

## ***Response to Reviewer #2***

We thank Prof. Stocker for his comments, which are addressed as explained below. The Referee's text is reported in *Italic* and our responses in roman.

5

*This paper contrasts predictions of contrasting models of ecosystem transpiration and assimilation during rain-free periods ("dry-down events") under different levels of elevated atmospheric CO<sub>2</sub> (eCO<sub>2</sub>), modulated also by simultaneous changes in temperature and relative humidity of the ambient air. The models are formulated in a simplified form to find analytical solutions, while maintaining some essential feedbacks and resolving principles that are founded on observations and*

10 *understanding of ecosystem mechanisms. This is a different approach compared to the approach taken in global vegetation models and the land components of Earth System Models. The advantage of reduced-form approaches as presented here is that emerging ecosystem behaviour can be linked directly to assumptions and mathematical properties of the model, and thus offer insights into "what matters most".*

15 Indeed, this is our main goal—capturing with closed form solutions the essential relations between climatic and soil conditions, and gas exchange rates.

*In my understanding, the most important result of the present study is that, irrespective of the chosen model, eCO<sub>2</sub>-driven acclimation of leaf-level physiology and ecosystem-level structure (morphology) interact in such a way that a reduction of*

20 *leaf-level transpiration is compensated by increased ecosystem foliage area (L) to fully exploit a constrained (limiting) resource - here water (Abstract l. 29 and Conclusions l., 736). This is a useful insight for understanding observed vegetation greening and streamflow trends in water-limited regions of the Earth and it is interesting that this is a consistent prediction that appears not to be subject to model formulation. I think the paper by Manzoni et al. could be a useful contribution to the literature if the authors manage to convincingly address a few major issues that I would like to raise in the following. I*

25 *should disclaim that I did not verify the (extensive) algebra presented in the manuscript. Please excuse me for the limited time I can invest in this review.*

We thank Prof. Stocker for his support and constructive criticism, which is addressed below. Our conclusions support the hypothesis that plants allocate resources to exploit limiting water. Another important—albeit methodological rather than

30 conceptual—result is that a full solution (i.e., including solving for the marginal water use efficiency) of the stomatal optimization is required to recover the observed effect of elevated atmospheric CO<sub>2</sub> concentration on gas exchange. This

finding resolves a long-standing controversy on the use and implementation of stomatal optimization models (as also noted by the reviewer in point (2) below).

35 MAJOR

(1) While the interaction of physiological and structural adjustments are presented as a key conclusion, it is unclear to what extent this is a model prediction or an assumption. Apparently, changes in  $L$  are prescribed to increase under  $eCO_2$ . This seems to imply that this interaction is not founded on first principles but is a direct reflection of an assumption that is built  
40 into all models (although admittedly an assumption with demonstrated empirical support, Fig. 3). Similarly, the conclusion that vegetation physiology and structure adjusts in such a way as to fully exploit the limiting resource, seems to be an assumption, rather than a prediction. The Cowan & Farquhar (1977) approach to predicting physiological responses subject to  $A-\lambda E$  maximisation starts with the presumption of a constrained amount of plant-accessible water and therefore has to predict that this amount of water be transpired over the course of a dry-down (“imposing the condition that all  
45 available water is used by the end of the dry period”, l. 670).

Regarding leaf area changes, they are assumed to be a function of atmospheric  $CO_2$  concentration for all the models. In this sense, the models do not predict changes in leaf area, but changes in gas exchange after accounting for prescribed variations in leaf area. The rationale for this choice is that leaf area varies over time scales longer than those typical of stomatal  
50 responses to environmental fluctuations (including soil moisture), so that we can ‘separate the temporal scales’ of the two problems—stomatal regulation (a variable to be solved for) and leaf area adjustment (assumed). It is also correct that we assumed that plants consume soil available water during a ‘typical’ dry period. (We comment below in the response to another comment about plants ‘knowing’ how long a dry period is.) This assumption sets only a boundary for the optimization problem, but does not offer clues on the trajectory of stomatal conductance during the dry period—that is the  
55 actual solution of the optimization problem. As it turns out, stomatal closure during a dry period occurs only when transpiration becomes water supply limited, otherwise a time-invariant soil moisture throughout the dry period represent the optimal trajectory. This is also the only trajectory that maximizes the cumulative net assimilation over that period. These trajectories (time-invariant or decreasing stomatal conductance) are not assumed, but are the result of the optimization problem solution.

60

Similarly for the PETA model (as I understand Donohue et al., 2013 - please clarify if not the same principles are applied in Donohue et al., 2017): Foliage area changes are predicted subject to “known” changes in leaf-level water use efficiency and a constrained amount of available water (corresponding to precipitation). In other words, all available water is assumed to be consumed. (But then, I must be misunderstanding something here, given that  $L$  changes are prescribed also in  
65 the PETA model here.) A clarification of these points would help to better understand the relevance of assumptions and the

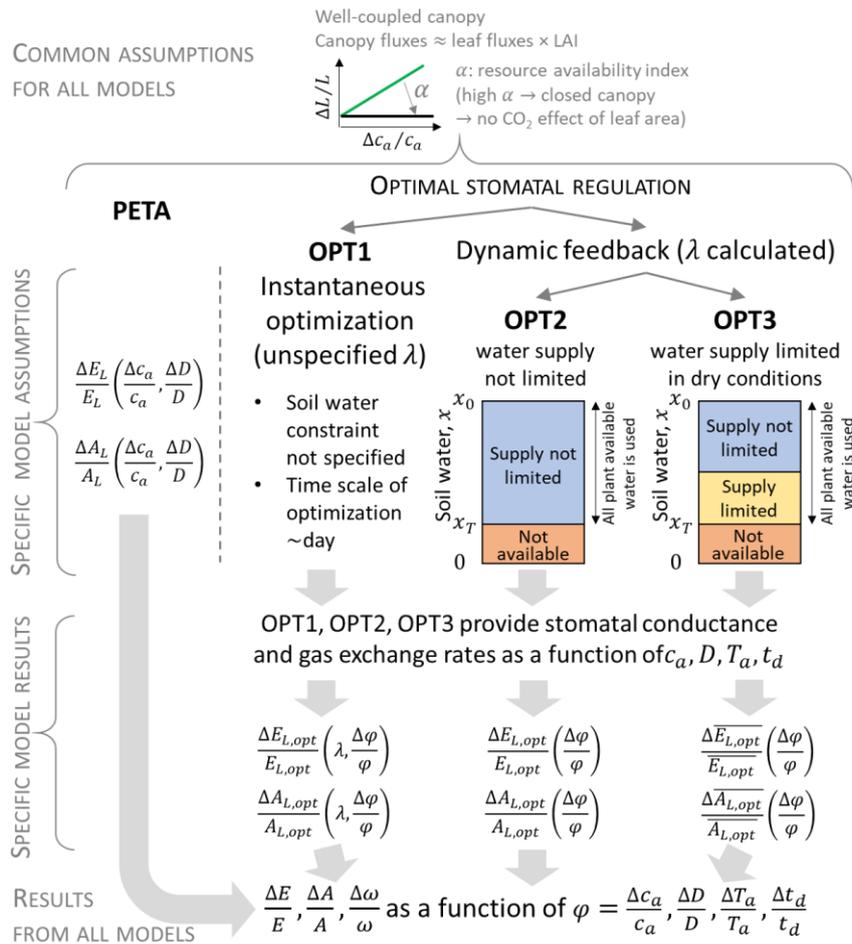
*purely predictive ability of the model (and therefore what can defensibly be presented as a conclusion from the present study).*

70 Also according to our interpretation, in the PETA model leaf area changes depend on how leaf-level water use efficiency varies with atmospheric CO<sub>2</sub> concentration and vapor pressure deficit (VPD). However, there are some differences between the approaches by Donohue et al. (2013) and Donohue et al. (2017). In the earlier paper, long-term canopy transpiration was assumed to be constrained by a fixed amount of precipitation, which implies  $dE_l/E_l = -dL/L$  (using the notation in Table 1 of the submitted manuscript;  $E_l$ : leaf-level transpiration rate,  $L$ : leaf area index). With this constraint, leaf area changes can be directly related to net assimilation, VPD, and atmospheric CO<sub>2</sub> concentration changes, which allows partitioning 75 variations in leaf area among the different climatic drivers. In the later paper, the resource availability index  $\alpha$  is introduced, and leaf area is expressed as an empirical function of  $\alpha$  without a specific constraint on canopy transpiration. In fact, their Figure 1 shows that canopy transpiration varies mildly with  $\alpha$  (Donohue et al., 2017). This is the PETA formulation we used; therefore, also in our contribution canopy transpiration is not assumed to be fixed (e.g., see variations in canopy transpiration with increasing atmospheric CO<sub>2</sub> in our Figure 5b).

80 We agree with the reviewer that the differences between model assumptions vs. predictions were not presented clearly and propose to revise the manuscript in the following ways:

- Improved Figure 1, where we can spell out more clearly what are the assumptions, and what are the results of each model (Figure R1).
- Added clarifications in Section 2.2 where describing the PETA model: “Equation (8) also shows that canopy 85 transpiration can vary unless both leaf-level transpiration and leaf area index are constant. This result of the PETA model differs from a key assumption of the stomatal optimization model (Sections 2.3.2 and 2.3.3).” and in Section 2.3.2 where describing the solution of the stomatal optimization problem: “It is important to emphasize that this specific stomatal conductance trajectory is not a result of our assumption that all available water is used—it is rather the solution that best balances the water consumption rate over time to maximize net assimilation.”

90



**Figure R1. Revised conceptual representation of the models used to assess gas exchange responses.**

(2) Another potentially useful insight of the present study is that realistic stomatal responses to  $e\text{CO}_2$  can be predicted only with stomatal optimality models with a “dynamic feedback” (OPT2 and OPT3), but not with an “instantaneous optimality” model (OPT1). In the models investigated here, this “instantaneous optimality”-based prediction follows from a constant  $\lambda$ , and given a constrained amount of transpirable water. This result is then used to argue that OPT1 (representing in general an “instantaneous optimality” model) is not realistic. This could be misunderstood as a demonstration of a general uselessness of similar “instantaneous optimality”-based models (e.g., Prentice et al., 2014; Sperry et al., 2016; Wolf et al., 2016). However, not all “instantaneous optimality” models are based on optimising C assimilation for a constrained amount of transpiration, and stomatal conductance is (in line with observations) predicted by several “instantaneous optimality” models to decline with rising  $\text{CO}_2$  (Stocker et al., 2020, see their Eq. C1; Joshi et al., 2021 Fig. 2). Hence, the presentation of instantaneous vs. dynamic feedback optimality models runs the danger of creating a straw-man argument. A clarification

and intuitive explanation of why stomatal conductance is predicted to increase by OPT1 but not in other “instantaneous optimality”-based models, seems needed.

We totally agree. There is value in instantaneous optimization approaches (which should perhaps be called ‘instantaneous maximization’ since no optimal trajectories are determined), though it might be important to remind readers that in general instantaneous optimization does not guarantee that organism fitness is maximized over a specified time frame (dry down as in our case or more in general life span). In other words, using resources now at very high rate might maximize gains today, but result in stress and possibly higher mortality tomorrow. The problem is even more complicated when competition among mutants or species is considered—but even in that case a ‘local’ maximization does not guarantee an optimal (or evolutionary stable) outcome. Yet, models defining instantaneous gains and costs and maximizing the difference between these two terms can be highly effective and could represent a pragmatic solution for vegetation modelling (though not an ‘optimal’ one in a mathematical sense).

If encouraged to revise our manuscript, we can provide a more balance comparison with other approaches (including acknowledgement of their value), starting by clearly stating in the Discussion (Section 4.4) that: “For example, the optimization model based on Prentice et al. (2014) correctly predicts the observed short-term decrease in stomatal conductance under elevated atmospheric CO<sub>2</sub> (Eq. (C1) in Stocker et al., 2020) without invoking leaf area adjustments.” Moreover, we can be more comprehensive in the Introduction, referring to synthesis papers where optimization models are compared and including a statement to explain that in this manuscript we focus on a specific type of optimization-based models—those formulated as an optimal control problem (see our response to a minor comment below).

Regarding an intuitive explanation of the counterintuitive optimal response of stomatal to elevated CO<sub>2</sub> concentration with the OPT1 formulation, we can explain in these terms (Discussion Section 4.4): “These erroneous responses arise because at low CO<sub>2</sub> concentration a small increase in stomatal conductance results in large net CO<sub>2</sub> assimilation gains compared to the higher water losses, resulting in the counterintuitive opening of stomata as atmospheric CO<sub>2</sub> concentration is increased.”

(3) *The discussion of the investigated models in the context of the extensive literature on other modelling approaches to simulating physiology in response to soil moisture dry-downs is relatively slim. The single statement referring to such alternatives on l. 680 (“While these approaches are more physiologically accurate and their predictions compare well with observed trends, they do not guarantee that the water use is optimal over the whole optimization period.”) does not do it justice in my view. I want to avoid a more fundamental debate over the “constrained water” assumption of Cowan & Farquhar (1977) (How can a plant know in advance how long the current dry-down will last? How can it know how to optimally make use of available water from now until the [future] end of the dry-down? Why wouldn’t it be advantageous for a competitor to consume water immediately rather than save it for the future?), but the justification of not discussing alternative modelling approaches by stating that “they do not guarantee that the water use is optimal over the whole optimization period” seems unfair - particularly in view of the argument that I want to avoid getting into ;-).*

We see the point of the reviewer, and as also mentioned above, we recognize that a more balanced discussion is warranted.  
140 This is entirely feasible in the Discussion without altering significantly the structure, mostly expanding Section 4.4.

Regarding the fundamental question of the time scale over which plants can be expected to maximize carbon gains, we can be more open in a revised manuscript. We assumed that this time scale corresponds to an ‘average’ dry down duration, which can be criticised. Assuming that plants maximize carbon gains instantaneously is equally problematic in our view, because it implies that plants ‘know’ that saving resources today is not particularly useful (whereas it might be). In a way,  
145 with all optimization or maximization approaches, we are trying to reverse-engineering how evolution programmed plants to respond to environmental changes. Plants cannot foresee the future, but it is fair to assume that their responses are tuned to the environmental where they live, which—as a first order approximation—can be represented by the average conditions experienced over several generations. On these grounds, a dry down duration or the expectation that net gain maximization today will not harm the plant tomorrow are equally strong—and equally reasonable—assumptions. Depending on  
150 environmental conditions and competition patterns, one or the other might prove more useful for prediction.

*(4) The present manuscript is heavy on algebra. I understand that this is central to the reasoning of the presented analysis, but I recommend that all efforts be made that this manuscript can be read and its reasoning intuitively understood without deciphering the algebra. In general, reasonable efforts should be made to reduce the algebra, possibly relegating parts to  
155 the Appendix, while still maintaining the essential descriptions. Sorry that my point here is not more specific, but I recommend that the presentation of the science be presented to appeal to the widest possible audience.*

The methodological section with heavier mathematical derivations is Section 2.3“Optimal stomatal control models”. We can move most of the material from that section to an Appendix, to leave a streamlined text only presenting the results in an  
160 intuitive way. Following this approach, we would leave the equations for optimal stomatal conductance in the main text, together with their explanations; the actual mathematical derivations would become Appendix B. Figure 2 would remain in the main text as it illustrates the meaning of the optimal stomatal conductance solution. We would leave instead Sections 2.1 and 2.2 as they are, since they provide basic equations or final results (the latter for the PETA model). With these changes, only five equations would remain in Section 2.3, facilitating the reading of the Methods for a wide audience.

165

*MINOR*

*The specific scientific question and scope of the manuscript and the model investigation is not immediately clear. The last sentence of the abstract points to the essence being the coordination of physiology and morphology in their response to  
170 eCO<sub>2</sub>. Then, the question is stated more precisely as “... but it is not clear if and under which conditions these two effects balance out.” (l. 44). Is this the central question? Does the paper answer this question? If so, could an answer to that*

*question be given more clearly in the Conclusion section? The introduction shifts attention to other questions (l. 65, l. 105-107) and answers to these make up much of the Conclusions section instead. This comes at the cost of a not so well-defined scope overall.*

175 The ambiguity probably derived from the double scope of this contribution. On the one hand, we highlight some implications of the optimization model that have been previously overlooked (a methodological issue); on the other hand—with both PETA and optimization models—we address the scientific question of how plants respond to elevated atmospheric CO<sub>2</sub>. Both aspects are relevant. In the submitted abstract we state our central question (formulated as a research gap): “The net effect of elevated CO<sub>2</sub> on leaf- and canopy-level gas exchange thus remains unclear.” The concluding sentence of the abstract answers this question: “coordination of physiological and morphological characteristics in vegetation to maximize resource use (here water) under altered atmospheric conditions.” The same question is brought up at the beginning of the Introduction: “This increase in the canopy-level evaporating surface area could counterbalance the reduction in transpiration caused by stomatal closure at the leaf level, but it is not clear if and under which conditions these two effects balance out.” However, we agree with the reviewer that later in the Introductions the questions seem to have changed. We propose to rephrase them as follows, including a statement of our further methodological aim to compare model predictions and discuss the limitations of the PETA and optimization models:

- 185 “1. How do physiological (stomatal conductance) and morphological (leaf area) adjustments combine to determine leaf and canopy gas exchange rates under atmospheric CO<sub>2</sub> concentrations?
2. How do physiological and morphological adjustments determine gas exchange responses to combined changes in CO<sub>2</sub> concentration and atmospheric or soil drought?

190 By comparing the predictions of the PETA and optimization models, we provide a theoretical perspective on these questions, while also identifying advantages and limitations in these different modelling approaches.”

In the conclusions, we can more clearly answer these questions: “Both models predict low sensitivity of canopy transpiration rates to a changing climate, indicating that morphological adjustments (leaf area increase) compensate physiological adjustments (stomatal closure).” The very last sentence in the Conclusions re-states the same message in slightly different terms: “Overall, these results imply that physiological and morphological traits acclimate to changing environmental conditions in a coordinated manner to ensure that limiting resources such as water are used efficiently.” We hope that with these changes, the overall scope is clearer.

200 *The different model variants could be better linked with specific hypotheses about controls and mechanisms determining stomatal responses to eCO<sub>2</sub>. Are there specific questions to be answered by comparing predictions from the different models?*

Perhaps the main conceptual difference regarding CO<sub>2</sub> responses is between optimization model OPT1 and the other two variants, OPT2 and OPT3. The former should allow capturing short-term responses for fixed  $\lambda$  (but it does not, as discussed in the manuscript), whereas the latter two should be able to capture long term responses that include morphological changes

205

(because  $\lambda$  varies with atmospheric CO<sub>2</sub> via changes in leaf area). The differences between these model variants are presented in Fig. 3 and commented upon in the Discussion. Due to the nature of the optimization approach—in practice a way to avoid a description of detailed physiological processes thanks to an ecological ‘goal’ function—it is difficult to link these model variants to specific mechanistic hypotheses, so we do not have a clear answer to this comment.

210

*l. 39 “stimulates plant growth and thus increases leaf area”: Is increased leaf area a consequence of stimulated growth?*

Yes, that seems to be the case. Higher net assimilation provides resources for faster growth, and hence also higher leaf area (assuming resources are invested both above and belowground).

215 *l. 41 “open canopies”: A dependence of the eCO<sub>2</sub> effect on leaf area subject to initial leaf area (open canopy) is mentioned throughout the manuscript. In view of canopies being open due to water limitation, nutrient limitations, low temperatures, or simply due to young age, is often not specified, but may be relevant for responses and certainly for underlying mechanisms. Could references to “open canopies” be made more specific throughout?*

This is a good point, also raised by Reviewer #2 in relation to the meaning of the resource availability index  $\alpha$ . Essentially, 220 in the PETA formulation by Donohue et al. (2017),  $\alpha$  represents how much an ecosystem can increase its leaf area index in response to elevated atmospheric CO<sub>2</sub>. As the reviewer points out, why a canopy is open is important to understand its potential leaf area changes, and to interpret our results (though results are based on a fixed  $\alpha$  value, and so are not affected by how we define  $\alpha$ ). To clarify, we propose to interpret  $\alpha$  as a measure of forest age ( $\alpha$  increasing with age) or canopy status with respect to the maximum leaf area index expected for a site ( $\alpha$  decreasing if the canopy is close to its maximum LAI). 225 Clarifications to this regard can be added in Section 2.2: “This index represents how far vegetation is from the maximum  $L$  expected for that location—high  $\alpha$  indicates an old stand or in general a stand with  $L$  close to the maximum, where additional leaf area increases are not possible (see also Sect. **Error! Reference source not found.**)”

*l. 73-74: “The model is based ...” Add: ... and the assumption that vegetation in water-limited regions makes full use of a 230 constrained flux of water (~precipitation).*

Only the 2013 formulation of the PETA hypothesis relies on this assumption, and we use the 2017 version, so it might be confusing to include this assumption here. Please see also our response to comment (1) above.

*l. 83: “Stomatal optimization” models are referred to as models relying on the “Lagrange multiplier”  $\lambda$ . This seems to be a 235 too narrow definition of “stomatal optimization”. In my understanding, models that predict stomatal responses to changes along the soil-plant-atmosphere continuum may be considered here too (e.g., Sperry et al., 2016; Wolf et al., 2016).*

To address this comment, we can cite other examples, even though they do not perform a formal optimization through time. To avoid a long list of references, while also clarifying our focus, we would add in the Introduction: “Among the numerous

240 optimization-based models available (Mencuccini et al., 2019; Wang et al., 2020, and references therein), we focus here on those formulated as an optimal control problem in which stomatal conductance is solved through time.”

*Table 1: Confusing use of ‘T’ in  $T_a$  and  $T_d$ , while the two ‘T’ are different variables with different units.  $E_{SR}$  not explained.*

245 We can change the symbol used for the dry period duration to  $t_d$ ; we apologize for the confusion on the symbol  $E_{SR}$ , which appeared by mistake as it was changed (not everywhere apparently) to  $E_w$ .

*l. 147: Spell out that  $c_i:c_a$  is assumed to remain constant under  $eCO_2$ .*

250 This is only an approximation for the denominator of the  $A-c_i$  curve, not an assumption. In other words, the  $A-c_i$  curve is linearized, without losing the dependence of  $c_i:c_a$  on atmospheric  $CO_2$  concentration (Eq. (10) in Katul et al., 2010). A clarification can be added by explaining: “As a result,  $A_L$  is a linear function of  $c_i$ , but the slope of the relation decreases with increasing atmospheric  $CO_2$  concentration; moreover, this approximation allows retaining variations in  $c_i/c_a$  with  $c_a$  (Katul et al., 2010).”

255 *l. 150: “ $A_L$  is a linear function of  $c_i$  but with a declining slope at high  $CO_2$  concentration”. This seems to be a contradiction in itself. Either it’s linear or has a varying slope.*

The slope of the linearized  $A-c_i$  curve depends on  $c_a$ , so it can change as atmospheric  $CO_2$  increases, but this was not clear in the way we had formulated the sentence. Combining this and the previous comment, we propose to amend this sentence as: “As a result,  $A_L$  is a linear function of  $c_i$ , but the slope of the relation decreases with increasing atmospheric  $CO_2$  concentration; moreover, this approximation allows retaining variations in  $c_i/c_a$  with  $c_a$  (Katul et al., 2010).”

260

## References

- 265 Donohue, R. J., Roderick, M. L., McVicar, T. R., and Farquhar, G. D.: Impact of CO<sub>2</sub> fertilization on maximum foliage cover across the globe's warm, arid environments, *Geophys. Res. Lett.*, 40, 3031–3035, <https://doi.org/10.1002/grl.50563>, 2013.
- Donohue, R. J., Roderick, M. L., McVicar, T. R., and Yang, Y. T.: A simple hypothesis of how leaf and canopy-level transpiration and assimilation respond to elevated CO<sub>2</sub> reveals distinct response patterns between disturbed and undisturbed vegetation, *J. Geophys. Res.-Biogeosciences*, 122, 168–184, <https://doi.org/10.1002/2016jg003505>, 2017.
- 270 Joshi, J., Stocker, B. D., Hofhansl, F., Zhou, S., Dieckmann, U., and Prentice, I. C.: Towards a unified theory of plant photosynthesis and hydraulics, *bioRxiv*, 2020.12.17.423132, <https://doi.org/10.1101/2020.12.17.423132>, 2021.
- Katul, G., Manzoni, S., Palmroth, S., and Oren, R.: A stomatal optimization theory to describe the effects of atmospheric CO<sub>2</sub> on leaf photosynthesis and transpiration, *Ann. Bot.*, 105, 431–442, 2010.
- Mencuccini, M., Manzoni, S., and Christoffersen, B.: Modelling water fluxes in plants: from tissues to biosphere, *New Phytol.*, 222, 1207–1222, <https://doi.org/10.1111/nph.15681>, 2019.
- 275 Prentice, I. C., Dong, N., Gleason, S. M., Maire, V., and Wright, I. J.: Balancing the costs of carbon gain and water transport: testing a new theoretical framework for plant functional ecology, *Ecol. Lett.*, 17, 82–91, <https://doi.org/10.1111/ele.12211>, 2014.
- 280 Stocker, B., Wang, H., Smith, N., Harrison, S., Keenan, T., Sandoval, D., Davis, T., and Prentice, I.: P-model v1.0: an optimality -based light use efficiency model for simulating ecosystem gross primary production, *Geosci. MODEL Dev.*, 13, 1545–1581, <https://doi.org/10.5194/gmd-13-1545-2020>, 2020.
- Wang, Y., Sperry, J. S., Anderegg, W. R. L., Venturas, M. D., and Trugman, A. T.: A theoretical and empirical assessment of stomatal optimization modeling, *New Phytol.*, n/a, <https://doi.org/10.1111/nph.16572>, 2020.