From functional to mechanistic: coordination between turgor loss point and traits related to drought tolerance in herbaceous plants

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Abstract

We addressed the recent plea for a use of traits with a direct mechanistic link to drought tolerance to be considered in trait-based and global change ecology. On 122 herbaceous species covering a broad range of water availability conditions in temperate grasslands, we demonstrated the feasibility of the use of turgor loss point (π_{tlp}), a key leaf drought tolerance trait that becomes operational for large-scale studies via the novel osmometry method. We investigated the coordination of this mechanistic trait with other commonly used proxies of drought tolerance. π_{tlp} was not or very weakly coordinated with the first two leading dimension of the global spectrum of form and function, plant height and specific leaf area. π_{tlp} was tightly coordinated with intrinsic water use efficiency and leaf dry matter content. We suggest that stratification by plant functional types and/or accounting for species phylogeny might help to identify trait relationships that may be transferable among different systems.

Introduction

Climate change is predicted to increase the occurrence of extreme drought events, altering the water available for plant growth (Dai 2013; IPCC 2014). Mechanistic understanding of plant growth and survival, as well as changes in community assembly and species distribution in response to changes in water availability can be understood through physiological 'response' traits directly linked to plant water economics (also termed 'hydraulic traits'; Engelbrecht *et al.* 2007; Suding *et al.* 2008; Reich 2014; Griffin-Nolan *et al.* 2018). Unfortunately, those physiological traits have been mostly omitted in large-scale species distribution and community-scale trait studies because their measurement is time- and labor-intensive, and unfeasible under field conditions (for reviews see Bartlett *et al.* 2012b; Griffin-Nolan *et al.*2018). To this end, different morphological response traits (e.g., specific leaf area) have been used as proxies. However, the relationship between these proxies and precipitation (as a dominant measure of water availability for plants) on the community scale is weak, highlighting the need for a more careful trait selection (for review see Griffin-Nolan *et al.* 2018).

A turning point could arise from a recent development of a fast and feasible osmometry method to determine leaf osmotic potential at turgor loss point (π_{tlp}), one of the key physiological response traits. The osmometry method, originally developed on woody species (Bartlett *et al.* 2012a) has been subsequently validated for herbaceous species (Griffin-Nolan *et al.* 2019; Májeková *et al.* 2019), and thus can be applied across a wide range of functional types. Leaves maintaining a more negative π_{tlp} remain turgid at lower soil water potentials allowing them to maintain critical physiological processes (growth and photosynthetic assimilation of CO_2) under drier conditions (Scholander *et al.*1965; Schulze *et al.* 1987; Kubiske & Abrams 1990; Kramer & Boyer 1995; Koide *et al.* 2000; Mitchell *et al.* 2008; Bartlett *et al.* 2012b). This leaf-level dehydration tolerance (*sensu* Volaire 2018) trait can be scaled-up to a whole-plant drought tolerance (*sensu* Noy-Meir 1973), a link well documented in woody species (Lenz *et al.* 2006; Baltzer *et al.* 2008; Bartlett *et al.* 2012b, 2016b). It is important to note that we are referring to drought tolerance as the ability of plants to maintain growth under decreased water availability (*sensu* Noy-Meir 1973), as opposed to drought avoidance and drought escape). π_{tlp} also clearly relates to other leaf-level hydraulic traits, such as leaf water potential at 50% loss of hydraulic and stomatal conductance (Bartlett *et al.* 2016a; Farrell *et al.* 2017; Griffin-Nolan *et al.* 2019).

A question that remains open is the degree of coordination between π_{tlp} as the trait mechanistically linked to drought tolerance and functional traits commonly used as proxies for drought tolerance in trait-based and global change ecology. Coordination among physiological and morphological aspects of plant's phenotype on both leaf and whole-plant level is important for understanding the mechanisms governing plant's responses to water availability and their adaptive nature. To this end, we have selected four traits spanning a spectrum of 'hard' to 'soft' traits on the leaf- and whole-plant level: intrinsic water-use efficiency (iWUE) captured by carbon stable isotope $\delta^{13}C$, leaf dry matter content (LDMC), specific leaf area (SLA, inverse of leaf mass per area, LMA) and plant height.

Water use efficiency represents a ratio of net CO_2 assimilation to stomatal conductance (Farquhar *et al.* 1989; Seibt*et al.* 2008). If water is the main limiting factor, iWUE should increase with decreasing water availability (Araya *et al.* 2010). The carbon stable isotope $\delta^{13}C$ in leaves has been often used as a surrogate of iWUE (Farquhar *et al.* 1989; Seibt*et al.* 2008). Leaf $\delta^{13}C$ is an integrated, long-term measure of the ratio between internal and ambient CO_2 concentrations (C_i/C_a) that reflects the stomatal conductance to CO_2 and thus also drought-induced stomatal closure. This is because plants preferentially assimilate lighter ^{12}C and increasingly use heavier^{13}C when CO_2 (and thus the abundant lighter isotope) is less available. Such a situation typically occurs in leaves with closed stomata under drought stress (Farquhar *et al.*1989).

Specific leaf area (SLA; inverse of leaf mass per area, LMA), has been the most commonly used proxy in relation with water availability gradients, especially in large-scale studies along very long gradients (Westoby 1998; Westoby *et al.* 2002; Wright *et al.* 2004; Díaz *et al.* 2016; Griffin-Nolan *et al.* 2018). It also represents the second axis of the global spectrum of form and function (Díaz *et al.* 2016), an important trait of the leaf economics spectrum (Wright *et al.* 2004), as well as a prominent trait for assessment of plant strategies (Westoby 1998; Pierce *et al.* 2013). Decrease in SLA and increase in leaf dry matter content (LDMC) under decreasing water availability can be explained by the decrease in leaf expansion rates achieved by formation of smaller cells and/or tighter packed cells with less air space in between and/or thicker cell walls in order to reduce water requirements under drought (Garnier *et al.* 2001; Poorter *et al.* 2009). Though LDMC and SLA are often considered interchangeable (e.g. Pierce *et al.* 2013), when leaf thickness as the third player is being considered (Vile *et al.* 2005), their relationship is hyperbolic (Garnier *et al.* 2001; Vendramini*et al.* 2002), potentially resulting in SLA and LDMC diverging in their response to the stress considered (e.g. Hodgson *et al.*2011).

Plant height is the leading dimension of the global spectrum of form and function (Díaz *et al.* 2016), a prominent trait defining plant strategies (Westoby 1998; Pierce *et al.* 2013) and as Westoby*et al.* (2002) states: "[...] the quantitative trait that has been adopted by virtually everyone doing comparative plant ecology". A decrease in plant height along gradients of decreasing water availability relies on two assumptions that act together in the field conditions. First is the simple premise that plant growth and biomass production are reduced under water limitation (Schulze *et al.* 1987). Second is the 'hydraulic limitation hypothesis' (Ryan*et*

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al. 2006), which states that with increasing height increases also the difficulty to supply leaves with water and the risk of embolism, leading to stomata closure, decrease in photosynthesis, and less carbon available for growth.

An important factor to be considered when investigating the coordination among different traits possibly related to the same function (i.e. water availability) is whether such a relationship is biophysically based, and thus transferable to all plant species, or whether it is reflecting the adaptations typical for a certain plant functional type (PFT) or a phylogenetic clade. If two traits are related in the same way within different PFTs, or the relation holds when accounting for phylogenetic relatedness between species, it would suggest that whenever there was an evolutionary change in one trait in one direction, it coincided with an evolutionary change of a second trait in the same direction due to adaptations to the same selection pressure ('selective correlation'; Stebbins 1950; Felsenstein 2004).

Here, we screened 122 temperate grassland species from different plant functional types (forbs and graminoids) across a broad range of water availability conditions in European temperate grasslands (from dry through mesic to wet grasslands). We assessed the relationships between π_{tlp} and other traits related to plant drought tolerance on the leaf and the whole-plant level. We hypothesized that higher physiological leaf-level dehydration tolerance (more negative π_{tlp}) will be coordinated with (1) physiological trait represented by higher intrinsic water use efficiency (iWUE) measured as $\delta^{13}C$; (2) lower values of the specific leaf area (SLA) associated with less acquisitive species within the leaf economics spectrum; (3) leaf-level morphological trait represented by higher leaf dry matter content (LDMC); (4) plant stature represented by smaller maximum vegetative height. We further investigated whether the aforementioned relationships are biophysically-based, and therefore general, or driven by the differences inherent in the major plant functional types and/or inherent in different phylogenetic clades.

Materials and Methods

Plant material and localities

122 herbaceous species were considered from two major plant functional types (forbs, n = 94; graminoids, n = 28) across a broad range of water availability conditions in European temperate grasslands (from dry through mesic to wet grasslands; Supporting Information S1). Species were sampled in the area of the White Carpathians Mts. (SE Czech Republic) and in Ohrazení (S Czech Republic), In both areas, the mean annual temperature is 7–9 °C, mean annual precipitation is 600–920 mm and they have been traditionally managed by mowing once a year in June or July (for details see Lepš 2014; Mudrák *et al.* 2019). These grasslands are renowned for high species diversity (Chytrý et al. 2015). The dominant species in the White Carpathian Mts. meadows are *Bromus erectus* , *Molinia arundinacea* , and *Carex montana* , with common forbs such as *Agrimonia eupatoria* , *Centaurea jacea* , *Geranium sanguineum* , *Hypericum maculatum* , *Leucanthemum vulgare* agg. and *Vincetoxicum hirundinaria* . In the Ohrazení meadow, the dominant grasses are *Molinia caerulea* and *Holcus lanatus* , alongside with about 10 species of sedges and common forbs such as *Angelica sylvestris* , *Betonica officinalis* , *Galium boreale* , *Potentilla erecta* , *Ranunculus acris* and *Lychnis flos-cuculi* .

Trait measurements

π_{tlp} was measured for all 122 species in late May to early June 2016 following the protocol described in detail in (Májeková *et al.* 2019). Prior to measurements, plants were rehydrated overnight in dark cool room with aboveground parts sealed in a plastic bag. For the 101 species from the White Carpathians, specific leaf area (SLA; m^2 kg^{-1}), leaf dry matter content (LDMC; mg g^{-1}) and vegetative height (height; cm) were measured on rehydrated plants at the same time as π_{tlp} following the standardized protocol (Pérez-Harguindeguy *et al.* 2013). For a subset of 20 species from the White Carpathians, $\delta^{13}C$ was measured in a previous campaign in autumn 2006 (described in detail in de Bello *et al.* 2012). For the 37 species from Ohrazení, SLA, LDMC, height and $\delta^{13}C$ were measured in a previous campaign in June 2013 (described in detail in (Mudrák *et al.* 2019) following the standardized protocol (Pérez-Harguindeguy *et al.* 2013).

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Statistical analyses

We used a one-way ANOVA to test for the differences in traits between the plant functional types (PFTs; forb and graminoid). We tested the coordination between π_{tlp} and other functional traits (height, SLA, LDMC and δ^{13}C) with standardised major axis regression, using the package *SMATR* (Warton et al., 2012). First, we considered a model with only traits (hereafter 'All'). Second, we added PFTs with comparison of slopes of the trait relationships between the forbs and graminoids. Third, we considered the phylogenetic relatedness among species. For this, an age-calibrated phylogeny of species (Durka & Michalski 2012) was used to test whether traits exhibited a phylogenetic signal by calculating Pagel's lambda (Pagel 1999) using the package *phytools* (Revell 2012). Since all traits (except for SLA) showed a phylogenetic signal, we applied phylogenetically independent contrasts (Felsenstein 1985) implemented in the package *ape* (Paradis *et al.* 2004) to account for phylogenetic relatedness among species. The contrasts where then used instead of the raw data in the standardised major axis analyses, with intercept forced through zero (Garland *et al.* 1992). It needs to be noted that the PFTs considered here as forbs and graminoids are phylogenetically monophyletic and these two PFTs mainly reflect phylogeny (i.e. a graminoid clade vs. 'the rest'). The goal was not to use phylogeny as addition to the PFTs, but rather as two alternative methods that are commonly used in trait-based ecology. Height was $\log_{10}$-transformed for all analyses to meet the assumption of the homogeneity of variances. All analyses were performed in the R software (R Core Team 2016).

Results

π_{tlp} was considerably more negative in graminoids than in forbs (one-way ANOVA; $F_{1,120} = 26.86$, $P < 0.001$; Fig. 1a). LDMC was significantly higher in graminoids than in forbs (one-way ANOVA; $F_{1,120} = 87.66$, $P < 0.001$; Fig. 1b). There was no difference between the PFTs in their SLA values (one-way ANOVA; $F_{1,120} = 2.72$, $P = 0.1$; Fig. 1c). Graminoids were significantly taller than forbs (one-way ANOVA; $F_{1,120} = 33.41$, $P < 0.001$; Fig. 1d). No difference was observed in the δ^{13}C values (one-way ANOVA; $F_{1,52} = 0.004$, $P = 0.95$; Fig. 1e).

π_{tlp} was strongly negatively related to LDMC (more negative π_{tlp}, i.e. higher leaf-level dehydration tolerance, was coordinated with higher LDMC) and to δ^{13}C (more negative π_{tlp} coordinated with less negative δ^{13}C, i.e. higher intrinsic water-use efficiency) on all levels: (a) when considering all species together (Table 1a, Fig. 2a,e), (b) within the plant functional types with the slopes of the relationships being not significantly different (Table 1b, Fig. 2a,e), (c) as well as after accounting for phylogenetic relatedness (Table 1c, Fig. 2b,f).

The positive relationship between π_{tlp} and SLA (more negative π_{tlp} coordinated with lower SLA) only emerged weakly for forbs (Table 1b, Fig. 2c) and then more strongly after accounting for phylogenetic relatedness (Table 1c, Fig. 2d). π_{tlp} was weakly negatively related to vegetative height (more negative π_{tlp} coordinated with taller species) when all species were considered together (Table 1a, Fig. 2g). However, the relationship disappeared completely within PFTs (Table 1b, Fig. 2g), as well as after accounting for species phylogenetic relatedness (Table 1c, Fig. 2h).

Discussion

On an extensive set of 122 herbaceous species covering a broad range of water availability conditions in European temperate grasslands (from dry through mesic to wet grasslands), we documented a coordination between a physiological trait directly linked to water availability (turgor loss point, π_{tlp}) and traits commonly used as proxies of drought tolerance: intrinsic water use efficiency (iWUE), leaf dry matter content (LDMC), specific leaf area (SLA) and maximum plant height (height). By investigating the relationships across species, within major plant functional types (PFTs, forbs and graminoids) and by accounting for plant phylogenetic relatedness, we were able to distinguish the relationships that were strong and more general (π_{tlp}–LDMC and π_{tlp}–iWUE) against those that were weaker (π_{tlp}–SLA) or that could even be misleading at a first glance (π_{tlp}–height).

π_{tlp} is coordinated with intrinsic water-use efficiency

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Higher leaf level dehydration tolerance (more negative π_{tlp}) was coordinated with higher intrinsic water use efficiency (less negative $\delta^{13}C$) on all levels considered. This may suggest that though plants adjust their osmotic potential in order to keep stomata open, the mechanism has a limited capacity in temperate plants. This may occur when the intensity of the environmental stress (drought expressed by soil water potential) is not stable but fluctuates deeply below the capacity of the osmotic adjustment. During these episodes, stomata are closed, and the assimilated carbon has less negative $\delta^{13}C$ (higher iWUE). The existence of the coordination across our species can be interpreted perhaps as a continuous run (i.e. developing osmotic adjustment) of the drought-exposed plants out of the dehydration stress, which they never quite win (if they won, they would keep stomata open, i.e. lower iWUE), but which is sufficient enough to maintain growth and ensure survival in the more stressful conditions.

π_{tlp} is coordinated with leaf dry matter content

Stronger leaf dehydration tolerance (i.e., more negative π_{tlp}) was tightly coordinated with LDMC, morphological trait commonly used as a proxy of plant's drought resistance. One explanation is that because low osmotic potential is a response to drought stress (e.g. Májeková *et al.* 2019), water limitation simultaneously results in smaller cell wall expansion, smaller cells and/or more cell walls, i.e. greater LDMC (Poorter *et al.* 2009). Our results are in line with previous evidence on coordination between these two leaf-level traits in herbaceous species: on 33 C4 grassland species (Liu & Osborne 2015) and on 19 tallgrass prairie species including C3 and C4 graminoids, forbs and shrubs (Griffin-Nolan *et al.* 2019). The fact that we found the coordination between π_{tlp} and LDMC to be strong on all levels, together with the previous evidence including also C4 species, suggests that this coordination is biophysically-based and transferable to other systems, plant functional types and plant families.

π_{tlp} is only weakly coordinated with specific leaf area

Our results imply that the mechanistic link between SLA, plant water economics and soil water availability could not be simply assumed on any given scale without further stratification. The coordination between π_{tlp} and SLA appeared only weakly ($P = 0.045$) in forbs after plants were stratified by their PFTs and then more strongly when their phylogenetic relatedness was accounted for. Our results fit well into the so far ambiguous evidence regarding the coordination between SLA and π_{tlp}. While a weak relationship between more negative π_{tlp} and higher SLA was documented in a big compiled tree dataset (Zhu *et al.* 2018), no was found on a more local spatial scale for both trees (Maréchaux *et al.* 2015, 2019) and herbs (Májeková *et al.* 2019). Moreover, recent evidence found no relationship between SLA and water availability gradients at different spatial and taxonomical scales (for reviews see Bartlett *et al.* 2012b; Griffin-Nolan *et al.* 2018). This together suggest that SLA should be considered very carefully as a proxy reflecting the response of plants to water availability.

π_{tlp} is not coordinated with plant height

The relationship between π_{tlp} and plant height represents a nice example on how a coordination between two traits could be misinterpreted without a further stratification by PFTs or accounting for plant phylogenetic relatedness. When considering all species without any stratification, a weak, but significant, negative relationship appears between π_{tlp} and plant height, suggesting that higher leaf dehydration tolerance (more negative π_{tlp}) is coordinated with taller stature. A potential explanation would offer itself, i.e. that shorter plants growing under taller plants would be sheltered from direct irradiance, and therefore experience less dehydration. However, a closer examination reveals that, in our case, the relationship is driven purely by the pronounced differences between forbs and graminoids in both traits considered and disappears within the PFTs and after accounting for plant phylogenetic relatedness. It seems that the risk of embolism, driving the relationship between plant height and hydraulic traits in woody species (Ryan *et al.* 2006; Liu *et al.* 2019), does not play a major role in the shorter, relatively to trees, herbaceous species. Indeed, a height of 1 m generates only –0.01 MPa of gravitational potential, which is negligible when compared with osmotic potential. Rather, plant height in grassland plants seems to be under different and potentially more important selection pressures than water availability, such as competitive ability for light (Keddy & Shipley 1989),

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thus resulting in little coordination among height and π_{tlp}.

Critical remarks: the use of single traits as proxies of function

One needs to carefully consider that the leaf is a multifunctional organ, and thus bulk leaf traits might not always capture exactly the response to a single abiotic factor in question. For instance, a plant's response to water availability measured through SLA, a very popular leaf economics spectrum trait, can be simultaneously confounded by light availability, nutrient availability, or herbivory resistance (Walters & Reich 1999; Sack 2004; Poorter *et al.* 2009; Markesteijn *et al.* 2011; John *et al.* 2017). As advocated by Hodgson *et al.* (2011), one needs to carefully consider how valid the use of such a trait is as a stand-alone proxy of a single function.

Here, we would like to reinforce this view by tentatively proposing three advices. First, we advocate the use of traits known to be directly and mechanistically linked to the factor in question (Griffin-Nolan *et al.* 2018). In case of water availability, we suggest using π_{tlp}, which is a hydraulic trait feasible to measure on a large number of individuals. Second, if leaf traits are measured that could reflect multiple functions of the leaf, i.e. SLA and/or LDMC, we suggest coupling them with traits known to be mechanistically linked to the factor in question. In case of water availability, this would be π_{tlp}, intrinsic water use efficiency, or any other hydraulic trait. This should be done on at least a subset of species, in order to validate (e.g. by simple correlation) that the morphological traits reflect the function in question.

Third, both approaches can be further reinforced by coupling the quantitative measures with the plant functional types. Indeed, if a relationship (with another trait, or environmental factor) is driven only by the differences in functional types, but does not hold within the types, one might ask whether the trait considered really reflects the function being investigated. For instance, here we showed that stratification by simple plant functional types and/or accounting for species phylogeny might help to identify relationships that are biophysically-based, and therefore potentially better transferable among different systems. Such would be the coordination between π_{tlp} and LDMC, where the strength of the relationship holds true within PFTs, after accounting for phylogenetic relatedness, as well as in other functional groups such as C4 grasses (Liu & Osborne 2015). On the other hand, the need for verifying the relationship is in our case highlighted on the relationship between π_{tlp} and plant vegetative height, which disappeared when further stratifying by PFTs or accounting for phylogenetical relatedness.

Conclusion

Here, we addressed the recent plea for a use of traits that would have a more direct mechanistic link to plant drought resistance and water availability to be considered in community trait ecology and global change ecology (Brodribb 2017; Griffin-Nolan *et al.* 2018; Volaire 2018). We demonstrated the feasibility of the use of a physiological drought tolerance leaf-level trait, π_{tlp}, that becomes operational via the novel osmometry method for large-scale studies across different plant functional types. In herbaceous plants, π_{tlp} was tightly coordinated with intrinsic water use efficiency and with leaf dry matter content. However, π_{tlp} was not coordinated with plant height, which is the leading dimension of the global spectrum of form and function, and only weakly to SLA, which represents the second most important axis (Díaz *et al.* 2016). This implies that another important axis related to drought tolerance and water availability has so far been largely omitted in the trait-based ecology. Finally, stratification either by simple plant functional types and/or by accounting for species phylogeny might help to identify relationships that are biophysically-based and under the same selection pressure, and therefore potentially better transferable among different systems.

Acknowledgements

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Supporting Information

Additional supporting information may be found in the online version of this article.

Appendix S1. List of species, their plant functional type (PFT) and locality of sampling.

Table 1. Results of the standardized major axis of the relationships between turgor loss point (π_{tlp}, MPa, n = 122) and leaf dry matter content ('LDMC', mg g^{-1}, n = 122), specific leaf area ('SLA', m^2 kg^{-1}, n = 122), intrinsic water use efficiency measured by carbon stable isotopes ('δ^{13}C', n = 57) and vegetative height (log$_{10}$-transformed; 'Height', n = 122) in models (a) with only traits, (b) with traits and plant functional types ('PFTs'), and (c) based on data after accounting for phylogenetic independent contrasts. In (b) 'LR' denotes the Bartlett-corrected likelihood ratio for slopes comparison between the PFTs under the null hypothesis that slopes are equal. Significant relations are highlighted in bold.

		LDMC	SLA	δ^{13}C	Height
(a) Model with traits	(a) Model with traits	(a) Model with traits	(a) Model with traits	(a) Model with traits	(a) Model with traits
R^2	R^2	0.314	0.011	0.125	0.033
P	*P*	**<0.001**	0.239	**0.009**	**0.045**
(b) Model with traits and PFTs	(b) Model with traits and PFTs	(b) Model with traits and PFTs	(b) Model with traits and PFTs	(b) Model with traits and PFTs	(b) Model with traits and PFTs
Forb	R^2	0.168	0.042	0.130	0.000
	P	**<0.001**	**0.045**	**0.036**	0.978
Graminoid	R^2	0.174	0.023	0.241	0.009
	P	**0.027**	0.441	**0.028**	0.638
Slope comparison	LR	0.004	1.427	1.436	0.143
	P	0.950	0.232	0.231	0.704
(c) Model with phylogenetic independent contrasts	(c) Model with phylogenetic independent contrasts	(c) Model with phylogenetic independent contrasts	(c) Model with phylogenetic independent contrasts	(c) Model with phylogenetic independent contrasts	(c) Model with phylogenetic independent contrasts
R^2	R^2	0.145	0.096	0.116	0.007
P	*P*	**<0.001**	**<0.001**	**0.015**	0.368

Figure 1. Differences between the plant functional types in (a) turgor loss point (π_{tlp}), (b) vegetative height (log$_{10}$-transformed), (c) specific leaf area (SLA), (d) intrinsic water use efficiency measured by carbon stable isotopes (δ^{13}C) and (e) leaf dry matter content (LDMC) and. Bars are mean + 1 standard error.

Figure 2. Standardized major axis of the relationships between turgor loss point (π_{tlp}) and leaf dry matter content (LDMC), specific leaf area (SLA), intrinsic water use efficiency measured as carbon stable isotopes ratio (δ^{13}C) and vegetative height (log$_{10}$-transformed). Left panel presents actual data and right panel presents data after accounting for the phylogenetical relatedness ('PIC'; phylogenetic independent contrasts). Purple dots and lines denote forbs, green dots and lines denote graminoids, grey dashed line is based on all data. 'All', all data combined; 'F', forbs; 'G', graminoids; 'F = G', slopes of the relationship do not differ significantly between forbs and graminoids.
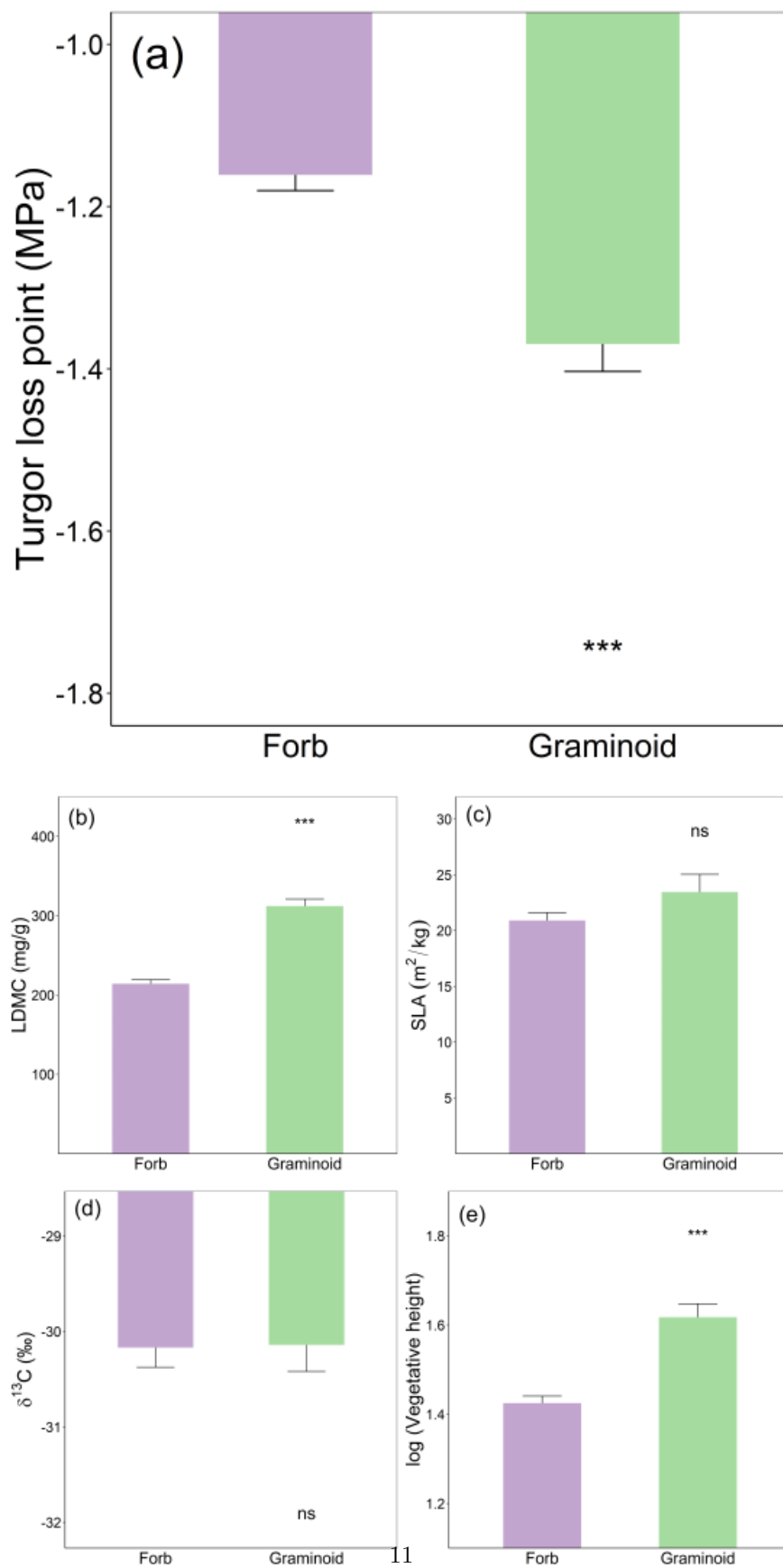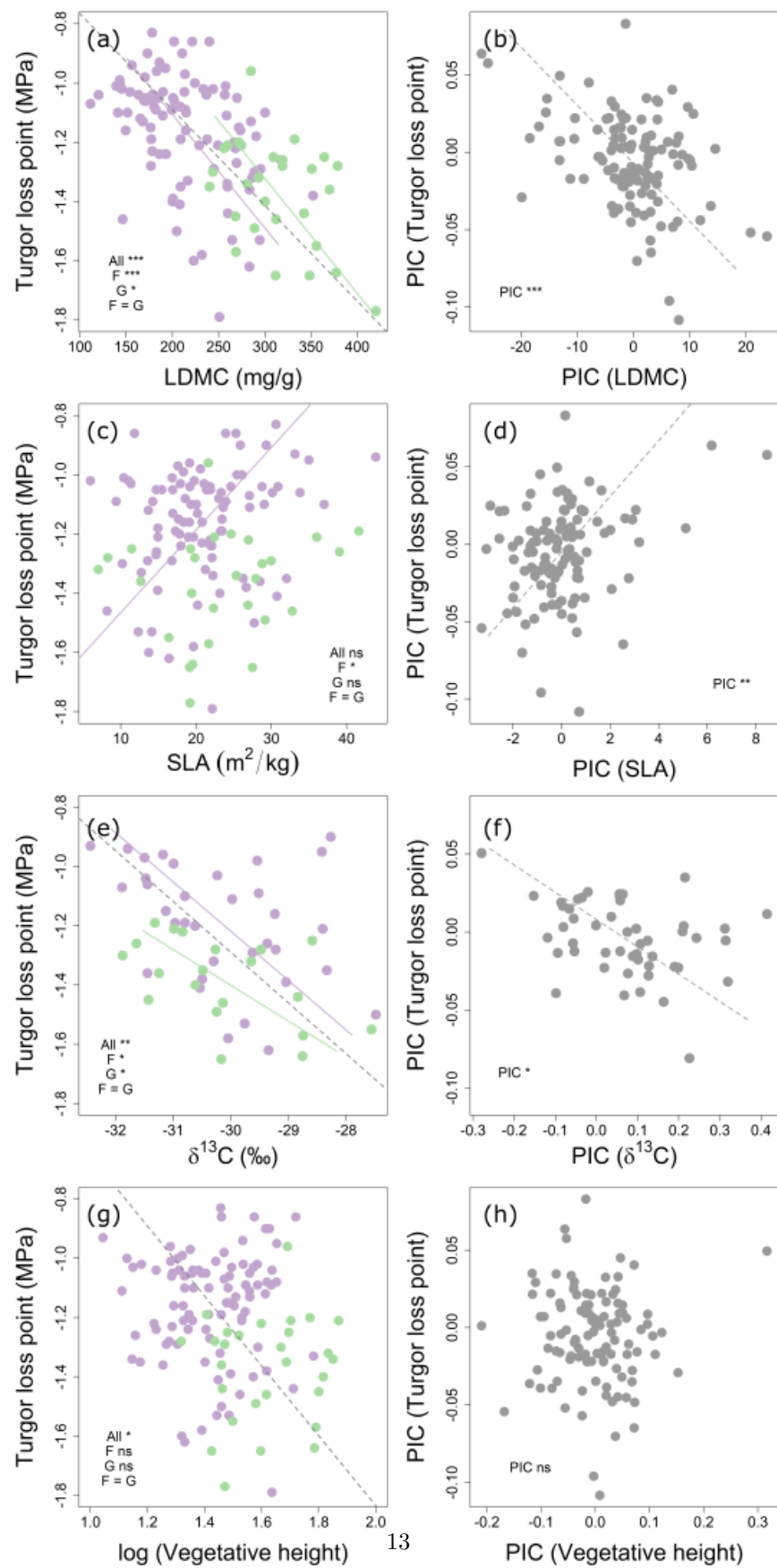
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Figure 1

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Figure 2