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The role of genus and life span in predicting seed and vegetative trait variation and correlation in *Lathyrus*, *Phaseolus*, and *Vicia*

Sterling A. Herron[1] | Matthew J. Rubin[2] | Matthew A. Albrecht[3] |
Quinn G. Long[4] | Marissa C. Sandoval[5] | Allison J. Miller[1]

[1]Saint Louis University, Department of Biology, 3507 Laclede Avenue, St. Louis, Missouri 63103, USA

[2]Donald Danforth Plant Science Center, 975 North Warson Road, St. Louis, Missouri 63132, USA

[3]Missouri Botanical Garden, Center for Conservation and Sustainable Development, 4344 Shaw Boulevard, St. Louis, Missouri 63110, USA

[4]Shaw Nature Reserve, 307 Pinetum Loop Road, Gray Summit, Missouri 63039, USA

[5]University of California, Berkeley, Rausser College of Natural Resources, 319 Wellman Hall, Berkeley, California 94704, USA

Correspondence

Allison J. Miller and Sterling A. Herron, Saint Louis University, Department of Biology, 3507 Laclede Avenue, St. Louis, Missouri 63103, USA.
Email: allison.j.miller@slu.edu and sterling.herron@slu.edu

Abstract

Premise: Annual and perennial life history transitions are abundant among angiosperms, and understanding the phenotypic variation underlying life span shifts is a key endeavor of plant evolutionary biology. Comparative analyses of trait variation and correlation networks among annual and perennial plants is increasingly important as new herbaceous perennial crops are being developed for edible seed. However, it remains unclear how seed to vegetative growth trait relationships correlate with life span.

Methods: To assess the relative roles of genus and life span in predicting phenotypic variation and trait correlations, we measured seed size and shape, germination proportion, and early-life-stage plant height and leaf growth over 3 mo in 29 annual and perennial, herbaceous congeneric species from three legume genera (*Lathyrus*, *Phaseolus*, and *Vicia*).

Results: Genus was the strongest predictor of seed size and shape variation, and life span consistently predicted plant height and leaf number at single time points. Correlation networks revealed that annual species had significant associations between seed traits and vegetative traits, whereas perennials had no significant seed-vegetative associations. Each genus also differed in the extent of integration between seed and vegetative traits, as well as within-vegetative-trait correlation patterns.

Conclusions: Genus and life span were important for predicting aspects of early-life-stage phenotypic variation and trait relationships. Differences in phenotypic correlation may indicate that selection on seed size traits will impact vegetative growth differently depending on life span, which has important implications for nascent perennial breeding programs.

KEYWORDS

Fabaceae, Leguminosae, life history strategy, perennial grain, phenotypic correlation, phenotypic integration, relative growth rate, resource allocation, trade-offs, trait correlation network

Life history strategy in plants involves complex patterns of reproductive, growth, and survival trait allocation and trade-offs, which can shed light on past adaptive drivers and future evolutionary trajectories (Stearns, 1992). Describing ecological, genetic, and phenotypic life history patterns and commonalities among diverse taxa is thus fundamental in advancing evolutionary biology (Friedman and Rubin, 2015; Díaz et al., 2016). Annuals and perennials are two broadly

recognized life span categories in plants (Friedman, 2020). Annual plants grow, reproduce, and senesce within one year, whereas perennial plants live for multiple years and vary from short-lived, herbaceous individuals to expansive, ancient clonal colonies (e.g., *Populus*) and/or large woody individuals (e.g., *Sequoia*). Although traits associated with annual-perennial differences are well defined in some model systems (e.g., *Arabidopsis*; Albani and Coupland, 2010), this

comparative framework has garnered renewed attention with increasing focus on understudied, wild species as sources of novel crop breeding material, particularly perennial species closely related to annual crops, such as corn, rice, and wheat (Lundgren and Des Marais, 2020). Furthermore, life span transitions are common in many angiosperm systems (e.g., Castillejinae: Tank and Olmstead, 2008; core Pooideae: Lindberg et al., 2020; Saxifragales: Soltis et al., 2013), which allows for parallel investigation of phenotypic and genetic commonalities in life history evolution across diverse groups.

Several broad life history frameworks have been proposed to interpret the range of phenotypic variation and trait correlation in plants. Classic plant life history theory balances an individual's survival with reproduction as modes of achieving fitness (Cole, 1954; Gadgil and Solbrig, 1972; Charnov and Schaffer, 1973). Derivations of this have described systems of interconnected phenotypes that span predictable trait networks or spectra (Grime, 1977; Wright et al., 2004; Díaz et al., 2016). Evidence for differences in traits associated with life span comes largely from leaf and shoot traits (Garnier, 1992; Garnier and Laurent, 1994; Atkinson et al., 2016), reproductive traits (Pitelka, 1977; Vico et al., 2016), and root traits (Roumet et al., 2006). Nevertheless, it has long been recognized that plant species with different life histories display diverse trait systems that are not always predicted by generalized models; these traits are ultimately determined by the species' distinct set of environmental stressors and evolutionary history (Thompson and Hodkinson, 1998; Crews and DeHaan, 2015). Herbaceous perennials have been recognized as particularly variable in life history trait combinations (Grime, 1977; Kitchen, 1994), and several studies have found evidence for herbaceous perennial species with acquisitive and ruderal traits (i.e., swift resource acquisition and fast growth and maturation to produce seeds before environmental disturbance), including comparable reproductive biomass and relative growth rate to annuals (Verboom et al., 2004; González-Paleo and Ravetta, 2015; Vico et al., 2016).

Understanding how traits are correlated is crucial to advancing life history theory. For example, relationships between trait variation in different plant organs, or phenotypic integration, helps shed light on whether or not key traits were consistently selected together as a functional module, if functional trade-offs are occurring, or if the traits in question remain functionally independent (Murren, 2002). Furthermore, trait relationships can reveal fundamental selective constraints on plant phenotypic diversity. Previous studies in herbaceous species found negative relationships between vegetative growth and some aspects of reproduction (e.g., flowering meristems in an annual; Geber, 1990) but generally positive relationships between vegetative size traits and size of seeds produced within a species (Geber, 1990) and among species (in herbaceous perennials; Kleyer et al., 2019). Other studies reported that life span plays an important role in predicting functional reproductive-vegetative trait relationships, including the association of flowering time with seed size and plant height

(Bolmgren and Cowen, 2008; Du and Qi, 2010; Segrestin et al., 2020). However, to our knowledge, few studies have focused on congeneric annual-perennial differences in correlations of seed size with vegetative plant size and growth rate. Here, we build on previous life history studies by investigating how relationships between seed and vegetative traits may shift with life span in multiple genera within a single family.

Due to its life history diversity, as well as its economic, ecological, and agricultural importance, the legume family (Fabaceae Lindl.) is an excellent system in which to study phenotypic patterns associated with life span (Ciotir et al., 2019). Similar to other systems, annual legume species tend to have greater relative allocation to sexual reproduction in percent energy and percent dry biomass than perennial congeners (Pitelka, 1977; Turkington and Cavers, 1978). Studies have also explored other functionally important traits in congeneric annual and perennial legumes, including seed mass (Pitelka, 1977; Marshall et al., 1985; Kelly and Hanley, 2005; Ward et al., 2011; Herron et al., 2020); specific leaf area (den Dubbelden and Verburg, 1996; Roumet et al., 2000); photosynthetic rate (Pitelka, 1977; Roumet et al., 2000); and growth rate (den Dubbelden and Verburg, 1996; Kelly and Hanley, 2005). In these studies, life span was commonly not the focus, or it was unclear whether annuals and perennials differed significantly. These studies also tended to focus on two or three species per genus, while a much greater range of annual and perennial species diversity exists within genera and across Fabaceae (Ciotir et al., 2019).

Here, we focus on a panel of annual and herbaceous perennial species from the agriculturally important Fabaceae genera *Lathyrus* L. (grass pea), *Phaseolus* L. (common bean), and *Vicia* L. (vetch) in order to characterize phenotypic diversity associated with life span and expand current understanding of trait variation and correlation associated with different life history strategies. We asked the following questions: (1) What is the relative importance of genus and life span in predicting seed and vegetative trait variation? (2) How do seed and vegetative trait correlations differ across genera and life spans? We addressed these questions by quantifying seed size, germination, and early-life-stage vegetative growth in congeneric, wild herbaceous annual and perennial species grown in a common environment.

MATERIALS AND METHODS

Plant material

We obtained 80 accessions representing annual and perennial species within *Lathyrus*, *Phaseolus*, and *Vicia* from the U.S. Department of Agriculture (USDA) National Plant Germplasm System (Western Regional Plant Introduction Station, Pullman, Washington, USA) in spring 2017. Most accessions consisted of seeds harvested from plants

grown at germplasm facilities (accession information is available in Appendix S1). When possible, we obtained accessions of annual and perennial species from closely related phylogenetic groups based on published molecular phylogenies of each genus (Appendix S2). For *Lathyrus*, five annual and five perennial species were studied; for *Phaseolus*, four annual and five perennial species; and for *Vicia*, five annual and five perennial species (Table 1). One to seven accessions were used for each species, with origins from diverse geographic areas when possible. All accessions used were designated as "wild" by the USDA (except one accession of *Vicia americana*, which was later determined to be wild), meaning that seed collection took place in a natural population outside of cultivation, although this may also include naturalized populations that escaped from cultivation in some cases. Seeds were stored in a desiccator at 3–4.5°C and 36% relative humidity prior to the start of the experiment. For each accession used in this study, life span and cultivation status were derived from the online USDA description in the Germplasm Resources Information Network (GRIN-Global), which was checked against literature sources (Freytag and Debouck, 2002; Wu et al., 2010; Schaefer et al., 2012; more information on life span variation is available in Appendix S2).

Seed size and shape measurements

In this study, seeds are the initial juvenile stage in plants later measured for vegetative growth. A pool of seeds for each accession was first weighed to the nearest 0.1 mg and divided by the number of weighed seeds for that accession to estimate mean single seed mass. Seeds were then imaged on an EPSON DS-50000 scanner (Nagano, Japan), and images were formatted to 400 dpi and analyzed in ImageJ (Schneider et al., 2012). Severely damaged seeds and seeds in a nonstandardized orientation (only occurring for some bilaterally symmetric seeds of *Lathyrus cicera*) were removed from analysis. In ImageJ, seed images were cropped and converted to binary ("Make Binary" function) or, if the contrast was not well defined, the image was converted to 8-bit grayscale and a binary threshold ("Threshold" function) was applied and the threshold value adjusted for the highest seed contrast and lowest noise (shadows). Remaining pixel holes within seeds were removed using the "Fill Holes" function, and erroneous gaps on the perimeter of seeds were filled using the "Convex Hull" function (gift wrapping algorithm) or traced in manually when this was not adequate. Small accessory structures still attached to the seeds (primarily funicles) were removed from the image. From this final image we extracted four size parameters per seed: length (Feret's diameter, or the maximum distance between any two points along the perimeter of the seed), width (minimum distance between two points along the perimeter of the seed, perpendicular to the length), perimeter, and area; and two shape parameters: circularity and roundness. Circularity is calculated as $4\pi \times$ (area)/

(perimeter)^2 and represents the extent to which the seed shape approximates a circle, ranging from 0 to 1. Roundness is calculated as $4 \times$ (area)/($\pi \times$ (major axis length)^2) and is the inverse of the seed's aspect ratio (length to width ratio of the best-fitting ellipse). The majority of the seeds measured for size and shape were then used in germination experiments.

Germination measurements

Following weighing and scanning, seeds were surface-sterilized in 6% sodium hypochlorite aqueous solution for 5 to 6.5 min, then rinsed with reverse osmosis water and patted dry (Frehner and Conn, 1987; Galasso et al., 1997). To break physical dormancy characteristic of many legumes (Baskin and Baskin, 2014), seeds were scarified using P100 and P60 grade sandpaper; malformed and severely damaged seeds were excluded from germination analyses. All scarified seeds were surface-sown in unsterilized quartz sand (Fairmount Santrol Handy Sand, Chesterland, Ohio, USA; ~34 mL) in 20 mm deep plastic Petri dishes; seeds were oriented horizontally (morphology allowing, hilum parallel lengthwise to substrate surface) in a grid pattern. Dishes were initially watered to saturation and were remoistened when dry. Seeds were germinated in 12:12 h light:dark conditions inside a temperature-controlled incubator, with 20:10°C and 25:15°C light:dark temperature settings for temperate and subtropical/tropical species, respectively, in order to expose the species to temperatures similar to their native range. *Vicia* seeds were incubated at temperate settings, *Phaseolus* seeds were incubated at tropical settings, and *Lathyrus* seeds were divided into both settings depending on the species (*Lathyrus* annuals included both temperate and subtropical species, perennials only temperate) (Appendix S1). Each accession had two or three replicate Petri dishes and 5–26 seeds per replicate (Appendix S1). Replicate dishes were randomized when placed in the incubator and re-randomized after each germination check.

Germination was defined as an extension of the radicle past the seed coat or, in rare cases, backwards emergence of the seedling due to the radicle pushing against the seed coat. Germinated and imbibed seeds were counted beginning 1 d following placement on the substrate, then every several days up to 10–12 d, and then at 3 wk and/or 4 wk, until all imbibed, viable seeds germinated (Baskin and Baskin, 2014). From the germination counts up to 4 wk, days to 50% germination (T_{50}) was calculated as a measure of germination time, using the "PROBIT" procedure in SAS version 9.4, which calculates a maximum likelihood estimate of germination timing with a default maximum iteration of 50 (University Edition; SAS Institute, 2017). The final total of germinated seeds from all replicates was used to calculate germination proportion by accession. Accession age (years in frozen storage prior to this study, ranging from 2 to 30 yr) was the only covariate for germination T_{50} and proportion. See Appendix S3 for additional details on T_{50} and germination

TABLE 1 Sampling summary of seed, germination, and vegetative traits in annual and perennial species of *Lathyrus*, *Phaseolus*, and *Vicia*

Genus	Life span	Species[a]	Seed size Accessions	Seeds	Germination Accessions	Seeds	Vegetative growth Accessions	Plants
Lathyrus L.	Annual	*L. annuus* L.	1	89	1	60	1	12
		L. aphaca L.	3	82 (62–99)	2	60 (59–60)	2	13 (11–15)
		L. cicera L.	3	69 (61–77)	3	57 (52–60)	3	11 (9–12)
		L. hirsutus L.	4	74 (40–93)	4	50 (37–57)	4	10 (4–13)
		L. odoratus L.	1	61	1	51	1	12
	Perennial	*L. japonicus* Willd.	3	94 (49–151)	3	54 (44–60)	3	15 (8–19)
		L. latifolius L.	4	71 (62–80)	4	59 (56–60)	4	20 (18–22)
		L. pratensis L.	3	155 (121–174)	3	60 (60)	3	18 (18–19)
		L. sylvestris L.	4	80 (63–100)	4	56 (45–60)	4	16 (12–19)
		L. tuberosus L.	1	101	1	56	1	8
Phaseolus L.	Annual	*P. acutifolius* A. Gray	6	57 (46–68)	6	52 (42–61)	6	13 (9–15)
		P. filiformis Benth.	6	63 (53–74)	6	53 (43–60)	6	15 (10–20)
		P. lunatus L.	2	29 (25–33)	2	27 (24–29)	2	14 (13–15)
		P. vulgaris L.	7	35 (25–52)	7	33 (23–49)	7	10 (7–13)
	Perennial	*P. angustissimus* A. Gray	2	58 (57–59)	2	54 (53–54)	2	25 (21–29)
		P. leptostachyus Benth.	4	52 (39–68)	4	46 (34–60)	4	19 (17–20)
		P. maculatus Scheele[b]	1	37	1	35	1	16
		P. parvulus Greene	1	41	1	38	1	22
		P. polystachios (L.) Britton, Sterns & Poggenb.	2	30 (26–34)	2	28 (25–30)	2	21 (19–22)
Vicia L.	Annual	*V. benghalensis* L.	1	63	1	57	1	11
		V. ervilia (L.) Willd.	3	59 (52–64)	3	53 (46–58)	3	14 (13–15)
		V. hirsuta (L.) Gray	2	128 (118–138)	2	57 (55–58)	1	10
		V. sativa L.	3	69 (61–75)	3	59 (57–60)	3	13 (11–15)
		V. villosa Roth	4	64 (53–83)	4	54 (48–60)	4	12 (8–14)
	Perennial	*V. americana* Muhl. Ex Willd.	1	124	1	58	1	31
		V. cracca L.	4	88 (67–117)	4	59 (56–60)	4	19 (17–21)
		V. dumetorum L.	1	59	–	–	1	13
		V. sepium L.	1	90	1	60	1	20
		V. tenuifolia Roth	2	81 (77–85)	2	60 (60)	2	32 (26–38)

Notes: "Seeds" and "plants" respectively refer to the average seed number and plant number among accessions studied for each species, with the range of seed/plant number among accessions in parentheses, where applicable. The maximum sampling is shown (after removal of any problematic data and outliers), where seed size collectively includes seed mass and ImageJ seed measurements, germination includes germination proportion and T_{50}, and vegetative growth includes height and/or leaf number measurements on a plant at one or both dates (DAP-21 and DAP-35). A trait-by-trait summary of accession-level sampling is in Appendix S1. Also, some accessions were dropped before use in the accession-level PCA (see statistical methods)—these included six accessions: one each of *Lathyrus annuus*, *L. aphaca*, *L. cicera*, *Phaseolus acutifolius*, *Vicia dumetorum*, and *V. hirsuta* (Appendix S1).

[a]Some species' accessions include subspecific rankings. These include *Lathyrus japonicus* subsp. *maritimus* (1 accession), *Phaseolus acutifolius* var. *acutifolius* (3), *P. acutifolius* var. *tenuifolius* (3), *P. maculatus* subsp. *ritensis* (1), *P. polystachios* subsp. *polystachios* (1), *P. polystachios* subsp. *sinuatus* (1), *P. vulgaris* var. *aborigineus* (all accessions), *Vicia tenuifolia* subsp. *tenuifolia* (1), and *V. tenuifolia* subsp. *dalmatica* (1).

[b]The USDA has since changed the taxonomic status of *P. maculatus* subsp. *ritensis* to *P. ritensis*, but we retain its original taxonomic status here (*sensu* Freytag and Debouck, 2002).

proportion methodology. A subset of vigorous seedlings from at least two Petri dish replicates of each accession was planted soon after germination (see next section).

Vegetative growth measurements

Using seeds germinated in the experiment above, a subset of seedlings was planted in Ball Professional Growing Mix (no peat; West Chicago, Illinois, USA) within 38-cell trays (cell diameter 4.95 cm, cell depth 12.65 cm) as soon as possible following germination, usually with all 38 cells filled with seedlings of different accessions. For each accession, 4–38 seedlings (mean = 15.3 ± 5.6) were transplanted and measured in total and were arranged in replicate groups across different trays (1–7 replicates per accession, mean = 3.6; Appendix S1). From 19 June to 25 August 2017, plants were grown in hoophouse A-6 at the Missouri Botanical Garden (St. Louis, Missouri, USA), which was covered in a light shade cloth; day-night temperatures ranged from 14°C to 40°C. Typically plants were watered daily. Beginning at the first week of measurement, 150 ppm 15-5-15 NPK aqueous fertilizer was applied approximately weekly, and climbing plants were trained around thin bamboo poles for taller plants (usually *Phaseolus*) or 18-inch hyacinth sticks for shorter plants. Growth trays were spatially randomized in the hoophouse on 1 August. Pesticides were applied on 15 August in the hoophouse to control thrips, consisting of an aqueous solution of Pylon (0.08% v/v), Mavrik (0.08% v/v), and Tristar (0.52% v/v). A subset of plants had to be transferred on 25 August to the Saint Louis University Biology Department greenhouse, where accessions of a mix of annual and perennial *Lathyrus* (10 accessions) and *Vicia* (3 accessions) had their second measurement (DAP-35) taken over 2 wk (Appendix S1). Here, plants were watered every few days and temperatures were similar to the previous hoophouse, but lighting was more heterogeneous. Due to any impact of this transfer, linear models were also tested with these accessions dropped (Appendix S4).

Vegetative growth was measured nondestructively for each individual plant over a 2 wk period. Planting date in the hoophouse was used as a baseline for vegetative measurements, since seedlings showed most growth development after this date. All seedlings were first assessed for plant height, leaf number, vigor, and reproductive status beginning 20–22 d after planting (DAP-21; time point 1) in the hoophouse, with the same measurements taken at 34–37 d after planting (DAP-35; time point 2) to determine growth rate on an individual plant level, as well as the height and leaf number measurements at the respective single time points (height and leaf number DAP-21 and DAP-35, i.e., measures of vegetative size). Plant height was measured from ground level to the base of the shoot apex on the tallest stem. Leaf number was counted for each node on the same stem measured for height, up to the most recently developed, countable node, at least ~2 mm from the next node near the shoot apex. If a node was leafless, a leaf was counted only if there was a clear remaining structure indicating that a leaf was present, usually stipules. For all *Phaseolus* species in this study, two unifoliate eophylls are present at the first true node, and thus two leaves were counted for that node for all individuals, even if they had dehisced. The leaves of *Lathyrus aphaca* are reduced to tendrils, and the enlarged stipules are functionally the main photosynthetic organ; thus, each pair of stipules was counted as one leaf (Sharma and Kumar, 2012). Absolute growth rate (AGR) was calculated on a per day basis by dividing the difference in height and leaf number by the number of days elapsed between the two time points (as defined in Rees et al., 2010; Pommerening and Muszta, 2016). Relative growth rate (RGR) was calculated for height and leaf number by taking the difference of the natural log of the trait at both dates and dividing by the days elapsed: ln(trait DAP-35)−ln(trait DAP-21)/(time point 2−time point 1) (as in Perez-Harguindeguy et al., 2013). The vigor covariate was a qualitative assessment of the impacts of environmental and endogenous factors on plant health; this was scored categorically as "low" (damaged or unhealthy, with clear impact on growth), "medium" (some damage or somewhat unhealthy, with some impact on growth), or "high" (little or no damage and robust growth). The lowest vigor observed between the two dates was assigned as the covariate for AGR and RGR analyses. Plants with severe damage were removed from analysis; if the damage occurred by DAP-21, all vegetative data were excluded for that plant, and if the damage occurred by DAP-35, only DAP-35, AGR, and RGR data were excluded. If either height or leaf number AGR/RGR was negative, all vegetative data were removed for that plant. Given the possible preferential allocation of energy to reproductive structures, we scored the reproductive status of the plant ("reproductive" or "nonreproductive") at both dates measured (if either date was "reproductive," that was assigned to AGR and RGR as well). Lastly, for AGR and RGR, height at DAP-21 was included as a covariate to help control for the effect of different developmental sizes at the start of the growth period. Each of these covariates was tested as a random effect in our linear models (statistical analysis section). Any accessions were dropped for a vegetative trait if fewer than three plants remained following final data filtering.

Statistical analyses

Statistical analyses were performed in R version 3.6.1 (R Core Team, 2019), and graphics were completed using "ggplot2" (Wickham, 2016). Principal component analysis (PCA) was computed on data scaled to unit variance using the "prcomp" function (base R). PCA was first implemented for accession-level data for all traits in the full data set and then for individual-level seed and vegetative data sets for each genus separately. Eigenvalues were derived using the "get_eigenvalue" function from the R package factoextra (Kassambara and Mundt, 2020).

The PCA variable plot was created using the R package ggbiplot (Vu, 2011). Accessions and individuals with much reduced data (about half of the traits missing) or that had removed outliers (see last paragraph of this section) were excluded entirely from PCAs (noted in Appendix S1). Specifically in the case of individual-level vegetative data, individuals were dropped if measurement data were missing from an entire date (DAP-21 or DAP-35), and thus growth rate was missing as well. In the case of accessions and individuals with few traits with missing data (one to three), missing values were imputed using a regularized iterative PCA algorithm ("imputePCA" function in the R package missMDA; Josse and Husson, 2016), for which the number of components were estimated using generalized cross-validation ("estim_ncpPCA" function). For the accession-level PCA, imputation was necessary for seven out of 1258 data values (0.6%).

Linear mixed models were employed on a trait-by-trait basis to assess contribution of genus, life span, species, and covariates to phenotypic variation using the "lmer" function in the R package lme4 (Bates et al., 2015). For all ImageJ seed measurements and vegetative traits, models were tested on individual seed- and plant-level data, respectively. For germination T_{50}, the model was tested on individual replicate-level data. The remaining trait models used accession-level data. Each model included genus, life span, genus × life span, and species (nested within genus × life span) as the main fixed effects and accession nested within species as a random effect (except for principal components, seed mass, and germination proportion, since they were measured at the accession level). For vegetative traits, replicate nested within accession was additionally included in the model as a random effect. Trait models were assessed using a type III analysis of variance (ANOVA) with Satterthwaite's method for degrees of freedom, with the exception of traits consisting of only accession-level data, which were assessed using a type I ANOVA (and the base R "lm" function). Type III ANOVA significance was assessed using the R package afex (Singmann et al., 2020) in conjunction with lme4. Using the R package emmeans, adjusted means for each genus-life span combination were derived from our models ("emmeans" function) and post hoc custom contrasts were conducted using a Bonferroni correction ("contrast" function; Lenth, 2020). The mentioned covariates for germination and vegetative measurements were added as random effects for the appropriate trait model. Significance of random effects was assessed by taking the likelihood ratio test of the model following sequential removal of each random term, using the "rand" function in the R package lmerTest (Kuznetsova et al., 2017); nonsignificant random effects were dropped from the final model (the "reduced model"; see Appendix S5 for the final significance of random effects). In some cases, certain accessions or species were dropped from models in order to assess their influence where potential confounding factors were present (Appendix S4).

To identify phenotypic relationships in the full data set and within each genus and life span subgroup, bivariate trait correlations were calculated. Using scaled and centered accession-level data, Pearson correlation coefficients and P-values were generated for each bivariate trait correlation using the "rcorr" function in the R package Hmisc (Harrell, 2020); this function removes missing values in pairs instead of removing an entire accession that has a missing value, thus retaining the maximum amount of data. Bivariate correlations and their significance were visualized using network plots generated with the R package igraph (arranged with the "tkplot" function; Csardi and Nepusz, 2006). Correlation matrices were also visualized with the "corrplot" function in the R package corrplot (Wei and Simko, 2017). Accessions with reduced sampling replication that exhibited trait values that were statistical outliers in their genus (defined as being >1.5× the interquartile range below or above the first and third quartiles, respectively) were removed from all analyses of that trait (models, PCA, and correlations). This resulted in dropping one Phaseolus and Vicia accession for germination T_{50} (both with $n = 2$ replicates), and one Lathyrus accession ($n = 1$ replicate) for most vegetative traits (Appendix S1).

RESULTS

We investigated variation and correlation in seed, germination, and early-life-stage vegetative growth traits in 80 accessions of annual and perennial Lathyrus, Phaseolus, and Vicia species. Genus was the strongest predictor of seed size and shape variation, and life span consistently predicted plant height and leaf number at single time points. Broad correlation networks revealed that annual species had significant associations between seed traits and vegetative traits, whereas perennials had no significant seed-vegetative associations. Each genus also differed in their patterns of integration between seed and vegetative traits, as well as within-vegetative trait correlation patterns.

Genus and life span predict trait variation

Principal component analyses identified key phenotypic differences among genera and between life spans, and identified correlated groups of traits (Figure 1). The first two principal components (PC1 and PC2) explained 42.2% and 24.7% of variance, respectively. Seed size traits clustered tightly (Figure 1A) and loaded positively on both PC1 and PC2. Seed shape (circularity and roundness) loaded negatively onto PC1 (Figure 1A, Appendix S6), suggesting that seed size and seed shape are negatively correlated. Vegetative traits were less tightly grouped: height at DAP-21 and DAP-35 and leaf number at DAP-21 loaded positively onto PC1, whereas leaf number at DAP-35 and AGR and RGR for both height and leaf number loaded negatively onto PC2 (Figure 1A, Appendix S6). Germination T_{50} loaded positively onto PC2, opposite of vegetative traits, whereas germination proportion loaded positively onto PC1 in conjunction with vegetative traits (Figure 1A, Appendix S6).
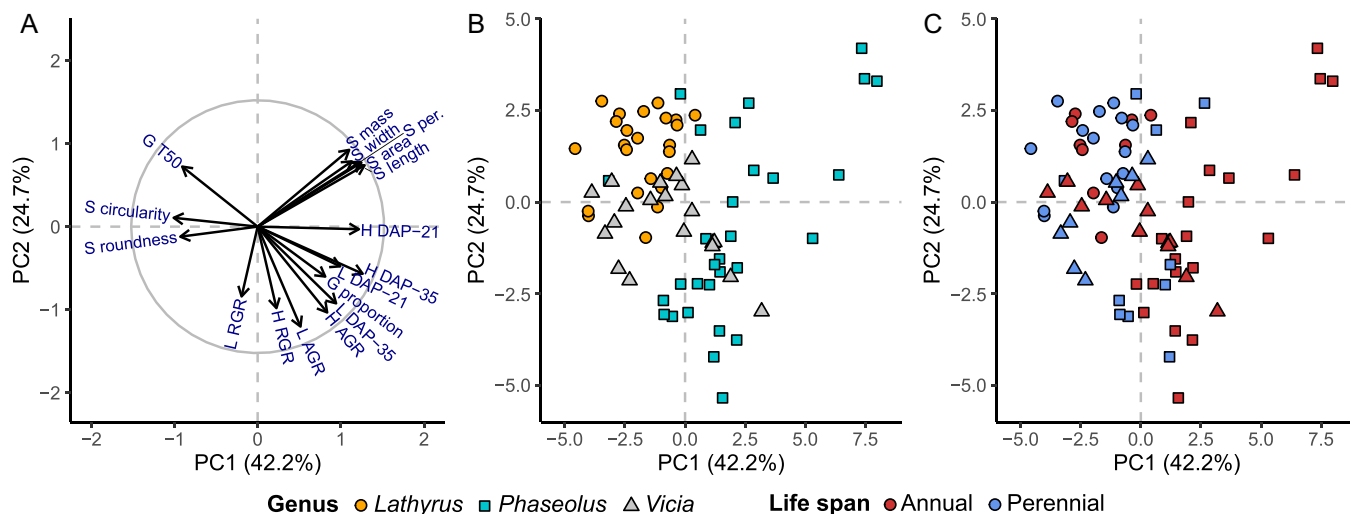
FIGURE 1 Principal component analysis (PCA) for the full data set accession means. (A) Variable correlation circle, which shows the relative representation of each variable on the first two principal components (specifically, the correlation between the variable and the principal components), where distance of each variable's arrow from the origin indicates increasing representation of that trait in the PCA in a particular region of principal component space. It also shows the correlations between variables, where variables spaced more closely are positively correlated. S, seed (perim. = perimeter); G, germination; H, height; L, leaf. AGR is absolute growth rate, and RGR is relative growth rate. Variable labels were sometimes adjusted slightly from arrow tips to allow complete visualization. (B) Individual accession data points mapped to the same principal component space with color and shape corresponding to genus. (C) Individual accession data points with color corresponding to life span and shape to genus

Phaseolus occupied the largest area in PC1 and PC2, followed by *Vicia* then *Lathyrus* (Figure 1B). Annuals occupied a greater area of principal component space than perennials, with much of this area occupied by *Phaseolus* annuals (Figure 1C). Among genera, *Phaseolus* accessions tended to occupy space in PC1 and PC2 showing greater seed size and greater height and leaf number at DAP-21 and DAP-35, as well as higher growth rate in some cases; *Lathyrus* and *Vicia* tended to have smaller, more circular seeds with a delayed T_{50} and smaller vegetative traits (Figure 1B; Appendix S6). Comparing annual and perennial species within genera, annuals showed greater values on PC1 than perennials for *Phaseolus* and *Vicia*, indicating greater seed size and single-time-point vegetative size (Figure 2A–C, Appendix S6). However, the mean difference in PC1 was significant only for *Phaseolus*, and *Lathyrus* showed the opposite pattern (Figure 2C, Appendix S7). No significant, consistent differences were detected for PC2 between life spans within genera, indicating a general lack of difference in AGR and RGR between life spans (Figure 2D–F; Appendices S6 and S7). For PC3, all three genera exhibited a significantly higher mean value in annuals than perennials, again indicating greater single-time-point vegetative size but lower RGR (loading more strongly than in PC2) in annuals compared to perennials (Figure 2G–I; Appendices S6 and S7). Consistent with Figures 1 and 2, our linear models also identified genus as the strongest predictor of PC1 and PC2, and life span as the strongest predictor of PC3 (Table 2).

Principal component analyses of seed and vegetative data highlighted similar seed trait loading patterns in each genus; however, differences among genera existed in vegetative trait loadings and in how annual and perennial species separated in

phenotypic space (Appendices S8, S9, S10, S11, and S12). In each of the three genera, seed size traits were tightly correlated and loaded positively onto PC1, while seed shape (circularity and roundness) loaded positively onto PC2, approximately orthogonal to seed size (Appendices S8, S9, S10, and S11). In general, annual and perennial species did not separate consistently in principal component space across genera with respect to seed size; however, *Phaseolus* perennials had more distinctly circular/round seeds than annuals (Appendices S8, S9, and S10). By contrast, vegetative traits showed less consistent and less closely grouped loading patterns in PCAs across genera (Appendices S8, S9, S10, and S12). Similar to seed PCAs, *Lathyrus* and *Vicia* had more overlap in annual-perennial vegetative variation than *Phaseolus*. Annual *Phaseolus* species had consistently greater single-time-point vegetative size than perennial species (Appendices S8, S9, and S10). Nevertheless, two annual *Vicia* species, *V. villosa* and *V. benghalensis*, showed distinctly greater single-time-point vegetative size than other *Vicia* species (Appendix S10). These data revealed important species-level variation underlying the broad life span patterns within each genus.

In our linear mixed models, genus was the strongest predictor of seed trait variation (Table 2). Genus was significant for all seed traits except seed mass, whereas life span was not significant for any seed trait (Table 2). Despite lack of life span significance, post hoc tests revealed that all seed size traits except seed mass were significantly larger in annual *Phaseolus* species than in perennial *Phaseolus* species (Appendix S7). For seed circularity and roundness, there was a significant genus × life span interaction for *Phaseolus* where both traits were significantly greater in perennials than in annuals, but roundness was significantly lower in perennials than in annuals for
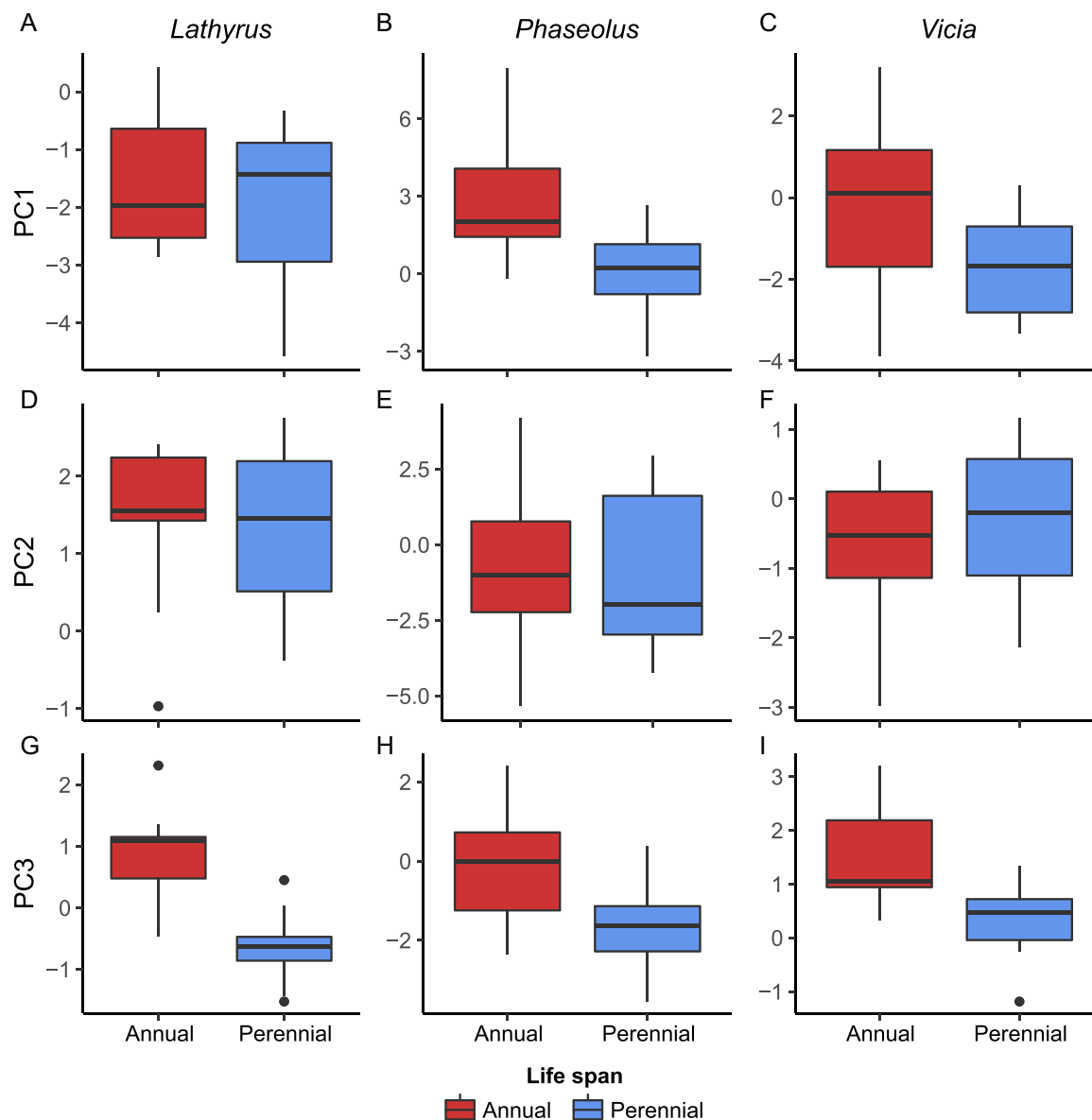
FIGURE 2 Boxplot representing the median values of annual and perennial accessions for each genus for the first three principal components: PC1 (A–C), PC2 (D–F), and PC3 (G–I) (as displayed in Figure 1C). The thick black line represents the median, box hinges represent the 25th and 75th percentiles (interquartile range; IQR), and the whiskers represent 1.5 * IQR. Data beyond 1.5 * IQR are shown as outlier points

Lathyrus (Table 2 and Appendix S7). Linear models revealed a significant genus and life span effect for germination T_{50}, while only genus was significant for germination proportion (Table 2). Perennials in all genera had delayed mean T_{50} compared to annuals, but this was significant only for *Lathyrus* (Appendix S7). Nevertheless, germination results should be interpreted with caution, as the genus effect cannot be dissociated from effects of incubator settings (see Materials and Methods).

By contrast, life span was the most consistent predictor of vegetative trait variation at single time points (Table 2). Life span was a significant predictor for height and leaf number DAP-21 and DAP-35, with more variation attributable to life span than to genus (nonsignificant) for height (Table 2).

For leaf number DAP-21 and DAP-35, life span explained a similar amount of variation as genus, and both effects were significant (Table 2). For all genera, perennials had lower mean height and leaf number at DAP-21 and DAP-35 than congeneric annuals; this difference was significant for *Phaseolus* (height and leaf number) and for *Vicia* (leaf number only; Appendix S7). Vegetative growth rate patterns (AGR and RGR) did not show consistent patterns with respect to life span differences within each genus, and neither genus nor life span was significant for these traits in the linear models (Table 2). *Lathyrus* showed a significant difference in leaf number RGR (perennials greater than annuals) and *Phaseolus* in height AGR and RGR (annuals greater than perennials; Appendix S7). Although nonsignificant, in *Lathyrus* both

TABLE 2 Analysis of variance (ANOVA) table of the final, reduced linear mixed models for the full data set, including accession-level principal components (Figure 1) and seed, germination, and vegetative growth traits

	Trait	Genus	Life span	Genus × life span	Species	Accession
(a)	PC1	$F_{2,47} = \textbf{58.61}^{***}$	$F_{1,47} = \textbf{24.89}^{***}$	$F_{2,47} = \textbf{5.33}^{**}$	$F_{21,47} = \textbf{4.33}^{***}$	NA[a]
	PC2	$F_{2,47} = \textbf{24.39}^{***}$	$F_{1,47} = 0.02$	$F_{2,47} = 0.36$	$F_{21,47} = \textbf{5.98}^{***}$	NA[a]
	PC3	$F_{2,47} = \textbf{21.93}^{***}$	$F_{1,47} = \textbf{47.85}^{***}$	$F_{2,47} = 0.37$	$F_{21,47} = \textbf{2.12}^{*}$	NA[a]
(b)	Seed mass	$F_{2,49} = 2.04$	$F_{1,49} = 3.49$	$F_{2,49} = 0.95$	$F_{23,49} = \textbf{5.38}^{***}$	NA[a]
(c)	Seed length	$F_{2,50.8} = \textbf{21.80}^{***}$	$F_{1,50.7} = 0.71$	$F_{2,50.8} = 0.41$	$F_{23,50.7} = \textbf{7.84}^{***}$	LRT = **4708.51*****
	Seed width	$F_{2,50.8} = \textbf{15.01}^{***}$	$F_{1,50.7} = 0.27$	$F_{2,50.8} = 1.81$	$F_{23,50.8} = \textbf{10.24}^{***}$	LRT = **4029.91*****
	Seed perimeter	$F_{2,50.8} = \textbf{21.00}^{***}$	$F_{1,50.7} = 0.51$	$F_{2,50.8} = 0.19$	$F_{23,50.8} = \textbf{8.92}^{***}$	LRT = **4737.46*****
	Seed area	$F_{2,50.8} = \textbf{19.09}^{***}$	$F_{1,50.7} = 0.41$	$F_{2,50.8} = 0.14$	$F_{23,50.7} = \textbf{6.93}^{***}$	LRT = **5113.95*****
	Seed circularity	$F_{2,49.9} = \textbf{53.58}^{***}$	$F_{1,49.2} = 1.07$	$F_{2,49.9} = \textbf{6.30}^{**}$	$F_{23,49.5} = \textbf{4.97}^{***}$	LRT = **538.28*****
	Seed roundness	$F_{2,50.3} = \textbf{27.43}^{***}$	$F_{1,49.7} = 0.12$	$F_{2,50.3} = \textbf{19.08}^{***}$	$F_{23,49.9} = \textbf{4.78}^{***}$	LRT = **674.03*****
(d)	Germination T$_{50}$	$F_{2,44.5} = \textbf{9.07}^{***}$	$F_{1,44.5} = \textbf{7.22}^{*}$	$F_{2,44.5} = 2.35$	$F_{20,44.9} = 1.35$	LRT = **81.64*****
(e)	Germination proportion	$F_{2,50} = \textbf{21.62}^{***}$	$F_{1,50} = 1.44$	$F_{2,50} = 1.62$	$F_{22,50} = 0.87$	NA[a]
(f)	Height DAP-21	$F_{2,47.3} = 1.56$	$F_{1,47.6} = \textbf{9.05}^{**}$	$F_{2,47.3} = 1.63$	$F_{22,47.1} = 0.85$	LRT = **222.99*****
	Leaf number DAP-21	$F_{2,53.0} = \textbf{20.83}^{***}$	$F_{1,60.5} = \textbf{22.03}^{***}$	$F_{2,52.9} = 2.70$	$F_{23,46.9} = \textbf{6.06}^{***}$	LRT = **69.38*****
(g)	Height DAP-35	$F_{2,49.0} = 1.07$	$F_{1,49.5} = \textbf{12.74}^{***}$	$F_{2,48.9} = 1.73$	$F_{23,48.1} = \textbf{1.96}^{*}$	LRT = **158.00*****
	Leaf number DAP-35	$F_{2,52.0} = \textbf{17.16}^{***}$	$F_{1,54.1} = \textbf{13.19}^{***}$	$F_{2,51.6} = 3.00$	$F_{23,48.5} = \textbf{9.37}^{***}$	LRT = **41.05*****
(h)	Height AGR	$F_{2,48.1} = 0.26$	$F_{1,48.8} = 1.53$	$F_{2,47.9} = 0.58$	$F_{22,46.8} = \textbf{2.51}^{**}$	LRT = **103.65*****
	Height RGR	$F_{2,45.5} = 0.33$	$F_{1,46.9} = 0.17$	$F_{2,45.4} = 1.10$	$F_{22,44.0} = \textbf{2.70}^{**}$	LRT = **54.46*****
	Leaf number AGR	$F_{2,49.5} = 1.04$	$F_{1,51.1} = 0.06$	$F_{2,49.1} = 0.23$	$F_{22,46.5} = \textbf{3.00}^{***}$	LRT = **43.24*****
	Leaf number RGR	$F_{2,49.3} = 1.31$	$F_{1,52.4} = 3.05$	$F_{2,49.3} = 1.21$	$F_{22,47.1} = 1.64$	LRT = **30.27*****

Notes: Letters denote separate models with different random effects; the main effects are the same for all traits. The accession effect and other significant random effects from the reduced model were included; the accession effect is represented by the likelihood ratio test statistic (LRT). ANOVAs are all type III with the exception of principal components, seed mass, and germination proportion (type I), due to the data consisting of only accession-level means with no significant random effects. Additional random effect significance is listed in Appendix S5. Significant values are bolded (at least $P < 0.05$). When non-whole numbers, denominator degrees of freedom were rounded to the first decimal.

[a]Accession could not be used as a random effect in these trait models due to the data set consisting of accession-level means.

$^{*}P < 0.05$; $^{**}P < 0.01$; $^{***}P < 0.001$.

height and leaf AGR and height RGR were also greater in perennials; in *Vicia* height and leaf number AGR were greater in annuals than in perennials, but height and leaf number RGR were greater in perennials (nonsignificant; Appendix S7). In addition, species was a significant predictor for most traits, with the exception of germination T$_{50}$, germination proportion, height at DAP-21, and leaf number RGR (Table 2). Several random effects controlled for in the models were significant (Appendix S5), while dropping data with potentially confounding factors from the models resulted in minimal change in significance (Appendix S4).

Genus and life span patterns in trait correlations

Correlation network plots revealed dynamic seed and vegetative trait correlations which reflected unique genus and life span–specific patterns (Figures 3 and 4). Considering the full data set network and commonalities among each subgroup, there were always strong, significant positive correlations among all seed size traits, which were always significantly negatively correlated with seed circularity and/or roundness (Figure 3A–F; Appendix S13). All significant correlations among vegetative growth traits were positive, with the exception of height and leaf number RGR (Figure 3A–F). Seed size traits were more commonly significantly correlated to single-time-point height and leaf number (positive) than to AGR or RGR (Figure 3A–F). Despite commonalities, considerable variation in magnitude and significance in trait integration also existed between annuals and perennials and among genera.

Annual and perennial species differed in their seed trait to vegetative growth correlations and their overall connectivity among traits (Figure 3B, C). Annuals showed significant positive relationships between all seed size traits and height DAP-21 and DAP-35, as well as significant negative correlations between seed shape and all single-time-point
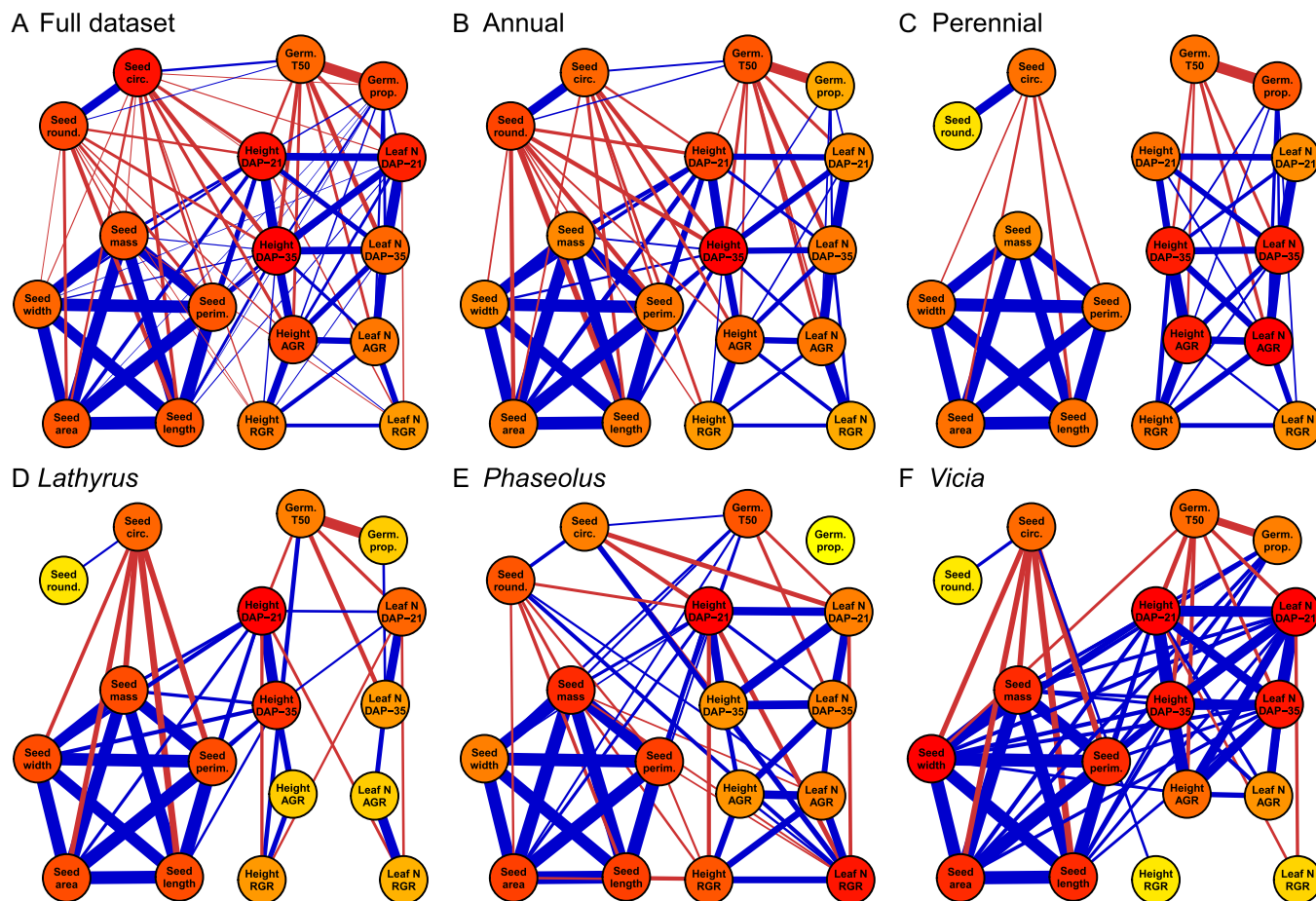
FIGURE 3 Correlation network plots for the full data set (A) and for the data set subgroups: (B) annuals, (C) perennials, (D) *Lathyrus*, (E) *Phaseolus*, and (F) *Vicia*. This represents bivariate correlations between all traits for accession-level data of each subgroup. Presence of lines (edges) between trait nodes indicates a significant correlation between those traits (Pearson; $P < 0.05$); in order to magnify the differences in correlation, edges display the square of the Pearson correlation (r^2). Blue signifies a positive correlation and red a negative correlation; line thickness corresponds to the strength of the correlation. Node color signifies degree (the number of significant trait connections to that node), which ranges from yellow (low) to red (high); node color is in relation to the maximum number of connections for that subgroup and so is not directly comparable across subgroups

height and growth rate traits, whereas perennials lacked any significant correlation between seed and vegetative traits (Figure 3B, C; Appendices S14 and S15). Annuals also displayed more significant correlations between seed size and seed shape traits (negative) than perennials, and annuals showed significant positive correlations between seed shape and germination T$_{50}$, whereas seed and germination traits were not connected for perennials (Figure 3B–C; Appendices S14 and S15). However, both annual and perennial subgroups showed significant positive correlations among most vegetative traits (Figure 3B, C; Appendices S14 and S15). Perennials generally had stronger positive correlations between height and leaf traits compared to annuals, as well as between single-time-point height and height AGR/RGR (Figure 3B, C; Appendices S14 and S15). Within the life span networks, single-time-point height and leaf number displayed some of the highest numbers of significant connections to other traits (Figure 3B, C).

The three genera differed in the extent of integration between seed and vegetative traits, as well as the extent of

correlation among vegetative traits (Figure 3D–F). *Lathyrus* showed the least number of significant seed to vegetative trait correlation, with only height DAP-21 and DAP-35 positively correlated to seed size (Figure 3D; Appendix S16). *Lathyrus* also showed the least connectivity among vegetative traits, with height and leaf trait groups having few significant correlations with each other (Figure 3D; Appendix S16). *Phaseolus* showed significant positive correlations between seed size and height at DAP-21 and significant negative correlations between seed size and height/leaf RGR, with the opposite pattern between seed shape traits and vegetative traits (Figure 3E; Appendix S17). *Phaseolus* also showed predominantly positive correlations between vegetative traits, and it had more significant seed and vegetative trait connections to RGR than the other genera (Figure 3E; Appendix S17). *Vicia* showed the highest number of significant correlations between seed and vegetative traits, with height and leaf number DAP-21 and DAP-35 significantly positively correlated with all seed size traits, but there was minimal correlation between AGR/RGR and seed traits (Figure 3F; Appendix S18).
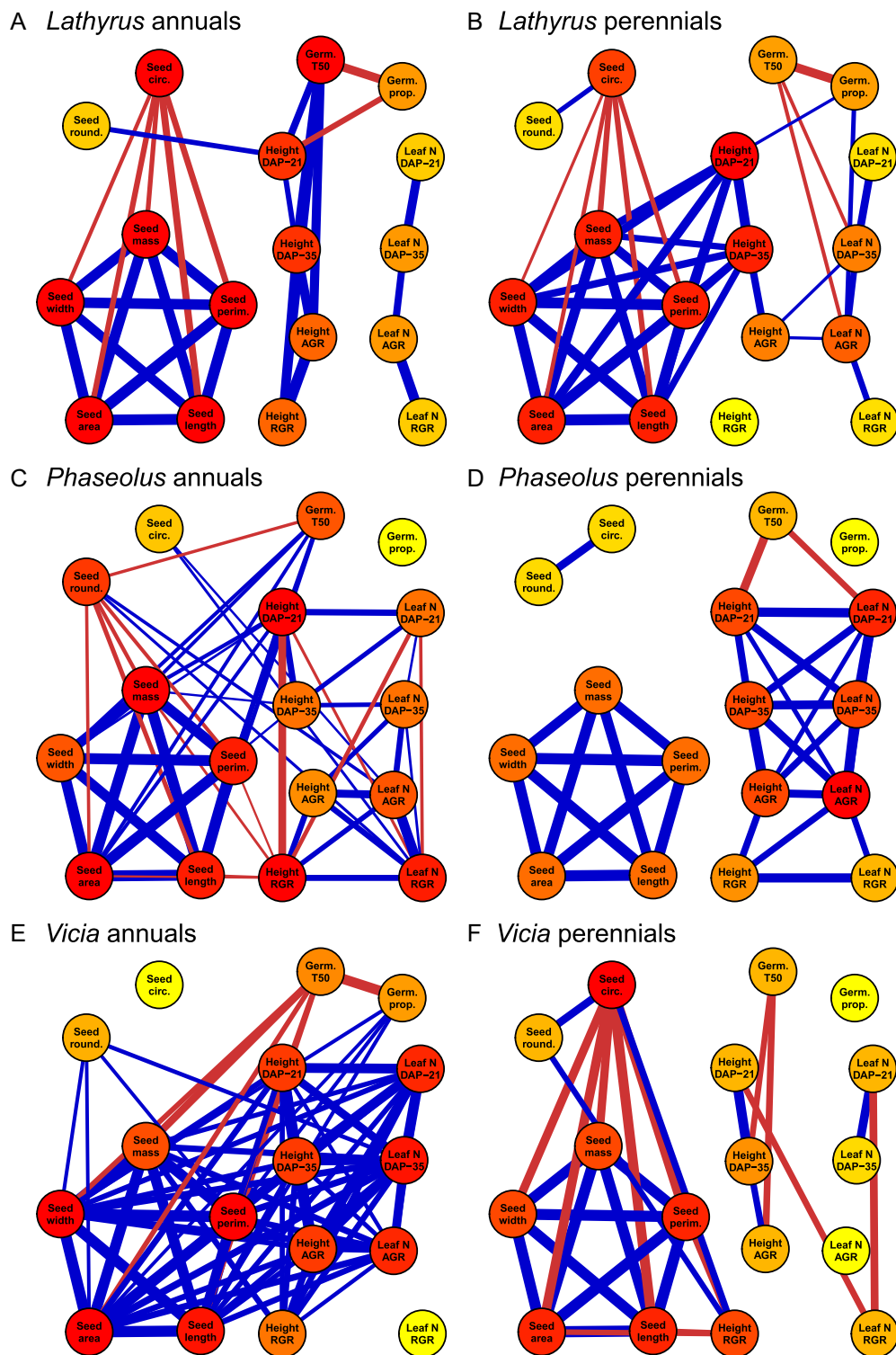
FIGURE 4 Correlation network plots for each genus × life span combination, including (A) *Lathyrus* annuals, (B) *Lathyrus* perennials, (C) *Phaseolus* annuals, (D) *Phaseolus* perennials, (E) *Vicia* annuals, and (F) *Vicia* perennials. This represents bivariate correlations between all traits for accession-level data of each subgroup; only significant correlations are shown. See Figure 3 for a description of plot features.

Vicia also showed the most robust cluster of significant positively correlated vegetative traits, with the exception of RGR (Figure 3F; Appendix S18). Lastly, seed size was significantly negatively correlated to seed circularity for *Lathyrus* and *Vicia*, whereas for *Phaseolus* seed size traits

were significantly negatively correlated with seed roundness but not circularity (Figure 3D–F; Appendices S16, S17, and S18). For all three genera, height DAP-21 showed a consistently high number of significant correlations to other traits (Figure 3D–F).

In order to investigate whether broad patterns of trait correlations in annuals and perennials were consistent within each genus, we generated network plots of bivariate correlations within each genus × life span subgroup (Figure 4). The pattern of significant seed-vegetative correlations in annuals but not in perennials was generally upheld in *Phaseolus* and *Vicia*, whereas the opposite was true for *Lathyrus* (Figure 4A–F). *Lathyrus* annuals had only one significant correlation (positive) between seed roundness and height DAP-21, whereas *Lathyrus* perennials had numerous correlations between seed size traits and height DAP-21 and height DAP-35 (Figure 4A, B). *Phaseolus* annuals showed several significant correlations between seed traits (shape and size) and vegetative traits, whereas *Phaseolus* perennials showed no significant seed-vegetative correlations; additionally, seed size and shape were decoupled in *Phaseolus* perennials (Figure 4C, D). *Vicia* annuals showed the most robust seed-vegetative integration, mainly significant positive correlations between seed size and all vegetative traits except leaf number RGR; for *Vicia* perennials, the only significant seed-vegetative correlation was from height RGR to seed length and circularity (Figure 4E, F).

DISCUSSION

Life history strategy and life span classification (annual, perennial) is associated with various aspects of species reproductive and vegetative biology, but the role of life span in predicting seed to vegetative trait correlation is not well characterized. Here, we examined trait variation and correlation of seed size and shape, and the germination and vegetative growth characters of seedlings derived from those seeds, in annual and perennial congeners from three herbaceous legume genera. We found that while genus was a strong predictor of seed size and shape, life span most consistently predicted single-time-point vegetative size. Patterns of trait correlation between and among seed size and vegetative traits differed by life span and genus. Specifically, annual species typically had more significant seed to vegetative trait correlations than perennial species.

Evolutionary basis of genus and life span differences

In this study, genus was consistently the strongest predictor of seed size and shape variation, which illustrates the importance of phylogenetic context in understanding some aspects of life history variation (Silvertown and Dodd, 1996). Annual-perennial overlap in species-level variation further demonstrates that seed size and shape variation does not consistently separate by life span group (Appendices S8, S9, and S10). Mazer (1989) similarly found a greater amount of variation in seed mass explained by family-level classification (30%) than life history type (22%)

in a large study of 10 families of Indiana Dune angiosperms. However, they still found life history to be significant, which was attributed to the distinctly large seeds of tree species (Mazer, 1989). Our study builds on this previous work in that it focuses on herbaceous species of three legume genera and quantifies multiple dimensions of seed size and shape on an individual seed level (instead of seed mass class), without specificity to a particular habitat (Mazer, 1989). Importantly, Mazer (1989) also noted that life history was usually not a significant predictor of seed mass within a family, and there was not a significant difference in seed mass among annuals and herbaceous perennials in their study. Our lack of clear life span signal in seed size (but see post hoc tests for *Phaseolus*) is also consistent with a meta-analysis of ~3000 congeneric comparisons of annual and perennial species (Vico et al., 2016).

While genus predicted seed traits, our data demonstrated that life span consistently predicted single-time-point vegetative size. In this study, annual species displayed greater mean height and leaf number measured at 21 d and 35 d from planting compared to congeneric perennial species in the first year of growth; this was significant for both traits in *Phaseolus* and for leaf number in *Vicia* (Appendix S7). Greater growth in annuals is consistent with the predicted pattern for annuals and perennials according to the acquisitive-conservative resource economics spectrum (e.g., Roumet et al., 2006; González-Paleo and Ravetta, 2015). Vegetative growth is commonly measured in terms of RGR, for which life span is usually a significant predictor (e.g., Garnier, 1992; Atkinson et al., 2016; Simpson et al., 2021); however, in this study neither life span nor genus was a significant predictor of either AGR or RGR in terms of plant height and leaf number. This could reflect the timing of growth measurements (made at the seedling stage), since species have different trajectories of RGR over their life span, with RGR typically decreasing with time as the plant becomes larger (Turnbull et al., 2008). Also, while RGR is typically measured using successive destructive biomass harvests on different plants (Perez-Harguindeguy et al., 2013), we nondestructively measured height and leaf number growth on the same individual plants, which may result in different patterns.

Annual species exhibited significant correlations between seed and vegetative traits (plant height and leaf number), but the same correlations were not significant in perennial species (Figure 3B, C). These data point to a potentially important decoupling of vegetative and seed trait variation in perennial species, at least in traits measured in the early life span and under controlled conditions. Correlational decoupling has consistently been found between floral and vegetative traits, attributed to strong directional selection for a trait module as independent from other organismal size variation (Berg, 1960); however, this pattern is variable across diverse life history types and environments (Armbruster et al., 1999; Pélabon et al., 2013). Furthermore, different patterns of phenotypic integration may occur when assessed across species (evolutionary integration), rather than within a species, as is commonly the scope of

study (Klingenberg, 2014). Plant size and seed size are often significantly positively correlated in broad interspecific meta-analyses (Leishman et al., 1995; Moles et al., 2004; Díaz et al., 2016); greater vegetative size may allow greater total reproductive output through increased seed size and/ or number. These studies often include herbaceous and woody species with a wide range of life history strategies; however, our data indicate that, when examining specific life history categories such as herbaceous perennials, unique patterns may emerge.

Other multispecies studies including herbaceous perennials have focused on relationships between vegetative traits and traits of seeds produced by that individual or from individuals in separate studies, a slightly different approach than our study, which examined correlations between seed traits and the vegetative traits expressed by the individuals germinated from those seeds. Kleyer et al. (2019), studying predominantly herbaceous perennials in a northwestern European flora, also found no significant correlation between plant height and mass of seeds produced by that plant. Similarly, a meta-analysis of separate studies representing 526 species in Sweden found no significant relationship between plant height and seed mass in herbaceous perennials, whereas that relationship did exist for herbaceous annuals, woody perennials, and the full data set (Bolmgren and Cowen, 2008). Our study extends these results and demonstrates similar correlation patterns between source seeds and the plants derived from them, grown in a common environment.

Considering within-genus life span trait correlations, annual species in *Phaseolus* and *Vicia* again exhibited significant seed-to-vegetative correlations whereas perennials did not, and the opposite was true for plant height in *Lathyrus* (Figure 4). Despite decreased sample size for these intrageneric patterns, this finding further reinforces the importance of phylogenetic context in interpreting life span–associated trait patterns and provides additional support for observed differences in trait correlation in annual and perennial plants. The consistent seed-to-vegetative correlations in annuals may reflect repeated directional selection for a similar, disturbed habitat, and consequently more constrained and predictable phenotypic correlations among species. By contrast, herbaceous perennial species may be less selectively constrained as a group and thus distinctly variable in their expressed trait combinations. Variability among the herbaceous perennial species studied may mask signatures of potential developmental correlations among traits; it remains to be determined whether seed-vegetative decoupling extends to individual herbaceous perennial species.

Future considerations

Additional biological variables across the range of species studied must also be considered. For example, the correlation between seed size and vegetative plant size will likely change and potentially weaken with increased time from germination and depends on the relative growth rate of the species in question (Benjamin and Hardwick, 1986). Phenotypic integration is likewise sensitive to growth environment and developmental stage (Murren, 2002); it remains to be seen if the observed phenotypic correlations are robust in each species' resource-limited native environment and across a longer period of growth, from germination to maturation. Given the wide range of species used in this study, it not feasible to pinpoint precise physiological variables affecting the seed-vegetative size relationship; studies of phenotypic integration across multiple biological scales, from individual to population to species, will assist in more precise biological interpretation. These results should also be extended to belowground traits, including roots and storage organs, as well as reproductive output by the end of the growing season, phenotypes that are integral to life history strategy. Altogether, there remains ample opportunity to expand current understanding of life span–associated phenotypic integration in terms of developmental and temporal effects, with extension to additional plant organ systems, and under different environmental conditions.

Furthermore, there is considerable intraspecific variation in life span and its associated traits; this was evidenced by the significant accession term in all of our linear models. For simplicity, we adopted the broad titles of annual and perennial in this study, but life span is not a simple binary or categorical trait, and life span alone cannot be expected to encompass the biological complexity of all underlying life history traits. Examples of additional life span–associated trait variation may include reproductive patterns (e.g., semelparous and iteroparous), growth determinacy vs. indeterminacy, clonality, length of the juvenile phase, and mating system (autogamous, allogamous, and mixed), among many others (Friedman, 2020). Life span is also inherently context dependent: both annual and perennial populations can occur within a single species across its range, often due to variation in environmental disturbance (e.g., *Mimulus guttatus*; Friedman et al., 2015), *Oryza perennis* (Morishima et al., 1984), and *Zostera marina* (Reynolds et al., 2017). We are also limited in our ability to know and control for the maternal growth environment and selection history for these accessions, which may include either direct or indirect artificial selection for those collected in feral populations and as agricultural weeds. Thus, in order to gain a more precise understanding of life span–associated phenotypes in these species, a thorough assessment of variation and correlation within and across many diverse populations is needed.

Relevance to perennial breeding goals

With increasing interest in de novo domestication of wild species and breeding of hybrid herbaceous perennial crops to reduce the ecological impacts of agriculture (Glover et al., 2010; Ryan et al., 2018), broadening our understanding of trait networks in herbaceous perennials is essential. Evidence to date highlights potential seed yield-vegetative trade-offs in

emergent perennial crops (e.g., González-Paleo et al., 2016; Pastor-Pastor et al., 2018); however, the challenge of co-selecting for negatively correlated traits has been accomplished in modern plant breeding, and breeders have successfully selected for both high seed yield and sustained perenniality in rice (DeHaan et al., 2005; Huang et al., 2018). Our results suggest less phenotypic integration between some seed and vegetative traits in herbaceous perennials compared to annuals, which could allow for relatively unconstrained selection for seed and vegetative yield simultaneously. This could be particularly important for grain crops that are dual-purposed as forage, where vigorous vegetative growth is also favorable—for example, *Thinopyrum intermedium* (Pugliese, 2017) and *Silphium integrifolium* (Vilela et al., 2020). It remains to be seen if the seed-vegetative decoupling extends to later lifetime vegetative characteristics such as shoot and root biomass allocation and total reproductive yield, and if these relationships hold when mature plants are measured over multiple years under field conditions.

CONCLUSIONS

Despite the prevalence of annual-perennial transitions in angiosperms, we know relatively little about how life span classification is associated with phenotypic correlations across ecologically and agriculturally important traits in herbaceous plants. Our results show that in three legume genera, seed variation was primarily explained by genus and single-time-point vegetative size variation was primarily explained by life span. Further, annual species showed stronger seed to vegetative trait correlation in early growth than herbaceous perennial congeners. These findings call for further investigation into how these trait correlations differ throughout the life span of perennial plants, as well as into correlations among other important life history traits, particularly allocation to reproductive and perennating organs. This study highlights that both life span and phylogenetic context are key in predicting phenotypic variation, and there remain numerous underexplored systems in which to expand understanding of life history.

AUTHOR CONTRIBUTIONS
S.A.H. and A.J.M. designed the study. S.A.H. implemented the research and wrote the manuscript. M.A.A., Q.G.L., A.J.M., M.J.R., and M.C.S. assisted in critical review and writing of the manuscript. M.J.R. assisted in germination calculations and statistical methods and interpretation. M.A.A. and Q.G.L. assisted in crafting germination experiments and provided the necessary resources. M.C.S. assisted in data acquisition.

DATA AVAILABILITY STATEMENT
All phenotypic data (individual seed, germination, vegetative growth, and accession-level data) are available on Figshare (https://doi.org/10.6084/m9.figshare.16640998.v1). R and SAS code and the associated files are available on GitHub (https://github.com/SAHerron/Legume_Life_span_Genus_Effects.git).

ORCID
Sterling A. Herron http://orcid.org/0000-0003-1738-0874
Matthew J. Rubin http://orcid.org/0000-0001-5484-2324
Matthew A. Albrecht http://orcid.org/0000-0002-1079-1630
Allison J. Miller http://orcid.org/0000-0002-2722-9361

REFERENCES
Albani, M. C., and G. Coupland. 2010. Comparative analysis of flowering in annual and perennial plants. *In* Current topics in developmental

biology, vol. 91, 323–348. Elsevier Science Publishing Co Inc, San Diego, United States.

Armbruster, W. S., V. S. Di Stilio, J. D. Tuxill, T. C. Flores, and J. L. Velásquez Runk. 1999. Covariance and decoupling of floral and vegetative traits in nine neotropical plants: a re-evaluation of Berg's correlation-pleiades concept. *American Journal of Botany* 86: 39–55.

Atkinson, R. R. L., E. J. Mockford, C. Bennett, P. A. Christin, E. L. Spriggs, R. P. Freckleton, K. Thompson, et al. 2016. C4 photosynthesis boosts growth by altering physiology, allocation and size. *Nature Plants* 2: 1–5.

Baskin, C. C., and J. M. Baskin. 2014. Seeds: Ecology, biogeography, and evolution of dormancy and germination, 2nd ed. Academic Press, San Diego, California, USA.

Bates, D., M. Mächler, B. Bolker, and S. Walker. 2015. Fitting linear mixed-effects models using lme4. *Journal of Statistical Software* 67: 1–48.

Benjamin, L. R., and R. C. Hardwick. 1986. Sources of variation and measures of variability in even-aged stands of plants. *Annals of Botany* 58: 757–778.

Berg, R. L. 1960. The ecological significance of correlation pleiades. *Evolution* 14: 171–180.

Bolmgren, K., and P. D. Cowan. 2008. Time - size tradeoffs: A phylogenetic comparative study of flowering time, plant height and seed mass in a north-temperate flora. *Oikos* 117: 424–429.

Charnov, E. L., and W. M. Schaffer. 1973. Life-history consequences of natural selection: Cole's result revisited. *The American Naturalist* 107: 791–793.

Ciotir, C., W. Applequist, T. E. Crews, N. Cristea, L. R. DeHaan, E. Frawley, S. A. Herron, et al. 2019. Building a botanical foundation for perennial agriculture: Global inventory of wild, perennial herbaceous Fabaceae species. *Plants, People, Planet* 1: 375–386.

Cole, L. C. 1954. The population consequences of life history phenomena. *The Quarterly Review of Biology* 29: 103–137.

Crews, T. E., and L. R. DeHaan. 2015. The strong perennial vision: A response. *Agroecology and Sustainable Food Systems* 39: 500–515.

Csardi, G., and T. Nepusz. 2006. The igraph software package for complex network research. *InterJournal, Complex Systems* 1695: 1-9.

DeHaan, L. R., D. L. Van Tassel, and T. S. Cox. 2005. Perennial grain crops: A synthesis of ecology and plant breeding. *Renewable Agriculture and Food Systems* 20: 5–14.

den Dubbelden, K. C., and R. W. Verburg. 1996. Inherent allocation patterns and potential growth rates of herbaceous climbing plants. *Plant and Soil* 184: 341–347.

Díaz, S., J. Kattge, J. H. C. Cornelissen, I. J. Wright, S. Lavorel, S. Dray, B. Reu, et al. 2016. The global spectrum of plant form and function. *Nature* 529: 167–171.

Du, G., and W. Qi. 2010. Trade-offs between flowering time, plant height, and seed size within and across 11 communities of a QingHai-Tibetan flora. *Plant Ecology* 209: 321–333.

Frehner, M., and E. E. Conn. 1987. The linamarin β-glucosidase in Costa Rican wild lima beans (*Phaseolus lunatus* L.) is apoplastic. *Plant Physiology* 84: 1296–1300.

Freytag, G. F., and D. G. Debouck. 2002. Taxonomy, distribution, and ecology of the genus *Phaseolus* (Leguminosae-Papilionoideae) in North America, Mexico, and Central America. *Sida, Botanical Miscellany* 23: 1–300.

Friedman, J. 2020. The evolution of annual and perennial plant life histories: ecological correlates and genetic mechanisms. *Annual Review of Ecology, Evolution, and Systematics* 51: 461–481.

Friedman, J., and M. J. Rubin. 2015. All in good time: Understanding annual and perennial strategies in plants. *American Journal of Botany* 102: 497–499.

Friedman, J., A. D. Twyford, J. H. Willis, and B. K. Blackman. 2015. The extent and genetic basis of phenotypic divergence in life history traits in *Mimulus guttatus*. *Molecular Ecology* 24: 111–122.

Gadgil, M., and O. T. Solbrig. 1972. The concept of r- and K-selection: evidence from wild flowers and some theoretical considerations. *American Naturalist* 106: 14–31.

Galasso, I., G. Sonnante, D. G. Tota, and D. Pignone. 1997. Comparison of molecular cytogenetic and genetic analyses in accessions of the two biotypes of *Vicia benghalensis* L. *Annals of Botany* 79: 311–317.

Garnier, E. 1992. Growth analysis of congeneric annual and perennial grass species. *Journal of Ecology* 80: 665–675.

Garnier, E., and G. Laurent. 1994. Leaf anatomy, specific mass and water-content in congeneric annual and perennial grass species. *New Phytologist* 128: 725–736.

Geber, M. A. 1990. The cost of meristem limitation in *Polygonum arenastrum*: Negative genetic correlations between fecundity and growth. *Evolution* 44: 799–819.

Glover, J. D., J. P. Reganold, L. W. Bell, J. Borevitz, E. C. Brummer, E. S. Buckler, C. M. Cox, et al. 2010. Increased food and ecosystem security via perennial grains. *Science* 328: 1638–1639.

González-Paleo, L., and D. A. Ravetta. 2015. Carbon acquisition strategies uncoupled from predictions derived from species life-cycle. *Flora* 212: 1–9.

González-Paleo, L., A. E. Vilela, and D. A. Ravetta. 2016. Back to perennials: Does selection enhance tradeoffs between yield and longevity? *Industrial Crops & Products* 91: 272–278.

Grime, J. P. 1977. Evidence for the existence of three primary strategies in plants and its relevance to ecological and evolutionary theory. *American Naturalist* 111: 1169–1194.

Harrell, F. E. 2020. Hmisc: Harrell miscellaneous. R package version 4.4-1. Website: https://CRAN.R-project.org/package=Hmisc

Herron, S. A., M. J. Rubin, C. Ciotir, T. E. Crews, D. L. Van Tassel, and A. J. Miller. 2020. Comparative analysis of early life stage traits in annual and perennial *Phaseolus* crops and their wild relatives. *Frontiers in Plant Science* 11: 1–14.

Huang, G., S. Qin, S. Zhang, X. Cai, S. Wu, J. Dao, J. Zhang, et al. 2018. Performance, economics and potential impact of perennial rice PR23 relative to annual rice cultivars at multiple locations in Yunnan Province of China. *Sustainability* 10: 1086.

Josse, J., and F. Husson. 2016. missMDA: A package for handling missing values in multivariate data analysis. *Journal of Statistical Software* 70: 1–31.

Kassambara, A., and F. Mundt. 2020. factoextra: Extract and visualize the results of multivariate data analyses. R package version 1.0.7. Website: https://CRAN.R-project.org/package=factoextra

Kelly, C. K., and M. E. Hanley. 2005. Juvenile growth and palatability in co-occurring, congeneric British herbs. *American Journal of Botany* 92: 1586–1589.

Kitchen, S. G. 1994. Perennial forb life-history strategies on semiarid rangelands: Implications for revegetation. USDA Forest Service General Technical Report INT-GTR-313, Intermountain Research Station, Provo, Utah, USA.

Kleyer, M., J. Trinogga, M. A. Cebrián, R. Ejrnæs, A. Trenkamp, C. Fløjgaard, D. C. Albach, et al. 2019. Trait correlation network analysis identifies biomass allocation traits and stem specific length as hub traits in herbaceous perennial plants. *Journal of Ecology* 107: 829–842.

Klingenberg, C. P. 2014. Studying morphological integration and modularity at multiple levels: concepts and analysis. *Philosophical Transactions of the Royal Society B: Biological Sciences* 369: 33–35.

Kuznetsova, A., P. B. Brockhoff, and R. H. B. Christensen. 2017. lmerTest Package: Tests in linear mixed effects models. *Journal of Statistical Software* 82: 1–26.

Leishman, M. R., M. Westoby, and E. Jurado. 1995. Correlates of seed size variation: a comparison among five temperate floras. *Journal of Ecology* 83: 517–530.

Lenth, R. 2020. emmeans: Estimated marginal means, aka least-squares means. R package version 1.4.7. Website: https://CRAN.R-project.org/package=emmeans

Lindberg, C. L., H. M. Hanslin, M. Schubert, T. Marcussen, B. Trevaskis, J. C. Preston, and S. Fjellheim. 2020. Increased above-ground resource allocation is a likely precursor for independent evolutionary origins of annuality in the Pooideae grass subfamily. *New Phytologist* 228: 318–329.

Lundgren, M. R., and D. L. Des Marais. 2020. Life history variation as a model for understanding trade-offs in plant–environment interactions. *Current Biology* 30: R180–R189.

Marshall, D. L., N. L. Fowler, and D. A. Levin. 1985. Plasticity in yield components in natural populations of three species of *Sesbania*. *Ecology* 66: 753–761.

Mazer, S. J. 1989. Ecological, taxonomic, and life history correlates of seed mass among Indiana Dune Angiosperms. *Ecological Monographs* 59: 153–175.

Moles, A. T., D. S. Falster, M. R. Leishman, and M. Westoby. 2004. Small-seeded species produce more seeds per square metre of canopy per year, but not per individual per lifetime. *Journal of Ecology* 92: 384–396.

Morishima, H., Y. Sano, and H. I. Oka. 1984. Differentiation of perennial and annual types due to habitat conditions in the wild rice *Oryza perennis*. *Plant Systematics and Evolution* 144: 119–135.

Murren, C. J. 2002. Phenotypic integration in plants. *Plant Species Biology* 17: 89–99.

Pastor-Pastor, A., A. E. Vilela, and L. González-Paleo. 2018. The root of the problem of perennials domestication: is selection for yield changing key root system traits required for ecological sustainability? *Plant and Soil* 435: 161–174.

Pélabon, C., N. C. Osler, M. Diekmann, and B. J. Graae. 2013. Decoupled phenotypic variation between floral and vegetative traits: distinguishing between developmental and environmental correlations. *Annals of Botany* 111: 935–944.

Perez-Harguindeguy, N., E. Garnier, S. Lavorel, H. Poorter, P. Jaureguiberry, W. K. Cornwell, J. M. Craine, et al. 2013. New handbook for standardised measurement of plant functional traits worldwide. *Australian Journal of Botany* 61: 167–234.

Pitelka, L. F. 1977. Energy allocations in annual and perennial lupines (*Lupinus*: Leguminosae). *Ecology* 58: 1055–1065.

Pommerening, A., and A. Muszta. 2016. Relative plant growth revisited: Towards a mathematical standardisation of separate approaches. *Ecological Modelling* 320: 383–392.

Pugliese, J. Y. 2017. Above- and belowground response to managing Kernza (*Thinopyrum intermedium*) as a dual-use crop for forage and grain. M.Sc. thesis, Ohio State University, Columbus, Ohio, USA.

R Core Team. 2019. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria.

Rees, M., C. P. Osborne, F. I. Woodward, S. P. Hulme, L. A. Turnbull, and S. H. Taylor. 2010. Partitioning the components of relative growth rate: How important is plant size variation? *American Naturalist* 176: E152–E161.

Reynolds, L. K., J. J. Stachowicz, A. R. Hughes, S. J. Kamel, B. S. Ort, and R. K. Grosberg. 2017. Temporal stability in patterns of genetic diversity and structure of a marine foundation species (*Zostera marina*). *Heredity* 118: 404–412.

Roumet, C., E. Garnier, H. Suzor, J. L. Salager, and J. Roy. 2000. Short and long-term responses of whole-plant gas exchange to elevated CO_2 in four herbaceous species. *Environmental and Experimental Botany* 43: 155–169.

Roumet, C., C. Urcelay, and S. Díaz. 2006. Suites of root traits in species growing differ between annual and perennial species growing in the field. *New Phytologist* 170: 357–367.

Ryan, M. R., T. E. Crews, S. W. Culman, L. R. DeHaan, R. C. Hayes, J. M. Jungers, and M. G. Bakker. 2018. Managing for multifunctionality in perennial grain crops. *BioScience* 68: 294–304.

SAS Institute. 2017. Base SAS®, version 9.4. SAS Institute Inc., Cary, North Carolina, USA.

Schaefer, H., P. Hechenleitner, A. Santos-Guerra, M. M. De Sequeira, R. T. Pennington, G. J. Kenicer, and M. A. Carine. 2012. Systematics, biogeography, and character evolution of the legume tribe Fabeae with special focus on the middle-Atlantic island lineages. *BMC Evolutionary Biology* 12: 1–19.

Schneider, C. A., W. S. Rasband, and K. W. Eliceiri. 2012. NIH Image to ImageJ: 25 years of image analysis. *Nature Methods* 9: 671–675.

Segrestin, J., M. Navas, and E. Garnier. 2020. Reproductive phenology as a dimension of the phenotypic space in 139 plant species from the Mediterranean. *New Phytologist* 225: 740–753.

Sharma, V., and S. Kumar. 2012. Stipules are the principal photosynthetic organs in the papilionoid species *Lathyrus aphaca*. *National Academy Science Letters* 35: 75–78.

Silvertown, J. W., and M. Dodd. 1996. Comparing plants and connecting traits. *Philosophical Transactions of the Royal Society of London, B, Biological Sciences* 351: 1233–1239.

Simpson, K. J., R. R. L. Atkinson, E. J. Mockford, C. Bennett, C. P. Osborne, and M. Rees. 2021. Large seeds provide an intrinsic growth advantage that depends on leaf traits and root allocation. *Functional Ecology* 35: 2168–2178.

Singmann, H., B. Bolker, J. Westfall, F. Aust, and M. S. Ben-Shachar. 2020. afex: Analysis of factorial experiments. R package version 0.27-2. Website: https://CRAN.R-project.org/package=afex

Soltis, D. E., M. E. Mort, M. Latvis, E. V. Mavrodiev, B. C. O'meara, P. S. Soltis, J. G. Burleigh, and R. R. De Casas. 2013. Phylogenetic relationships and character evolution analysis of Saxifragales using a supermatrix approach. *American Journal of Botany* 100: 916–929.

Stearns, S. C. 1992. The evolution of life histories. Oxford University Press, Oxford, UK.

Tank, D. C., and R. G. Olmstead. 2008. From annuals to perennials: Phylogeny of subtribe Castillejinae (Orobanchaceae). *American Journal of Botany* 95: 608–625.

Thompson, K., and D. J. Hodkinson. 1998. Seed mass, habitat and life history: A re-analysis of Salisbury (1942, 1974). *New Phytologist* 138: 163–167.

Turkington, R. A., and P. B. Cavers. 1978. Reproductive strategies and growth in four legumes. *Canadian Journal of Botany* 56: 413–416.

Turnbull, L. A., C. Paul-Victor, B. Schmid, and D. W. Purves. 2008. Growth rates, seed size, and physiology: Do small-seeded species really grow faster? *Ecology* 89: 1352–1363.

Verboom, G. A., H. P. Linder, and W. D. Stock. 2004. Testing the adaptive nature of radiation: Growth form and life history divergence in the African grass genus *Ehrharta* (Poaceae: Ehrhartoideae). *American Journal of Botany* 91: 1364–1370.

Vico, G., S. Manzoni, L. Nkurunziza, K. Murphy, and M. Weih. 2016. Trade-offs between seed output and life span-a quantitative comparison of traits between annual and perennial congeneric species. *New Phytologist* 209: 104–114.

Vilela, A. E., L. González-Paleo, D. A. Ravetta, E. G. Murrell, and D. L. Van Tassel. 2020. Balancing forage production, seed yield, and pest management in the perennial sunflower *Silphium integrifolium* (Asteraceae). *Agronomy* 10: 1471.

Vu, V. Q. 2011. ggbiplot: A ggplot2 based biplot. R package version 0.55. Website: http://github.com/vqv/ggbiplot

Ward, P. R., J. A. Palta, and H. A. Waddell. 2011. Root and shoot growth by seedlings of annual and perennial medic, and annual and perennial wheat. *Crop and Pasture Science* 62: 367–373.

Wei, T., and V. Simko. 2017. R package "corrplot": Visualization of a correlation matrix. R package version 0.84. Website: https://CRAN.R-project.org/package=corrplot

Wickham, H. 2016. ggplot2: Elegant graphics for data analysis. Springer-Verlag, New York, New York, USA. Website: https://ggplot2.tidyverse.org

Wright, I. J., P. B. Reich, M. Westoby, D. D. Ackerly, Z. Baruch, F. Bongers, J. Cavender-Bares, et al. 2004. The worldwide leaf economics spectrum. *Nature* 428: 821–827.

Wu, Z. Y., P. H. Raven, and D. Y. Hong [eds.]. 2010. Flora of China, vol. 10 (Fabaceae). Science Press, Beijing, China, and Missouri Botanical Garden Press, St. Louis, Missouri, USA. Website: http://www.efloras.org/browse.aspx?volume_id=2010 [accessed 10 April 2020].

SUPPORTING INFORMATION

Additional supporting information may be found in the online version of the article at the publisher's website.

Appendix S1. Accession descriptive metadata and sample size for each trait.

Appendix S2. Summary of the phylogenetic relationships among species for each genus and relevant notes for select species.

Appendix S3. Additional details regarding germination T_{50} and proportion methodology.

Appendix S4. Analysis of variance table of each trait model with subsets of the data removed to test for influence on significance, as well as notes on other specific changes to analyses.

Appendix S5. Table of all significant random effects in addition to accession for each trait model.

Appendix S6. Variable loadings for principal components 1–5 of the full accession-level data set principal component analysis (Figure 1).

Appendix S7. Table of adjusted means of each trait for annuals and perennials of each genus and post hoc custom contrasts.

Appendix S8. Principal component analyses of the full individual-level seed and vegetative trait data sets for *Lathyrus*.

Appendix S9. Principal component analyses of the full individual-level seed and vegetative trait data sets for *Phaseolus*.

Appendix S10. Principal component analyses of the full individual-level seed and vegetative trait data sets for *Vicia*.

Appendix S11. Variable loadings for principal components 1 and 2 of the full individual-level seed data set principal component analysis, parsed by genus (corresponding to Appendices S8, S9, and S10).

Appendix S12. Variable loadings for the full individual-level vegetative data set principal component analysis, parsed by genus (principal components 1–3 for *Lathyrus*, principal components 1 and 2 for *Phaseolus* and *Vicia*; corresponding to Appendices S8, S9, and S10).

Appendix S13. Correlation matrix for the full data set, showing Pearson correlation coefficients between every combination of traits, using accession-level data.

Appendix S14. Correlation matrix for the annual data subgroup, showing Pearson correlation coefficients between every combination of traits, using accession-level data.

Appendix S15. Correlation matrix for the perennial data subgroup, showing Pearson correlation coefficients between every combination of traits, using accession-level data.

Appendix S16. Correlation matrix for the *Lathyrus* data subgroup, showing Pearson correlation coefficients between every combination of traits, using accession-level data.

Appendix S17. Correlation matrix for the *Phaseolus* data subgroup, showing Pearson correlation coefficients between every combination of traits, using accession-level data.

Appendix S18. Correlation matrix for the *Vicia* data subgroup, showing Pearson correlation coefficients between every combination of traits, using accession-level data.