Photoperiod shows strong relationships with leaf and wood traits but not woody plant budburst

Deirdre Loughnan[1], Faith A M Jones[1], and E M Wolkovich[1]

January 4, 2024

[1] Department of Forest and Conservation, Faculty of Forestry, University of British Columbia, 2424 Main Mall Vancouver, BC Canada V6T 1Z4.

Corresponding Author: Deirdre Loughnan deirdre.loughnan@ubc.ca

Research questions

1. Do phenological cue-trait relationships change across transects? Populations?
2. How do budburst cues relate to functional traits in temperate woody species?
3. How do shrub and tree species differ in their cue-trait relationships?

Materials and Methods

Field sampling

1. General pop description
   (a) Combined *in situ* trait data with budburst data from two growth chamber cutting experiments
   (b) Both datasets span latitudinal gradient of 4-6° for eastern and western communities respectively
   (c) Traits were measured across four eastern populations — Harvard Forest, Massachusetts, USA (42.55°N, 72.20°W) and St. Hippolyte, Quebec, Canada (45.98°N, 74.01°W) and four western population — Smithers (54.78°N, 127.17°W) and E.C. Manning Park (49.06°N, 120.78°W)
   (d) Growth chamber study — cuttings collected from the most southern and northern populations in each transect
2. Describing spp
   (a) Across populations — measured diverse assemblage of species across the understory and canopy layer
   (b) Selected angiosperm spp that were most abundant in forest communities—using closely related species and congeners between eastern and western communities
   (c) Focus on angiosperm spp only—leafout likely under more similar environmental controls than gymnosperm, conifer, species—previous work has shown similar cue responses with phylogeny

Functional traits

1. General sampling info
   (a) Traits were measured in the summer prior to each growth chamber study.
   (b) Eastern transect: traits measured summer 2015—May 28 to August 12, 2015
   (c) Western transect: traits measured May 29 to July 30, 2019
   (d) For each species present at a populations—measured traits for 1-10 healthy, adult, individuals
2. Trait sampling — structural/wood traits
   (a) Measured a total of five traits: height, DBH, SSD, LMA, LNC
   (b) Traits were measured according to methods outlined in —Perez-Harguindeguy2013.
   (c) Height was measured using a range finder (TruePulse200L).
   (d) Diameter at breast height was measured 1.42 m from the ground.
   (e) Wood volume was measured within 12 hours of collection — 10 cm sample taken from tree branches or shrub primary stems — measured using the water displacement method
   (f) Branch segments were dried at 105°C for 24h and weighted.
3. Trait sampling — leaf traits
   (a) Leaf traits were measured for five haphazardly selected, fully expanded, and hardened leaves
   (b) High resolution scans of leaf area taken using Canon flatbed scanner (CanoScan Lide 220) within 12 hours of collection
   (c) Leaf area was then quantified using the ImageJ software (version X)
   (d) Leaves were dried for 48h at 70°C and weighed using a precision balance.

Growth chamber study

1. General information
   (a) For growth chamber study—collected branch cuttings from our highest and lowest latitude populations in each transect
   (b) For both growth chamber studies — 8 distinct treatments:
   (c) two levels of chilling — no additional chilling or 30 days at 4°C = eastern study, and 30 days or 70 days of chilling at 4°C for our western study — all dark
   (d) two levels of forcing—a cool regime of 15:5°C and a warm regime of 20:10°C
   (e) two photoperiods of either 8 or 12 hours
   (f) Observations of budburst stage made every 1-3 days for each sample for several months
   (g) For detailed discussion of study sample collection and methods see Loughnan et al. (in prep)

Statistical Anlaysis

1. Introduce approach and why it is useful:
   (a) Our analysis combined the trait data with the budburst data from the growth chamber study
   (b) Built joint model for each trait—directly models the relationship between leaf and structural traits and budburst
   (c) Approach carries through uncertainty between trait and phenology data—combines observational trait data with experimental phenology data
2. Begin to describe the model:
   (a) hierarchical linear model—partitioning variation of individual observations (i) of a given trait value ($y_{\text{trait}[i]}$) to the effects of species (*sp id*), and population-level differences arising from transects (*transect id*) or the interaction between transects and latitude (*transect* $\times$ *latitude*), and residual variation or 'measurement error' (σ_{trait})
   (b) Transect included as a categorical variable and latitude as a continuous variable
   (c) Most traits were modeled using their natural units—except LMA was rescaled by 100
3. describe trait part of model
   (a) Model gives unique estimates for each species and species-level variance ($\alpha_{\text{sp[sp id]}}$)—partial pooling—controls for variation in the number of trait estimates per spp and trait variability
   (b) Estimate for differences between transect ($\beta_{transect}$) and the interaction between transects and latitude ($\beta_{transect\times latitude}$)
   (c) These species-level estimates of traits become predictors of species-level estimates of each cue— ($\beta_{\text{force[sp]}}$, $\beta_{\text{chill[sp]}}$, $\beta_{\text{photo[sp]}}$)

   Trait model:

$$\mu_{trait} = \alpha_{\text{grand trait}} + \alpha_{\text{sp[sp id]}} + \beta_{\text{transect}} \times transect + \beta_{\text{transect}\times\text{latitude}} \times (transect \times latitude) \tag{1}$$

$$\alpha_{Sp} \sim normal(0, \sigma_{sp})$$
$$y_{trait} \sim normal(\mu_{trait}, \sigma_{trait})$$

   trait-cue relationship:

$$\begin{aligned} \beta_{\text{chill[sp]}} &= \alpha_{\text{chill}[sp]} + \beta_{\text{trait.chill}} \times \alpha_{\text{trait sp[sp]}} \\ \beta_{\text{force[sp]}} &= \alpha_{\text{force}[sp]} + \beta_{\text{trait.force}} \times \alpha_{\text{trait sp[sp]}} \\ \beta_{\text{photo[sp]}} &= \alpha_{\text{photo}[sp]} + \beta_{\text{trait.photo}} \times \alpha_{\text{trait sp[sp]}} \end{aligned} \tag{2}$$

4. Explain phenology part of model
   (a) Across each spp—get esti of overall effect of each trait on cue ($\beta_{\text{trait.chill}}$, $\beta_{\text{trait.force}}$, $\beta_{\text{trait.photo}}$)
   (b) and estimate of species-level cue variation not explained by traits ($\alpha_{\text{chill}[sp]}$, $\alpha_{\text{force}[sp]}$, $\alpha_{\text{photo}[sp]}$) = how well trait effects explain species-level differences
   (c) From this we can estimate individual species responses to cues (C_i, F_i, P_i, respectively—*z*-scored)—with residual variation across species ($\alpha_{\text{pheno[sp]}}$) and observations ($\sigma_{\text{pheno}}$)

(d) Partial pooling for residual variation across species and variation in cues not attributed to the trait

$$\mu_{pheno} = \alpha_{\text{pheno[sp]}} + \beta_{\text{chill[sp]}} \times C_i + \beta_{\text{force[sp]}} \times F_i + \beta_{\text{photo[sp]}} \times P_i \tag{3}$$
$$y_{\text{pheno[i]}} \sim normal(\mu_{pheno}, \sigma_{\text{pheno}})$$

$$\begin{aligned} \alpha_{\text{pheno}} &\sim normal(\mu_{\alpha_{\text{pheno}}}, \sigma_{\alpha_{\text{pheno}}}) \\ \alpha_{\text{force}} &\sim normal(\mu_{\alpha_{\text{force}}}, \sigma_{\alpha_{\text{force}}}) \\ \alpha_{\text{chill}} &\sim normal(\mu_{\alpha_{\text{chill}}}, \sigma_{\alpha_{\text{chill}}}) \\ \alpha_{\text{photo}} &\sim normal(\mu_{\alpha_{\text{photo}}}, \sigma_{\alpha_{\text{photo}}}) \end{aligned} \tag{4}$$

5. General info about model checks etc.:
   (a) weakly informative priors
   (b) validated model priors using prior predictive checks
   (c) model was coded in the Stan programming language and fit using the rstan package cite rstan2018
   (d) Four chains (6000-8000 total sampling iterations), and all models met basic diagnostic checks, including no divergences, high effective sample size (n_{eff}), and $\hat{R}$ close to 1—LMA model still has low ESS—issue with neff—otherwise this is true
   (e) 90% UI given in text

Results

1. General info about study—numbers from methods
   (a) Across populations — measured 47 species — 28 eastern transect and 22 western transect
   (b) Eastern transect: 13 shrubs and 15 trees
   (c) Western transect: 18 shrubs and 4 trees
   (d) Three species occurred in both transect
   (e) Measured traits of 1428 unique individuals between the two transects: height ($n = 1317$), DBH ($n = 1220$), SSD ($n = 1359$), LMA ($n = 1345$), LNC ($n = 1351$)
   (f) Across our two growth chamber studies we made observations of 4211—studies spanning 82 and 113 days for our eastern and western studies respectfully.
   (g) Using a joint modeling approach we then combined our trait data with budburst data from 4211 cuttings across the same 47 species sampled in the field.
2. Most traits did not differ with population or latitude
   (a) Most of our traits showed negligible trait variation across populations—neither between the two transects (main effect of transect only) or by latitudes within each transect (an interactive effect between transect and latitude)
   (b) Leaf mass area differed by latitude within transects (0.3, UI: 0.2,0.4)—eastern species LMA increased with latitude—generally greater than western species—Fig. 1 d

   (c) LNC similarly showed a weak interaction between latitude and transect (0.3, UI: 0.2,0.4)—higher latitude populations in eastern transect had lower LNC than lower latitude populations and western species—Fig. 1 e
   (d) But our wood and structural traits showed no differences across populations or transects (Table 2 - 3)
   (e) Across each of our models—species level differences exceeded population level variation—with the exception of SSD, most trait models showed degress of species level variation ranging from 0.9 (UI: 0.8, 1.2) for LNC up to to 5.7 (UI: 4.7, 6.9) for DBH
   (f) Fig. 1

3. Only a subset of our traits related to at least one budburst cue—but responses were generally weak
   (a) Of our three cues—photoperiod strongest relationship with other traits
   (b) Our two structural traits = similar cue relationships—taller plants with greater DBH = stronger photoperiod responses = earlier bb under longer daylengths (-1.7, UI: -2.9, -0.5 for height and -2.3, UI: -3.4, -1.1 for DBH)
   (c) No relationship between cues and SSD (Table 3)
   (d) We also did not find a relationship between between wood ring porosity with cues—see similar cue estimates across each of the four ring types 3.

4. Only one of our two leaf traits showed moderate responses to photoperiod.
   (a) LMA showed a moderate response to photoperiod with low LMA species advancing in bb with longer photoperiods (-7.5, UI: -10.9, -4.1)
   (b) LNC had no responses to any of our three cues

5. Traits varied in whether temperature had the strongest effects—with some traits showing stronger than expected photoperiod cues
   (a) Most models estimated chilling followed by forcing as the strongest cues of budburst
   (b) LMA model—estimating photoperiod cues (-7.5, UI: -10.9, -4.1) that were slightly stronger than forcing cues (-7.1, UI: -13.0, -1.6)—while effects of chilling were weak (90% UI crossing zero) (Table 4)
   (c) In contrast—LNC (Table 5) and SSD models (Table 3) both showed weak photoperiod effects on budburst (90% UI crossing zero)
   (d) By including additional traits in modeling cue responses—found the relative importance of different cues in shaping budburst varied substantially
   (e) This—paired with strong species-level variation—will lead to varying estimated responses of species relative budburst dates across the different trait models
   (f) ref Fig. 4 and Fig. 5

6. Including the effects of multiple traits provides novel insights into potential tradeoffs between growth and environmental cues
   (a) Estimates of bb using our model parameters show clear differences in species timing between trees and shrubs for some traits but not all (Fig. 6)
   (b) Height showed strong correlations between bb timing and trait values—shrub spp were estimated to bb earlier—especially under stronger cues—tree spp were later
   (c) But this was not the case for leaf traits—LNC showed no distinct separation between species functional groups (Fig. 6)

Discussion

1. Our study = one of the first to combine trait data with phenological cue responses for the same individuals and across species distributions.
    (a) Includes plant communities in eastern and western deciduous forests of North America
    (b) Samples collected from multiple populations—modelling approach that accounts for variation across populations and transects
    (c) Joint modelling approach = use sp-level estimates for traits to understand phenological cue responses and budburst timing
    (d) Taking a community-level approach—with woody tree and shrub species = different growth strategies and presumably suites of traits
2. Regardless of trait effects—our joint modeling approach still estimated phenological cue responses in line with previous work.

Figures

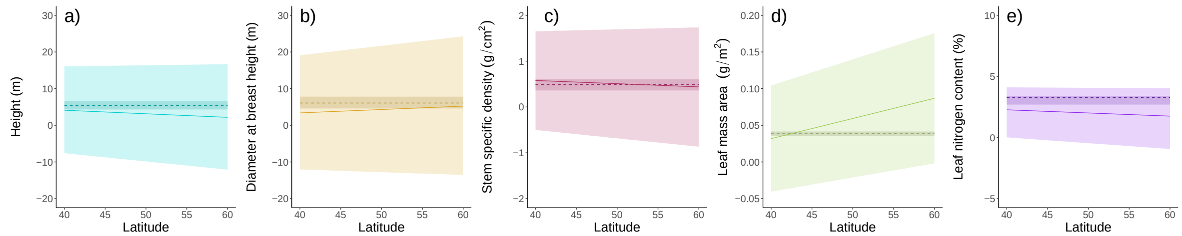

Figure 1: We found geographic differences for four of our functional traits, with the direction of the relationship varying across traits. Of our traits a. height, b. diameter at base height, c. leaf mass area, and e. leaf nitrogen content showed strong interactions between latitude and transect, while d. stem specific density showed no effects of geography. Dashed lines represent the western transect and solid lines the eastern transect.

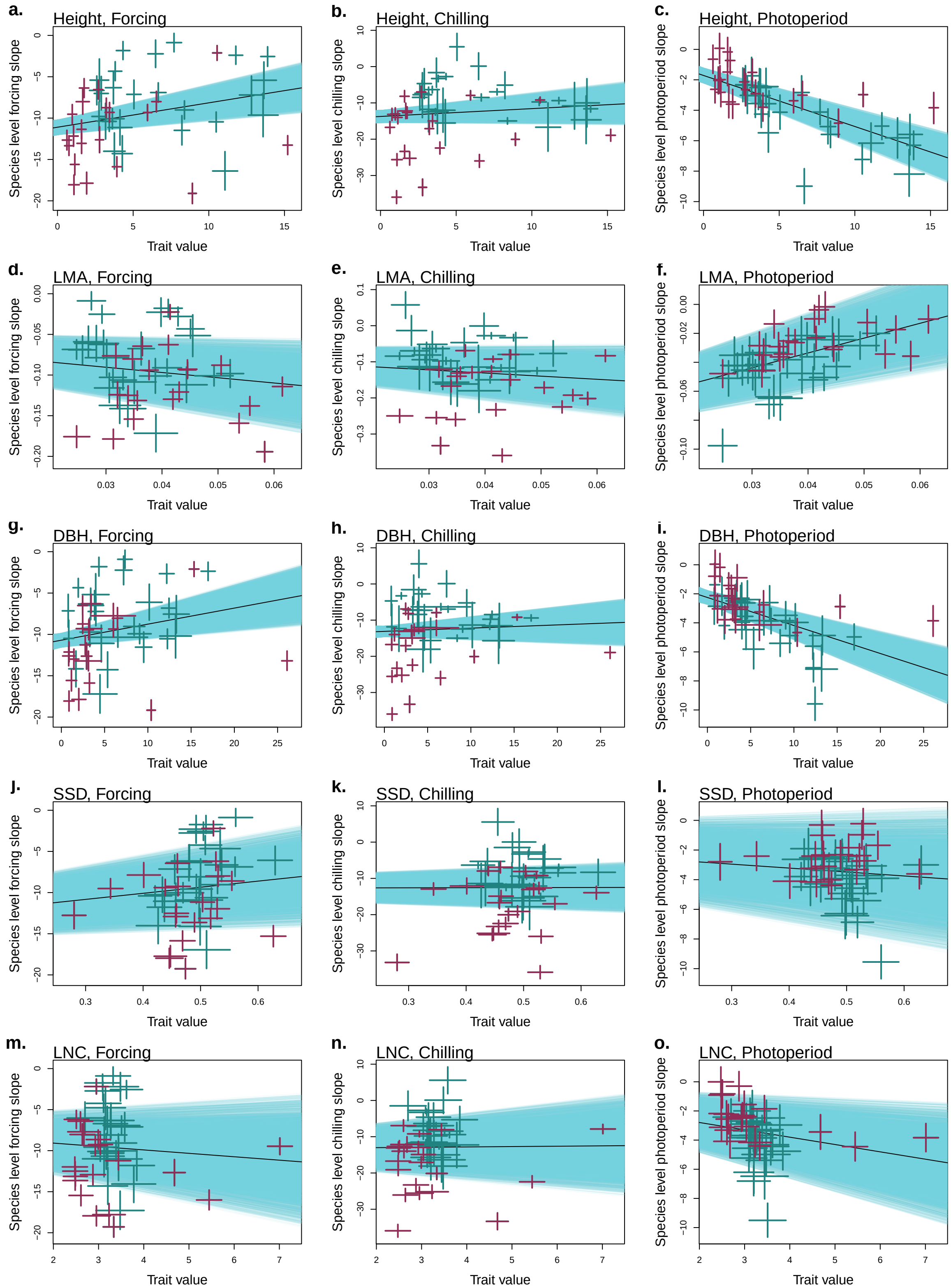

Figure 2: Relationship between species traits and cue responses, for height (a-c), leaf mass area (d-f), diameter at breast height (g-i), stem specific density (j-l), and the leaf nitrogen content (m-o). Point colours representing different species groups, with tree species are depicted in maroon and shrub species in teal.

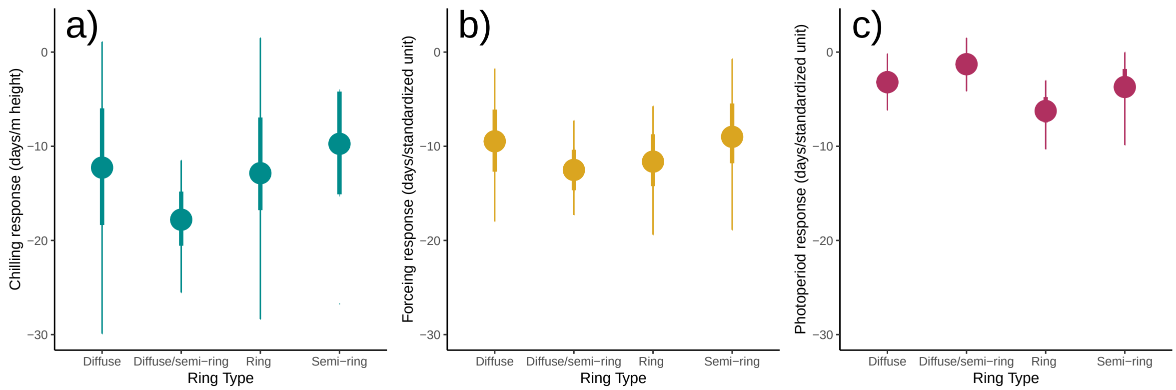

Figure 3: Despite species wood structures causing differing growth strategies, we did not find this trait to correlate with differences in cue responses across species. Thinner lines represent the 90% UI and thicker lines the 50% UI. Here we show the results for height only.

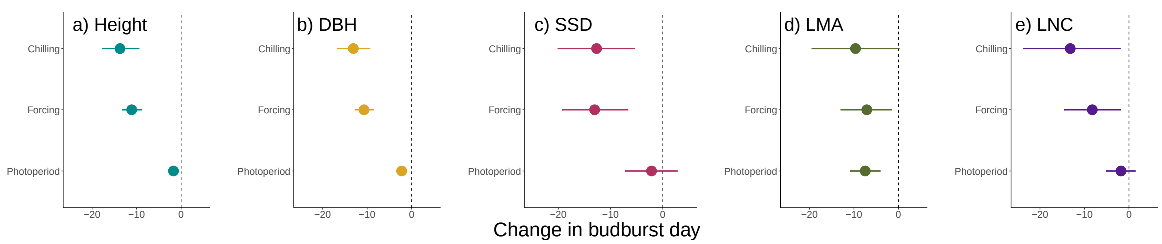

Figure 4: We found fairly consistent estimates for budburst cue responses to chilling, forcing, and photoperiod for each of our trait models: a. height, b. diameter at breast height, c. stem specific density, d. leaf mass area, and e. leaf nitrogen contnet. Lines represent 90% uncertainty intervals.

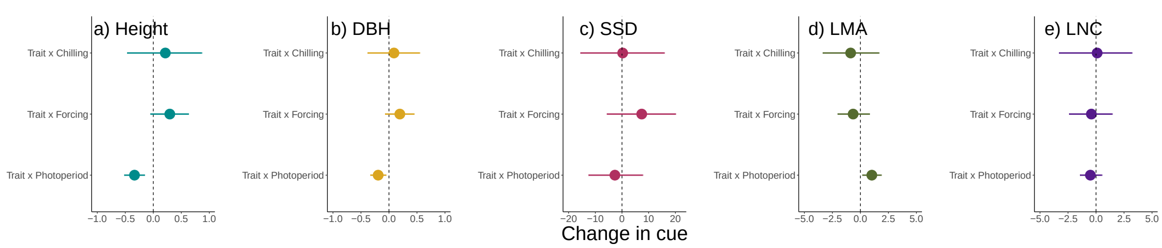

Figure 5: The relationships between traits and cue responses varied considerably across each of our trait models, a. height, b. diameter at breast height, c. stem specific density, d. leaf mass area, and e. leaf nitrogen content, and for individual cues. Lines represent 90% uncertainty intervals. Note the differences in the scale of the x-axis.

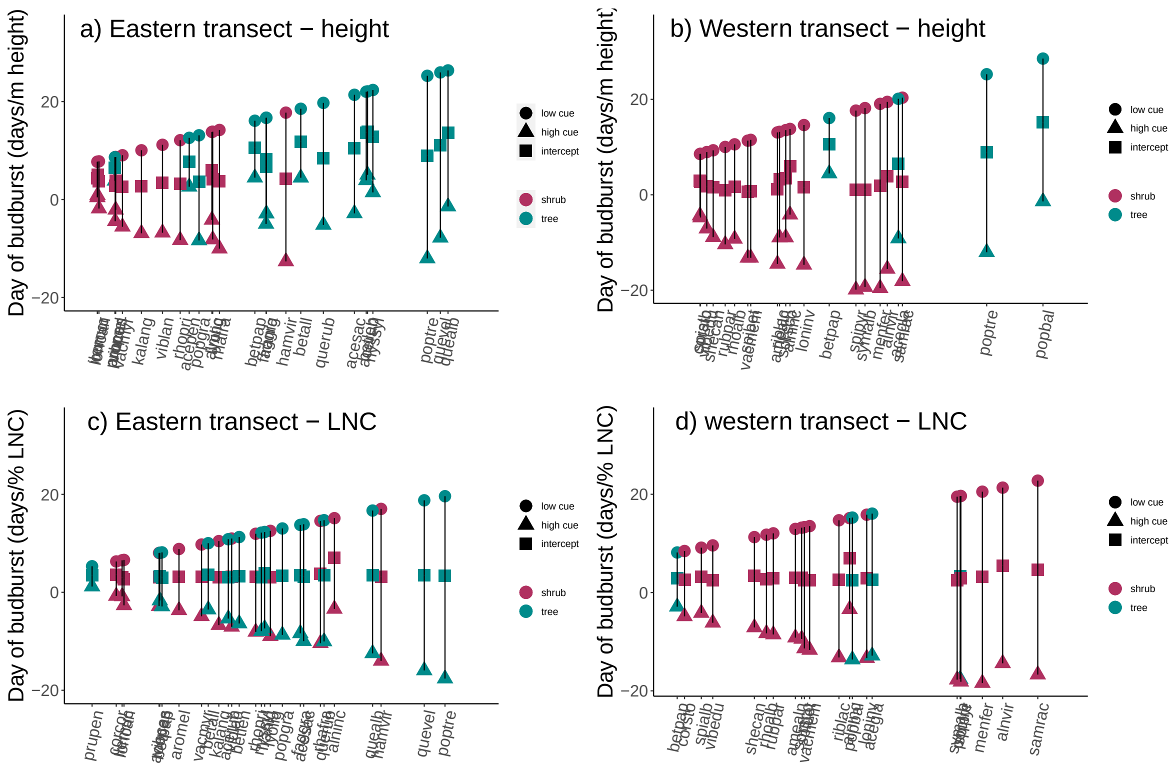

Figure 6: We found budburst estimates differed between our full model (intercept plus cues, depicted as triangles for high cues and as circles for low cues), versus the intercepts only model (without cues, shown as squares). Species are ordered in increasing budburst dates for both the eastern (a and c) and western (b and d) populations, spanning from early budbursting shrubs, in red, to late budbursting trees in blue. For traits such as height (a and b) we found distinct partitioning of budburst across shrub and tree species, but this was not the case for all traits, with our model of leaf nitrogen content showing highly mixed budburst order of shrub and tree species (c and d).

Tables

Table 1: Summary output from a joint Bayesian model of height and budburst phenology in which species are partially pooled. The effect of transect is modeled as a categorical variable and latitude as continuous in interaction term with transect. The model includes photoperiod and forcing as continuous variables, with all three cues z-scored to allow comparisons across cues.

	mean	5%	25%	75%	95%
Transect	2.60	-3.00	0.40	4.90	8.30
Transect x latitude	-0.10	-0.20	-0.10	-0.00	0.00
Forcing	-11.10	-13.30	-12.00	-10.20	-8.80
Chilling	-13.70	-17.80	-15.50	-12.10	-9.50
Photoperiod	-1.70	-2.90	-2.20	-1.20	-0.50
Trait x forcing	0.30	-0.10	0.20	0.40	0.60
Trait x chilling	0.20	-0.50	-0.00	0.50	0.90
Trait x photoperiod	-0.30	-0.50	-0.40	-0.30	-0.20

Table 2: Summary output from a joint Bayesian model of DBH and budburst phenology in which species are partially pooled. The effect of transect is modeled as a categorical variable and latitude as continuous in interaction term with transect. The model includes photoperiod and forcing as continuous variables, with all three cues z-scored to allow comparisons across cues.

	mean	5%	25%	75%	95%
Transect	-6.40	-13.70	-9.40	-3.40	1.00
Transect x latitude	0.10	-0.10	0.00	0.20	0.30
Forcing	-10.70	-12.80	-11.60	-9.90	-8.60
Chilling	-13.10	-16.70	-14.60	-11.60	-9.40
Photoperiod	-2.30	-3.40	-2.70	-1.80	-1.10
Trait x forcing	0.20	-0.10	0.10	0.30	0.40
Trait x chilling	0.10	-0.40	-0.10	0.30	0.50
Trait x photoperiod	-0.20	-0.30	-0.20	-0.10	-0.10

Table 3: Summary output from a joint Bayesian model of SSD and budburst phenology in which species are partially pooled. The effect of transect is modeled as a categorical variable and latitude as continuous in interaction term with transect. The model includes photoperiod and forcing as continuous variables, with all three cues z-scored to allow comparisons across cues.

	mean	5%	25%	75%	95%
Transect	0.40	-0.10	0.20	0.60	0.90
Transect x latitude	-0.00	-0.00	-0.00	-0.00	0.00
Forcing	-13.00	-19.30	-15.70	-10.50	-6.70
Chilling	-12.70	-20.10	-15.60	-9.70	-5.40
Photoperiod	-2.20	-7.20	-4.20	-0.00	2.80
Trait x forcing	7.40	-5.70	2.00	13.00	20.10
Trait x chilling	0.30	-15.60	-5.90	6.50	15.90
Trait x photoperiod	-2.70	-12.60	-7.00	1.50	7.80

Table 4: Summary output from a joint Bayesian model of LMA and budburst phenology in which species are partially pooled. The effect of transect is modeled as a categorical variable and latitude as continuous in interaction term with transect. The model includes photoperiod and forcing as continuous variables, with all three cues z-scored to allow comparisons across cues.

	mean	5%	25%	75%	95%
Transect	-11.70	-15.30	-13.20	-10.20	-8.00
Transect x latitude	0.30	0.20	0.20	0.30	0.40
Forcing	-7.10	-13.00	-9.30	-4.80	-1.60
Chilling	-9.60	-19.50	-13.50	-5.70	0.20
Photoperiod	-7.50	-10.90	-8.90	-6.00	-4.10
Trait x forcing	-0.60	-2.00	-1.20	-0.10	0.80
Trait x chilling	-0.90	-3.40	-1.90	0.10	1.70
Trait x photoperiod	1.00	0.20	0.70	1.40	1.90

Table 5: Summary output from a joint Bayesian model of LNC and budburst phenology in which species are partially pooled. The effect of transect is modeled as a categorical variable and latitude as continuous in interaction term with transect. The model includes photoperiod and forcing as continuous variables, with all three cues z-scored to allow comparisons across cues.

	mean	5%	25%	75%	95%
Transect	0.00	-0.80	-0.30	0.40	0.90
Transect x latitude	-0.00	-0.00	-0.00	-0.00	-0.00
Forcing	-8.20	-14.50	-10.80	-5.80	-1.80
Chilling	-13.20	-23.80	-17.70	-8.80	-2.00
Photoperiod	-1.80	-5.20	-3.10	-0.40	1.40
Trait x forcing	-0.40	-2.40	-1.20	0.40	1.40
Trait x chilling	0.10	-3.30	-1.20	1.50	3.20
Trait x photoperiod	-0.50	-1.40	-0.90	-0.10	0.50