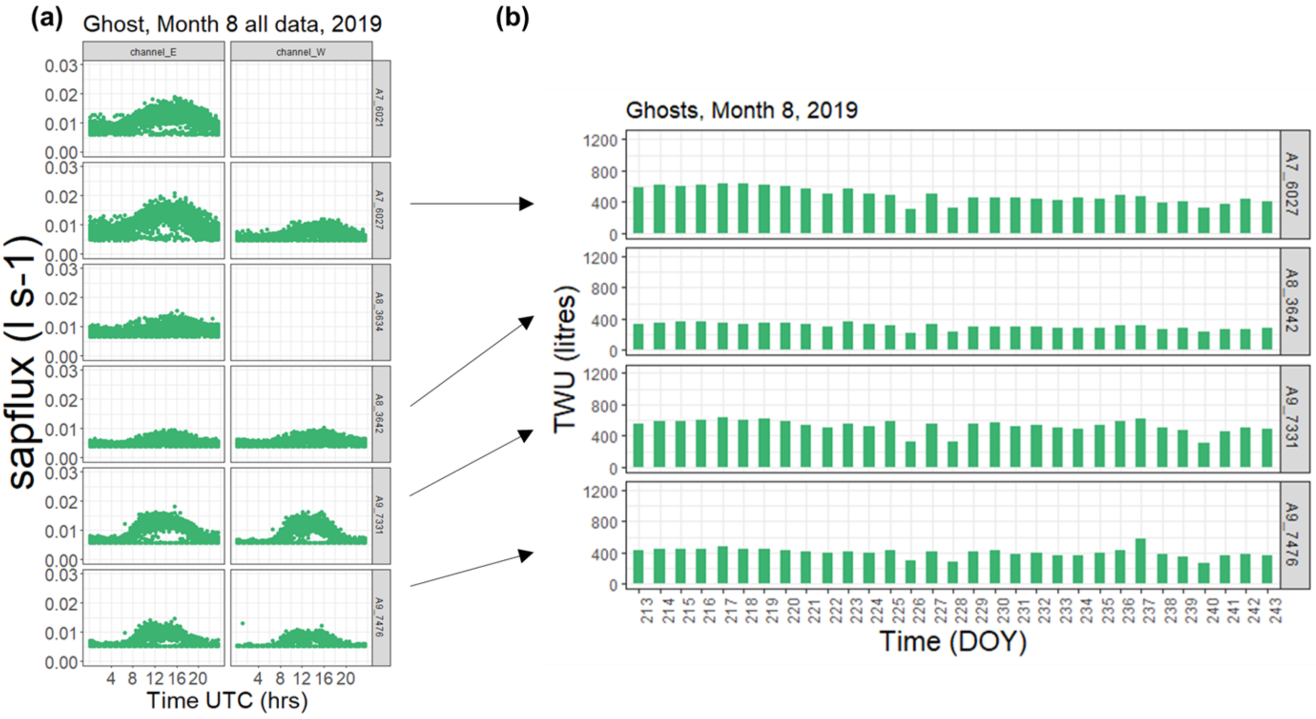

Figure 3: Example *Ghost* Array xylem sap responses in August 2019. (a) diel (24hour) tree sap flux for all days in August 2019 are superimposed. E (left) and W (right) facing probesets for six *Ghost* trees show circumferential imbalance in xylem flux. All data for the individual month is superimposed across time-of-day sampling (hours, UTC). Frequency of sampling is every 0.5 hrs. Faulty probeset positions are shown blank. (b) Example of accumulative daily diurnal water usage (TWU) per tree totalled for E and W facing probesets across month 8 2019 for four *Ghost* trees having both E and W probesets functioning with the other two *Ghost* trees omitted due to faulty probesets. Time DOY.

Diel sap flux patterns in August 2019 (before filtering to eliminate nocturnal data) are shown in Figure 4(a) as an example, for the *Ghost* arrays with East– and West–facing probesets in each column. The sap flux data still show minimum threshold levels (which vary by tree size) determined by the post heat pulse sampling period. Once again it is noticeable that there is often circumferential imbalance in xylem sap flux in the East (lefthand column of Fig. 4(a)) and West (righthand column of Fig. 4(a)) probeset position data, which reflect the asymmetry in growth ring width around the stem typical in these old growth trees. The blank panels represent faulty probes (in two *Ghost* trees) corrected by autumn 2019. To compare individual tree responses across the leaf-on seasons, further data filtering is required. We filter the half-tree sap flux parameters using the solar azimuth and solar radiation parameters captured from the FACE control instrumentation (solar azimuth > -6°, solar radiation > 0 W m^{-2}) to give just daylight (diurnal) data. Where both probesets in a tree are providing good data, a mean whole tree sap flux is then derived and accumulated into TWU (Fig. 4(b)) as we had sufficient tree data to not include results where probesets had failed. In future analysis we could use half-tree data once we understand the proportion of sap flux and TWU exhibited by each probeset (e.g. following failure of contact with sapwood of a previously functioning probeset).

The TWU data reported here compare well to results from other studies (Table S4: David et al., 2013; Sánchez-Pérez et al., 2008; Tatarinov et al., 2005; Baldocchi et al., 2001).

(modified Section 2.9 old LL410-411)

Data processing, visualization and analysis

… points for outliers are used. ANOVA models to test hypotheses use the functions *anova and summary.* Function *autoplot* from the *ggfortify* library is used to check the assumptions of normality of the residuals.

Results and Discussion (RC1, RC2)

(additional figures and tables)

ANOVA testing of hypotheses

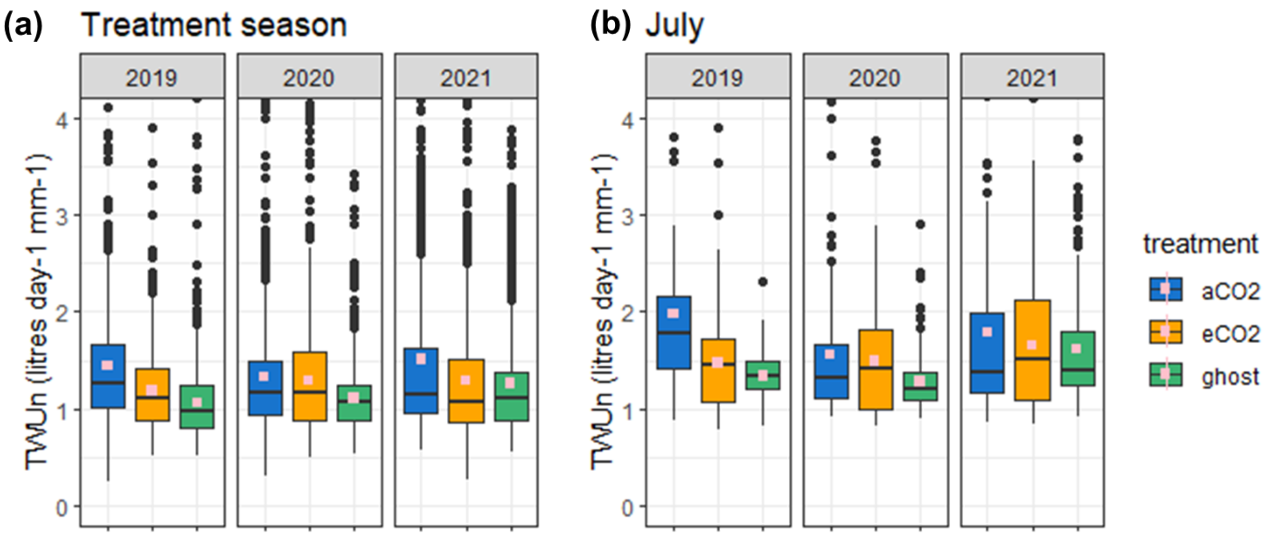

Figure 4: Treatment comparison of TWU. For years 2019- 2021 the TWU$_n$ (litres d-1 mm^{-1}) data is shown for the three treatment types. The distributions are shown as box and whisker plots showing median and interquartile range (IQR, 25%ile to 75%ile) with whiskers calculated as 1.5 x IQR from the hinge and points for outliers. Mean values, calculated from the entire range of data, are shown as spots (pink). (a) The season data April to October is combined for each year. (b) July for each year is shown.

	2019 **p value**	**2019** **%**	**2020** **p value**	**2020** **%**	**2021** **p value**	**2021** **%**
Season	< 0.001	-19%	p>0.05, actual value 0.079	-3%	<0.001	-13.9%
July only	< 0.001	-26%	p>0.05, actual value 0.37	-4.5%	p>0.05, actual value 0.19	-7.3%

Table 2: ANOVA model_CO$_2$ p-value and % difference summary for eCO2 TWU$_n$ compared with aCO2 TWU$_n$.

	2019 **p value**	**2019** **%**	**2020** **p value**	**2020** **%**	**2021** **p value**	**2021** **%**
Season	< 0.001	+37%	< 0.001	+20%	<0.001	+20%
July only	< 0.001	+48%	< 0.001	+22%	p>0.05, actual value 0.071	+9.9%

Table 3: ANOVA model_inf p-value and % difference summary for aCO2 TWU$_n$ compared with ghost TWU$_n$.

(revisions to old LL571-572)

... The seasonality of *TWU* and TWU_n are different in the two years, but both exhibit similar general responses to daylength within each treatment season.

(revisions to old LL643-651)

VWC at the start of each annual treatment season affects canopy usage during early leaf-on, but we have seen that reduction of soil moisture across leaf-on season appears not to affect total water usage. In our forest, as reported elsewhere from previous research, oak trees are most likely using deeper > 1 m depth water resources by a combination of hydraulic recharge from deep tap roots and capillarity from the perched water table. Our TWU results indicate that, other than on wet/ cloudy days, oaks do not diminish their xylem sap flux and water usage across the treatment season, or respond to depletion of shallow soil moisture, during very dry periods. For example, during the most pronounced continuous dry period of the observation period (June to July 2018, Fig. 12), there appears to be no inter-year difference in median *Ghost* tree diurnal sap flux (Fig. 6(a)) or trends in 95%ile sap flux (Fig. S5), or median TWU ((Fig. S6) in all trees, despite depletion of shallow soil moisture…

Appendix B: Limitations of the time-out characteristic and outliers

(revisions to old LL 743. Addition of a sentence)

The limitations of the time-out characteristic and the effect this places on HPC data are recorded in several references (e.g. see Tranzflo (New Zealand) manual). The limitations impact on our choice of data processing (e.g. diurnal versus diel) and feed through into the statistics we report. These limitations also introduce a truncation effect at lower heat velocities so that the distribution of the resulting raw and processed data is not symmetrical.

Appendix C: Details of xylem sap flow measurements and calculations

In each research array a datalogger and multiplexer (CR1000+AM25T, Campbell Scientific, Logan, Utah, USA) was used for year-round 24-hour capture of raw data from sap flux HPC probesets manufactured by Tranzflo (New Zealand), soil and throughfall measurement devices.

The logger was programmed for data capture using CRBasicEditor under LoggerNet (versions to 4.6.2), also by Campbell Scientific, Logan, Utah, USA. We tested our prototype installation set-up in mid-summer 2017 to determine if we could capture the expected range of heat velocities and applied similar capture programs to all array loggers.

Each target oak tree had two probesets, East and West facing using long (7 cm four-sensor) probes. Each probeset was inserted at a stem height between 1.1 and 1.3 m and contained a central heat pulse probe and two measurement probes (each containing four thermocouples for long probes respectively) upstream and downstream of the heater (Fig. 2). Transducers were positioned radially in the stem (to suit the ring-porous characteristics and bark thickness of old growth *Q.robur*). Each probeset was protected from natural heating by reflective insulation covers during the treatment season.

During monitoring, a heater pulse of duration 1.5 to 2.5 secs was applied half-hourly through a heater box (one per tree) to the heater probes. The pulse duration was dependant on the number of heaters pulsed simultaneously. A 2 second pulse was standard for the two oak per array (four long probeset) configuration. Each thermocouple pair in the upstream and downstream positions takes up to 330sec

(5.5 minutes) to reach a differential heat balance point and this time determines the minimum detectable heat velocity, for a time just within this timeout period. The thermocouple datalogger sampling rate of 0.5 secs determines the maximum detectable speed (minimum time-to-balance), which, given normal interference levels, is adequate for deriving maximum heat velocity. 16 differential thermocouple configurations are sampled per array in one 6 minute timeslot every 30 minutes, giving time-to-balance t_z data in seconds.

Data collection problems, due to logger earthing and sap probe misconnections at manufacture, caused data loss early in the project. Contact with sapwood was maintained for all oak trees from installation to March 2021, when two out of the 36 probesets failed.

Raw file processing

Logger data from the nine C1000 FACE research loggers were collated by array and transducer type (i.e. 7 cm probeset datasets for oaks only) using ‘R’, then combined into year files for further data processing.

Xylem sap flux calculations.

Following quality checks, each stage of calculation to produce wound-corrected sap velocity and sap flux density at each transducer position (four per probeset) was performed in stages (see Table A2). Table A2 lists the methodology and equations along with associated literature sources for each stage. At stage 3 (Table A2), the Green and Clothier (1988) polynomial factors were used for wound compensation. For stage 4, the conversion factor c1 was derived (Eq.(A4) and Eq.(A5)) to calculate xylem sap velocity from heat velocity in oakwood (Edwards and Warwick, 1984; Marshall, 1958). Measurement of wet and dry woodcores and microcores previously described provided data for derivation.

Figure C1 shows further details of positioning of peak sap flux through the sapwood in two trees. Figure C1(a) pools results from both trees. The diurnal maxima from the larger tree are larger than those for the smaller tree. Figure C1(b) pools probeset results from the larger tree, E facing (top) and W facing (bottom), illustrating stem imbalance.

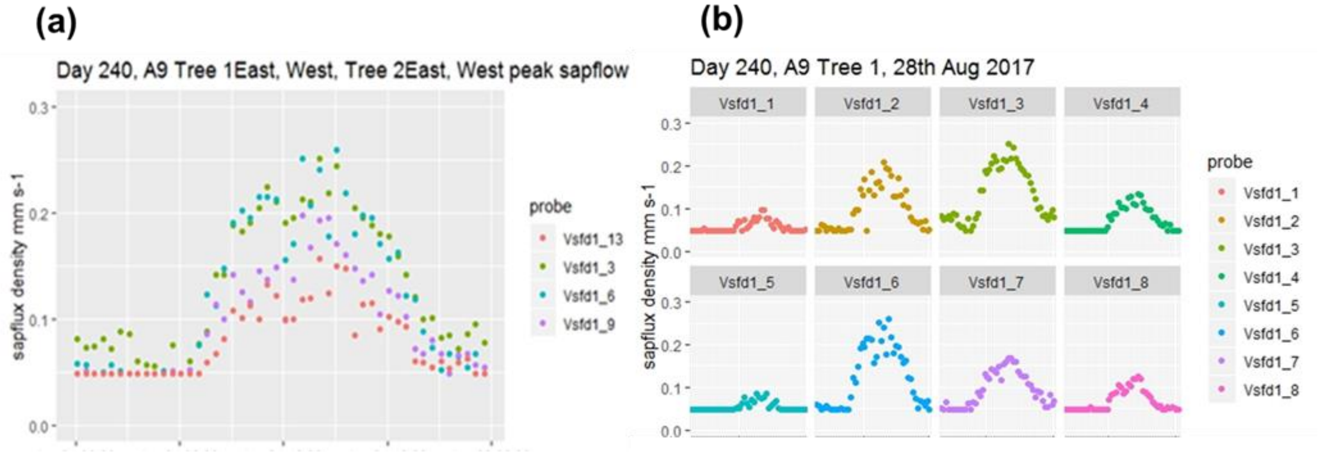

Figure C 1: a) Example Stage 4 output showing peak point sap flux density in two trees for one sunny day in August 2017. Tree 1 (vsfd1_3 and vsfd1_6) bark radius is larger than Tree 2 (vsfd1_9 and vsfd_13). b) Example Stage 4 output showing changes to point sap flux density across the active xylem for E facing

(top) and W facing (bottom) probesets of one tree (Tree 1) on the same day in August 2017. The lefthand probe position is nearest to the bark and the righthand probeset position is nearest to the heartwood. Note the peak value occurs at different sensor positions for the two probesets.

The nocturnal/ pre-dawn response for the smaller tree in 1(a) (vsfd1_9 and vsfd_13)) and the less vigorous thermocouple positions in the larger tree in Figure C1(b) (vsfd1_1, vsfd_4, vsfd1_5 and vsfd_8) have their minima determined by the previously mentioned time-out limit (i.e. t_z of 330 secs). These minima do not affect the processing of diurnal values but influence nocturnal value accuracy of the lowest point sap flux density (see Appendix B). The radial pattern of sap flux density increases in amplitude to a peak position within the probeset measurement zone and then decreases again towards the heartwood boundary as depth from the cambium increases (Fig. C1(a) and (b)), a characteristic of these ring porous oak species. The radial amplitude patterns vary across seasons.

Converting point xylem sap flux data to whole tree water usage.

An adapted simple integration method (Hatton et al. 1990), based on a weighted average approach was used where the point sap flux density is weighted by the areas of the annular rings associated with each r_z. Fig. C1. Hatton et al. (1990) consider their method, in comparison with alternatives (e.g. fitting a least-squares polynomial), a simpler and more accurate approach for estimation of the volume flux.

(end Appendix C)