Range-wide phenotypic variation in seed and wing traits of Cyclocarya paliurus across environmental gradients in China

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Abstract

Cyclocarya paliurus is a wind-dispersed tree of high medicinal and economic value, yet range-wide variation in seed traits and its environmental correlates remain poorly quantified. We sampled 139 trees from 30 natural populations spanning the subtropical range in China and measured six traits (seed diameter, thickness, diameter-to-thickness ratio, wing diameter, volume, and thousand-kernel weight). Using a nested linear mixed framework, we partitioned variance within and among populations, calculated phenotypic differentiation (Vst), and related traits to geography and climate. Coefficients of variation ranged from 0.01–55.83%, with the highest mean CV for seed volume and the lowest for seed diameter. On average, 77% of the total variance was among populations (mean Vst ≈ 82%), indicating strong population differentiation. Altitude and thermal regime were the primary correlates of wing morphology and seed mass: populations at higher elevations and cooler sites tended to have larger wings and heavier seeds. PCA and k-means clustering resolved six geographically coherent phenotypic groups. Our results provide a quantitative baseline for climate-informed seed sourcing and conservation planning, and suggest that environmental filtering acts on coordinated suites of seed and wing traits rather than on single traits in isolation.

Introduction

Cyclocarya paliurus (Batal) Iljinskaja, commonly called “sweet tea tree”, the sole species in its genus in the family Juglandaceae and is widely distributed at 420–2500 m elevation in mountainous subtropical regions of China (Deng et al. 2017; Fang et al. 2006; Liu et al. 2018). Its leaves have long been consumed by local communities as food (Birari & Bhutani 2007), and have been used in traditional Chinese medicine (TCM) formulations as well as in functional foods (Cao et al. 2017; Fang et al. 2011). The species is also cultivated for fine timber and landscaping because of its attractive foliage and samaras. Over the past two decades, a series of studies on *C. paliurus* has demonstrated hypolipidemia and antidiabetic activities, among other therapeutic effects (Xiao et al. 2014; Yang et al. 2016), consistent with the enrichment of bioactive constituents in the leaves—including cyclocariosides, flavonoids, steroids, saponins, and related compounds (Li et al. 2017). Consequently, interest in exploiting these values has intensified, and large quantities of tender leaves are required for tea and medicinal uses. However, plantations dedicated to leaf production remain insufficient. At present, *C. paliurus* is propagated primarily from seed, which exhibits deep dormancy and typically germinates ~ 2 years after sowing under natural conditions (Fang et al. 2006). Recent work on cultivation —covering dormancy-breaking treatments, nursery practices, and environmental effects on leaf metabolites—has made tangible progress. (Deng et al. 2015; Deng et al. 2017; Fu et al. 2015; Liu et al. 2017; Yang et al. 2017). Nevertheless, land reclamation, overharvesting (including repeated leaf collection), and over-excavation in recent decades have caused substantial loss and fragmentation of wild resources, destabilizing population structure. Clarifying the distribution and genetic diversity of C. *paliurus* is therefore essential to guide sustainable use and biodiversity conservation. In particular, a comprehensive assessment of seed-trait phenotypic diversity requires larger datasets that integrate environmental covariates and broader sampling across the climatic regions where *C. paliurus* predominates.

Phenotypic diversity—variation in observable traits among individuals and populations—is a core dimension of biodiversity and reflects the combined influences of genetic background and environment (Liang et al. 2015). Plant phenotypic variation is an outcome of genotype reaction norms: environmentally induced phenotypes can arise and, under selection, may become genetically assimilated through shifts in reaction norms. Natural populations harbor ample genetic variation for plastic responses, enabling evolution by natural selection and other mechanisms (Pigliucci et al. 2006). Among plant traits, seeds and fruits—central to sexual reproduction—integrate dispersal potential and maternal allocation, thereby capturing key aspects of fitness and ecological strategy. Compared with many vegetative traits, seed traits often show high stability and have been widely used to infer patterns of genetic diversity and among-population differentiation (Diao et al. 2014; Liang et al. 2015; Pigliucci et al. 2006; Roberto et al. 2009).

Dispersal of *C. paliurus* seeds is wind-driven, and dispersal patterns are shaped by topography and local wind regimes. Previous studies have focused mainly on seed quality and dormancy-breaking methods (Fang et al. 2006), with relatively little attention to seed-trait variation across the highly heterogeneous habitats occupied by the species.

Although phenotypic variation in *C. paliurus* has been investigated, most studies relied on geographically limited materials (She et al. 2009), and disentangling environmental effects from genetic differentiation remains challenging. Recognizing these constraints, we examined the drivers of seed-trait variation to improve understanding of the species' adaptability.

Our goal was to assemble a range-wide seed collection representing diverse origins in China and to provide a comprehensive view of phenotypic variation in *C. paliurus*. To our knowledge, our dataset (139 accessions) is the largest compiled to date. Specifically, we aimed to (i) quantify phenotypic variation in seed traits, (ii) examine inter-trait correlations, (iii) test associations between seed traits and environmental variables and characterize within- and among-population variation, and (Stephanie et al.) partition trait variance attributable to geographic origin. These results are expected to clarify distribution and variation patterns under contrasting environments and to support germplasm grouping for breeding targets, as well as the development of conservation and utilization strategies in China.

Materials and methods

Plant materials

The investigations were conducted in natural forests across the entire geographical range of *C. paliurus* in China. 139 seed samples were collected from 30 natural populations (Fig. 1) in sub-tropical regions and the distance between any two populations is more than 100 km, with the exception of Z1, Z2, and Z3, which are at least 30 km apart. Samples from each population were collected at least 100 m apart; all sampling sites are within nature reserves. To reduce variation among individuals due to potential effects of position on seed traits, we collected seed at the upper middle crown of each individual tree and the seeds were removed to laboratory and air-dried until use. The environmental factors, including geographical and climatic parameters, were recorded at each sampling site for meteorological analysis. All the corresponding voucher samples were deposited in the Gene Bank of *C. paliurus*, Nanjing Forestry University, Nanjing, P. R. China.

Seed traits and environmental factor measurements

The seed traits measured on a single-seed basis were seed diameter, seed thickness, seed diameter/thickness (the ratio of seed diameter to thickness), seed wing diameter, seed volume and 1000-kernel weight. 50 seeds in quadruplicate were selected randomly from each sample. Seed diameter, seed thickness, and wing diameter were first measured with a digital caliper (± 0.01 mm). Then the wing and kernel of each seed was separated and their 1000-kernel weights were measured to the nearest 0.0001 g on an electronic balance. The traits calculated on single seed basis were seed diameter, seed thickness, diameter/thickness ratio (seed diameter divided by thickness), seed wing diameter, seed volume and 1000-kernel weight. Finally, the seeds were placed in a measuring cylinder with sand and the seed volume was calculated from the difference in volume before and after immersion. The environmental data (including annual average temperature, annual average precipitation and annual average sunshine hours) of the 30 natural populations were retrieved using a 30-year climatic database (1971–2000). Geographical parameters including longitude, latitude and altitude were measured for each sampled individual using GPS (Table 1).

Table 1
Geographical and climatic information of the 30 natural populations of *C. paliurus* investigated

Code	Populations	Latitude (°N)	Longitude (°E)	MAT	Altitude /m	MASH	MAP	n
A1	Jixi, Qingliangfeng, Anhui	30°08'50"	118°53'41"	15.0	680	1926.4	1480.0	11
A2	Jingde, Anhui	30°13'34"	118°27'00"	15.5	610	1784.1	1307.6	5
A3	Qimen, Guniujiang, Anhui	29°56'11"	117°42'44"	11.9	400	1800	1762.9	3
A4	Shucheng, Anhui	31°29'00"	116°52'30"	15.6	790	1969	1027.7	4
F1	Pucheng,Kuangshan,Fujian	27°55'43"	118°35'46"	13.2	786	1738.7	1200.0	2
F2	Sanming, Mingxi, Fujian	26°20'07"	117°10'03"	16.5	574	1900	1737.0	13
Z1	Jiumucun, Anji, Zhejiang	30°34'36"	119°38'22"	11.6	774	2009	1476.0	1
Z2	Longwangshan, Anji, Zhejiang	30°40'00"	119°41'00"	14.0	425	1851	1400.0	4
Z3	Meiwuling, Anji, Zhejiang	30°40'12"	119°41'13"	16.5	678	1862	1600.0	3
Z4	Fenghua, Zhejiang	29°45'41"	121°13'04"	16.0	513	1850	1602.0	8
Z5	Lishui, longquan, Zhejiang	28°05'40"	119°11'19"	12.3	1216	1783.2	1415.7	6
Z6	Tiantongsi, Ningbo, Zhejiang	29°48'03"	121°47'33"	16.2	60	1850	1500.0	2
Z7	Wencheng, Zhejiang	27°32'24"	119°06'36"	18.5	959	1725	1698.0	6
H1	Enshi, Hefeng, Hubei	29°52'47"	110°05'10"	14.8	1155	1960	1400.0	1
H2	Wufeng, Yichang, Hubei	30°17'00"	110°31'48"	16.7	688	1533	1893.9	9
H3	Yongsun, Jishou, Hunan	28°57'53"	109°50'53"	15.8	730	1266.3	1365.9	4
H4	Nanzhao, Henan	33°28'35"	112°25'05"	12.7	880	1897.9	900.0	5
H5	Shangcheng, Henan	31°42'54"	115°32'58"	15.4	930	1763.1	1241.4	1
J1	Fenyi, Dagangshan, Jiangxi	27°49'37"	114°40'56"	18.0	450	1737.1	1590.0	3
J2	Xiushui, Jiujiang, Jiangxi	29°05'07"	114°31'08"	16.5	854	1700	1450.0	1
S1	Lueyang, Shaanxi	33°20'00"	106°10'24"	13.2	1250	1526.2	860.0	6
S2	Leshan, Muchuan, Sichuan	28°58'00"	103°47'00"	12.9	1200	965.3	1533.2	15
S3	Qingchuan, Guangyuan, Sichuan	32°35'00"	105°15'36"	21.5	825	900	1340.0	3
G1	Longlin, Baise, Guangxi	24°45'36"	105°20'00"	17.1	1798	1475	1200.0	6
G2	Tianlin, Baise, Guangxi	24°20'18"	106°15'24"	20.3	1445	1377.7	1115.0	3
G3	Longsheng, Guilin, Guangxi	25°49'12"	109°58'24"	16.4	606	1244	1544.0	4
G4	Jianghe, Qiandongnan,	26°42'12"	108°22'48"	15.4	1178	1236.3	1200.0	2

MAT: mean annual temperature (℃); MASH: mean annual sunshine hours (h); MAP: mean annual precipitation (Stephanie et al.);

n = number of individuals

Code	Populations	Latitude (°N)	Longitude (°E)	MAT	Altitude /m	MASH	MAP	n
	Guizhou							
G5	Liping, Qiandongnan, Guizhou	26°20'24"	109°09'36"	15.6	727	1317.5	1425.9	2
G6	Shiqian, Tongren, Guizhou	27°32'00"	108°15'36"	16.7	1239	1232.9	966.0	2
G7	Yinjiang, Tongren, Guizhou	27°58'24"	108°30'36"	15.1	1032	1255	1100.0	4
MAT: mean annual temperature (℃); MASH: mean annual sunshine hours (h); MAP: mean annual precipitation (Stephanie et al.);								
n = number of individuals								

Statistical analysis

We studied the phenotypic variation of *C. paliurus* seed traits using a nested design, which is an appropriate alternative to study the genetic structure of a population. Because of its random structure, all individuals contribute information (Guan et al. 2016; Pereira et al. 2001). The following linear mixed model was used to investigate the variance components for phenotypic variation in *C. paliurus* seed traits across the 30 populations.

Y_{ijk}= μ + P_i + $T_{j(i)}$+ $\epsilon_{(ij)k}$

where Y_{ijk} is *k*th observation of the seed phenotypic traits within the *j*th individual seed sample of *i*th population, *μ* is the overall mean, P_i is the effect of the *i*th population (fixed), $T_{j(i)}$is the effect of the *j*th tree within the *i*th population (random), and $\epsilon_{(ij)k}$ is the residual error. $V_{st}$was calculated as V_{st} = $\delta t/s^2/(\delta t/s^2 + \delta s^2)$, where $\delta t/s^2$is the variance component among populations, and δs^2 is the variance component within population(Liang et al. 2015). For straightforward interpretation, the variance components were expressed as percentages of the total variance.

For each population, values of the seed phenotypic traits were reported as mean ± SD (standard deviation), the coefficient of variation (*CV*) according to the formula *CV* = 100%× SD/mean were calculated. Pearson's correlation coefficient was used to assess the correlations between seed traits and geographical and environmental parameters and among different seed traits. One-way analysis of variance (ANOVA) was performed to analyze the seed phenotypic traits of different populations. K-means clustering was undertaken to further examine the seed phenotypic traits of different *C. paliurus* populations and the cluster plot was constructed using the first two principal components (Dim1 and Dim2). All the above statistical analysis was performed with R 3.5.2.

Results

Variation in C. paliurus seed phenotypic traits

Seeds of *C. paliurus* from different natural populations clearly varied in morphological characteristics. The ANOVA results showed that most seed traits, including seed diameter ($F = 312.2$, $P < 0.01$), seed thickness ($F = 111.4$, $P < 0.01$), seed diameter/thickness ($F = 137.00$, $P < 0.01$), seed wing diameter ($F = 495.68$, $P < 0.001$), seed volume ($F = 18.22$, $P < 0.001$) and seed 1000-kernel weight ($F = 705.148$, $P < 0.001$) were significantly different among the 30 populations and most seed traits, including seed diameter ($F = 52.8$, $P < 0.01$), seed thickness ($F = 37.8$, $P < 0.01$), seed diameter/thickness ($F = 48.00$, $P < 0.01$), seed wing diameter ($F = 43.69$, $P < 0.01$), seed volume ($F = 3.859$, $P < 0.001$) and seed 1000-kernel weight ($F = 95.375$, $P < 0.001$) were also significantly different within populations.

The variance analysis of seed phenotypic traits of 30 *C. paliurus* natural populations is presented in Fig. 2. The seed diameter (Fig. 2a) and seed diameter/thickness (Fig. 2c) of Enshi natural population (H1) in the central region of the distribution of *C. paliurus* were the largest, which were 1.13 cm and 1.59. The smallest seed diameter and seed diameter/thickness were found in Fenhua population (Z4), which were only 0.77cm, 1.03 and in the eastern of the distribution of *C. paliurus*. As for seed thickness (Fig. 2b), the thickest seed (0.91cm) was observed from Leshan natural population (S2), while the thinnest seed (0.58cm) was observed from Xiushui population (J2), which was approximately 36% lower than that of the Leshan population (S2). The seeds of Lishui natural population (Z5) showed the best performance in the seed wing diameter (Fig. 2d) and seed 1000-kernel weight (Fig. 2f), which were the largest (5.65cm) and heaviest (227.7g) seeds sources of these collections. The smallest wing diameter (3.06cm) was found in Longwangshan population (Z2) and the lightest seeds (93.38g) came from the Qingchuan population (S3). The seed volume (Fig. 2e) varied significantly among the populations, ranging from 0.13 (Tiantongsi population (Z6)) to 0.34cm^3 (Leshan population(S2)).

The seed traits of *C. paliurus* in different populations exhibited a wide variation range, with *CV* ranging from 0.01–55.83% (Table 2). The average *CV* of the seed volume among 30 populations exhibited the largest variation (36.24%), while the seed diameter exhibited the lowest variation (10.68%).The variation in seed phenotypic traits among the 30 populations was relatively high (17.53%); the average variation within populations was largest in Lueyang natural population (S1) (26.99%) and lowest in Enshi natural population (H1) (8.77%).

Table 2
Coefficient of variance of seed phenotypic traits in natural *C. paliurus* populations

Code	SD	ST	SDT	SWD	SV	SKW	Mean
A1	7.65	12.68	13.12	13.88	33.66	14.86	15.98
A2	10.95	10.95	11.41	11.03	38.88	12.75	16.00
A3	16.16	13.94	17.55	10.74	55.83	11.77	21.00
A4	14.16	17.17	14.81	8.97	33.85	20.65	18.27
F1	8.17	12.41	14.56	12.28	26.02	0.94	12.40
F2	11.53	18.13	17.91	14.46	45.99	27.40	22.57
Z1	10.17	11.87	16.22	10.97	24.00	0.17	12.23
Z2	10.46	12.41	16.86	30.87	29.93	8.22	18.13
Z3	9.30	14.84	13.44	9.14	32.14	17.50	16.06
Z4	10.26	14.04	13.10	12.90	26.22	18.79	15.89
Z5	11.11	12.56	11.87	13.48	54.81	13.65	19.58
Z6	7.94	13.14	11.42	8.93	52.84	27.19	20.24
Z7	9.67	15.40	13.23	31.63	23.09	3.86	16.15
H1	12.42	11.13	15.73	10.30	3.01	0.02	8.77
H2	10.08	17.10	16.62	13.31	32.12	26.32	19.26
H3	13.71	10.52	16.00	9.65	39.52	20.91	18.38
H4	19.89	13.81	22.77	11.98	30.89	13.48	18.80
H5	8.75	9.22	12.67	11.72	40.23	0.01	13.77
J1	13.98	11.03	17.79	11.59	48.91	12.39	19.28
J2	7.88	13.26	9.30	11.99	33.15	0.01	12.60
S1	11.89	27.78	21.83	18.40	54.26	27.78	26.99
S2	9.89	32.97	16.11	13.16	18.90	12.86	17.32
S3	5.67	12.08	12.10	9.01	25.37	5.03	11.54
G1	9.74	16.13	15.19	24.77	53.19	17.38	22.73
G2	11.75	16.90	12.15	18.06	25.04	20.39	17.38
G3	9.20	16.28	17.07	11.94	37.49	17.38	18.23
G4	14.12	12.43	17.63	8.47	40.54	39.34	22.09
G5	5.84	12.50	14.50	13.54	46.72	18.57	18.61

SD = seed diameter (cm); ST = seed thickness (cm); SDT = seed diameter/thickness; SWD = seed wing diameter (cm);

SV = seed volume (cm^3); SKW = 1000-kernel weight (g)

Code	SD	ST	SDT	SWD	SV	SKW	Mean
G6	7.80	13.18	14.98	7.47	39.24	1.37	14.01
G7	10.16	17.22	13.34	24.49	41.42	22.77	21.57
Mean	10.68	14.77	15.04	13.97	36.24	14.46	17.53
SD = seed diameter (cm); ST = seed thickness (cm); SDT = seed diameter/thickness; SWD = seed wing diameter (cm);							
SV = seed volume (cm^3); SKW = 1000-kernel weight (g)							

The Pearson correlation matrix of seed traits (Fig. 3) of 139 samples revealed a high positive association of seed diameter with seed diameter/thickness (0.56), seed wing diameter (0.49), seed 1000-kernel weight (0.57), seed thickness (0.36) and seed volume (0.36). The seed thickness showed high positive association with seed wing diameter (0.40), seed volume (0.52), seed 1000-kernel weight (0.43) and high negative association with seed diameter/thickness (-0.56). A high positive association of seed wing diameter with seed volume (0.38) and seed 1000-kernel weight (0.71) was found. Also, A high positive association between seed volume and seed 1000-kernel weight (0.36) was observed.

Phenotypic differentiation of C. paliurus seeds

The phenotypic differentiation coefficients (*Vst*) are expressed as the proportion of the variance in the seed traits among different populations to total genetic variation. For a better understanding of phenotypic variance of *C. paliurus* among 30 natural populations, the *Vst* was measured using six seed traits based on nested variance component analysis (Table 3), and the results showed the variation of the *Vst* of the six seed traits in seed phenotypic traits of the 30 natural populations was relatively high, ranging from 74.67–91.9% with an average of 82.41%. The average *Vst* of the seed wing diameter among 30 populations exhibited the largest variation (91.9%), while seed diameter/thickness exhibited the lowest variation (74.06%). The average *Vst* of the seed traits was 77.32% among the 30 natural populations, but 16.33% within the populations. Therefore, this finding revealed that variation and diversity of seed traits among the populations were significantly greater than within the populations.

Table 3
Variance component and phenotypic differentiation coefficient (*Vst*) among the C. paliurus populations

Seed traits	Variance component			Variance component proportion (%)			V_{st} (%)
	Among populations $\delta t/s^2$	Within populations δs^2	Errors $\delta^2 e$	Among populations $P_{t/s}$	Within populations p_s	Errors p_{se}	
seed diameter	2.07	0.35	0.006	85.30	14.43	0.27	85.53
seed thickness	1.76	0.60	0.02	74.17	25.16	0.66	74.67
diameter/thickness	3.09	1.08	0.02	73.66	25.80	0.54	74.06
seed wing diameter	155.85	13.74	0.31	91.73	8.09	0.18	91.90
seed volume	0.06	0.01	0.04	50.96	12.58	36.46	80.20
1000-kernel weight	32989	4452	0.15	88.11	11.89	0.0004	88.11
Mean	5525.305	744.63	0.091	77.32167	16.325	6.351733	82.41167

Relationship between seed traits and environmental factors

Generally, seed traits that exhibited large differences within and among populations may be affected by climate conditions and geographical environment. Analysis of seed traits comparison with environmental data (Fig. 3) revealed seed 1000-kernel weight was significantly negatively correlated with mean annual temperature (-0.34), while it was positively correlated with altitude (0.23). The seed wing diameter showed high negative association with longitude (-0.31), mean annual sunshine hours (-0.37) and mean annual temperature (-0.27) and high positive association with altitude (0.38). The seed diameter was significant positively correlated with altitude (0.33), but negatively correlated (-0.28) with longitude and mean annual temperature (-0.26). The seed thickness was found positively association with altitude (0.24) and negative association with longitude (-0.22) and mean annual precipitation (-0.22). A positive association between seed diameter/thickness and mean annual precipitation (0.22) were observed. The seed volume showed weaker associations with environmental factors.

The cluster plot (Fig. 4) for the 30 populations was obtained through K-means cluster analysis using the most significant components (Dim1 and Dim2), which showed 30 populations divided into six distinct clusters. Cluster I included one population from Hubei(H1); cluster II included the two populations from Jiangxi(J2) /Hubei (H1); cluster III included the six populations from Sichuan (S1), Hunan (H3), Henan (H4), Fujian (F1), Zhejiang (Z7), Guizhou (G2); cluster IV included the six populations from Zhejiang (Z1, Z2, Z4), Anhui (A1, A4) and Sichuan (S3); cluster V included the two populations from Zhejiang (Z5) and Sichuan (S2); cluster VI included the thirteen populations from Guizhou (G1, G3, G4, G5, G6, G7), Hubei(H2), Henan(H5), Zhejiang (Z3, Z6), Fujian(F2), Jiangxi (J1) and Anhui (A2).

Discussion

Range-wide diversity and population differentiation in seed traits

Across 30 natural populations, we documented substantial phenotypic variation in seed traits, with significant differences both among and within populations (Results). At the population level, the mean coefficient of variation (CV) was 17.53%; the among-population variance component was greatest for seed volume (36.24%) and smallest for seed diameter (10.68%); within-population variation peaked at Lueyang (S1) and was lowest at Enshi (H1). This hierarchical partitioning indicates that population differentiation is a prominent feature of seed phenotypes in Cyclocarya paliurus. Conceptually, such patterns are consistent with spatially heterogeneous selection in the Q_ST–F_ST framework when phenotypic differentiation exceeds neutral expectations, while recognizing that explicit genomic benchmarks and common-garden designs are required for formal inference(Chung et al. 2023; Leinonen et al. 2013; Whitlock 2008). Placing our magnitudes in a broader context, range-wide tree studies commonly find that functional traits map onto geography and climate: in Populus trichocarpa, multi-trait datasets from common gardens revealed high phenotypic variance and clear clines with environment, with moderate–high heritabilities for many traits(Klein et al. 2025b; McKown et al. 2014). At macroecological scales, seed size shows systematic geographic/climatic structure across woody floras (e.g., latitude, energy, water), indicating that seed phenotypes often track environmental structure at large spatial extents(Moles et al. 2006). Recent trait syntheses further identify diaspore mass as a primary axis in the global spectrum of plant form and function(Diaz et al. 2022), providing an updated macroecological context for our size-related variance components. Finally, the marked within-population variation we observed (e.g., S1) is ecologically plausible in mountainous landscapes where maternal environments and microhabitat heterogeneity inflate seed variance; classic syntheses and experiments show that maternal conditions during seed development shift seed mass and related traits(Callejas-Diaz et al. 2022; Donohue 2009).

Environmental gradients as primary correlates of wing morphology and seed mass

Trait–environment analyses (Fig. 3) identified altitude and thermal regime as the main correlates of wing diameter and seed mass (TKW). The observed tendency for larger wings at higher elevations and heavier seeds at cooler, less energy-rich sites is mechanistically plausible. Aerodynamic experiments demonstrate that autorotating samaras generate elevated lift via a stable leading-edge vortex (LEV), which lowers terminal velocity and prolongs time aloft—advantages in complex mountain winds (Jung & Rezgui 2024; Lentink et al. 2009). Wind-tunnel tests across 36 species further show that wing loading (mass per wing area)—not terminal velocity alone—best predicts the lift-off threshold for secondary wind dispersal, emphasizing the joint roles of wing area and mass, i.e., the wing–mass coupling we observe (Tian et al. 2023). Mechanistic models highlight canopy turbulence and updrafts as key enablers of long-distance dispersal, favoring designs that either stay aloft longer (larger wings) or secure establishment under short growing seasons (greater maternal provisioning → heavier seeds)(Soons & Ozinga 2005).

At the same time, cross-taxon comparisons caution against universal directions: the seed-mass–dispersal trade-off is strong among species but often weak or inconsistent within species once cone/wing traits are modeled explicitly(Wyse & Hulme 2021, 2022). This supports a mechanism-anchored view in which elevational and thermal regimes filter combinations of wing and mass, rather than acting on single traits in isolation—an interpretation consistent with our results (Fig. 3).

Multivariate trait syndromes and geographically coherent phenotypic groups

Multivariate analyses demonstrated that seed-size traits (diameter, thickness, volume, TKW) loaded primarily on one axis, whereas wing morphology loaded on another, and K-means clustering recovered geographically coherent phenotypic groups (Fig. 4). This structure indicates that locally favored trait syndromes—no single traits—underpin dispersal and early establishment strategies across environments. In *P. trichocarpa*, multiple traits covary along climatic/geographic axes, and trait space maps onto environmental space, reinforcing that environmental filtering acts on coordinated suites of traits (McKown et al. 2014)). From a theory standpoint, the global spectrum of plant form and function is low-dimensional, with variation collapsing onto a size axis (encompassing diaspore/seed mass) and a resource-economics axis(Diaz et al. 2022), predicting that filters will act on trait combinations. Samara flight work shows that descent velocity can be robust to sizeable perturbations in mass or wing area when performance is described by unified drag formulations rather than simple correlations, implying is performance surfaces in trait space (Lentink et al. 2009). Consistent with this robustness, recent wind-tunnel experiments in Acer demonstrate near-constant descent performance despite large morphological perturbations(Schaeffer et al. 2024), and, at the biogeographical scale, samara-bearing species in China show clear geographic–climatic structuring(Du et al. 2022), paralleling our clusters' alignment with regional environments. Such robustness helps explain why our PCA reveals coordinated axes and our clusters align with regional climates—multiple morphological routes can achieve comparable dispersal outcomes, with local windscapes and thermal regimes determining which routes are favored (Fig. 4).

Conclusion

Our range-wide assessment of Cyclocarya paliurus seed phenotypes reveals three consistent signals. First, seed-trait diversity is high and strongly structured among populations: across 30 natural populations the mean CV was 17.53%, the among-population component was greatest for seed volume (36.24%) and smallest for seed diameter (10.68%), and within-population variation peaked at Lueyang (S1) and was lowest at Enshi (H1) (Results). Second, altitude and thermal regime emerged as the primary correlates of wing diameter and seed mass (TKW) (Fig. 3). Third, multivariate analyses resolved a size axis (diameter, thickness, volume, TKW) distinct from a wing-morphology axis, and K-means recovered geographically coherent phenotypic groups (Fig. 4). These patterns are mechanistically coherent with samara aerodynamics and trait-based theory. Larger wings at higher elevations and heavier seeds at cooler, less

energy-rich sites align with LEV-based lift and wing-loading constraints that prolong time aloft while securing establishment under short growing seasons(Lentink et al. 2009; Tian et al. 2023). The separation—and coordinated covariation—of size and wing axes, together with geographically structured clusters, indicates that environmental filtering acts on trait combinations rather than single traits, consistent with the low-dimensional global spectrum in which diaspore/seed mass is a primary axis(Diaz et al. 2022). Recent wind-tunnel work further shows that different wing–mass combinations can deliver near-equivalent descent performance, explaining why multiple morphological routes can be favored in different windscapes(Schaeffer et al. 2024).

Implications and next steps. These findings provide an operational basis for climate-informed seed sourcing and seed-zone delineation in restoration and ex situ conservation—e.g., prioritizing large-wing/heavy-seed populations for cooler, windier sites, with contrasting syndromes matched to warmer targets(Török et al. 2024; Vitt et al. 2022). To distinguish local adaptation from phenotypic plasticity and to translate mechanisms into guidelines, we recommend (i) common-garden/reciprocal-transplant tests spanning thermal and wind regimes; (ii) aerodynamic assays quantifying wing loading and flight performance under controlled turbulence; and (iii) genomic contrasts (e.g., Q_ST–F_ST; climate-GWAS) to benchmark phenotypic divergence against neutral structure and to identify adaptive loci (Chung et al. 2023; Klein et al. 2025a; Leinonen et al. 2013; Whitlock 2008). Linking flight performance to seedling establishment across climates will close the mechanism-to-fitness loop and refine climate-smart transfer for this socio-economically important species.

Declarations

Author Contribution

W.Y. and Y.Ce. designed the study and carried out field sampling. W.Y. performed seed trait measurements and data analysis. Y.Ce. contributed to data processing and visualization. Y.Cu. assisted with data curation and figure preparation. X.L. and S.F. supervised the research, provided guidance on methodology, and revised the manuscript critically. W.Y. drafted the initial version of the manuscript. All authors discussed the results, contributed to manuscript revisions, and approved the final version for submission.

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Data Availability

The datasets generated and analyzed during the current study are available from the corresponding authors on reasonable request.

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Figures

Figure 1

Geographic location of the 30 natural populations of *Cyclocarya paliurus* investigated in this study.

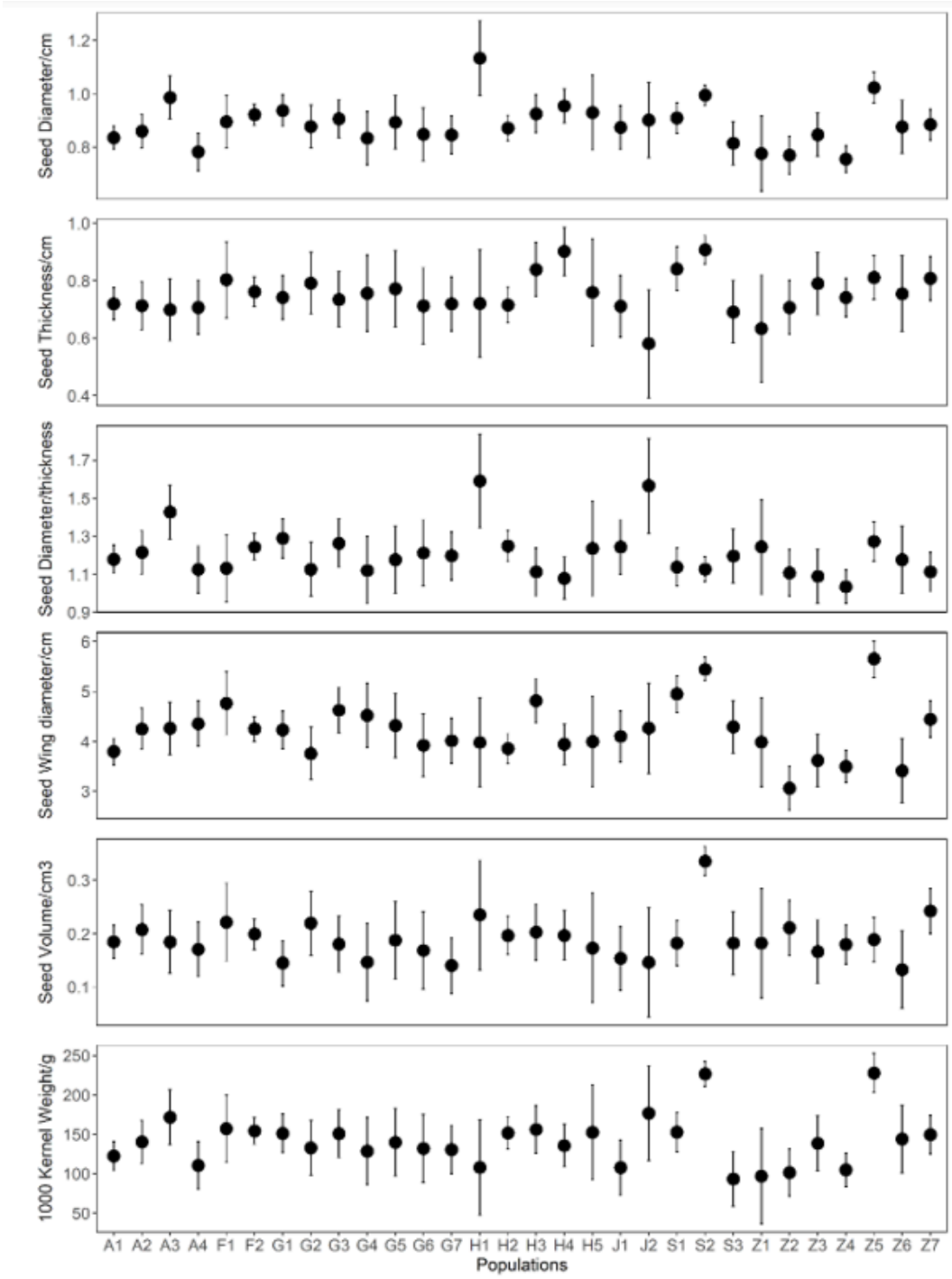

Figure 2

Variance analysis of six seed phenotypic traits (seed diameter, seed thickness, diameter/thickness ratio, wing diameter, seed volume, and 1000-kernel weight) among and within natural populations of *C. paliurus*.

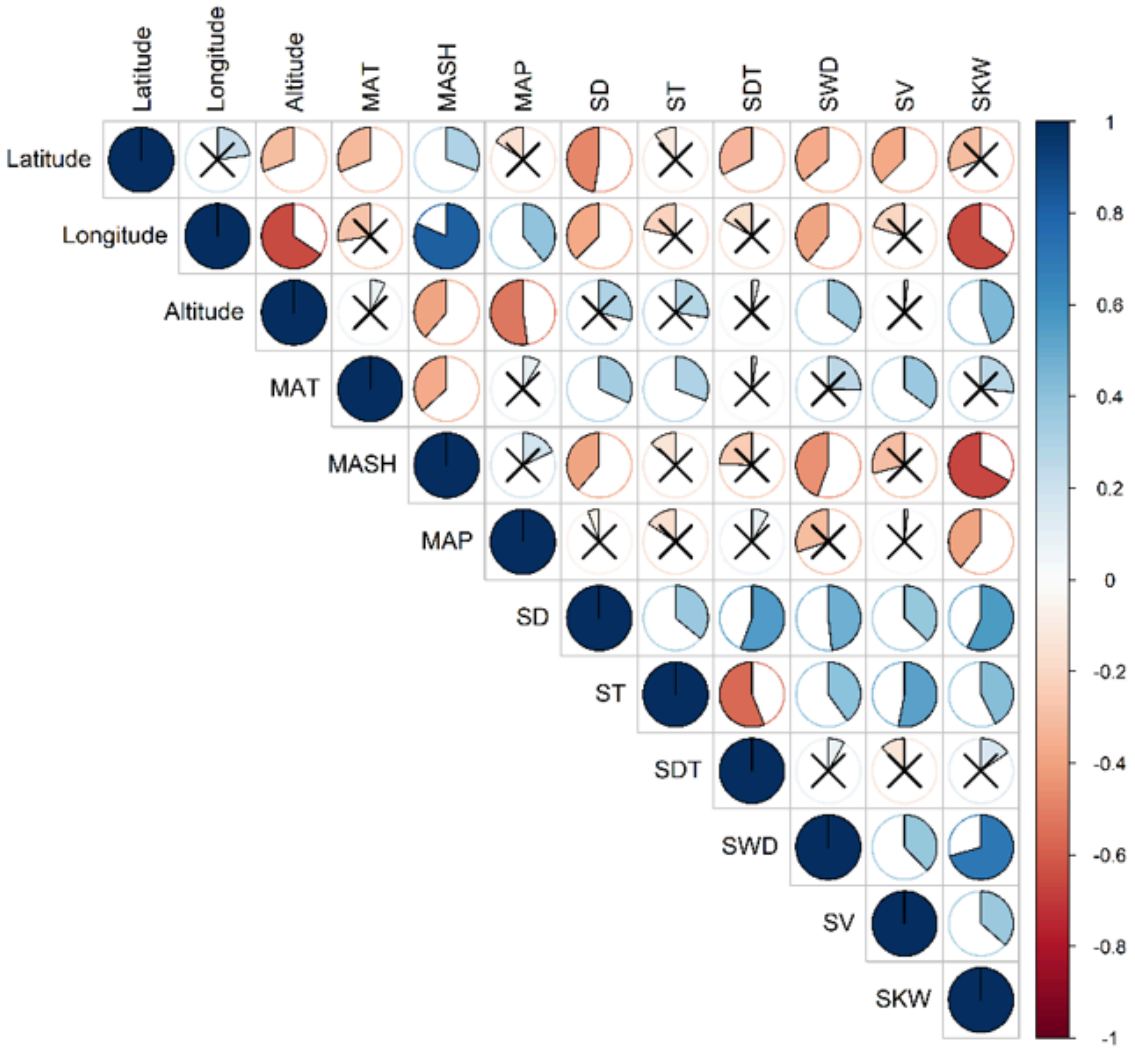

Figure 3

Correlation analysis between seed phenotypic traits and environmental factors (MAT: mean annual temperature; MASH: mean annual sunshine hours; MAP: mean annual precipitation). Traits include SD = seed diameter (cm), ST = seed thickness (cm), SDT = diameter/thickness ratio, SWD = seed wing diameter (cm), SV = seed volume (cm^3), and SKW = 1000-kernel weight (g).

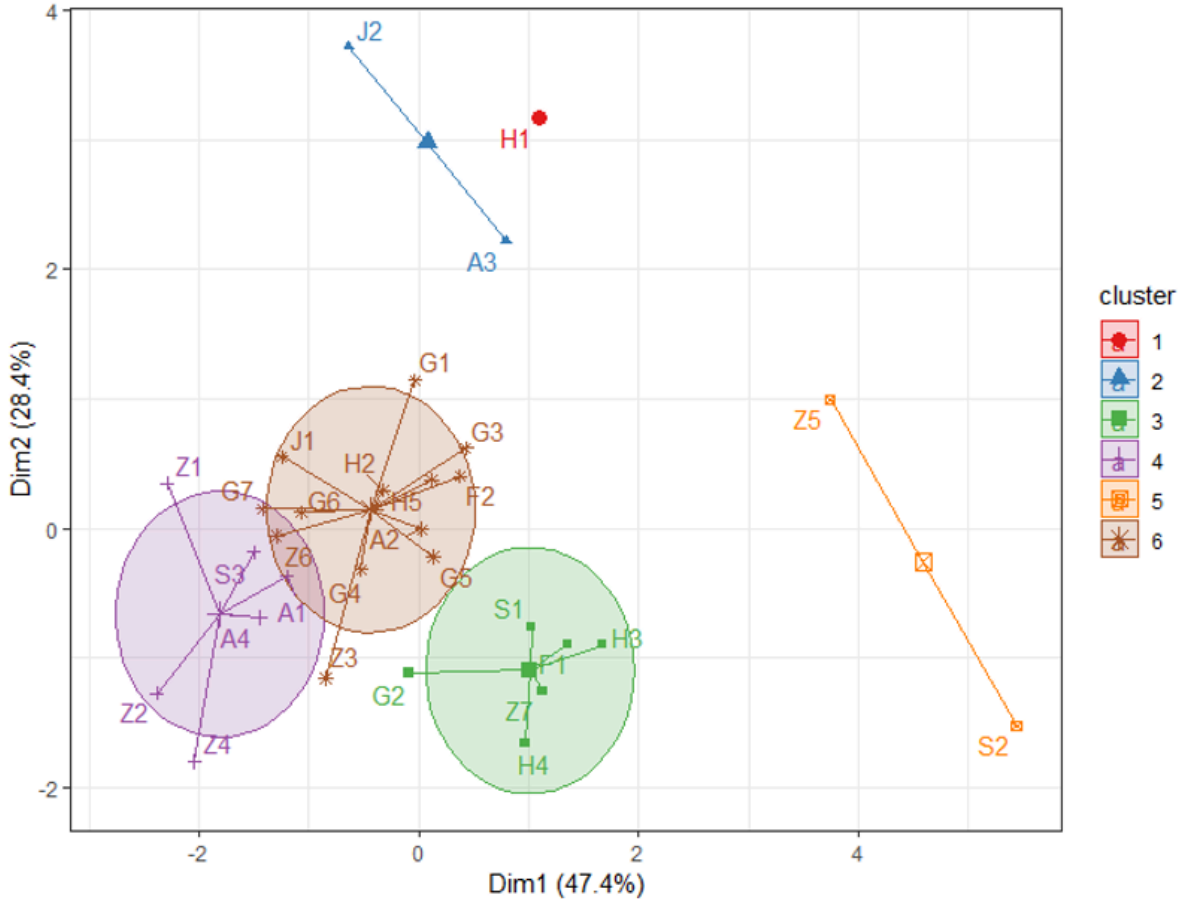

Figure 4

K-means clustering plot of 30 *C. paliurus* populations based on principal components of seed phenotypic traits, showing six distinct phenotypic groups.