Unravelling the phenology-phylogeny tangle.

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Results & Discussion

Most species are sensitive to all three primary cues—forcing, chilling, and photoperiod (Figs. 1, Supporting Table **??**)(see also Laube et al., 2014; Ettinger et al., 2020)—with sensitivity to chilling approximately five-fold greater than sensitivity to photoperiod, on average (phenological advances of 7.2 days per standardized unit vs 1.4 days, for chilling and photoperiod, respectively; see Table **??**). However, these average sensitivities estimated across species, fail to capture the large differences in species' responses to chilling and forcing (Figs. 1, Supporting Table **??**).

By allowing species responses to vary, based on a model including their shared evolutionary history, we found species differences dwarfed differences between cues, especially temperature cues (Fig. 1). The largest in magnitude cue—chilling—varied 25-fold between species, while variation to forcing varied 6-fold. These results suggest the strong focus on comparing cues may miss the largest variation. Robust phenological forecasts must then account for both the complexity of multiple cues and species-level variation in responses to them.

Differences across clades & cues

The large differences across species generally occurred coherently within clades, producing striking differences between clades. Oaks and beeches (Fagaceae), elms (Ulmaceae) and buckthorns (Rhamnaceae) are highly sensitive to chilling while rhododendrons (Ericaceae), butterfly bushes (Scrophulariaceae) or spindles (Celastraceae) show little to no response to chilling (Fig. 1 a). Similar clade-level variation was observed for forcing, where some of these clades—e.g., Ericaceae, Rhamnaceae, Ulmaceae, or Fagaceae—were particularly sensitive (advancing their budburst more than 10 days per standardized unit of forcing) and others such as the Sapindaceae, Cornaceae or Juglandaceae families show little response (Fig. 1 b).

Some species responded strongly to both temperature cues (chilling and forcing), which could suggest the existence of syndromes where the genetic basis for responses to one cue (e.g., forcing) has been been selected for alongside responses to another cue (e.g. chilling). We might expect, for example, early-season species to be sensitive to multiple cues as mistiming phenology could result in exposure to harsh conditions, leading to tissue loss or death (Sakai and Larcher, 1987), and sensitivity to multiple cues likely provides greater insurance against mistiming (Wolkovich and Donahue, 2021). Alternatively or additionally, linkage or pleiotropism among loci associated with sensitivity to different cues (Nakagawa et al., 2005) could induce correlated cues. However, the correlation in species sensitivities across cues was weak ($r = 0.31$; between forcing an chilling) and some genera, such as *Tilia* and Ericaceae[genus?], displayed strong responses to forcing but weak responses to chilling, while others, such as XXXXExamplesXXXX, show strong responses to chilling but weak responses to forcing (Fig. 1). Species sensitivity to one cue, thus, does not constrain sensitivity

to another cue, and selection might operate independently on responses to different cues (Bonamour et al., 2019).

In contrast to both chilling and forcing, species-level responses to photoperiod were almost uniform—present across nearly all species. This provides novel insight on a large debate over the prevalence of photoperiod cues in temperate trees, where previous experiments (Basler and Körner, 2012; Zohner et al., 2016) and modeling exercises (e.g., Hunter and Lechowicz, 1992; Schaber and Badeck, 2003) suggested important variability across species that may constrain the responses of certain species to warming (Way and Montgomery, 2015). Our results suggest variability is constrained to a very limit set of species in Fagaceae, which have been heavily studied, including *Fagus sylvatica* (e.g., Basler and Körner, 2012; Zohner et al., 2016; Kramer et al., 2017). These same few species are also where most evidence of local adaptation in photoperiod cues for spring phenology comes from (e.g., Kramer et al., 2017), in contrast with common garden studies of other species, which found little evidence of local adaptation in spring (but not fall) phenology (Aitken and Bemmels, 2016). The uniformity of response to photoperiod that we found supports this latter view of generally low local adaptation in photoperiod cues for spring phenology: given that studies spanned a range of latitudes (with many species studied in only a narrow latitudinal range), if local adaptation was high in photoperiod cues we would expect more variability across species.

Our results suggest that while chilling and forcing are the dominant cues in terms of the magnitude of their effect sizes, most species are also sensitive to photoperiod, highlighting the potential importance of this cue to both climate change responses in the future, and through evolutionary history. In plants, photoperiod regulates a number of fundamental processes including growth, flowering, stress tolerance, and circadian rhythm (Serrano-Bueno et al., 2017; Singh et al., 2017), with photoperiodic sensing and adaptation to shifting daylength likely occurred early in the evolution of plants, with origins in the green algae (Serrano-Bueno et al., 2017). It is possible, therefore, that phenological responses to photoperiod reflect ancestral sensitivities with origins in deep time. Large plasticity in responses to additional environmental cues (e.g., temperature) for a given photoperiod cue (e.g., Kramer, 1995) may allow species to track interannual variation in climate with little directional selection on photoperiod sensitivities. Alternatively, photoperiod might thus provide a reliable calibration of the underlying biological clock (Jackson, 2009) upon which seasonal variation in climate modifies the tempo.

Phylogenetic structure of phenological cues

Variation—or lack thereof—in cues across species and clades, alongside related estimates from our modeling approach, provides possible insights into the evolution of cues across the phylogeny. While responses to each cue were phylogenetically structured, the strength of phylogenetic conservatism in response differs between cues (Fig. 3). Responses to temperature (forcing and chilling) were moderately structured ($\lambda = 0.65$

and $\lambda = 0.54$, for forcing and chilling, respectively), with closely related species exhibiting more similar sensitivities than distantly related species. Phylogenetic structure in species responses to photoperiod was comparatively weaker ($\lambda = 0.39$) (see Fig. **??**, Table **??**), emphasizing again that photoperiod diverges from other climate cues. In addition to be weaker and more uniform across species, sensitivity to photoperiod appears to also be less evolutionarily constrained than sensitivities to temperature cues. While our approach is unique in focusing on species cues, our results support previous work showing phylogenetic signal in plant phenological timing observed from floras or long-term data **?**Davies et al. (2013)

What might drive phylogenetic structure in species temperature responses? Differences between species in their responses represent shifts in the slope of the relationship between the observed phenology and the cue. Thus, the observed phylogenetic structure in temperature responses (forcing and chilling) would be consistent with an interaction with a non-measured trait that moderates responses, and which also covaries with phylogeny (Davies et al., 2019). This fits fundamentally with the idea that early-season phenology plays a critical role in shaping species temporal niches (**?**) and thus should covary with a suite of life-history traits, including whether species are early-active with other rapid return on investment trait, or start later in the season but have higher competitive abilities (e.g., **??**). Geography may drive structured phylogenetic variation, especially in regions where clades have radiated such that close relatives have experienced the same suite of environmental cues and thus been subject to similar selection regimes. Conversely, if species are not generally geographically constrained, as may be more likely the case, especially across deep time (Fitzpatrick and Turelli, 2006), then we might expect phylogenetic structure in phenology to also weaken, especially when aggregating across locations with different cues (Davies et al., 2013).

Weak phylogenetic signal in photoperiod sensitivity (Fig. 3) might seem at odds with observations that distantly related species respond more similarly (and less variably) to photoperiod than they do to forcing or chilling. However, somewhat counterintuitively, both uniform and random responses can manifest as low phylogenetic signal when indexed by Brownian motion expectations (see Wiens et al.). Rapid local adaptation within species might thus erase the phylogenetic structure in photoperiod responses, but would seem at odds with the uniformity in species' responses. Alternatively, if responses to photoperiod evolved early in plants, as we suggest above, and subsequent selection on photoperiod sensitivity was constrained by stabilizing selection operating on other life-history attributes sensitive to photoperiod (XXCITE), we would predict both low interspecific variation and weak phylogenetic signal in responses, matching observations. This latter interpretation is also consistent with our estimates of lower σ for photoperiod responses (CITE FIGURE). Here, as in more traditional phylogenetic comparative methods, σ represents the rate of evolution, and thus our model suggests photoperiod responses are also evolving slower than temperature responses. However, we also reveal (see Appendix XXX) that an early burst model of evolution, in which trait variation accumulates

rapidly early in the history of a clade and then slows through time, consistent with our interpretation of photoperiod evolution.

Phylogenetic conservatism (high λ) and slow evolutionary rates (low σ) in traits has sometimes been interpreted as indicative of evolutionary constraints to adaptive change (cite Wiens et al. and others cites XXX). If this were the case, we might then suggest that species with strong forcing response might be more vulnerable to future warming spring temperatures because phylogenetic signal in responses to forcing is strong, while species may be able to better adapt to changes in chilling as λ is lower, and σ is higher. However, this is misleading as estimates of lambda are independent from the rate of evolution, and macroevolutionary rates estimated on phylogenetic trees are integrate across millions of years of evolutionary history, and thus do not necessarily inform us of maximum possible rates of evolution over shorter timescales. Indeed, there is accumulating evidence for rapid evolution to warming climate ... (cites XXXX). We highlight these parameter here to improve our understanding of the evolutionary history of species phenological cues, and to underscore the importance of correctly modelling species differences in ecological forecasts.

Forecasting species-level responses

Not accounting for phylogeny had an effect on model coefficients (model slopes for forcing and chilling shifted by 7.2% and 3.7%, respectively; Fig. 4) and shifted cross-species variance in their responses to forcing (Var $\beta_{phylo} = 8.74$; Var $\beta_{non-phylo} = 5.01$), chilling (Var $\beta_{phylo} = 23.45$; Var $\beta_{non-phylo} = 17.47$), and photoperiod responses (Var $\beta_{phylo} = 0.82$; Var $\beta_{non-phylo} = 0.93$). Counterintuitively, induced reductions in cross-species variance, far from increasing estimation accuracy could lead to increased type-II error by failing to detect actual relationships among cue and responses that would only emerge when phylogeny is correctly accounted for (see Supporting Information XX). Either ignoring ($\lambda = 0$) or overestimating phylogenetic structuring of predictors ($\lambda = 1$) can bias model coefficients (predictors with high λ are biased in non-phylogenetic models and those with low λ are biased if Brownian Motion is imposed). Importantly, not accounting for phylogeny increased the uncertainty around each individual species estimation of their responses to forcing and chilling (see Fig. SXX in Supporting Information), which could lead to less precise predictions and forecasts of phenology for individual species although overall model accuracy might still appear reasonable (see Appendix XX in Supporting Information).

Phylogenetic hierarchical models such as those used here, have the potential to inform on which clades will be more sensitive to different axes of climate change—e.g., changes in cold temperatures over winter or in warm temperatures in spring and summer. For example, in standard non-phylogenetic models, we would not have identified oaks (genus *Quercus*) as being among the most sensitive taxa to forcing and chilling (see e.g., (Ettinger et al., 2020)), but we show that species within this genus advance phenology by 2 days per standard unit of forcing and 4 days per standard unit of chilling. Ignoring phylogeny and species-level variation

5

may (i) introduce bias into estimated model coefficients, (ii) underestimate true variability in species biological responses and, (iii) increase uncertainty around species-level estimates. Our integrative phylogenetic approach allows us to move beyond crude classifications based upon functional groups (e.g., !REFS) and species complexes (e.g. Ettinger et al. (2020)), to make improved ecological forecasts for individual species. Our model, highlighting strong phylogenetic structure in species response, also highlights the potential for predicting phenological sensitivities in unmeasured species. While phylogenetic imputation must be done with extreme care (Molina-Venegas et al., 2018), we can make predictions with reasonable accuracy for close relatives of well studied species when there is strong phylogenetic structure in species responses. Notably this is likely to be the case for many temperate woody plant clades, for which we have multiple experimental observations at varying treatment levels for well-known drivers of phenology.

Accurate forecasts of phenology remain elusive; recent records have suggested an apparent decline in species phenological sensitivity to increasing temperatures (Fu et al., 2015; Piao et al., 2017), but such observations may simply reflect non-linear responses to warming trends (Wolkovich et al., 2021), perhaps reflecting the complex interactions, trade-offs and synergies among multiple cues. While all species appear sensitive to all cues, forcing and chilling dominating over photoperiod, and species-level variation in responses is large, of a magnitude similar to, or greater than, the difference in average effect sizes between cues. Phylogenetic conservatism in species phenology has been well documented (Davies et al., 2013; Rafferty and Nabity, 2017; Joly et al., 2019), and may thus help explain species differences. Here, using a novel hierarchical Bayesian model, we allow for phylogenetic non-stationarity in species sensitivities (allowing slopes to vary across species; see (Davies et al., 2019)) to generate species-specific estimates of responses to shifting cues. Uniquely, our model additionally provides information on the evolutionary of species sensitivities, and how they may have been configured along evolutionary time. Our results are consistent with an early origin of photoperiod sensitivity, followed by a history of stabilizing selection. Forcing and chilling have evolved more rapidly. However, we caution that macroevolutionary rates provide a poor guide to future adaptive potential over short timescales, and accurate ecological forecasting requires well-designed experiments analysed appropriately.

Chunks that need to fit into the forecasting section ....

Helpfully, the contrasts between temperature sensitivities and photoperiod sensitivities in both their variability across species and phylogenetic structuring allow for improved multi-species forecasts: (1) the large variability among species in temperature responses makes predicting species individual responses challenging, but the phylogenetic structure in responses lets us borrow information from close relatives to improve our predictions; (2) the weak phylogenetic structure in photoperiod responses indicates that phylogeny retains little information, but because species responses are generally more uniform, we can be more confident in assuming the mean species response across species, comforted in the knowledge that small errors will likely not have large impact given the relatively weak overall contribution of this cue.

The genus *Fagus* is recognized as being particularly sensitive to photoperiod (Fu et al., 2019). Specifically, *Fagus sylvatica* is nearly five times more sensitive to photoperiod than most other measured tree species. The question arises as to whether species with outlying responses should be chosen as the model from which to extrapolate knowledge as done with *Fagus sylvatica* in the phenology literature (REFs for PEP75?!).

References

Aitken, S. N., and J. B. Bemmels. 2016. Time to get moving: assisted gene flow of forest trees. Evol Appl 9:271–90.

Basler, D., and C. Körner. 2012. Photoperiod sensitivity of bud burst in 14 temperate forest tree species. Agricultural and Forest Meteorology 165:73–81.

Bonamour, S., L. M. Chevin, A. Charmantier, and C. Teplitsky. 2019. Phenotypic plasticity in response to climate change: the importance of cue variation. Philosophical Transactions of the Royal Society B-Biological Sciences 374.

Davies, T., E. Wolkovich, N. Kraft, N. Salamin, and S. E. Travers. 2013. Phylogenetic conservatism in plant phenology. Journal of Ecology 101:1520–1530.

Davies, T. J., J. Regetz, E. M. Wolkovich, and B. J. McGill. 2019. Phylogenetically weighted regression: A method for modelling non-stationarity on evolutionary trees. Global ecology and biogeography 28:275–285.

Ettinger, A., C. Chamberlain, I. Morales-Castilla, D. Buonaiuto, D. Flynn, T. Savas, J. Samaha, and E. Wolkovich. 2020. Winter temperatures predominate in spring phenological responses to warming. Nature Climate Change pages 1–6.

Fitzpatrick, B., and M. Turelli. 2006. The geography of mammalian speciation: Mixed signals from phylogenies and range maps. Evolution 60:601–615.

Fu, Y. H., S. Piao, X. Zhou, X. Geng, F. Hao, Y. Vitasse, and I. A. Janssens. 2019. Short photoperiod reduces the temperature sensitivity of leaf-out in saplings of *Fagus sylvatica* but not in horse chestnut. Global Change Biology 25:1696–1703.

Fu, Y. S. H., H. F. Zhao, S. L. Piao, M. Peaucelle, S. S. Peng, G. Y. Zhou, P. Ciais, M. T. Huang, A. Menzel, J. P. Uelas, Y. Song, Y. Vitasse, Z. Z. Zeng, and I. A. Janssens. 2015. Declining global warming effects on the phenology of spring leaf unfolding. Nature 526:104–107.

Hunter, A. F., and M. J. Lechowicz. 1992. Predicting the timing of budburst in temperate trees. Journal of Applied Ecology 29:597–604.

Jackson, S. D. 2009. Plant responses to photoperiod. New Phytologist 181:517–531.

Joly, S., D. F. Flynn, and E. M. Wolkovich. 2019. On the importance of accounting for intraspecific genomic relatedness in multi-species studies. Methods in Ecology and Evolution .

Kramer, K. 1995. Phenotypic plasticity of the phenology of 7 european tree species in relation to climatic warming. Plant Cell and Environment 18:93–104.

Kramer, K., A. Ducousso, D. Gomory, J. K. Hansen, L. Ionita, M. Liesebach, A. Lorent, S. Schueler, M. Sulkowska, S. de Vries, and G. von Wuehlisch. 2017. Chilling and forcing requirements for foliage bud burst of European beech (*Fagus sylvatica* L.) differ between provenances and are phenotypically plastic. Agricultural and Forest Meteorology 234:172–181.

Laube, J., T. H. Sparks, N. Estrella, J. Höfler, D. P. Ankerst, and A. Menzel. 2014. Chilling outweighs photoperiod in preventing precocious spring development. Global Change Biology 20:170–182.

Molina-Venegas, R., J. C. Moreno-Saiz, I. Castro Parga, T. J. Davies, P. R. Peres-Neto, and M. Á. Rodríguez. 2018. Assessing among-lineage variability in phylogenetic imputation of functional trait datasets. Ecography 41:1740–1749.

Nakagawa, H., J. Yamagishi, N. Miyamoto, M. Motoyama, M. Yano, and K. Nemoto. 2005. Flowering response of rice to photoperiod and temperature: a qtl analysis using a phenological model. Theoretical and Applied Genetics 110:778–786.

Piao, S., Z. Liu, T. Wang, S. Peng, P. Ciais, M. Huang, A. Ahlstrom, J. F. Burkhart, F. Chevallier, I. A. Janssens, et al. 2017. Weakening temperature control on the interannual variations of spring carbon uptake across northern lands. Nature climate change 7:359.

Rafferty, N. E., and P. D. Nabity. 2017. A global test for phylogenetic signal in shifts in flowering time under climate change. Journal of Ecology 105:627–633.

Sakai, A. K., and W. Larcher. 1987. Frost Survival of Plants. Ecological Studies. Springer-Verlag.

Schaber, J., and F. Badeck. 2003. Physiology-based phenology models for forest tree species in germany. International Journal of Biometeorology 47:193–201. International Conference on the Times they are a Changin, WAGENINGEN, NETHERLANDS, DEC 05-07, 2001.

Serrano-Bueno, G., F. J. Romero-Campero, E. Lucas-Reina, J. M. Romero, and F. Valverde. 2017. Evolution of photoperiod sensing in plants and algae. Current Opinion in Plant Biology 37:10–17.

Singh, R. K., T. Svystun, B. AlDahmash, A. M. Jönsson, and R. P. Bhalerao. 2017. Photoperiod-and temperature-mediated control of phenology in trees–a molecular perspective. New Phytologist 213:511–524.

Way, D. A., and R. A. Montgomery. 2015. Photoperiod constraints on tree phenology, performance and migration in a warming world. Plant, Cell & Environment 38:1725–1736.

Wolkovich, E., J. Auerbach, C. Chamberlain, D. Buonaiuto, A. Ettinger, I. Morales-Castilla, and A. Gelman. 2021. A simple explanation for declining temperature sensitivity with warming. Global Change Biology 27:4947–4949.

Wolkovich, E. M., and M. J. Donahue. 2021. How phenological tracking shapes species and communities in non-stationary environments. Biological Reviews .

Zohner, C. M., B. M. Benito, J. C. Svenning, and S. S. Renner. 2016. Day length unlikely to constrain climate-driven shifts in leaf-out times of northern woody plants. Nature Climate Change 6:1120–1123.
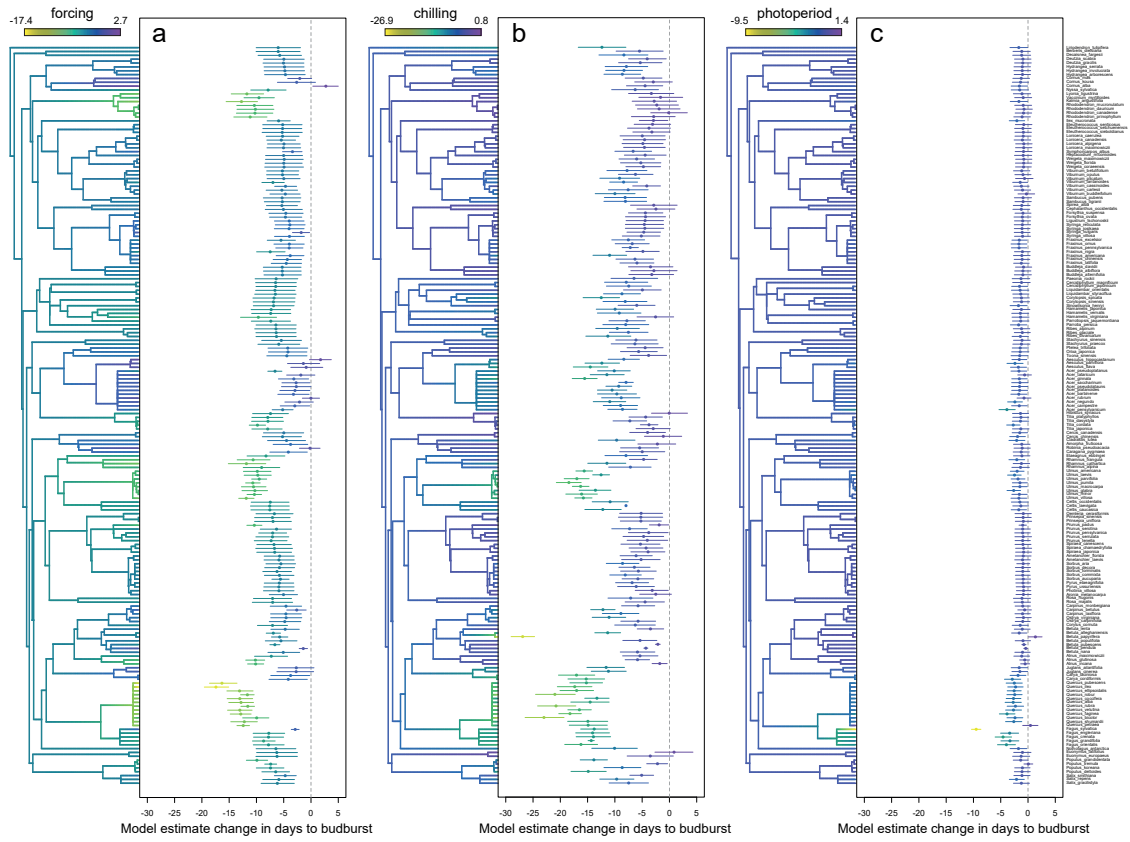
Tables and Figures

Figure 1: Phenological sensitivity to thee environmental cues, forcing (a), chilling (b) and photoperiod (c) measured in change in days to budburst per standardized unit (z-transformation) of the cues across 192 angiosperm species. The same phylogenetic tree is shown in each panel, colored acording to an estimation of ancestral character states, being the states at the tips the model slopes of our hierarchical phylogenetic model. Note that the color scale varies in each panel. Total tree depth is 81. My.
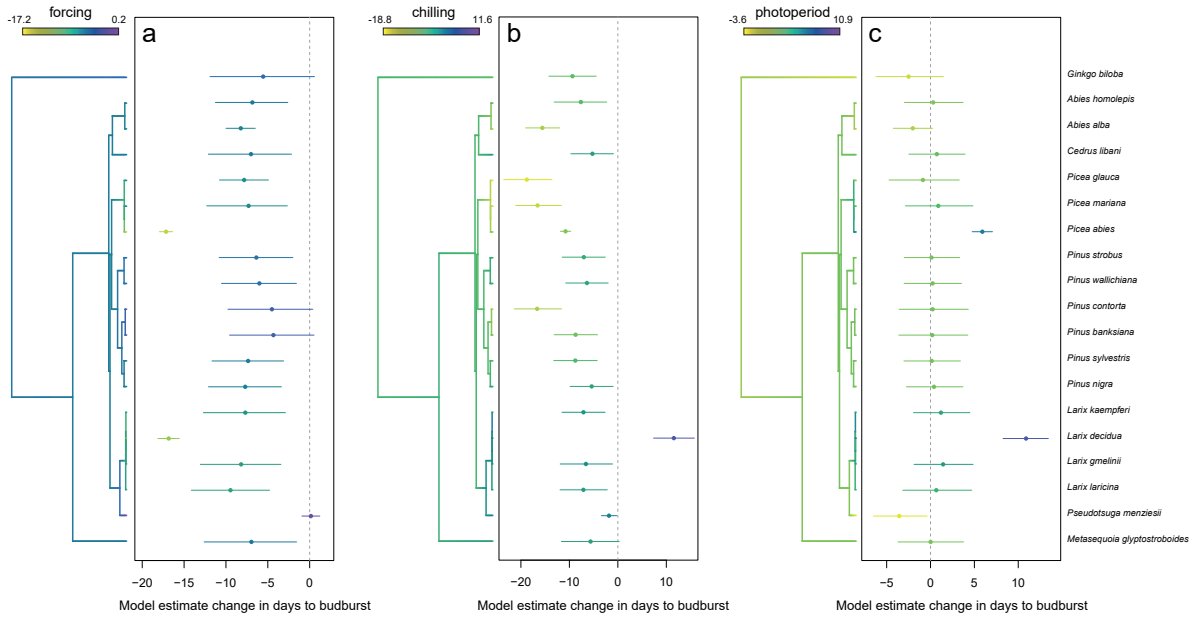
10

Figure 2: Phenological sensitivity to thee environmental cues, forcing (a), chilling (b) and photoperiod (c) measured in change in days to budburst per standardized unit (z-transformation) of the cues across 19 gymnosperm species. The same phylogenetic tree is shown in each panel, colored acording to an estimation of ancestral character states, being the states at the tips the model slopes of our hierarchical phylogenetic model. Note that the color scale varies in each panel. Total tree depth is 81. My.
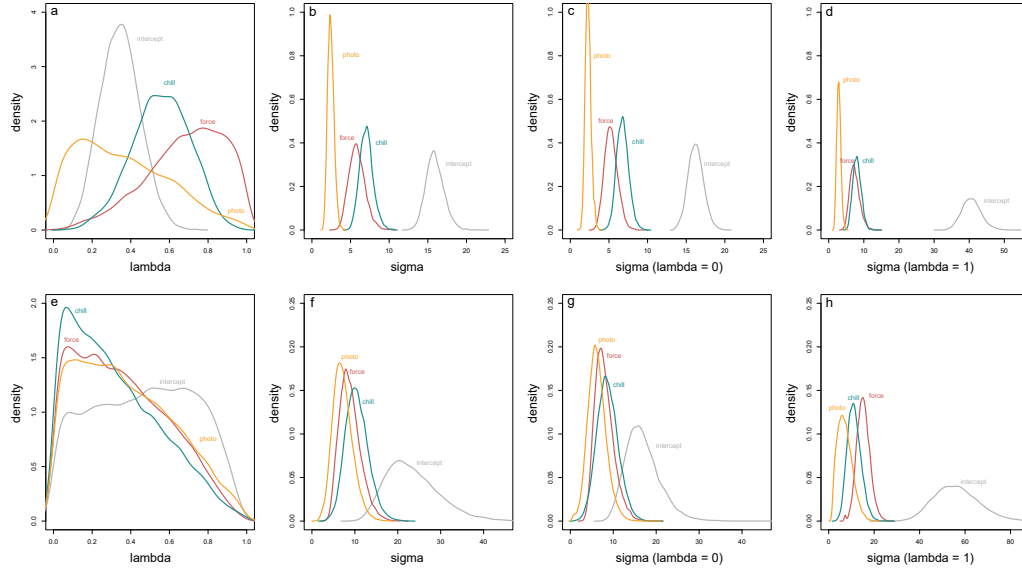
Figure 3: Density plots for the posterior distribution of phylogenetic signal measured by lambda for each cue included as a predictor in the model for angiosperms: forcing (red), chilling (blue), photoperiod (orange) and for the model intercept (grey). Panels correspond to angiosperms (a-d) and gymnosperms (e-h). Note that lambda estimations corresponding to panels c-d and g-h as they are constrained to be either equal zero or equal 1.
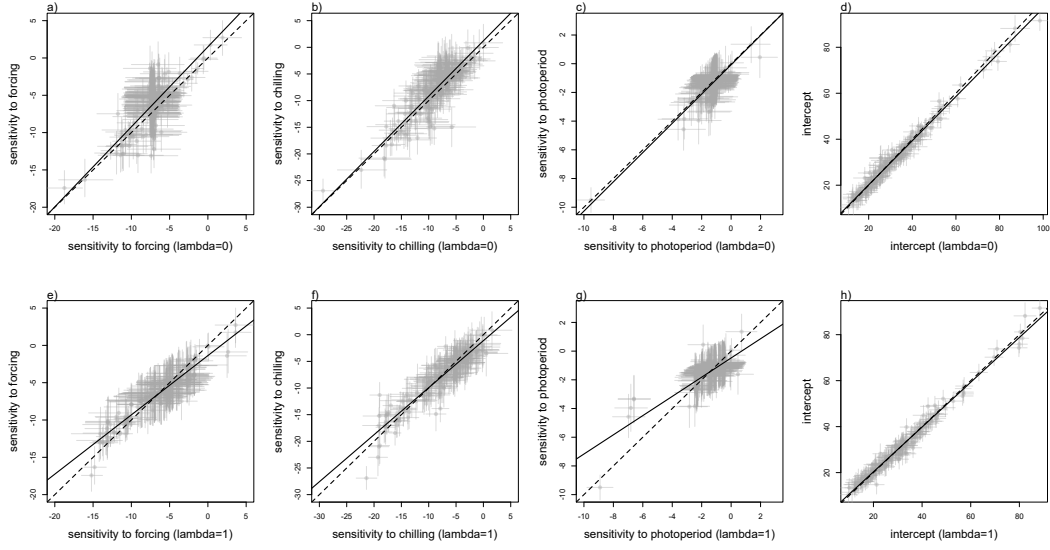
Figure 4: Correlations between model parameters as estimated by the full model and the models where lambda is constrained to be equal zero (upper row) or one (bottom row), for angiosperms. Panels correspond to sensitivity to forcing (a,e), to chilling (b,f), to photoperiod (c,g) and to model intercepts (d,h).

Figure 5: Correlations between model parameters as estimated by the full model and the models where lambda is constrained to be equal zero (upper row) or one (bottom row), for gymnosperms. Panels correspond to sensitivity to forcing (a,e), to chilling (b,f), to photoperiod (c,g) and to model intercepts (d,h).
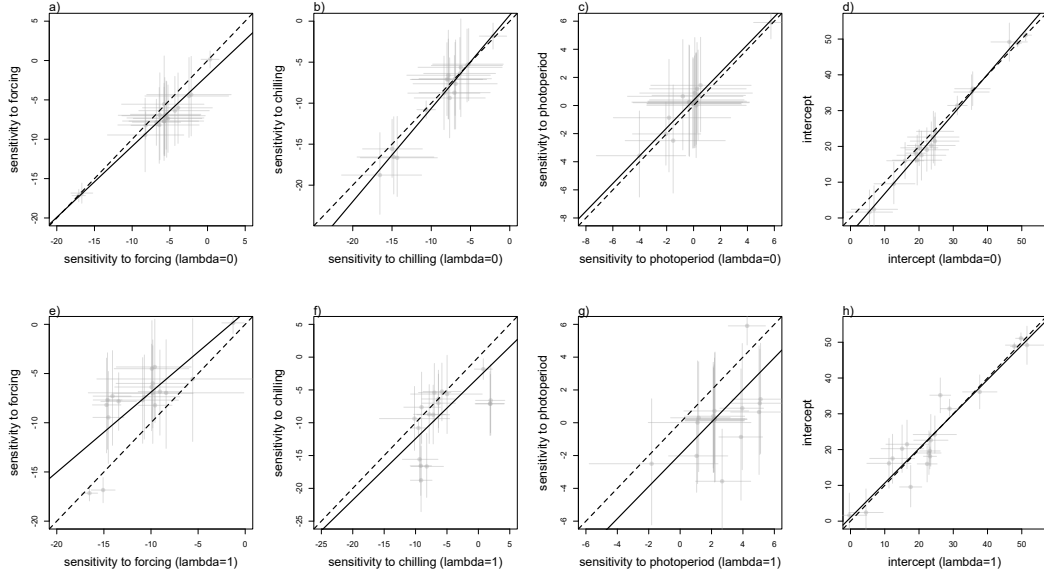
14

Table 1: Full model parameters estimated for 192 angiosperm species.

parameter	mean	sd	2.50%	50%	97.50%	n_eff
μ_α	30.57	3.41	23.68	30.59	37.14	5031.19
$\mu_{\beta forcing}$	-5.84	2.01	-9.72	-5.89	-1.79	2374.73
$\mu_{\beta chilling}$	-7.19	2.03	-11.15	-7.18	-3.18	3694.93
$\mu_{\beta photoperiod}$	-1.37	0.76	-2.92	-1.35	0.14	1565.41
λ_α	0.35	0.10	0.16	0.34	0.56	3416.51
$\lambda_{\beta forcing}$	0.68	0.20	0.23	0.71	0.98	185.35
$\lambda_{\beta chilling}$	0.56	0.15	0.25	0.56	0.83	738.57
$\lambda_{\beta photoperiod}$	0.36	0.24	0.02	0.33	0.88	296.51
σ^2_α	15.93	1.17	13.84	15.85	18.41	2988.37
$\sigma^2_{\beta forcing}$	5.84	1.04	4.03	5.78	8.15	502.74
$\sigma^2_{\beta chilling}$	7.05	0.87	5.48	7.02	8.92	1026.77
$\sigma^2_{\beta photoperiod}$	2.45	0.41	1.74	2.42	3.32	469.46
σ^2_y	12.81	0.18	12.47	12.80	13.17	4017.16

Table 2: Full model parameters estimated for 19 gymnosperm species.

parameter	mean	sd	2.50%	50%	97.50%	n_eff
μ_α	25.75	4.50	16.88	25.73	34.73	33151.86
$\mu_{\beta forcing}$	-5.92	3.80	-12.97	-6.05	1.90	16443.03
$\mu_{\beta chilling}$	-8.11	3.63	-15.31	-8.09	-0.94	21379.81
$\mu_{\beta photoperiod}$	-0.88	3.33	-8.01	-0.67	5.19	16301.93
λ_α	0.47	0.26	0.02	0.48	0.90	15934.03
$\lambda_{\beta forcing}$	0.36	0.23	0.02	0.33	0.84	14336.60
$\lambda_{\beta chilling}$	0.32	0.23	0.01	0.28	0.82	13230.88
$\lambda_{\beta photoperiod}$	0.37	0.24	0.02	0.34	0.88	11199.49
σ^2_α	23.47	6.20	13.87	22.59	37.81	18272.58
$\sigma^2_{\beta forcing}$	8.89	2.45	4.96	8.60	14.51	8126.51
$\sigma^2_{\beta chilling}$	10.47	2.66	5.78	10.30	16.17	8539.38
$\sigma^2_{\beta photoperiod}$	7.18	2.29	3.29	6.96	12.25	5625.69
σ^2_y	15.81	0.41	15.04	15.81	16.63	28640.16