

1 **Interaction of drought and frost in tree ecophysiology:**  
2 **rethinking the timing of risks.**

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14 GC, NMSP and HD developed the ideas presented in this viewpoint piece and wrote the  
15 manuscript, with contributions from CD, ND, HH and JMTR.

16  
17 **Conflict of interest disclosure**

18 The authors of this preprint declare that they have no financial conflict of interest with the  
19 content of this article. GC and NMSP are PCI FWS recommenders.

20  
21 **Keywords:** Abiotic stress, Acclimation, Carbon availability, Drought, Frost, Phenology, Risk  
22 assessment, Tree ecophysiology, Water balance

23 **Abstract (332 words)**

24 In temperate, boreal and alpine areas, The the edges of plant distribution are strongly affected  
25 mainly defined by abiotic constraints. For example, in temperate, boreal and alpine area, heat  
26 waves and drought are major constraints at low latitude and elevation whereas while cold and  
27 frost are key factors at high latitude and elevation. Over the next few decades, climate variability  
28 is expected to increase, enhancing the probability of extreme events and thus therefore, the  
29 potential stress imposed by abiotic constraints. Moreover, the probability likelihood of co-  
30 occurring and successive constraints, such as drought and frost, could increase in parallel. It is  
31 likely that an initial exposure to a first constraint would affect the vulnerability to a subsequent  
32 one. Three integrative physiological processes, namely water status, carbon status and the  
33 timing of phenological stages, are crucial to understanding maintain how trees will respond  
34 survival when facing to these abiotic constraints stress factors. Although these processes have  
35 largely been extensively studied alone, in response to a single constraint, their interaction has  
36 rarely been investigated. In this paper, we explored have examined how plant water and carbon  
37 status interact with the growth cycle and affect both the vulnerability and the exposure to  
38 climatic constraints via two different focuses: (i) How would the interaction of drought and  
39 frost and drought constraints modulate the vulnerability to a subsequent constraint? (ii) How  
40 vulnerability to a given constraint and phenology interact? In the light of numerous papers, we  
41 suggest that the interaction between frost and drought constraints should in the short-term  
42 influence water status and, in the longer term, the carbon status, both consequently affecting  
43 further vulnerability, potentially leading to a decline. This vulnerability can also be modulated  
44 by a shift in the ~~However,~~ annual phenological cycle induced could also be affected by a  
45 previous ~~constraint exposure and therefore modulate this direct mechanistic response.~~  
46 Furthermore, we have identified significant gaps of knowledge in the ecophysiological tree  
47 response to interacting stresses and therefore propose a framework for building a new  
48 generation of process-based models through three focus based on three major points:- (i) the

49 spatio-temporal variation in carbon ~~biomass~~hydrate composition, fluxes and allocation in  
50 relation to environmental drivers, (ii) the spatio-temporal variation in water content, water and  
51 osmotic potentials, (iii) the modulation of phenological processes in response to  
52 stress~~description of realistic organ temperature in relation to environment and phase transition.~~  
53 This framework can help to improve the current process based models and to identify  
54 interactions that needs to be better described in order to obtain ~~Improving the descriptive range~~  
55 ~~of these interrelations would subsequently allow a more~~ quantitative and dynamic ~~description~~  
56 view of drought and frost ~~resistance~~ vulnerabilities integrating the life history of the  
57 ~~tree~~individual plant.

58

## 59 Introduction

60 Abiotic (*e.g.* temperature, water, light, nutrients) and biotic factors (*e.g.* intra and  
61 interspecific competition, predation, parasitism) are the major drivers of plant distribution and,  
62 therefore, evolutionary change (Erwin 2009; Klanderud *et al.*, 2015). Although biotic factors  
63 drive the population dynamics in the ~~core~~entire range, abiotic factors ~~are more~~can be highly  
64 relevant at the rear and leading edges of the distribution (Hampe & Petit, 2005). Within the  
65 boreal, alpine, temperate and Mediterranean areas, plants are likely to be exposed to both  
66 drought and frost stress, although it may not happen every year. At the rear edge, drought stress  
67 is one of the critical abiotic factors (*e.g.* at low latitude and elevation in Europe; Loehle, 1998;  
68 Lines *et al.*, 2010; Ruiz-Benito *et al.*, 2013). At the leading edge (high latitude and elevation),  
69 cold and freezing temperatures usually limit the plant distribution (Gusta *et al.*, 1983; Körner,  
70 1998; Larcher, 2005; Charrier *et al.*, 2013a). Along a latitudinal gradient within the niche of a  
71 species<sup>2</sup>-niche, drought and frost constraints are clearly ~~divided-separated~~, *i.e.* episodes of mild  
72 or severe drought events are observed at-on the rear edge during the summer, and frost stress ~~at~~  
73 on the leading edge during the winter. This has led to define the autoecological approach to  
74 predict the distribution of plant species within frost-~~exposed~~ areas (boreal, alpine, temperate  
75 and Mediterranean areas), considering individual limitations, such as minimum temperature at  
76 the leading edge or water deficit at the rear edge (Guisan & Thuillier, 2005; Cheaib *et al.*, 2012).  
77 However, drought and frost can co-occur at high elevation when, late in the winter, air  
78 temperature increases while soil is still frozen, inducing the so-called winter drought  
79 (Tranquillini, 1979; Mayr & Charra-Vaskou, 2007; Charrier *et al.*, 2017). An alternative  
80 approach is thus required for taking the different involved processes into account, and, more  
81 importantly, for addressing their interaction to improve the actual species distribution models.

82 This is even more crucial since dramatic changes in climate are likely to increase exposure  
83 to abiotic constraints over all biomes (IPCC, 2012). Mean surface temperature increased at a  
84 rate of 0.2°C per decade over the last decades, leading to species migration at higher latitudes

85 and elevation (Parmesan & Yohe, 2003; Lenoir *et al.*, 2008). Ecosystem functioning is likely  
86 to be affected by these changes, however, through contradictory effects. On the one hand,  
87 increased productivity is expected in relation to longer growing season (early flushing dates and  
88 delayed senescence; Schaber & Badeck, 2005; Menzel *et al.*, 2006; Fu *et al.*, 2014; Keenan &  
89 Richardson, 2015). However, on the other hand, a substantial deterioration in various  
90 physiological processes is expected to be induced by *e.g.* an insufficient winter chilling that  
91 would alter plant development (Chuine *et al.*, 2016; Delpierre *et al.*, 2016), or increased climatic  
92 hazards such as drought (Porporato *et al.*, 2004; Allen *et al.*, 2010; Carnicer *et al.*, 2012) or late  
93 frosts events (Leinonen & ~~Hanninen~~Hänninen, 2002; Augspurger, 2013).

94 The extreme climatic events (*e.g.* drought, frost, heat spell and storm) play a major role in  
95 the distribution, survival and, therefore, in the adaptation of plants (Chapin *et al.*, 1993; van  
96 Peer *et al.*, 2004), as well as in the productivity and the ~~biodiversity~~composition of  
97 communities (Knapp *et al.*, 2002). The expected increase in climate variability will inevitably  
98 lead to an increase in the frequency, intensity and duration of extreme events (IPCC, 2012;  
99 Rummukainen, 2012; Seneviratne *et al.*, 2012). If the return period of two extreme events is  
100 divided by two (*e.g.* a 100-year episode occurring twice more often), the return period of the  
101 interaction between these two constraints should happen four times more often. How these  
102 extreme events will affect ecosystem functions is a key issue for climatologists, ecologists and  
103 modelers from IPCC (IPCC, 2014). In this regard, perennial plants, and especially trees, which  
104 are keystones structuring many ecosystems, sheltering biodiversity and being an important  
105 carbon stock, are the most likely to be affected by an increased exposure to abiotic constraints,  
106 even though they may not reach the lethal threshold at once (Chuste *et al.*, 2019). An accurate  
107 assessment of the emerging risks and their impacts on the physiology of trees is therefore  
108 urgently needed to predict dynamics of forest ecosystems and ~~in fine~~ultimately their influence  
109 on the water and carbon cycles at the global scale.

110 The risk of developing significant damages is only achieved at the crossing between climatic  
111 hazards (*e.g.* drought or frost constraint), stress exposure (*e.g.* low water potential or organ  
112 temperature) and tree vulnerability (*e.g.* low resistance to embolism or insufficient frost  
113 hardiness; [Breda-Bréda & Peiffer, 2014](#)). The prediction of emerging risks therefore needs  
114 accurate assessments of (i) the exposure to a given abiotic constraint (type, intensity, duration  
115 and frequency) and (ii) the spatio-temporal patterns of vulnerability and subsequent damages.  
116 The timing of critical phenological stages (*e.g.* bud burst date and leaf full expansion date with  
117 respect to frost and drought, respectively) would thus modulate risk and subsequent damages.

118 ~~However, despite it can can significantly affect the probability of interaction between~~  
119 ~~constraints, how~~ How the exposure to a given abiotic constraints at a given time instant would  
120 influence the vulnerability to the same or a different constraint in the future is a critical issue  
121 that has been only rarely investigated ([Miao \*et al.\*, 2009](#), [Anderegg \*et al.\*, 2013](#); [Batlori \*et al.\*,](#)  
122 [2017](#); [Hossain \*et al.\*, 2019](#)). ~~has not yet been characterized.~~

123 The vulnerability to abiotic constraints (which is the susceptibility to develop damages  
124 facing a given stress intensity) has been studied for decades via three different focuses, namely  
125 phenology (Fig. 1 Frame C; [Hänninen & Tanino, 2011](#)), water use (Fig. 1 Frame D, [Choat \*et\*](#)  
126 [al.](#), 2012; [Anderegg \*et al.\*, 2015](#)) and carbon availability (Fig.1 Frame B, [Sala \*et al.\*, 2012](#);  
127 [Hartmann & Trumbore, 2016](#)). The individual responses of each tree eco-physiological process  
128 has been integrated into process-based models such as PHENOFIT ([Morin & Chuine, 2005](#)),  
129 SUREAU ([Martin-StPaul \*et al.\*, 2017](#)) or CASTANEA ([Dufrene \*et al.\*, 2005](#)), for phenology,  
130 water use and carbon availability, respectively. Facing constraints, the response of each process  
131 may not be linear. Therefore, the response to simultaneous or successive constraints may  
132 generate chaotic-complex behavior. Integrating the physiological response to abiotic constraints  
133 is therefore needed to predict various forms of emergent behavior under new conditions  
134 imposed by climate change ([Bartholomeus \*et al.\*, 2011](#)).

135 This paper summarizes the state-of-the-art of tree responses to drought and frost constraints  
136 from the cellular to the individual scale and subsequent damages with the aim of emphasizing  
137 the undervalued role of their interactions at the short term scale (i.e. from the day to the year).  
138 Furthermore, different communities are interested in single process or sets of processes (*e.g.*  
139 hydraulic in response to drought, frost hardiness and damages, phenological cycles,...).  
140 ~~Another aim is therefore to bring scientists to make a step aside, having an integrative~~  
141 ~~perspective of plant's response to abiotic constraints.~~The influence of previous damages stress  
142 factors on plant vulnerability, is mainly relevant on woody plants and will be mainly explored  
143 at the annual level through two main questions (i) How would the interaction of drought and  
144 frost constraints modulate the vulnerability to a subsequent constraint? (ii) How do vulnerability  
145 to a given constraint and phenology interact? Addressing these questions will provide novel  
146 information about the interaction between climate and trees from a wider, multifactorial and  
147 temporal perspective. This framework would be crucial to understand and predict local  
148 mortality dynamics and ultimately to improve actual species distribution models.

149

#### 150 **vulnerability to a subsequent constraint?**

151 Over the last decades, ecophysiological studies have led to a comprehensive understanding  
152 on the direct relation between abiotic constraints and physiological processes (Frame #1 and  
153 #2; Fig. 1). ~~The onset of drought induced damages is mainly based on the hydraulic~~  
154 ~~component, whereas frost induced damages relies on the interaction between water and carbon~~  
155 ~~availability.~~From a physical perspective, both drought and frost constraints are related to a  
156 limited liquid water availability. Similar damages are generated at both the and cellular levels  
157 (i.e. plasmolysis and cell lysis; Levitt, 1980; Siminovitch & Cloutier, 1983); and -vascular  
158 levels (*i.e.* cavitation and embolism when a critical level of water potential is reached; Sperry  
159 & Sullivan, 1992; Charrier *et al.*, 2014). A critical factor in the response to both stresses is the

160 ~~accumulation of solutes to maintain solvation layer around macromolecules~~ ~~and~~ ~~inducing~~ ~~for~~ ~~instance~~ ~~the~~ ~~synthesis~~ ~~of~~ ~~dehydrins~~ ~~(Welling~~ ~~et~~ ~~al.~~ ~~, 2002)~~ ~~or~~ ~~the~~ ~~accumulation~~ ~~of~~ ~~solutes~~ ~~to~~ ~~maintain~~ ~~solvation~~ ~~layer~~ ~~around~~ ~~macromolecules~~. ~~Interestingly~~, the molecular response to both  
161 developed similar molecular responses to drought and frost constraints (Beck *et al.*, 2007),  
162 under the control of abscisic acid (ABA; Chandler & Robertson, 1994), ~~and~~-inducing, for  
163 instance, the synthesis of dehydrins (Welling *et al.*, 2002), ~~or~~ ~~the~~ ~~accumulation~~ ~~of~~ ~~solutes~~ ~~to~~  
164 ~~maintain solvation layer around macromolecules~~. Interestingly, the molecular response to both  
165 constraint involves the same regulatory mechanisms (*e.g.* Dehydration Responsive Elements  
166 containing Cold Binding Factors; Stockinger *et al.*, 1997; Baker *et al.*, 1994). The same  
167 pathways are thus activated in both cold and drought responses (Yamaguchi-Shinozaki &  
168 Shinozaki, 1994).

169 The onset of drought-induced damages is triggered by hydraulic failure and involves the  
170 interaction between water and carbon status (Frame 1 and reviews from Choat *et al.*, 2018;  
171 Martinez-Vilalta *et al.*, 2019, for extensive description of drought stress syndromes), as frost-  
172 induced damages relies on (Charrier *et al.*, 2013b). At the vascular level, Tree-species more  
173 vulnerable to winter embolism seem to be also more vulnerable to summer embolism (Charrier  
174 *et al.*, 2014). ~~Finally~~This may be related to ~~the~~ ability of air to propagate within xylem under  
175 high frost- or drought-related tension if the vulnerability to embolism seems to be essentially  
176 fixed phylogenetically, in relation to anatomy (through pit membrane membrane thickness;  
177 (Cochard *et al.*, 1992) Li *et al.*, 2016) ~~).~~ However, many tree species are able to restore their  
178 hydraulic conductivity, for instance to supply growth in spring. One refilling mechanism  
179 involves an osmoregulation via solutes compounds generating osmotic pressure in the lumen  
180 of the vessels during winter (Ewers *et al.*, 2001; Améglio *et al.*, 2002), and eventually summer  
181 (Nardini *et al.*, 2011). In conifers, the involved mechanism remains to be clarified, but many  
182 species seem able to do so (Sperry & Sullivan 1992; Sperry *et al.*, 1994; Mayr *et al.*, 2003;  
183 2014). Positive sap pressure has also been measured at the whole plant scale from the  
184 pressurization of the root system, notably in woody lianas (Priestley 1920; Sperry *et al.*, 1987;  
185 Charrier *et al.*, 2016).



186 Even if xylem conduits were completely refilled, residual bubbles within the xylem would  
187 expand at lower tension if the tree is exposed again to drought after winter or at higher freezing  
188 temperature occurring late in the winter (i.e. after the winter refilling process). Such a higher  
189 vulnerability after the initial stress exposure and release would occur through physical or  
190 chemical deformation of the pit ultrastructure (i.e. fatigue; Christensen-Dalsgaard & Tyree,  
191 2014).

192 However, despite high xylem embolism has been suggested to cause trees death (Brodrigg  
193 & Cochard, 2009), higher embolism than 50% and, even 90%, is frequently observed and  
194 recovered in over-wintering trees without impairing their survival suggesting that it does not  
195 constitute a lethal threshold by itself (Sperry *et al.*, 1994; Mayr *et al.*, 2003; 2006; 2019;  
196 Charrier *et al.*, 2013a). High xylem embolism rather seem to constitute an empirically  
197 correlated trait than a clear physiological lethal threshold (Mayr *et al.*, 2019).

198 ~~an osmoregulation via carbon compounds allows the resorption of the embolism during~~  
199 ~~winter (via stem pressure; Fig. 1 #3 Ewers *et al.*, 2001; Améglio *et al.*, 2002) and eventually in~~  
200 ~~summer (Nardini *et al.*, 2011).~~

201 Two alternative ~~hypotheses~~ hypothesis, although not ~~self-mutually~~ exclusive, ~~can should be~~  
202 taken into account to predict the interaction between stress factors ~~be introduced.~~ Legacy, is  
203 considered as a passive change of physiological status in response to previous stress exposure.  
204 We suggest that the modulation of either carbon and/or water pools would resulting in the  
205 modulation in-of vulnerability (McDowell *et al.*, 2008). ~~On the one hand, a~~ Altered water and  
206 carbon status would generate potential feedback physiological loops and trade-offs during  
207 successive abiotic ~~constraint~~ stress exposures (legacy effect). ~~and~~ memory, which is also  
208 called priming in plant defense facing biotic and also abiotic stress (Savvides *et al.*, 2016), is  
209 considered as an active process ~~through stress memory, or facilitated acclimations~~ shifting  
210 biological processes enhancing a basal level of resistance at the cost of reducing primary

211 metabolism such as growth (Bruce *et al.*, 2007; Walter *et al.*, 2013; Martinez-Medina *et al.*  
212 2016). It requires pre exposure to the stress factor in contrast with acclimation that occurs in  
213 anticipation. ~~On the one hand, altered water and carbon status would generate potential~~  
214 ~~feedback physiological loops and trade offs during successive abiotic constraint exposures~~  
215 ~~(legacy effect)~~. ~~On the other hand, i~~In long living organisms such as trees, the concept of  
216 memory is ~~still unclear~~ ommon because most of the studies dealing with memory have focused  
217 on short-term memory in annual plants. Some studies have evaluated how the vulnerability to  
218 a given constrain, *i.e.* frost or drought, is affected by a repeated exposure of the trees to it (Mayr  
219 *et al.*, 2003; Tomasella *et al.*, 2019). However, studies evaluating the feedback between  
220 constraints and vulnerability (*i.e.* the effect of drought on vulnerability to frost and vice versa)  
221 are very few (*e.g.* Kreyling, *et al.*, 2014; Sierra-Almeida *et al.*, 2016). We attempted to draw a  
222 generic picture as both constraints affect the same physiological processes (Fig. 1).

223 This interaction is likely to occur under montane/boreal climate. ~~A first path of interaction~~  
224 ~~between drought and frost is through water and solute content that both play a role at the organ~~  
225 ~~level. Indeed, drought constraint will reduce water content in all tree compartments, including~~  
226 ~~living cells (symplastic) and xylem conduits (apoplastic).~~ Such reduction in water content  
227 ~~would~~ increase in the solute concentration (*i.e.* lower osmotic potential) ~~that, consequently,~~  
228 would decrease the freezing point ( $-1.86^{\circ}\text{C}\cdot\text{mol}^{-1}\cdot\text{kg}^{-1}$ ; Hansen & Beck, 1988) in both living  
229 cells (Charrier *et al.*, 2013b) and the apoplast (Lintunen *et al.*, 2018). Soil water deficits can  
230 indeed initiate the early stages of frost hardiness in Douglas-fir (Timmis & Tanaka, 1976).  
231 ~~However, it should be noticed that t~~This effect is likely to be canceled during the transition from  
232 drought- to frost-exposed period, in case of a wet autumn period. ~~However, -changes in the~~  
233 water regime, as predicted by future climate predictions, could cause this dry summer and  
234 autumn conjunction to occur more -regularly.

235 Lower frost vulnerability of trees pre-exposed to drought is thus expected through two

236 potential side-effects (i) lower probability of ice formation under similar sub-zero temperatures  
237 and (ii) higher probability of ice nucleation within the apoplasm, pulling symplasmic water  
238 towards extracellular ice lattice ~~(when cryoprotective solutes can protect different~~  
239 ~~macromolecules as generally observed during frost acclimation, see above)~~. However, this  
240 effect may be balanced by a decrease in non-structural carbohydrates, due to photosynthesis  
241 limitation by stomatal closure, leading to similar, or even lower solute concentration in drought-  
242 exposed trees. In fact, both effects have been observed in winter frost damages following  
243 summer drought. Increasing frost hardiness is thus expected for relatively moderate dehydration  
244 ~~( $<30\%$  drought stress intensity; Fig. 3~~ as observed in Benzioni *et al.*, 1992; Kreyling *et al.*,  
245 2012; Sierra-Almeida *et al.*, 2016). It should also be noted that growth-related processes being  
246 more sensitive to water stress than photosynthesis (Fig. 2A), mild water stress would also  
247 promote transient increase in non-structural carbohydrates (McDowell, 2011; Granda &  
248 Camarero, 2017). However, would this transient increase result in higher reserves (potentially  
249 enhancing frost acclimation) or be allocated to other processes such as belowground growth?  
250 Finally, lower frost hardiness would be observed for ~~higher stress level ( $>50\%$  drought stress~~  
251 ~~level intensity)~~ in relation to carbon depletion (Fig. 3 right Wong *et al.*, 2009; Galvez *et al.*,  
252 2013; Fig. 3).

253 From the xylem/hydraulic point of view, the resulting vulnerability is frost-induced  
254 embolism is only generated when sap is under moderate tension (Mayr *et al.*, 2007). Freeze-  
255 thaw cycles will thus exacerbate the pre-existing tension promoting air seeding from vessels to  
256 vessels in dehydrated trees (Fig. 2B; Tyree & Sperry, 1989; Mayr *et al.*, 2007; Kasuga *et al.*,  
257 2015). Drought episodes during the growing season lead to the reduction in vessel diameter  
258 (Lovisolo & Schubert, 1998; Beikircher & Mayr, 2009) that would result in higher resistance  
259 to ~~FF-freeze-thaw induced~~ embolism. However, a reduced level of non-structural carbohydrates  
260 would limit the ability of the tree to refill embolized vessels during winter via active mechanism  
261 (*e.g.* stem pressure; Améglio *et al.*, 2001). ~~Even if xylem conduits were completely refilled,~~

262 ~~While this is a potential mechanism for avoiding hydraulic failure, it is not clear if this is a viable strategy for trees in temperate climates. The impact of this mechanism on~~  
263 would lie in the higher ability of sap to supercool and therefore avoid the phase shift from liquid  
264 to ice and the resulting formation of air bubbles (Lintunen *et al.*, 2018).

### 265 **Effect of frost on vulnerability to drought**

266 This interaction is likely to occur under temperate climate. ~~Although we have a relatively~~  
267 ~~clear vision of the potential effect of drought exposure on frost risks (cf. Fig. 2), m~~Major gaps  
268 of knowledge remain with respect to frost exposure onto drought vulnerability, although  
269 assumptions can be made. Overall, frost damages before drought exposure would mainly have  
270 negative effects for the trees, through limited hydraulic conductivity and/or biomass destruction  
271 although the affected organs as well as the consequences for the tree can be various. However,  
272 in our literature review, we did not find a single study that specifically explored the  
273 physiological consequences of frost damages during the following growing season and after,  
274 although Charrier ~~et al.~~ et al. (2018e2018a) highlighted a significant negative correlation  
275 between autumnal frost damages and fruit yield the following summer in walnut.

276 ~~When Low, but not necessarily frozen, soil temperature is too low (but not necessarily~~  
277 ~~frozen), the limits root water uptake becomes very limited (Améglio *et al.*, 2002). Thanks to~~  
278 ~~thermal inertia, the combination of cold soil and W~~warm and dry air can ~~thus~~ promote the  
279 aboveground dehydration and hydraulic failure. This type of phenomenon, called winter  
280 drought, is typical of late winter conditions at high altitudes (Mayr *et al.*, 2006; Charrier *et al.*,  
281 2017; Earles *et al.*, 2018). Winter drought- and ~~Furthermore, when the winter precipitation~~  
282 ~~regimes change from solid to liquid, the soil will reach lower temperature, damaging the root~~  
283 ~~system more frequently. This would alter the whole tree hydraulic architecture by decreasing~~  
284 ~~water supply and decreasing the ability to recover, ultimately increasing xylem embolism (Cox~~  
285 ~~& Malcom, 1997; Zhu *et al.*, 2000). This syndrome has been identified as pre-disposing dieback~~  
286 ~~for Yellow birch (Cox & Zhu, 2003). F~~freeze/thaw-induced embolism ~~and frost cracks~~ increase

287 the volume of air gas within the xylem tissue conduits, therefore facilitating the not only the entry spreading of air into through  
288 ~~the pit membrane thickness (Li et al., 2016). Such changes in the pit membrane thickness is likely to increase drought-~~  
289 ~~thaw events has been observed for different species when evaluating defrosted wood samples~~  
290 ~~(Li et al., 2016). Such changes in the pit membrane thickness is likely to increase drought-~~  
291 ~~vulnerability through air seeding (Charrier et al., 2014; Li et al., 2016). Such an increased~~  
292 ~~vulnerability would be detrimental to plant growth. Lower hydraulic conductivity would limit transpiration which should result in lower water use efficiency.~~  
293 When the winter precipitation regimes change from solid to liquid, the shorter snow cover  
294 duration is likely to expose soil to lower temperature, damaging the root system more frequently  
295 (Francon et al., 2020). This would alter the whole tree hydraulic architecture by decreasing  
296 water supply and decreasing the ability to recover, ultimately increasing xylem embolism (Cox  
297 & Malcom, 1997; Zhu et al., 2000). This syndrome has been identified as pre-disposing dieback  
298 for Yellow birch (Cox & Zhu, 2003).

299 Lower hydraulic conductivity, from winter embolism and limited spring refilling would limit  
300 plant transpiration, which should result in lower leaf water potential and stomatal conductance.  
301 As partially open stomata allow sufficient CO<sub>2</sub> diffusion while limiting the amount of transpired  
302 water, it is likely that water use efficiency would be increased. Depleting soil water content  
303 would be depleted more slowly which should thus delay drought onset and intensity. The same  
304 dynamic is expected. However, at the stand scale, the competition to water resources from other  
305 tree individuals or other plant species may delete this potential benefit after late frost damages,  
306 as Furthermore leaves would expand later in the season, when the probability of  
307 drought exposure (i.e. high evaporative demand combined with limited soil water availability)  
308 is higher although earlier soil water uptake has been limited. However, at the stand scale, the  
309 competition for water resources from other tree individuals or other plant species (less  
310 vulnerable to winter embolism or frost damages) may delete this potential benefit.

311

312 **Potential interaction between vulnerability to constraints and phenology**

313 The annual phenological cycle consists of developmental events related to the alternation of  
314 growth and dormancy of the trees. Longer growing seasons, such as predicted by vegetation  
315 models due to climate change, are expected to increase the carbon uptake and the ecosystem  
316 net primary production (Angert *et al.*, 2005, Delpierre *et al.*, 2009; Wolf *et al.*, 2016). However,  
317 this would be at the cost of longer periods of effective transpiration that would, ultimately,  
318 deplete soil water content and thus increase the exposure to drought stress. Although timing is  
319 a crucial parameter in the exposure to a seasonal abiotic constraint, the models predicting tree  
320 seasonality developed so far mostly consider non-stressing conditions (see Lang *et al.*, 1987;  
321 Delpierre *et al.*, 2016).

322 Photoperiod and temperature are key signals regulating plant phenology (Maurya &  
323 Bhalearao, 2017). In spring, ecodormancy release and growth (both primary and secondary) are  
324 accelerated by warm temperature, in some species in interaction with photoperiod (Laube *et al.*,  
325 2014). At the end of the growing season, growth cessation, bud set and endodormancy ~~is are~~  
326 mainly controlled by ~~decreasing~~ photoperiod and temperature through two separate, but  
327 temporally connected processes: one photosensitive (short day) and one thermosensitive (low  
328 temperature) process (Tanino *et al.*, 2010), ~~whereas budset and dormancy by warm and low~~  
329 night temperatures, respectively (Svystun *et al.*, 2019). Endodormancy is subsequently released  
330 by chilling temperature. Among these different stages, we can distinguish two different  
331 processes: temperature-promoted processes (*i.e.* occurring at a rate proportional to temperature  
332 such as ecodormancy release, primary and secondary growth, and budset) and signal-limited  
333 processes (*i.e.* occurring after a specific threshold has been reached such as growth cessation,  
334 endodormancy induction, leaf fall), which are indirectly affected by photoperiod and  
335 temperature. However, the effect of abiotic stress on the different phenological processes and,  
336 furthermore, how carry-over effects can modulate pluri-annual dynamics, have not been  
337 explored yet (Fu *et al.*, 2014). Thus, depending on the timing of the previous stage, the onset of

338 the following ones will be affected, leading to unpredictable behavior (Hänninen & Tanino,  
339 ~~2010~~2011). For instance, an increase in temperature would hasten or delay growth cessation  
340 depending on the diurnal dynamics of temperature (see Rohde *et al.*, 2011 and Kalcsits *et al.*,  
341 2009, respectively), affecting the subsequent stages (budset, endodormancy and budburst; Fig.  
342 4). Leaf senescence timing is positively correlated with budburst timing once removing the  
343 influence of autumn temperature (Fu *et al.*, 2014). This carry-over effect is, however, of second  
344 order (Liu *et al.*, 2019) and potentially hidden by the prominent role of environmental variables  
345 such as temperature and photoperiod (Vitasse *et al.*, 2009). Such an interacting process has been  
346 incorporated in a leaf senescence model (Delpierre *et al.*, 2009) by modulating the cold  
347 temperature sum leading to leaf senescence by the budburst date (~~Kennan-Keenan~~ &  
348 Richardson, 2015).

349 Drought stress affects radial (*i.e.* secondary) growth but its effects on primary growth is  
350 relatively unexplored. Drought exposure can ~~thus~~ significantly hasten the primary growth onset  
351 in *Erica multiflora*, although not in *Globularia alipum* (Bernal *et al.*, 2011). However, also no  
352 effect on growth cessation has been reported (Bernal *et al.*, 2011). One would expect that earlier  
353 growth onset would favor drought avoidance during springtime, but it could hasten soil water  
354 depletion and this may be at the cost of the alteration of the development of shoot, leaves and  
355 buds (Misson *et al.*, 2011). Autumnal drought is expected to induce earlier endodormancy,  
356 probably through the induction of ABA (Maurya & Bhalerao, 2017; Tylewicz *et al.*, 2018).  
357 Furthermore, higher temperatures associated to drought events would induce deeper dormancy  
358 (Heide, 2003; Tanino *et al.*, 2010; Rohde *et al.*, 2011). As this would result in later budburst  
359 the following year, these trees are likely to be more drought-exposed since they would expand  
360 their leaves during a period of more pronounced water deficit. Delayed dormancy and budburst  
361 are thus expected through synergistic combination between drought exposure and carry-over  
362 effect (Xie *et al.*, 2015).

363 The impact of frost events on phenology has essentially been reported in spring when it

364 affects developing organs such as flushing buds, flowers and new leaves. After a single  
365 damaging event, the resulting leaf area (*i.e.* post growth) can be reduced (up to more than 50%)  
366 and leaf full expansion delayed (16 to 34 days; Augspurger, 2013), exposing the tree to summer  
367 constraints, although leaf area and carbon reserves are reduced (Menzel *et al.*, 2015; d'Andrea  
368 *et al.*, 2019; 2020). During late summer, after growth cessation and dormancy induction, frost  
369 exposure may promotes the dormancy release and, as cold temperature, induces leaf fall (Rinne  
370 *et al.*, 1997). So depending on the timing, frost damages could delay or hasten the annual cycle  
371 (later leaf full expansion or earlier endodormancy release, after spring and autumn frost  
372 damages, respectively). However, frost events may induce both cellular and vascular damages  
373 in the distal organs, resulting in re-growth from more basal buds. After several years, the whole  
374 tree architecture may be affected resulting in smaller and denser canopy, which would be less  
375 frost exposed, but at the cost of canopy light transmittance and subsequent photosynthesis.

### 376 **A holistic modelling framework for tree stress physiology: a tentative outline**

377 A living tree is a functionally holistic system including constantly interacting growth units.  
378 Accordingly, the various physiological processes are linked to each other. Two different  
379 physiological variables such as water potential and frost hardiness, represent, at least partially,  
380 different manifestations of the same physiological phenomena (as described by the correlations  
381 between frost hardiness, water content and water potential).

382 Ecophysiological models have been developed separately for frost stress (Fuchigami *et al.*,  
383 1982; Leinonen 1996; Charrier *et al.*, 2018a) and drought stress (Sperry *et al.*, 1998; Zweifel *et*  
384 *al.*, 2005; Martin-StPaul *et al.*, 2017). By addressing the seasonal alternation of growth and  
385 dormancy, the annual phenological cycle also reflects major changes in the physiology of trees  
386 that could enhance or mitigate the vulnerability to stress factors. Accordingly, the integrated  
387 models of frost hardiness simulates the environmental responses of changes in frost hardiness  
388 according to the phenological stage (Kellomäki *et al.*, 1992; 1995; Leinonen 1996; Charrier *et*



389 al., 2018a). Seasonality is indeed located at the core of any modelling framework related to  
390 seasonal frost or drought stress factors. We first propose that existing ecophysiological models  
391 explicitly integrate seasonality weighing relevant parameters by the variables describing  
392 phenological processes (e.g. sum of growth degree days, sum of chilling units,...). Although  
393 not being mechanistic, these variables would unify the framework of the models, allowing  
394 further research into the deterministic relations between three highlighted processes  
395 (phenological, carbon and hydraulics). Such an integrated modeling approach should be used  
396 to the interaction between stresses factors through the following steps:

- 397 (i) Quantitative description of the variable(s) of interest e.g. loss of hydraulic conductivity,  
398 cellular lysis.
- 399 (ii) Identification of the relevant physiological drivers with special attention dedicated to the  
400 ones that interact with several variables of interest e.g. water or carbohydrate content.
- 401 (iii) Description of the relation between the drivers and the variable of interest e.g. relation  
402 between frost hardiness, tissue water content, carbohydrate and temperature (Poirier *et*  
403 al., 2010).
- 404 (iv) Experimentally-based description of the relation between physiological drivers and  
405 external climatic drivers to ensure realistic behavior.
- 406 (v) Mathematical description of the relationship between physiological driver and external  
407 climatic drivers e.g. carbohydrate content depending on air temperature (Charrier *et al.*,  
408 2018c).
- 409 (vi) Coupling the models obtained at Steps (ii) and (iv) and development of a model predicting  
410 the variable of interest with input data being climatic, state variables (such as specific  
411 parameters or initial values) and intermediate physiological drivers.

412 (vii) Simulate the tree response and compare with stress-related variable such as frost  
413 hardiness vs daily minimum temperature (Charrier *et al.*, 2018a).

414 Finally, such approach would easily simulate both legacy and memory effects (as a function of  
415 previous level of damages, water and carbon contents.

416

417 Exposures to drought and frost constraints ~~has exert a~~ potential feedbacks on the sensitivity  
418 to future constraints by affecting physiological components such as non-structural  
419 carbohydrates and water balance (*i.e. legacy* Fig. 2). Phenological processes has been for long  
420 only studied in non-stressed plants, although physiological seasonality is altered by stress (Fig.  
421 4). Major advances are needed in characterizing environmental control of phenological  
422 processes in trees during or after stress. Notably do critical sums of temperature (*e.g. chilling*  
423 or forcing temperature) vary with stress intensity, potentially explaining the residual variance  
424 in phenological predictions?

425 Phenological processes are likely to exhibit legacy through carbon balance *e.g. trophic*  
426 limitation of meristem growth (Bonhomme *et al.*, 2009), disturbed glycan deposition onto  
427 plasmodesmata (Rinne *et al.*, 2001). However, previous exposure to stress factors may also  
428 alter the rate of the future ecophysiological response through memory effect via accumulation  
429 of regulatory proteins, transcription factors or histone methylation (Bruce *et al.*, 2007; Walter  
430 *et al.*, 2013). As already performed in annual plants, there is a clear need for multi-constrained  
431 and longer term studies. ~~Although in woody plants have to face multi-annual constraints, these~~  
432 ~~mechanisms have been more thoroughly studied at relatively short time scale (up to one week)~~  
433 ~~and considering only one constraint (*i.e. drought or frost*).~~

434 To specifically deal with the interaction between constraints, we need to i) develop a systemic  
435 approach at the plant scale integrating ontogenic and histological differences as well as carbon

436 ~~and water uptake with hydraulic limitation for hydraulic processes (water uptake, transpiration) and carbon dynamics~~  
437 approach aimed to evaluate the crossing effects interactions between different constraints in the tree survival  
438 capacity both at short and a long time scale. Interesting insights have been brought by  
439 integrative studies, for instance, focusing on carbon availability and hydraulic failure facing  
440 drought (*e.g.* McDowell *et al.*, 2008). It is especially relevant to develop this type of approaches  
441 on different species exhibiting contrasted combinations of drought and frost tolerance,  
442 including provenances originating from the entire species' distribution range (core and edges)  
443 to unravel local adaptations (Kreyling *et al.*, 2014). The effect of different successive and  
444 concomitant constraints at different periods of the year should help to develop deterministic  
445 relationships between different physiological variables and processes in response to each of  
446 them. It would simulate the life history of the tree and modulated response through legacy and  
447 memory effects as a function of previous level of damages, water and carbon contents.  
448 Improving the descriptive range of these interrelations at the individual and population scales  
449 would subsequently allow quantitative and dynamic description of drought and frost resistance  
450 ~~integrating in interaction the life history of the tree.~~ This ~~is a pre-requisite to would~~ improve  
451 existing mechanistic models simulating these interacting processes in order to predict  
452 accurately the effect of cumulative stress on tree physiology and survival ~~as mentioned Frame~~  
453 3.

454

455 **Frame 1: Mechanisms leading to drought-induced damages**

456 Water stress results from an imbalance between inwards (root water absorption) and outwards  
457 water fluxes (transpiration and evaporation). Trees have developed resistance mechanisms to  
458 avoid water stress (maintaining high water potential; Fig. 1) by a tight control of both inwards  
459 (e.g. deep rooting) and outward water fluxes (e.g. stomatal closure, leaf shedding, low cuticular  
460 transpiration; Choat *et al.*, 2018; Martin-StPaul *et al.*, 2017). Maintaining xylem hydraulic  
461 function under high tension is also a critical trait avoiding dehydration, and ultimately  
462 desiccation, of living tissues (Fig. 1 #14; Sperry *et al.*, 1998, Brodribb & Cochard, 2009).  
463 However, ontogenic changes in xylem vulnerability to embolism may modulate thus the tree's  
464 resistance across seasons (Charrier *et al.*, 2018b).

465 In some species, leaves are more drought-sensitive than perennial organs (i.e. hydraulic  
466 vulnerability segmentation) and usually fall after the exposure to drought (Fig. 1 #6 Tyree *et*  
467 *al.*, 1993). Such a deciduous behavior can be too sudden to allow recycling nutrients (Fig. 1 #5  
468 Bréda *et al.*, 2006) but not in all cases (Marchin *et al.*, 2010). Following leaf fall, the  
469 reestablishment of a new cohort of leaves would mobilize non-structural carbohydrate reserves,  
470 which may not be restored before winter. Long-term drought induces stomatal closure to  
471 maintain the functionality of the hydraulic system. As respiration is maintained and even  
472 increased during warm spells, while carbon assimilation is reduced, carbon reserves steadily  
473 depletes (Fig. 2; McDowell *et al.*, 2008). Finally, two pools, namely solutes, including  
474 carbohydrates, and water, interact to maintain a solvation layer around biomolecules avoiding  
475 critical denaturation and related cellular death (Martinez Vilalta *et al.*, 2019).

## **Frame 2: Mechanisms leading to frost-induced damages**

Low temperature drives tree species distribution through different processes: including resistance to freezing temperature and to frost-induced embolism (Charrier *et al.*, 2013a), and the time to complete the annual cycle and seedling survival (Koerner *et al.*, 2016). In fully hydrated organs, ice formation can induce mechanical strains and frost cracks (Kubler, 1983; Cinotti, 1991). In moderately dehydrated organs, ice nucleation and propagation redistributes water towards nucleation sites (Ball *et al.*, 2006; Mayr & Charra-Vaskou, 2007) and likely provoke air-seeding within pits (Charrier *et al.*, 2014). However, frost-induced embolism in the distal parts would insulate the trunk from further dehydration (Fig. 1 #15; Lemoine *et al.*, 1999; Charrier *et al.*, 2017). Critical thresholds can be reached when the root water absorption could not compensate the evaporative demand during periods of sunny days alternating with freezing nights (*i.e.* winter drought; Tranquillini, 1979; Charrier *et al.*, 2017). At the cellular level, the low chemical potential of ice pulls water molecules from the cells towards extracellular ice lattice, causing pronounced cell dehydration and shrinkage (Dowgert and Steponkus, 1984; Charra-Vaskou *et al.*, 2016). To cope with seasonal frost stress, trees transiently increase their frost resistance in frost-exposed organs through the synthesis of cryoprotective solutes (Charrier *et al.*, 2013b). Cryoprotective solutes (*e.g.* carbohydrates, amino-acids or organic acids) can protect macromolecules by maintaining a solvation layer around it. Being the main substrate and energetic source, non-structural carbohydrates content in autumn is thus tightly related to maximum frost resistance reached during winter (Fig. 1 # 1; Palonen & Buszard, 1997; Morin *et al.*, 2007; Charrier *et al.*, 2013a; 2018c). As non-structural carbohydrates content usually increases with elevation through temperature limited-growth rather than by carbon availability (Hoch *et al.*, 2002; Shi *et al.*, 2008; Koerner, 2015), the increase in NSC at higher elevation would help plants to reach sufficient level of frost resistance in winter.

501 **References**

- 502 [Adams HD, Zeppel MJ, Anderegg WR, Hartmann H, Landhausser SM, Tissue DT, Huxman](#)  
503 [TE, Hudson PJ, Franz TE, Allen CD \*et al.\*, 2017. A multispecies synthesis of physiological](#)  
504 [mechanisms in drought induced tree mortality. \*Nature Ecology and Evolution\* 1: 1285](#)
- 505 Améglio, T., Ewers, F. W., Cochard, H., Martignac, M., Vandame, M., Bodet, C., & Cruiziat,  
506 P. (2001). Winter stem xylem pressure in walnut trees: effects of carbohydrates, cooling  
507 and freezing. *Tree Physiology*, 21: 387-394.
- 508 Améglio, T., Bodet, C., Lacoïnte, A., & Cochard, H. (2002). Winter embolism, mechanisms of  
509 xylem hydraulic conductivity recovery and springtime growth patterns in walnut and peach  
510 trees. *Tree Physiology*, 22: 1211-1220.
- 511 Améglio, T., Decourteix, M., Alves, G. *et al.*, (2004). Temperature effects on xylem sap  
512 osmolarity in walnut trees: evidence for a vitalistic model of winter embolism repair. *Tree*  
513 *Physiology*, 24: 785-793.
- 514 Anderegg, W. R., Plavcová, L., Anderegg, L. D., Hacke, U. G., Berry, J. A., & Field, C. B.  
515 (2013). Drought's legacy: multiyear hydraulic deterioration underlies widespread aspen  
516 forest die-off and portends increased future risk. *Global change biology*, 19(4), 1188-1196.
- 517 Anderegg, W. R., Flint, A., Huang, C. Y. *et al.*, (2015). Tree mortality predicted from drought-  
518 induced vascular damage. *Nature Geoscience*, 8: 367-371.
- 519 Angert, A., Biraud, S., Bonfils, C. *et al.*, (2005). Drier summers cancel out the CO<sub>2</sub> uptake  
520 enhancement induced by warmer springs. *Proceedings of the National Academy of*  
521 *Sciences of the United States of America*, 102: 10823-10827.
- 522 Augspurger, C. K. (2013). Reconstructing patterns of temperature, phenology, and frost damage  
523 over 124 years: spring damage risk is increasing. *Ecology*, 94: 41-50.
- 524 [Baker S.S., Wilhelm K.S., & Thomashow M.F. \(1994\). The 50- region of Arabidopsis thaliana](#)  
525 [cor15a has cis-acting elements that confer cold, drought, and ABA-regulated gene](#)  
526 [expression. \*Plant Molecular Biology\* 24: 701–713.](#)
- 527 Ball, M. C., Canny, M. J., Huang, C. X., Egerton, J. J. G., & Wolfe, J. (2006). Freeze/thaw-  
528 induced embolism depends on nadir temperature: the heterogeneous hydration hypothesis.  
529 *Plant, Cell & Environment*, 29: 729-745.
- 530 [Barigah, T. S., Charrier, O., Douris, M. \*et al.\*, \(2013a\). Water stress induced xylem hydraulic](#)  
531 [failure is a causal factor of tree mortality in beech and poplar. \*Annals of Botany\*, 112: 1431–](#)  
532 [1437.](#)
- 533 [Batllori, E., De Cáceres, M., Brotons, L., Ackerly, D. D., Moritz, M. A., & Lloret, F. \(2017\).](#)  
534 [Cumulative effects of fire and drought in Mediterranean ecosystems. \*Ecosphere\*. 8: e01906.](#)
- 535 Beck, E. H., Fettig, S., Knake, C., Hartig, K., & Bhattarai, T. (2007). Specific and unspecific  
536 responses of plants to cold and drought stress. *Journal of Biosciences*, 32: 501-510.
- 537 Beikircher B, & Mayr S (2009). Intraspecific differences in drought tolerance and acclimation  
538 in hydraulics of *Ligustrum vulgare* and *Viburnum lantana*. *Tree Physiol* 29:765–775
- 539 Benzioni, A., Palzkill, D. A., & Nelson, J. M. (1992). Flower bud dormancy, ABA  
540 concentration, and survival during frost of jojoba genotypes under water stress. *Journal of*  
541 *the American Society for Horticultural Science*, 117: 976-980.
- 542 Bernal, M., Estiarte, M., & Peñuelas, J. (2011). Drought advances spring growth phenology of  
543 the Mediterranean shrub *Erica multiflora*. *Plant Biology*, 13: 252-257.
- 544 Bréda, N., Huc, R., Granier, A., & Dreyer, E. (2006). Temperate forest trees and stands under  
545 severe drought: a review of ecophysiological responses, adaptation processes and longterm  
546 consequences. *Annals of Forest Science*, 63: 625-644.
- 547 Bréda, N., & Peiffer, M. (2014). Vulnerability to forest decline in a context of climate changes:  
548 new prospects about an old question in forest ecology. *Annals of Forest Science*, 71: 627-  
549 631.
- 550 Brodrigg, T. J., & Cochard, H. (2009). Hydraulic failure defines the recovery and point of death

551 in water-stressed conifers. *Plant physiology*, 149: 575-584.

552 ~~Burghardt, M., & Riederer, M. (2008). Cuticular transpiration. *Annual Plant Reviews, Biology*~~  
553 ~~of the Plant Cuticle. Blackwell publishing, 23: 292.~~

554 Carnicer, J., Brotons, L., Stefanescu, C., & Penuelas, J. (2012). Biogeography of species  
555 richness gradients: linking adaptive traits, demography and diversification. *Biological*  
556 *Reviews*, 87: 457-479.

557 ~~Chaar, H., & Colin, F. (1999). Impact of late frost on height growth in young sessile oak~~  
558 ~~regenerations. *Annals of forest science*, 56: 417-429.~~

559 ~~Cavender Bares, J., & Bazzaz, F. A. (2000). Changes in drought response strategies with~~  
560 ~~ontogeny in *Quercus rubra*: implications for scaling from seedlings to mature trees.~~  
561 ~~*Oecologia*, 124: 8-18.~~

562 Chapin III, F. S., Autumn, K., & Pugnaire, F. (1993). Evolution of suites of traits in response  
563 to environmental stress. *American Naturalist*, S78-S92.

564 Charra-Vaskou, K., Badel, E., Charrier, G. *et al.*, (2016). Cavitation and water fluxes driven by  
565 ice water potential in *Juglans regia* during freeze-thaw cycles. *Journal of Experimental*  
566 *Botany*, 67: 739-750.

567 Charrier, G., Bonhomme, M., Lacoite, A., & Améglio, T. (2011). Are budburst dates,  
568 dormancy and cold acclimation in walnut trees (*Juglans regia* L.) under mainly genotypic  
569 or environmental control?. *International Journal of Biometeorology*, 55: 763-774.

570 Charrier, G., Cochard, H., & Améglio, T. (2013a). Evaluation of the impact of frost resistances  
571 on potential altitudinal limit of trees. *Tree Physiology*, 33: 891-902.

572 Charrier G., Poirier M., Bonhomme M., Lacoite A., Améglio T. (2013b). Frost acclimation in  
573 different organs of walnut trees *Juglans regia* L.: How to link physiology and modelling?  
574 *Tree Physiology* 33: 1229-1241.

575 Charrier, G., Charra-Vaskou, K., Kasuga, J., Cochard, H., Mayr, S., & Améglio, T. (2014).  
576 Freeze-thaw stress: effects of temperature on hydraulic conductivity and ultrasonic activity  
577 in ten woody angiosperms. *Plant Physiology*, 164: 992-998.

578 Charrier, G., Ngao, J., Saudreau M., & Améglio, T. (2015) Effects of environmental factors and  
579 management practices on microclimate, winter physiology, and frost resistance in trees.  
580 *Frontiers in Plant Science*. 6: 259.

581 Charrier, G., Torres-Ruiz, J. M., Badel, E. *et al.*, (2016). Evidence for hydraulic vulnerability  
582 segmentation and lack of xylem refilling under tension. *Plant Physiology*, 172: 16571668.

583 Charrier, G., Nolf, M., Leitinger, G. *et al.*, (2017). Monitoring of freezing dynamics in trees: a  
584 simple phase shift causes complexity. *Plant Physiology*, 173: 2196-2207.

585 ~~Charrier, G., Chuine, I., Bonhomme, M., & Améglio, T. (2018a). Assessing frost damages using~~  
586 ~~dynamic models in walnut trees: exposure rather than vulnerability controls frost risks.~~  
587 ~~*Plant, Cell & Environment*, 41: 1008-1021.~~

588 Charrier, G., Delzon, S., Domec, J. C. *et al.*, (2018a2018b). Drought will not leave your glass  
589 empty: Low risk of hydraulic failure revealed by long-term drought observations in world's  
590 top wine regions. *Science Advances*, 4: eaao6969.

591 Charrier, G., Lacoite, A., & Améglio, T. (2018b2018c). Dynamic modeling of carbon  
592 metabolism during the dormant period accurately predicts the changes in frost hardiness  
593 in walnut trees *Juglans regia* L.. *Frontiers in Plant Science*, 9: 1746.

594 ~~Charrier, G., Chuine, I., Bonhomme, M., & Améglio, T. (2018c). Assessing frost damages using~~  
595 ~~dynamic models in walnut trees: exposure rather than vulnerability controls frost risks.~~  
596 ~~*Plant, Cell & Environment*, 41: 1008-1021.~~

597 Cheaib, A., Badeau, V., Boe, J. *et al.*, (2012). Climate change impacts on tree ranges: model  
598 intercomparison facilitates understanding and quantification of uncertainty. *Ecology*  
599 *letters*, 15: 533-544.

600 Choat B, Jansen S, Brodribb TJ *et al.*, (2012). Global convergence in the vulnerability of forests

601 to drought. *Nature*, 491: 752–5

602 Choat, B., Brodribb, T. J., Brodersen, C. R., Duursma, R. A., López, R., & Medlyn, B. E.  
603 (2018). Triggers of tree mortality under drought. *Nature*, 558: 531.

604 Christensen-Dalsgaard, K. K., & Tyree, M. T. (2014). Frost fatigue and spring recovery of  
605 xylem vessels in three diffuse-porous trees in situ. *Plant, Cell & Environment*, 37:  
606 10741085.

607 Chuine, I., Bonhomme, M., Legave, J. M., García de Cortázar-Atauri, I., Charrier, G., Lacoite,  
608 A., & Améglio, T. (2016). Can phenological models predict tree phenology accurately in  
609 the future? The unrevealed hurdle of endodormancy break. *Global Change Biology*. 22:  
610 3444-3460.

611 ~~Chuine, I., & Beaubien, E. G. (2001). Phenology is a major determinant of tree species range.  
612 *Ecology Letters*, 4: 500-510.~~

613 Cinotti, B. (1991). Investigation of the intrinsic properties of wood for explaining the  
614 occurrence of frost-crack in *Quercus petraea* Liebl. and *Quercus robur* L. *Annales des  
615 Sciences Forestières*, 48: 453-68.

616 Cochard, H., Cruiziat, P., & Tyree, M. T. (1992). Use of positive pressures to establish  
617 vulnerability curves: further support for the air-seeding hypothesis and implications for  
618 pressure-volume analysis. *Plant physiology*, 100: 205-209.

619 Cox, R. M., & Malcolm, J. W. (1997). Effects of duration of a simulated winter thaw on dieback  
620 and xylem conductivity of *Betula papyrifera*. *Tree Physiology*, 17: 389-396.

621 Cox, R. M., & Zhu, X. B. (2003). Effects of simulated thaw on xylem cavitation, residual  
622 embolism, spring dieback and shoot growth in yellow birch. *Tree Physiology*, 23: 615-624.

623 D'Andrea, E., Rezaie, N., Battistelli, A. ~~et al.~~ (2019). Winter's bite: Beech trees survive  
624 complete defoliation due to spring late-frost damage by mobilizing old C reserves. *New  
625 Phytologist*.

626 Delpierre, N., Soudani, K., Francois, C. *et al.*, (2009). Exceptional carbon uptake in European  
627 forests during the warm spring of 2007: a data-model analysis. *Global Change Biology*,  
628 15: 1455-1474.

629 Delpierre, N., Vitasse, Y., Chuine, I., Guillemot, J., Bazot, S., & Rathgeber, C. B. (2016).  
630 Temperate and boreal forest tree phenology: from organ-scale processes to terrestrial  
631 ecosystem models. *Annals of Forest Science*, 73: 5-25.

632 Dowgert, M. F., & Steponkus, P. L. (1984). Behavior of the plasma membrane of isolated  
633 protoplasts during a freeze-thaw cycle. *Plant Physiology*, 75: 1139-1151.

634 Dufrêne, E., Davi, H., François, C., Le Maire, G., Le Dantec, V., & Granier, A. (2005).  
635 Modelling carbon and water cycles in a beech forest: Part I: Model description and  
636 uncertainty analysis on modelled NEE. *Ecological Modelling*, 185: 407-436.

637 Earles, J. M., Stevens, J. T., Sperling, O., Orozco, J., North, M. P., & Zwieniecki, M. A. (2018).  
638 Extreme mid-winter drought weakens tree hydraulic-carbohydrate systems and slows  
639 growth. *New Phytologist*, 219: 89-97.

640 Erwin, D. H. (2009). Climate as a driver of evolutionary change. *Current Biology*, 19: R575–  
641 R583.

642 Ewers, F. W., Améglio, T., Cochard, H. *et al.*, (2001). Seasonal variation in xylem pressure of  
643 walnut trees: root and stem pressures. *Tree Physiology*, 21: 1123-1132.

644 Francon, L., Corona, C., Till-Bottraud, I., Choler, P., Carlson, B. Z., Charrier, G., ... & Lopez-  
645 Saez, J. (2020). Assessing the effects of earlier snow melt-out on alpine shrub growth: The  
646 sooner the better?. *Ecological Indicators*, 115: 106455.

647 Fu, Y. S., Campioli, M., Vitasse, Y. *et al.*, (2014). Variation in leaf flushing date influences  
648 autumnal senescence and next year's flushing date in two temperate tree species.  
649 *Proceedings of the National Academy of Sciences*, 111: 7355-7360.

650 Fuchigami, L. H., Weiser, C. J., Kobayashi, K., Timmis, R., & Gusta, L. V. (1982). A degree



651 growth stage (GS) model and cold acclimation in temperate woody plants. *Plant cold*  
652 *hardiness and freezing stress. Mechanisms and crop implications*, 2: 93-116.

653 Galvez, D. A., Landhäusser, S. M., & Tyree, M. T. (2013). Low root reserve accumulation  
654 during drought may lead to winter mortality in poplar seedlings. *New Phytologist*, 198:  
655 139-148.

656 [Ghesquière, M., Barre, P., Durand, J. L., Litrico, I., Sampoux, J. P., Mournet, P., & Volaire, F.](#)  
657 [\(2014\). Genetic Response to Climate Scenarios in \*Dactylis\* and \*Festuca\* of Temperate](#)  
658 [Versus Mediterranean Origin. In \*Quantitative Traits Breeding for Multifunctional\*](#)  
659 [Grasslands and Turf \(pp. 41-48\). Springer, Dordrecht](#)

660 Granda, E., & Camarero, J. J. (2017). Drought reduces growth and stimulates sugar  
661 accumulation: new evidence of environmentally driven non-structural carbohydrate use.  
662 *Tree Physiology*, 37: 997-1000.

663 Guisan, A., & Thuiller, W. (2005). Predicting species distribution: offering more than simple  
664 habitat models. *Ecology Letters*, 8: 993-1009.

665 Gusta, L. V., Tyler, N. J., and Chen, T. H. H. (1983). Deep undercooling in woody taxa growing  
666 north of the 40°C isotherm. *Plant Physiology*, 72: 122-128.

667 [Hacker, J., & Neuner, G. \(2008\). Ice propagation in dehardened alpine plant species studied by](#)  
668 [infrared differential thermal analysis \(IDTA\). \*Arctic, Antarctic, and Alpine Research\*, 40:](#)  
669 [660-670.](#)

670 Hänninen, H., & Tanino, K. (2011). Tree seasonality in a warming climate. *Trends in Plant*  
671 *Science*, 16: 412-416.

672 Hänninen, H. (1991). Does climatic warming increase the risk of frost damage in northern  
673 trees?. *Plant, Cell & Environment*, 14: 449-454.

674 [Hänninen, H. \(2016\). \*Boreal and temperate trees in a changing climate\*. Biometeorology.](#)  
675 [Springer, Dordrecht.](#)

676 [Hare, D. E., & Sorensen, C. M. \(1987\). The density of supercooled water. II. Bulk samples](#)  
677 [cooled to the homogeneous nucleation limit. \*The Journal of Chemical Physics\*, 87:](#)  
678 [4840-4845.](#)

679 Heide, O. M. (2003). High autumn temperature delays spring bud burst in boreal trees,  
680 counterbalancing the effect of climatic warming. *Tree Physiology*, 23: 931-936.

681 [Hoch, G., & Körner, C. \(2009\). Growth and carbon relations of tree line forming conifers at](#)  
682 [constant vs. variable low temperatures. \*Journal of Ecology\*, 97: 57-66.](#)

683 [Holtz, V., Bertrand, C. E., Anisimov, M. A., & Sengers, J. V. \(2012\). Thermodynamics of](#)  
684 [supercooled water. \*The Journal of Chemical Physics\*, 136: 094507.](#)

685 Intergovernmental Panel on Climate Change. (2012). Managing the Risks of Extreme Events  
686 and Disasters to Advance Climate Change Adaptation. A Special Report of Working  
687 Groups I and II of the Intergovernmental Panel on Climate Change [Field, C.B., V.

688 Intergovernmental Panel on Climate Change. (2014). Climate Change 2014—Impacts,  
689 Adaptation and Vulnerability: Regional Aspects. Cambridge University Press.

690 Kalcsits, L. A., Silim, S., & Tanino, K. (2009). Warm temperature accelerates short  
691 photoperiod-induced growth cessation and dormancy induction in hybrid poplar (*Populus*×  
692 spp.). *Trees*, 23: 971-979.

693 Kasuga, J., Charrier, G., Uemura, M., & Améglio, T. (2015). Characteristics of ultrasonic  
694 acoustic emissions from walnut branches during freeze-thaw-induced embolism  
695 formation. *Journal of Experimental Botany*, 66: 1965-1975.

696 Keenan, T. F., & Richardson, A. D. (2015). The timing of autumn senescence is affected by the  
697 timing of spring phenology: implications for predictive models. *Global Change Biology*,  
698 21: 2634-2641.

699 Kellomäki, S., Väisänen, H., Hänninen, H., Kolström, T., Lauhanen, R., Mattila, U., & Pajari,  
700 B. (1992). A simulation model for the succession of the boreal forest ecosystem.

701 Kellomäki, S., Hanninen, H., & Kolstrom, M. (1995). Computations on frost damage to Scots  
702 pine under climatic warming in boreal conditions. *Ecological Applications*, 5: 42-52.

703 Klanderud, K., Vandvik, V., & Goldberg, D. (2015). The importance of biotic vs. abiotic drivers  
704 of local plant community composition along regional bioclimatic gradients. *PloS one*, 10:  
705 e0130205.

706 Knapp, A. K., Fay, P. A., Blair, J. M. *et al.*, (2002). Rainfall variability, carbon cycling, and  
707 plant species diversity in a mesic grassland. *Science*, 298: 2202-2205.

708 Koerner, C. (2015). Paradigm shift in plant growth control. *Current Opinion in Plant Biology*,  
709 25, 107-114.

710 Koerner, C., Basler, D., Hoch, G. *et al.*, (2016). Where, why and how? Explaining the low-  
711 temperature range limits of temperate tree species. *Journal of Ecology*, 104: 10761088.

712 Koerner, C. (1998). A re-assessment of high elevation treeline positions and their explanation.  
713 *Oecologia* 115: 445-459.

714 ~~Kramer, K., & Hänninen, H. (2009). The annual cycle of development of trees and process-based  
715 modelling of growth to scale up from the tree to the stand. In *Phenology of ecosystem  
716 processes* (pp. 201-227). Springer, New York, NY.~~

717 Kreyling, J., Wiesenberg, G. L., Thiel, D. *et al.*, (2012). Cold hardiness of *Pinus nigra* Arnold  
718 as influenced by geographic origin, warming, and extreme summer drought. *Environmental  
719 and Experimental Botany*, 78: 99-108.

720 Kubler, H. (1983). Mechanism of frost crack formation in trees—a review and synthesis. *Forest  
721 Science*, 29: 559-568.

722 Larcher, W. (2005). Climatic constraints drive the evolution of low temperature resistance in  
723 woody plants. *Journal of Agricultural Meteorology*, 61: 189-202.

724 Laube, J., Sparks, T. H., Estrella, N., Höfler, J., Ankerst, D. P., & Menzel, A. (2014). Chilling  
725 outweighs photoperiod in preventing precocious spring development. *Global Change  
726 Biology*, 20: 170-182.

727 Leinonen, I. (1996). A simulation model for the annual frost hardiness and freeze damage of  
728 Scots pine. *Annals of Botany*, 78: 687-693.

729 Leinonen, I., & Hänninen, H. (2002). Adaptation of the timing of bud burst of Norway spruce  
730 to temperate and boreal climates. *Silva Fennica*, 36: 695-701.

731 Lemoine, D., Granier, A., & Cochard, H. (1999). Mechanism of freeze-induced embolism in  
732 *Fagus sylvatica* L. *Trees*, 13: 206-210.

733 Lenoir, J., Gégout, J. C., Marquet, P. A., De Ruffray, P., & Brisse, H. (2008). A significant  
734 upward shift in plant species optimum elevation during the 20th century. *Science*, 320:  
735 1768-1771.

736 Lens, F., Sperry, J. S., Christman, M. A., Choat, B., Rabaey, D., & Jansen, S. (2011). Testing  
737 hypotheses that link wood anatomy to cavitation resistance and hydraulic conductivity in  
738 the genus *Acer*. *New Phytologist*, 190: 709-723.

739 Levitt, J. (1980). *Responses of Plants to Environmental Stress, Volume 1: Chilling, Freezing,  
740 and High Temperature Stresses*. Academic Press.

741 Li, S., Lens, F., Espino, S. *et al.*, (2016). Intervessel pit membrane thickness as a key  
742 determinant of embolism resistance in angiosperm xylem. *IAWA Journal*, 37: 152-171.

743 Lines, E.R., Coomes, D.A., Purves, D.W. (2010) Influences of forest structure, climate and  
744 species composition on tree mortality across the eastern US. (ed Hector A). *PloS One*, 5,  
745 e13212

746 Lintunen, A., Mayr, S., Salmon, Y., Cochard, H., & Hölttä, T. (2018). Drivers of apoplastic  
747 freezing in gymnosperm and angiosperm branches. *Ecology and evolution*, 8: 333-343.

748 Liu, G., Chen, X., Fu, Y., & Delpierre, N. (2019). Modelling leaf coloration dates over  
749 temperate China by considering effects of leafy season climate. *Ecological modelling*, 394:  
750 34-43.

751 Loehle, C. (1998). Height growth rate tradeoffs determine northern and southern range limits  
752 for trees. *Journal of Biogeography*, 25: 735-742.

753 Lovisolo C, Schubert A (1998) Effects of water stress on vessel size and xylem hydraulic  
754 conductivity in *Vitis vinifera* L. *Journal of Experimental Botany* 49: 693–700

755 Marchin, R., Zeng, H., & Hoffmann, W. (2010). Drought-deciduous behavior reduces nutrient  
756 losses from temperate deciduous trees under severe drought. *Oecologia*, 163: 845-854.

757 [Martinez-Medina, A., Flors, V., Heil, M., Mauch-Mani, B., Pieterse, C. M., Pozo, M. J., ... &](#)  
758 [Conrath, U. \(2016\). Recognizing plant defense priming. \*Trends in Plant Science\*, 21: 818-](#)  
759 [822.](#)

760 [Martinez-Vilalta, J., Anderegg, W. R., Sapes, G., & Sala, A. \(2019\). Greater focus on water](#)  
761 [pools may improve our ability to understand and anticipate drought-induced mortality in](#)  
762 [plants. \*New Phytologist\*, 223: 22-32.](#)

763 Martin, StPaul, N., Delzon, S., & Cochard, H. (2017). Plant resistance to drought depends on  
764 timely stomatal closure. *Ecology Letters*, 20: 1437-1447.

765 Maurya, J.P., & Bhalerao, R.P. (2017). Photoperiod-and temperature-mediated control of  
766 growth cessation and dormancy in trees: a molecular perspective. *Annals of Botany*, 120:  
767 351-360.

768 Mayr, S., & Charra, Vaskou, K. (2007). Winter at the alpine timberline causes complex within  
769 tree patterns of water potential and embolism in *Picea abies*. *Physiologia plantarum*, 131:  
770 131-139.

771 Mayr, S., Gruber, A., & Bauer, H. (2003). Repeated freeze–thaw cycles induce embolism in  
772 drought stressed conifers (Norway spruce, stone pine). *Planta*, 217: 436-441.

773 Mayr, S., Hacke, U., Schmid, P., Schwienbacher, F., & Gruber, A. (2006). Frost drought in  
774 conifers at the alpine timberline: xylem dysfunction and adaptations. *Ecology*, 87:  
775 31753185.

776 Mayr, S., Cochard, H., Améglio, T., & Kikuta, S. B. (2007). Embolism formation during  
777 freezing in the wood of *Picea abies*. *Plant Physiology*, 143: 60-67.

778 Mayr, S., Schmid, P., Laur, J., Rosner, S., Charra-Vaskou, K., Dämon, B., & Hacke, U. G.  
779 (2014). Uptake of water via branches helps timberline conifers refill embolized xylem in  
780 late winter. *Plant Physiology*, 164: 1731-1740.

781 Mayr, S., Schmid, P., Beikircher, B., Feng, F., & Badel, E. (2019). Die hard: timberline conifers  
782 survive annual winter embolism. *New Phytologist*

783 McDowell, N., Pockman, W. T., Allen, C. D. *et al.*, (2008). Mechanisms of plant survival and  
784 mortality during drought: why do some plants survive while others succumb to drought?.  
785 *New Phytologist*, 178: 719-739.

786 McDowell, N. G. (2011). Mechanisms linking drought, hydraulics, carbon metabolism, and  
787 vegetation mortality. *Plant Physiology*, 155: 1051-1059.

788 Menzel, A., Helm, R., & Zang, C. (2015). Patterns of late spring frost leaf damage and recovery  
789 in a European beech (*Fagus sylvatica* L.) stand in south-eastern Germany based on repeated  
790 digital photographs. *Frontiers in Plant Science* 6.

791 Menzel, A., Sparks, T. H., Estrella, N. *et al.-et al.*, (2006). European phenological response to  
792 climate change matches the warming pattern. *Global Change Biology*, 12: 1969-1976.

793 Misson, L., Degueldre, D., Collin, C., Rodriguez, R., Rocheteau, A., Ourcival, J. M., &  
794 Rambal, S. (2011). Phenological responses to extreme droughts in a Mediterranean forest.  
795 *Global Change Biology*, 17: 1036-1048.

796 Morin, X., Améglio, T., Ahas, R. *et al.*, (2007). Variation in cold hardiness and carbohydrate  
797 concentration from dormancy induction to bud burst among provenances of three European  
798 oak species. *Tree Physiology*, 27: 817-825.

799 Morin, X., & Chuine, I. (2005). Sensitivity analysis of the tree distribution model PHENOFIT  
800 to climatic input characteristics: implications for climate impact assessment. *Global*

801 Change Biology, 11: 1493-1503.

802 Nardini, A., Gullo, M. A. L., & Salleo, S. (2011). Refilling embolized xylem conduits: is it a  
803 matter of phloem unloading?. *Plant Science*, 180: 604-611.

804 O'Brien, M. J., Leuzinger, S., Philipson, C. D., Tay, J., & Hector, A. (2014). Drought survival  
805 of tropical tree seedlings enhanced by non-structural carbohydrate levels. *Nature Climate*  
806 *Change*, 4: 710-714.

807 Palonen, P., & Buszard, D. (1997). Current state of cold hardiness research on fruit crops.  
808 *Canadian Journal of Plant Science*, 77: 399-420.

809 Parmesan, C., & Yohe, G. (2003). A globally coherent fingerprint of climate change impacts  
810 across natural systems. *Nature*, 421: 37-42.

811 Poirier, M., Lacoite, A., & Améglio, T. (2010). A semi-physiological model of cold hardening  
812 and dehardening in walnut stem. *Tree Physiology*, 30: 1555-1569.

813 Porporato, A., Daly, E., & Rodriguez-Iturbe, I. (2004). Soil water balance and ecosystem  
814 response to climate change. *The American Naturalist*, 164: 625-632.

815 ~~Pramsohler, M., Haeker, J., & Neuner, G. (2012). Freezing pattern and frost killing temperature~~  
816 ~~of apple (*Malus domestica*) wood under controlled conditions and in nature. *Tree*~~  
817 ~~*Physiology*, 32: 819-828.~~

818 Rinne, P., Hänninen, H., Kaikuranta, P., Jalonen, J. E., & Repo, T. (1997). Freezing exposure  
819 releases bud dormancy in *Betula pubescens* and *B. pendula*. *Plant, Cell & Environment*,  
820 20: 1199-1204.

821 Rinne, P. L., Kaikuranta, P. M., & Van Der Schoot, C. (2001). The shoot apical meristem  
822 restores its symplasmic organization during chilling-induced release from dormancy. *The*  
823 *Plant Journal*, 26: 249-264.

824 Rohde, A., Bastien, C., & Boerjan, W. (2011). Temperature signals contribute to the timing of  
825 photoperiodic growth cessation and bud set in poplar. *Tree Physiology*, 31: 472-482.

826 Ruiz-Benito, P., Lines, E.R., Gómez-Aparicio, L., Zavala, M.A., Coomes, D.A. (2013). Patterns  
827 and drivers of tree mortality in Iberian forests: climatic effects are modified by competition.  
828 *PloS One*.

829 Rummukainen, M. (2012). Changes in climate and weather extremes in the 21st century. *Wiley*  
830 *Interdisciplinary Reviews: Climate Change*, 3: 115-129.

831 Sala, A., Woodruff, D. R., & Meinzer, F. C. (2012). Carbon dynamics in trees: feast or famine?.  
832 *Tree Physiology*, 32: 764-775.

833 Savvides, A., Ali, S., Tester, M., & Fotopoulos, V. (2016). Chemical priming of plants against  
834 multiple abiotic stresses: mission possible?. *Trends in plant science*, 21: 329-340.

835 Schaber, J., & Badeck, F. W. (2005). Plant phenology in Germany over the 20th century.  
836 *Regional Environmental Change*, 5: 37-46.

837 Schuster, C., Kirchner, M., Jakobi, G., & Menzel, A. (2014). Frequency of inversions affects  
838 senescence phenology of *Acer pseudoplatanus* and *Fagus sylvatica*. *International Journal*  
839 *of Biometeorology*, 58: 485-498.

840 ~~Schwilk, D. W., & Ackerly, D. D. (2005). Is there a cost to resprouting? Seedling growth rate~~  
841 ~~and drought tolerance in sprouting and nonsprouting *Ceanothus* (*Rhamnaceae*). *American*~~  
842 ~~*Journal of Botany*, 92: 404-410.~~

843 Shi, P., Koerner, C., & Hoch, G. (2008). A test of the growth limitation theory for alpine tree  
844 line formation in evergreen and deciduous taxa of the eastern Himalayas. *Functional*  
845 *Ecology*, 22: 213-220.

846 Sierra-Almeida, A., Reyes-Bahamonde, C., & Cavieres, L. A. (2016). Drought increases the  
847 freezing resistance of high-elevation plants of the Central Chilean Andes. *Oecologia*, 113.

848 Siminovitch, D., & Cloutier, Y. (1983). Drought and freezing tolerance and adaptation in plants:  
849 some evidence of near equivalences. *Cryobiology*, 20: 487-503.

850 Sperry, J. S., & Sullivan, J. E. (1992). Xylem embolism in response to freeze-thaw cycles and

851 water stress in ring-porous, diffuse-porous, and conifer species. *Plant Physiology*, 100:  
852 605-613.

853 Sperry, J. S., Holbrook, N.M., Zimmermann, M. H., & Tyree, M. T. (1987). Spring filling of  
854 xylem vessels in wild grapevine. *Plant Physiology*, 83: 414-417.

855 Sperry, J. S., Nichols, K. L., Sullivan, J. E., & Eastlack, S.E. (1994). Xylem embolism in ring-  
856 porous, diffuse-porous, and coniferous trees of northern Utah and interior Alaska. *Ecology*,  
857 75: 1736-1752.

858 Sperry, J. S., Adler, F. R., Campbell, G. S., & Comstock, J. P. (1998). Limitation of plant water  
859 use by rhizosphere and xylem conductance: results from a model. *Plant, Cell &  
860 Environment*, 21: 347-359.

861 Stockinger, E. J., Gilmour, S. J., & Thomashow, M. F. (1997). *Arabidopsis thaliana* CBF1  
862 encodes an AP2 domain-containing transcriptional activator that binds to the Crepeat/DRE,  
863 a cis-acting DNA regulatory element that stimulates transcription in response to low  
864 temperature and water deficit. *Proceedings of the National Academy of Sciences*, 94: 1035-  
865 1040.

866 Svystun, T., Bhalerao, R. P., & Jönsson, A. M. (2019). Modelling *Populus* autumn phenology:  
867 The importance of temperature and photoperiod. *Agricultural and Forest Meteorology*,  
868 271: 346-354.

869 Tanino, K. K., Kalcsits, L., Silim, S., Kendall, E., & Gray, G. R. (2010). Temperature-driven  
870 plasticity in growth cessation and dormancy development in deciduous woody plants: a  
871 working hypothesis suggesting how molecular and cellular function is affected by  
872 temperature during dormancy induction. *Plant Molecular Biology*, 73: 49-65.

873 Timmis, R., & Tanaka Y. 1976. Effects of container density and plant water stress on growth  
874 and cold hardiness of Douglas-fir seedlings. *For. Sci.* 22:167-172.

875 ~~Wabba, G. 1990. Spline models for observational data.~~

876 Tranquillini W. 1979. Physiological ecology of the alpine timberline. - Ecological Studies 31,  
877 Springer Verlag Berlin, Heidelberg, New York.

878 Tylewicz, S., Petterle, A., Marttila, S. *et al.*, (2018). Photoperiodic control of seasonal growth  
879 is mediated by ABA acting on cell-cell communication. *Science*, eaan8576.

880 Tyree, M. T., Cochard, H., Cruiziat, P., Sinclair, B., & Ameglio, T. (1993). Drought-induced  
881 leaf shedding in walnut: evidence for vulnerability segmentation. *Plant, Cell &  
882 Environment*, 16: 879-882.

883 Tyree, M. T., & Sperry, J. S. (1989). Vulnerability of xylem to cavitation and embolism. *Annual  
884 Review of Plant Biology*, 40: 19-36.

885 ~~van Genuchten, M.T. (1980). A closed-form equation for predicting the hydraulic conductivity  
886 of unsaturated soils. *Soil science society of America journal*, 44: 892-898.~~

887 Vanoni, M., Bugmann, H., Nötzli, M., & Bigler, C. (2016). Drought and frost contribute to  
888 abrupt growth decreases before tree mortality in nine temperate tree species. *Forest  
889 Ecology and Management*, 382: 51-63.

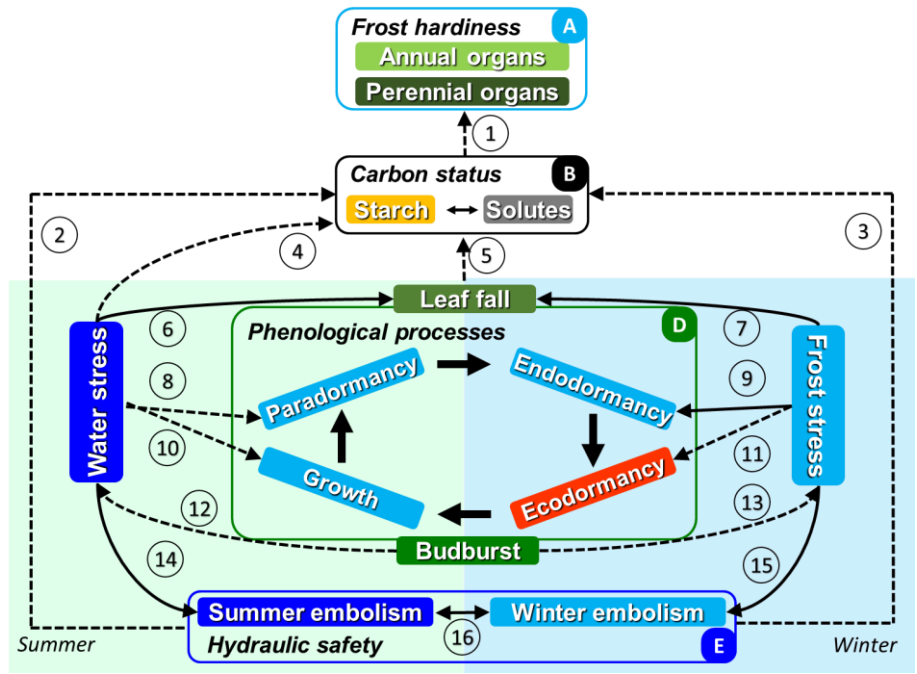
890 Vitasse, Y., Porté, A. J., Kremer, A., Michalet, R., & Delzon, S. (2009). Responses of canopy  
891 duration to temperature changes in four temperate tree species: relative contributions of  
892 spring and autumn leaf phenology. *Oecologia*, 161: 187-198.

893 Walter, J., Jentsch, A., Beierkuhnlein, C., & Kreyling, J. (2013). Ecological stress memory and  
894 cross stress tolerance in plants in the face of climate extremes. *Environmental and  
895 Experimental Botany*, 94, 3-8.

896 Wargo, P. M. (1981). Defoliation, dieback and mortality. The gypsy moth: research toward  
897 integrated pest management. Edited by CC Doane and ML McManus. US Dep. Agric.  
898 Tech. Bull, 1584, 240-248.

899 Wargo, P. M. (1996). Consequences of environmental stress on oak: predisposition to  
900 pathogens. *Annales des Sciences Forestières*, 53: 359-368.

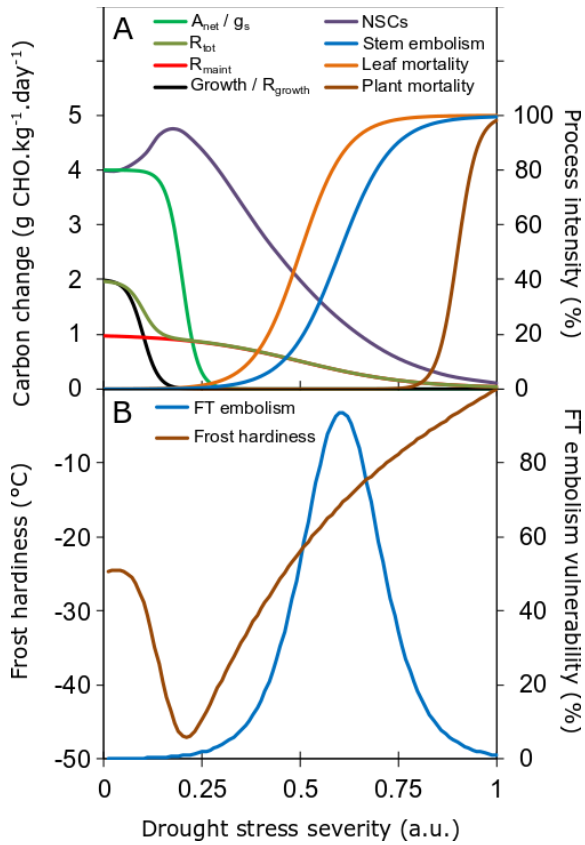
- 901 Welling, A., Moritz, T., Palva, E. T., & Junttila, O. (2002). Independent activation of cold  
902 acclimation by low temperature and short photoperiod in hybrid aspen. *Plant Physiology*,  
903 129: 1633-1641.
- 904 Wolf, S., Keenan, T. F., Fisher, J. B., Baldocchi, D. D., Desai, A. R., Richardson, A. D., ... &  
905 Peters, W. (2016). Warm spring reduced carbon cycle impact of the 2012 US summer  
906 drought. *Proceedings of the National Academy of Sciences*, 201519620.
- 907 Wong, B. L., Baggett, K. L., & Rye, A. H. (2009). Cold-season patterns of reserve and soluble  
908 carbohydrates in sugar maple and ice-damaged trees of two age classes following drought.  
909 *Botany*, 87: 293-305.
- 910 Xie, Y., Wang, X., & Silander, J. A. (2015). Deciduous forest responses to temperature,  
911 precipitation, and drought imply complex climate change impacts. *Proceedings of the*  
912 *National Academy of Sciences*, 112(12), 3633-3638.
- 913 Yamaguchi-Shinozaki, K., & Shinozaki, K. (1994). A novel cis-acting element in an  
914 Arabidopsis gene is involved in responsiveness to drought, low-temperature, or high-salt  
915 stress. *The Plant Cell*, 6: 251-264.
- 916 Zhu, X. B., Cox, R. M., & Arp, P. A. (2000). Effects of xylem cavitation and freezing injury on  
917 dieback of yellow birch (*Betula alleghaniensis*) in relation to a simulated winter thaw. *Tree*  
918 *Physiology*, 20: 541-547.
- 919 Zweifel, R., Zimmermann, L., & Newbery, D. M. (2005). Modeling tree water deficit from  
microclimate: an approach to quantifying drought stress. *Tree Physiology*, 25: 147-156.



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937

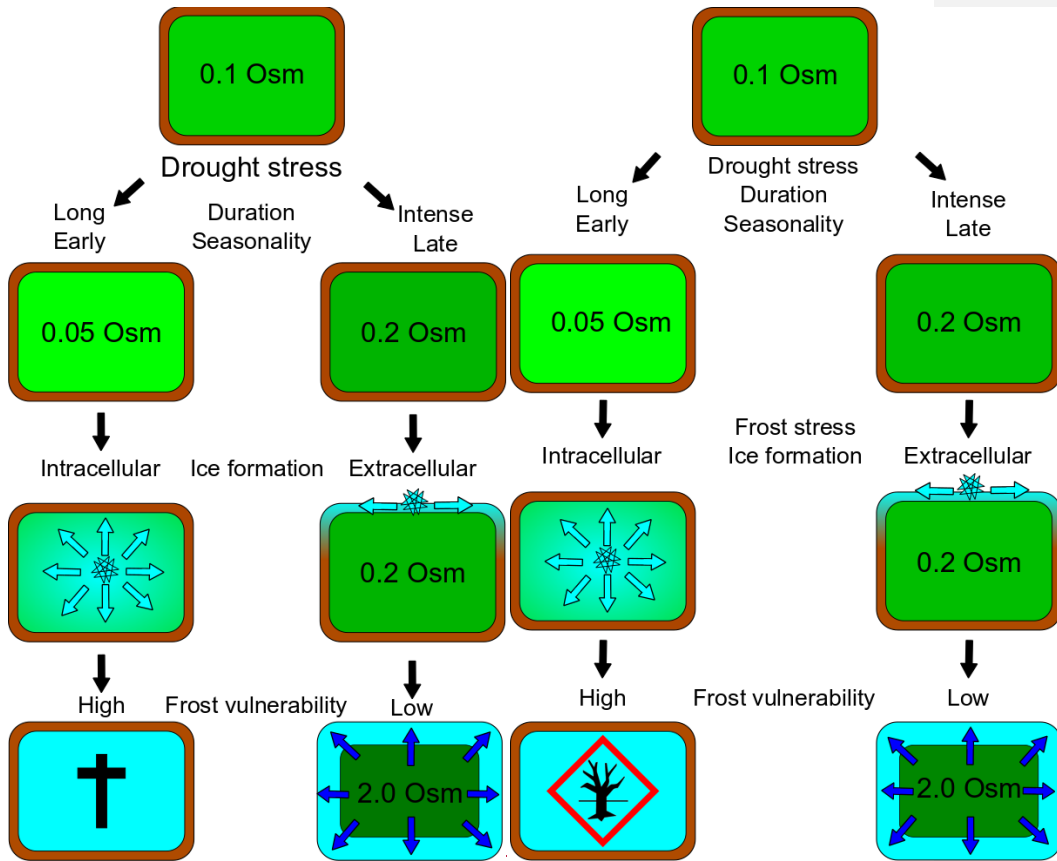
921 (mainly during winter period). Positive (arrowsolid) and negative (barsdashed) effects of water  
 922 stress (mainly during summer period) and frost stresses (mainly during winter period) are  
 923 expected on three functional components, namely carbon status, annual cycle phenological  
 924 processes and hydraulic architecture safety. Positive relations between timing events  
 925 (phenological stages such as leaf fall and budburst) and other processes indicate that earlier  
 926 event induce higher level, and vis-versa). All represented processes are interrelated either  
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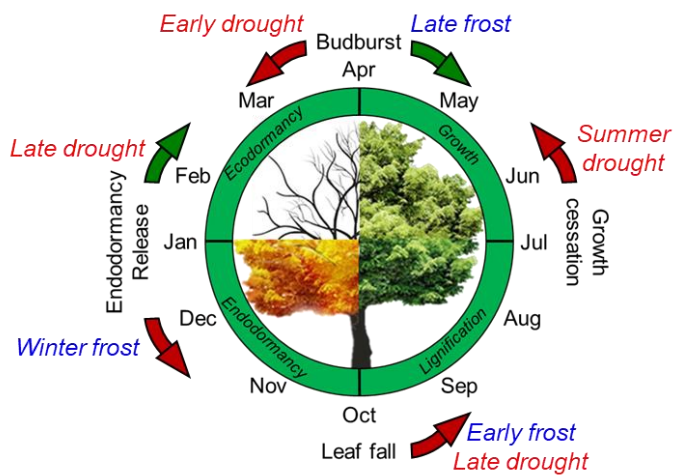
939 **Figure 2. A.** Processes and pools affected by drought stress severity-intensity (Photosynthesis  
 940  $A_{net}$ , stomatal conductance  $g_s$ , respiration (growth  $R_{growth}$ , maintenance  $R_{maint}$  and total  $R_{tot}$ ),  
 941 non structural carbohydrates (NSCs), stem embolism, leaf and plant mortality. Growth being  
 942 more sensitive to water stress than photosynthesis, and respiration, the NSCs pool initially  
 943 increases (McDowell, 2011). After photosynthesis declines, NSCs decline whereas leaf  
 944 mortality, embolism and plant mortality increase (Charrier *et al.*, 2018b). **B.** Potential effect  
 945 generated by drought stress on frost hardiness of living cell (through the non-linear relation  
 946 between NSC and water content) and vulnerability to freeze-thaw induced embolism.





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**Figure 3.** Alternative pathways that would explain why contrasted frost vulnerability vulnerabilities may differentially be affected by are observed after previous drought exposure, depending on how Osmolarity of intracellular sap, controlled notably by the ratio between soluble carbohydrates and water content, could be considered as the main driver the ratio between soluble carbohydrates and water content. On the right path, intense and/or late drought is expected to concentrate cell sap, increasing the probability of extra cellular ice nucleation. The low chemical potential of ice would pull water, further increasing cell osmolarity. ~~have been affected~~ On the left path, long and/or early drought prevents timely increase in solutes, through reduction in carbon reserves. Intracellular freezing through is expected to happen more often and at higher freezing temperature, inducing cellular damages.



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 960 **Figure 4.** Hastening (brown arrow) or delaying (green arrow) phenological stages in response  
 961 to drought and frost events. At the center is presented the typical seasonal phenological stages  
 962 in a deciduous tree in Northern hemisphere and, outside the potential effects of  
 963 drought and frost stress on hastening (brown arrow) or delaying (green arrow) phenological  
 964 stages depending on their timing. It should be noted that stress factors can have a lagged effect  
 965 onto phenology (e.g. late drought in late summer delaying winter dormancy release). Early and  
 966 late drought typically happen in May-June and September, respectively. Early and late frost  
 967 typically happen in September-October and April-May, respectively.