

## Research article

# Habitat preferences and functional traits drive longevity in Himalayan high-mountain plants

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Plant lifespan has important evolutionary, physiological, and ecological implications related to population persistence, community stability, and resilience to ongoing environmental change impacts. Although biologists have long been puzzled over the extraordinary variation in plant lifespan and its causes, our understanding of interspecific variability in plant lifespan and the key internal and external factors influencing longevity remains limited. Here, we demonstrate the concurrent impacts of environmental, morphological, physiological, and anatomical constraints on interspecific variation in longevity among > 300 vascular dicot plant species naturally occurring at an elevation gradient (2800–6150 m) in the western Himalayas. First, we show that plant longevity (ranging from 1 to 100 years) is largely related to species' habitat preferences. Ecologically stressful habitats such as alpine and subnival host long-lived species, while productive ruderal and wetland habitats contain a higher proportion of shorter-lived species. Second, longevity is influenced by growth form with monocarpic forbs having the shortest lifespan and woody shrubs having the highest. Small-statured cushion plants with compact canopies and deep roots, most found on cold and infertile alpine and subnival soils, had a higher chance of achieving longevity. Third, plant traits reflecting plant adaptations to stress and disturbance affect interspecific differences in plant longevity. We show that longevity and growth are negatively correlated. Slow-growing species are those that have a higher chance of reaching a high age. Finally, higher longevity was associated with high leaf carbon and phosphorus, low root phosphorus and nitrogen, and with large bark-xylem ratio. Our findings suggest that plant longevity in high elevation is intricately determined by a combination of habitat preferences and growth form, as well as the plant growth rate and physiological processes.

Keywords: adaptation strategies, alpine plants, environmental constraints, lifespan, longevity, plant functional traits



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## Introduction

Biologists have long been puzzled over the extraordinary variation in plant lifespan. Some plants live only a few weeks while others live for hundreds or even thousands of years (Kirkwood and Austad 2000, Silvertown 2013, Thomas 2013), and the evolutionary, physiological, and ecological importance of this variation is widely accepted (Munné-Bosch 2008, de Witte et al. 2012). For instance, lifespan is thought to relate to population persistence, community stability, and resilience (de Witte and Stöcklin 2010), with direct implications on the response to climate change (Morris et al. 2008). Yet, despite the extensive sampling over the last two decades (Schweingruber and Poschlod 2005, Schweingruber et al. 2013, 2020, Kipkeev et al. 2015, Doležal et al. 2018), data on plant age or even the potential maximum lifespan of plant species (longevity hereafter used synonymously) is still one of the least available variables in plant life history, particularly for herbaceous plants (McCarthy 1992, Roeder et al. 2021). A plant's ability to persist in harsh conditions depends on its ability to adapt to environmental constraints, which is reflected in its functional traits (Grime 1974, Körner 2021). Several studies on herbaceous plants have shown the influence of the environmental constraints determining the plant functional traits on the leaf lifespan (Adler et al. 2014, Zheng et al. 2022), while few other studies have shown the influence at whole plant level (Rosbakh and Poschlod 2018, Roeder et al. 2021). However, to comprehend plant persistence and life history, our understanding of interspecific variability of functional traits at the whole plant level as well as their association with environmental constraints driving longevity remains still limited in herbaceous plants (Ehrlén and Lehtilä 2002).

Historically, plant lifespan has been divided into three categories based on the number of growing seasons required to complete their life cycle: one (annual), two (biennial) and many (perennial). Due to the practical challenges of measuring age, plant size has often been used as a proxy of age and longevity in demographic studies of perennials and herbaceous species (Thomas 2013). Although useful in many ways, the use of plant size is very imprecise, especially for plants that grow in natural conditions for decades or centuries with little changes in overall size (Schweingruber and Wirth 2009). It could even be misleading when comparing populations growing under different environmental conditions and exhibiting contrasting growth rates, i.e. small individuals could be older than large individuals (Doležal et al. 2021). Recent advances in the anatomical determination of annual growth rings formed by secondary thickening of roots and stems allow us to overcome this challenge and make it possible to determine the exact age of individuals and their radial growth histories in many herbaceous dicot species (Dietz and von Arx 2005, von Arx et al. 2016, Doležal et al. 2018, Rosbakh and Poschlod 2018) and thus allow to explore the links between longevity and environmental drivers.

Two environmental drivers interact to influence longevity: environmental stress and disturbances (Grime 1974).

Grime (1974, 2006) defines stress as any factor that limits the rate of dry matter production of the vegetation due to its shortage or excess supply, e.g. light, moisture, temperature, etc. whereas a disturbance is any discrete event that disrupts an ecosystem or community and causes partial or total destruction of the vegetation, e.g. grazing, landslides, or erosion. Plants that can cope with environmental stress are expected to live longer whereas plants experiencing frequent disturbance are expected to have a short lifespan (Molisch 1938, Rosbakh and Poschlod 2018). Consequently, plant traits reflecting, directly or indirectly (functional markers), plant adaptations to stress and disturbances should inform about interspecific differences in plant longevity. In addition, some plant lineages may have evolved higher longevity together with these traits, so disentangling environmental drivers of longevity must account for the phylogenetic relatedness of species. However, the relative importance of different adaptive mechanisms for managing environmental stress and disturbances and hence promoting plant longevity is still poorly understood, especially in herbaceous plants.

Higher plant longevity is often associated with decreased environmental favorability (Nobis and Schweingruber 2013), which selects for specific adaptations such as in alpine plants (Lütz 2012). Low temperatures and soil nutrient deficiencies in mountain environments can potentially extend plant life by slowing meristematic growth (cell division and differentiation, Körner 2021) or by preferentially investing energy into somatic maintenance over sexual reproduction (Kirkwood 1977, Obeso 2002, Doležal et al. 2021). Long-lived plants thus often show slow vegetative growth (Schweingruber and Poschlod 2005, Büntgen et al. 2019), clonal multiplication, prolonged growth dormancy (Peñuelas and Munné-Bosch 2010, Klimešová et al. 2019), long-lived parenchymal cells (Doležal et al. 2018), specific resistance to pathogens (Larson 2001), or repair of somatic mutations that accumulate with age (de Pinto et al. 2012).

Because plants are sedentary organisms that inhabit the same place throughout their lives, their lifespan is strongly tied to the stability of the local environment and the different avoidance, resistance, and resilience strategies that plants utilize to survive disturbances (Rosbakh and Poschlod 2018, Munné-Bosch 2018). A habitat that is resource-limited but stable for a long time usually hosts long-lived species with slow growth and resource-conservative strategies characterized by low tissue and nutrient turnover rates (Adler et al. 2014, Büntgen et al. 2015). Conversely, fertile environments, where frequent or intense and unpredictable disturbances occur, select short-lived species with resource-acquisitive strategies (Herben et al. 2018). Although examples of long-lived individuals from stressful but stable habitats have been repeatedly demonstrated among trees (DeWoody et al. 2008, Schweingruber and Wirth 2009), differences in longevities across a large number of herbaceous plants have rarely been studied at a regional scale.

Functional traits characterize species' morphological, physiological, or anatomical adaptations and can be described at the level of organs, individuals, species, or

whole communities (Wright et al. 2004, Violle et al. 2007, Díaz et al. 2016, Liancourt et al. 2020). For instance, species with low tissue and nutrient turnover rates such as alpine perennial herbs, or persistence traits such as deep taproots or clonality can endure harsh environmental conditions and achieve longer lifespans (Adler et al. 2014). Species with higher growth and tissue turnover rates, like ruderal plants (Grime 1974), have a shorter lifespan (Salguero-Gómez et al. 2016). Integrating functional traits with species habitat and ecological preferences could provide much-needed mechanistic insight and a better understanding of interspecific variation in plant longevity.

This study aims to examine how environmental constraints and plant functional traits explain variation in plant longevity among 355 dicotyledonous species from a wide taxonomic spectrum and contrasting habitats of the northwestern Himalayas. This mountain region provides a well-suited natural laboratory to study the determinants of interspecific differences in longevity. Although overall dry, it encompasses a large diversity of plant species with contrasting traits, and a large diversity of habitats distributed along an extraordinarily long elevation gradient where plants can be found to 6150 m a.s.l., the world elevational limit for vascular plants (Doležal et al. 2018). Habitats from semideserts and steppes to an alpine and subnival zone were included, many of which are species-rich habitats threatened by current land use and climate changes (Doležal et al. 2016). These habitats cover wide thermal and hydrological gradients, which can manifest in different plant longevities due to different evolution and adaptation to specific environmental conditions. Therefore, we first analyzed how interspecific differences in longevity can be explained by species' habitat and ecological preferences related to thermal, hydrological, and disturbance gradients. We expected that stable habitats, such as alpine grasslands, would promote longevity, whereas unstable habitats, such as ruderal sites and alpine scree, would reduce longevity. Second, we explored how longevity variation is further influenced by plant size and growth form. We expected small-stature plants with compact canopies and deep taproots well adapted to cold and infertile soils such as alpine cushion plants to reach old age. Finally, we used plant functional traits to get more mechanistic insights into interspecific variation in plant longevity. We expected that plants with slow growth and resource-conservative strategies (e.g. with higher water and nutrient use efficiencies) would achieve higher longevity than species with fast growth and resource-acquisitive strategies (Adler et al. 2014, Salguero-Gómez et al. 2016).

## Material and methods

### Study area

The plants were collected in the Ladakh region of the northwestern Himalayas, India, on a territory of ca 80 000 km<sup>2</sup>, along an elevational gradient from 2650 to 6150 m. A large part of the studied area is covered by cold deserts and steppes,

salt lakes surrounded by wetlands, while scree, debris fields, and alpine grasslands form a belt above the steppe zone and subnival vegetation in the highest elevations (Dvorský et al. 2011). The 3 km elevation gradient correlates with a decrease in the mean annual/summer air temperatures from 5/18°C to -15/2.5°C (mean annual lapse rate is -7.19°C km<sup>-1</sup>) and an increase in annual precipitation from 50 to 250 mm (Dvorský et al. 2015, Macek et al. 2021). Due to the prevailing soil aridity and climate seasonality in the study area, perennial plants often have deep roots (Klimešová et al. 2011) and distinct annual growth rings (Doležal et al. 2018), which allow us to calculate their age and reconstruct their growth.

## Method

### Anatomical age and growth determination

Plant longevity was defined as the age of the existing, active (live) tissue of the oldest plant parts (root collars) in the oldest individuals collected for a given species (Fig. 1). Depending on the size of the population and the dominance of species in the habitat, target species were selected. From the target species 10–20 largest individuals each (7817 individuals in total) were collected in Ladakh during summer expeditions in 2012–2017 at various elevations covering most of the species (Supporting information). Plant individuals were first excavated from the soil, their height was measured and the roots were washed in water before the plant was separated into individual organs for further laboratory analyses. Plant age, radial growth, and anatomical structure were obtained for each individual according to standard protocols adapted from wood anatomy for herbs (dendrochronology, herbchronology) using growth rings in the secondary xylem of the root collars (Gärtner and Schweingruber 2013, Doležal et al. 2018). In the field, a segment of the oldest root part (about 5 cm long) was cut from each excavated plant and placed in a 50% aqueous ethanol solution to keep the root tissue soft and prevent mold growth. All annual rings of perennial plants should exist in the oldest plant tissues between the hypocotyl and the primary root (i.e. root collar zone), where the reaction to mechanical stress should be also minimal (Büntgen et al. 2015). Therefore, in the laboratory, we cut cross-sections from the root collars of each individual using a sledge microtome, stained with Astra Blue and Safranin and permanently fixed on a microscope slide with Canada Balsam. High-resolution cross-sectional microscopy images were taken using an Olympus BX53 microscope, an Olympus DP73 camera, and cellSense Entry 1.9 software. The age of each individual was estimated as the maximum number of annual rings counted along two radii for each cross-section. The age of the oldest individual of each target species was used as a proxy for maximal plant life span (hereafter longevity) and its functional traits as predictors of longevity (Fig. 2). Although longevity is sometimes used as a synonym for life expectancy (i.e. mean life span), we have used it as a synonym for maximum life span. The mean radial growth rate of the oldest individuals of each species was calculated from the annual growth ring widths measured along

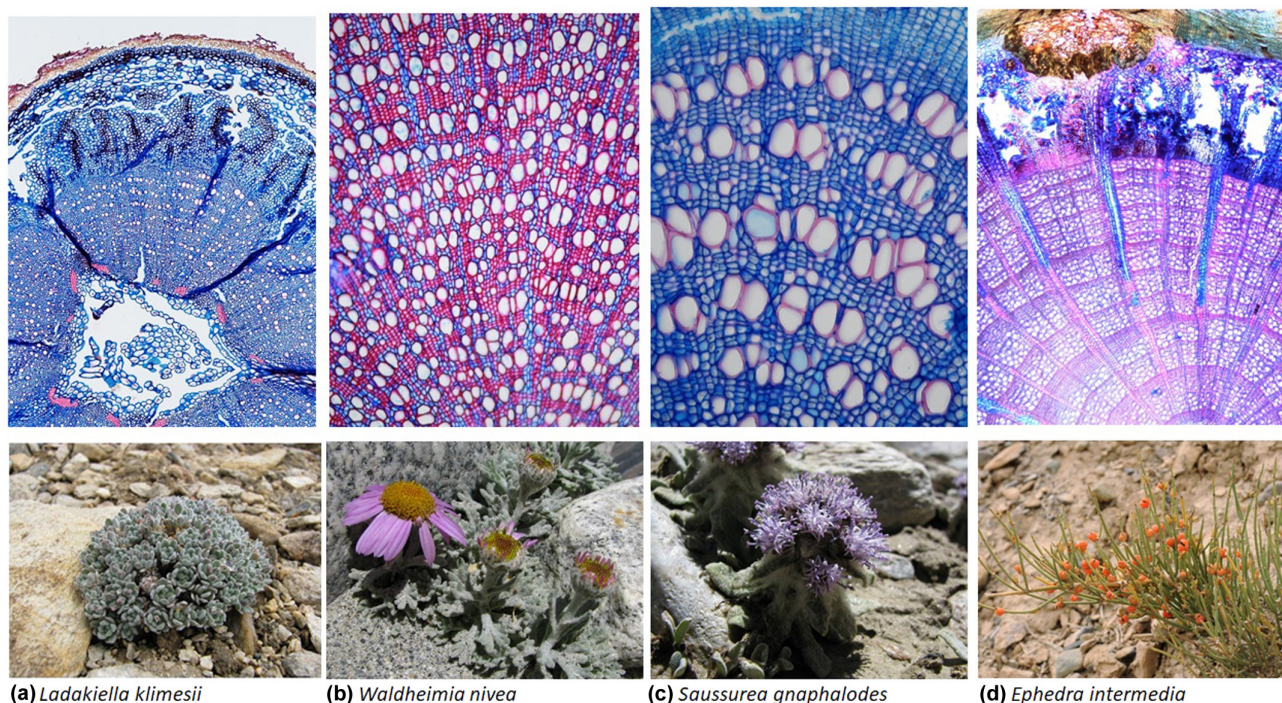


Figure 1. Examples of studied species and their double-stained cross-sections of root collars with growth rings.

two radii for each cross-section. Most of the species studied had the main root including the pith preserved, hence age was accurately determined for > 80% of all dicotyledonous species analyzed. Annual rings were generally well developed in the studied cushion plants due to the predominant ring-porous and semi-ring-porous xylem with prominent parts of early and latewood. However, the degree of distinctness of the rings varied with taxonomy, environmental conditions, and plant age, with large cushion plants, such as *Thylacospermum ceaspitosum*, having last rings indistinct, implying that the age

of these species is likely underestimated. We also recorded the proportion of three tissue types having different functions: sclerenchymatous tissue which provides mechanical support (shows lignification), parenchymatous tissue for carbohydrate storage (does not show lignification), and conductive tissue or xylem vessels for transporting water (Table 1).

#### Species' elevational optima and ecological indicator values

To understand how interspecific differences in plant longevity relate to species habitat preferences, we calculated for each

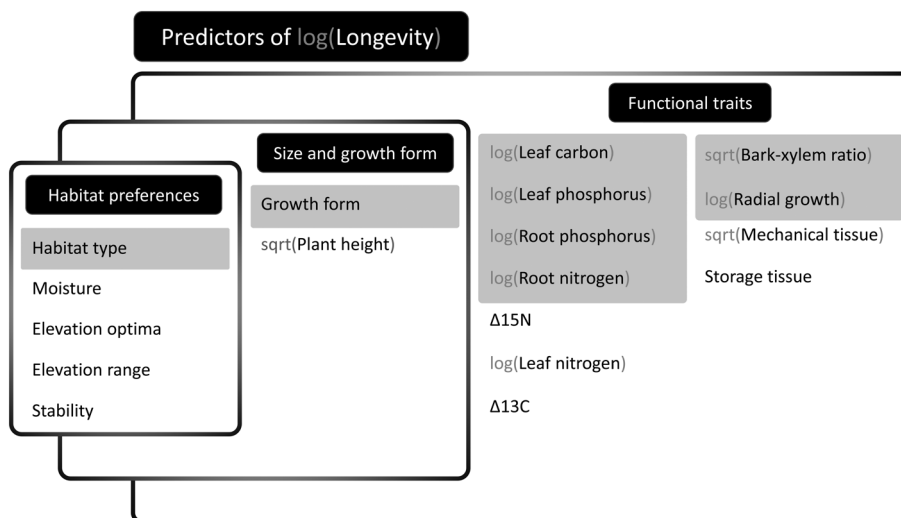


Figure 2. Variables used as predictors of plant longevity. Analyses were done in three steps, each time including selected predictors from previous steps as covariates. Selected predictors are highlighted in grey. 'sqrt' stands for square-root transformation and 'log' for transformation using the natural logarithm.

Table 1. List of functional traits used in this study, their abbreviations, and the trait average values at different habitats. The maximum value of the traits is shown within the parenthesis ( ).

Traits	Abbreviation	Ruderal	Semidesert	Steppe	Wetland	Scree	Alpine	Subnival
<b>Whole plant traits</b>								
Plant height (cm)	Height	39.7 (97)	31.09 (83)	36.61 (113)	22.64 (68)	29.21 (88)	23.57 (67)	6.93 (24)
Max. plant age (year)	Longevity	5.97 (47)	13.18 (101)	12.85 (70)	2.17 (8)	12.62 (46)	10.39 (70)	16.42 (50)
Radial growth rate (mm year <sup>-1</sup> )	Radial growth	0.27 (0.31)	0.25 (0.31)	0.24 (0.32)	0.22 (0.27)	0.24 (0.31)	0.23 (0.30)	0.19 (0.23)
<b>Organ traits</b>								
Leaf nitrogen content (%)	LNC	2.32 (4.86)	2.35 (3.68)	2.50 (5.01)	2.27 (4.09)	2.49 (5.63)	2.55 (7.28)	3.45 (6.82)
Leaf phosphorus content (%)	LPC	0.22 (0.73)	0.17 (0.24)	0.18 (0.42)	0.20 (0.58)	0.17 (0.32)	0.17 (0.34)	0.21 (0.33)
Leaf carbon content (%)	LCC	37.53 (45.1)	37.69 (46.7)	39.92 (47)	41.22 (46.8)	39.66 (46.9)	41.06 (48.1)	39.99 (47.9)
Leaf <sup>13</sup> C carbon isotope (‰)	Δ13C	-26.39	-23.82	-27.23	-27.55	-26.93	-27.00	-26.20
Leaf <sup>15</sup> N nitrogen isotope (‰)	Δ15N	5.42 (11.7)	4.77 (10.3)	3.15 (15.6)	3.95 (15.9)	1.40 (9.37)	1.03 (9.83)	2.68 (8.89)
Root nitrogen content (%)	RNC	1.01 (2.01)	0.97 (1.71)	1.06 (2.5)	1.05 (1.62)	1.08 (2.81)	1.09 (2.58)	1.81 (2.4)
Root phosphorus content (%)	RPC	0.14 (0.37)	0.11 (0.2)	0.11 (0.22)	0.14 (0.27)	0.12 (0.28)	0.13 (0.3)	0.18 (0.25)
Mechanical lignified tissue (%)	Lignified	42.0	38.0	29.1	30.1	24.9	31.0	8.9
Storage parenchymal tissue (%)	Parenchymatic	37.2	45.6	54.2	51.3	60.2	55.4	80.4
Conductive vessel tissue (%)	Conductive	20.8	16.4	16.6	18.6	14.8	13.7	10.7
Bark/xylem ratio	B/X ratio	0.48	0.60	0.66	1.56	0.84	0.88	0.86
<b>Growth forms</b>								
Monocarpic plants (%)	Monocarpic	63.64	27.78	12.09	25.00	5.88	10.96	10.53
Short epigeogenous rhizomes < 10 cm (%)	Srhizome	6.06	0.00	0.00	33.33	2.94	28.77	0.00
Long hypogeogenous rhizomes > 10 cm (%)	Lrhizome	3.03	0.00	5.49	16.67	4.41	9.59	0.00
Deep tap roots (%)	Taproot	6.06	44.44	59.34	4.17	63.24	30.14	15.79
Taproot with long belowground branches (%)	Pleocorm	18.18	11.11	13.19	12.50	17.65	10.96	36.84
Cushions with no adventitious roots (%)	Cushion	0.00	0.00	2.20	4.17	0.00	4.11	36.84
Woody plants (shrubs and subshrubs) (%)	Woody	3.03	16.67	7.69	4.17	5.88	5.48	0.00
<b>Species ecological information</b>								
Elevation optima (m a.s.l.)	Optima	3755	3631	4117	4146	4410	4486	5597
Elevation range (m)	Range	1667	1699	1657	1704	1624	1690	1554
Indicator value_Stability	Stability	1.85	1.72	1.91	2.04	1.74	2.11	1.68
Indicator value_Moisture	Moisture	1.70	1.00	1.37	2.83	1.74	2.07	2.00

species elevation optima using response curves fitted with Huisman–Olff–Fresco (HOF) models (Dvorský et al. 2017). Species response curves were derived from 4150 vegetation plots (each 100 × 100 m) sampled over the entire Ladakh between 1999 and 2014. The dataset contains > 122 000 records of the occurrence of vascular plant species along the exceptional elevational gradient from 2800 to 6150 m. Species' optima on the soil moisture and substrate stability gradients (ecological indicator values) were derived from the vegetation composition of 369 plots (each 100 m<sup>2</sup>) sampled in a stratified design to cover major vegetation types over the study area (Dvorský et al. 2011). The environmental variables were assessed in each plot using the following scales: stability of the soil/substrate surface (the inverse of disturbance frequency): 1) unstable (scree, dunes, solifluction soils), 2) partly stable (grasslands, steppes), 3) stable (rocky crevices, *Kobresia pygmaea* mats); soil moisture: 1) dry (substrate usually without visible traces of water), 2) mesic (with visible traces of water), 3) wet (water level regularly but transiently above soil surface), 4) permanent surface water (Dvorský et al. 2011, Klimešová et al. 2011, Doležal et al. 2016).

Each species were classified based on its spatial distribution into one of the seven main habitats: 1) ruderal vegetation around animal resting places, stables, and arable land near villages associated with 12% of the species studied, mostly monocarpic taxa; 2) semi-deserts at the lowest and driest elevations between 2800 and 4000 m (5% of species); 3) alpine steppes as the most common habitat type between 4000 and 5000 m (28% of species); 4) wetlands and salt marshes formed at the bottom of plains, around the shores of salt or brackish lakes and near mineral springs (8% of species); 5) alpine pastures occurring mainly along glacial streams and lakes on sandy muds well supplied with water, with the greatest species richness in Ladakh (23% of species); 6) alpine scree and boulder fields covering steep slopes with unstable substrate at higher elevations (19% of species); 7) subnival zone occupying the highest elevations up to the snow line (5% of species) (Dvorský et al. 2011, Doležal et al. 2018).

### Plant growth forms

To evaluate how interspecific differences in longevity relate to species plant growth forms, each species was classified into one of seven growth form categories based on woodiness, type of belowground organs (rhizomes versus primary tap-roots), connections between roots and shoots, rhizome length, adventitious roots formation, and canopy compactness (Klimeš and Klimešová 1999, Klimešová et al. 2011). These categories include 1) polycarpic non-clonal taprooted forbs predominating (41%) among studied herbaceous plants; 2) non-clonal taprooted herbs with underground branches connecting persistent taproot with aboveground shoots (so-called pleiocorm), accounted for 15% of the species; 3) clonal plants with short (<10 cm year<sup>-1</sup>) epigeogenous rhizomes or horizontal roots (10% among studied species), drawn into the soil by root contraction to serve as buds and

storage organs; 4) clonal plants with long hypogeogenous rhizomes or horizontal roots (6% among studied species), which spread > 10 cm year<sup>-1</sup> (Klimešová et al. 2011); 5) cushion plants (4% of the studied species); 6) woody shrubs (6% of species under study); 7) monocarpic plants (17% of studied species) (Table 1).

### Plant functional traits

To assess how interspecific differences in longevity relate to plant adaptive strategies, we measured a wide range of plant functional traits (Table 1, Fig. 2). We measured plant height as a proxy of plant stature and competitive ability. As a measure of the nutritional status of plants (Körner 2021), we measured carbon, nitrogen, and phosphorus concentrations in leaves (LCC, LNC and LPC), and nitrogen and phosphorus concentrations in roots (RNC and RPC). We measured stable carbon isotope signals ( $\delta^{13}\text{C}$ ) from leaves as a proxy for intrinsic water use efficiency (Farquhar et al. 1989, Liancourt et al. 2020). As a proxy of nutrient acquisition efficiency (Robinson 2001), we measured stable nitrogen isotope signals ( $\delta^{15}\text{N}$ ) from leaves (Yang et al. 2015). Leaf  $\delta^{15}\text{N}$  signals may reflect the dominant mode of nitrogen acquisition by plants, with a degree of symbiotic  $\text{N}_2$  fixation and mycorrhizal associations (e.g. ectomycorrhizal and arbuscular mycorrhizal species) that contribute most to the variation in the amount of  $\delta^{15}\text{N}$  (Craine et al. 2009). Phosphorus was determined after digestion in  $\text{HClO}_4$  using a spectrophotometer. The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ , as well as total carbon and nitrogen, were measured using an elemental analyzer coupled to an IRMS at the Stable Isotope Facility, UC Davis, USA.

### Species phylogeny

To assess the possible effect of phylogenetic relatedness on the relationships between longevity with various internal and external drivers, we reconstructed the phylogenetic tree (Fig. 3, 4) based on three molecular markers: matK, rbcL and ITS. Combined, these three loci cover protein-coding, RNA-coding, and noncoding sequences, as well as both plastid (matK, rbcL) and nuclear DNA (ITS). Variable mutational rates between loci maximize the ability to recognize major lineages along with species-level phylogenies. We acquired relevant sequences from NCBI GenBank for about one-third of the species. For the rest, we performed sequencing on material collected in the field. DNA was isolated from silica-gel dried leaf tissue using a Qiagen DNeasy Plant Mini Kit. For species for which molecular data were still missing (e.g. due to material degradation), relevant sequences of an unequivocal sister taxon were used instead. The L-INS-i algorithm implemented in the online version of MAFFT 6 (<http://mafft.cbrc.jp/alignment/server>) was employed to align the sequence datasets. The phylogenetic analysis itself was represented by the Bayesian inference, conducted in MrBayes ver. 3.1.2 (Ronquist and Huelsenbeck 2003). The tree inferred by MrBayes served as the basis for phylogenetic linear models in the subsequent analyses.

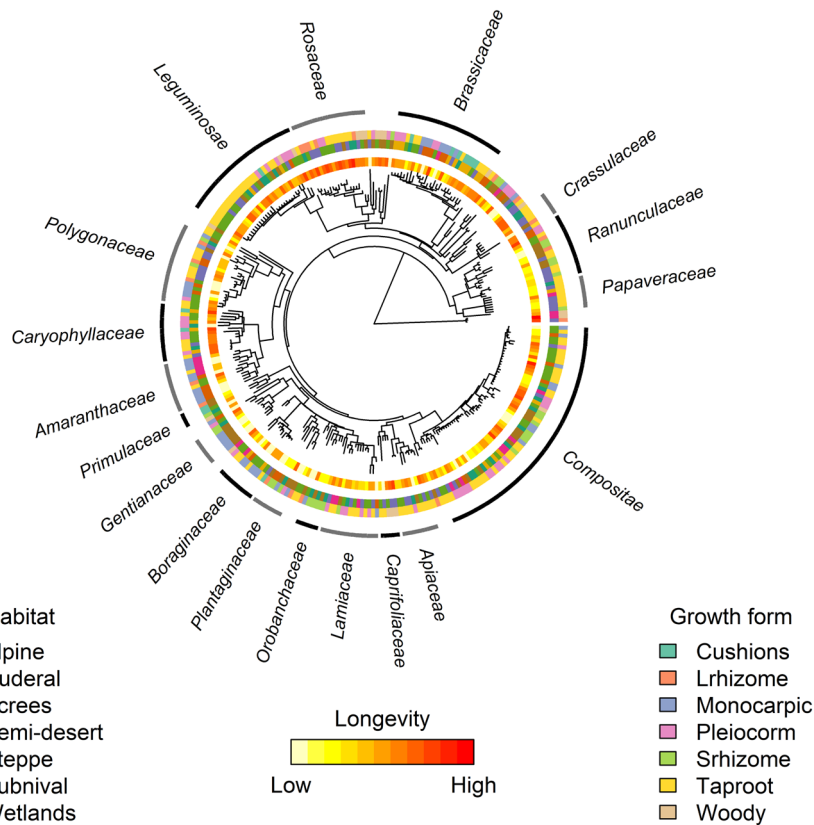


Figure 3. Phylogenetic tree with visualization of longevity, habitat and growth form. Longevity is shown in the inner circle, followed by habitat and growth form.

### Data analyses

To evaluate the effects of habitat preferences and plant traits on their longevity we used phylogenetic linear models. This model assumes that residual covariance is proportional to distance in the phylogenetic tree between respective species (Freckleton et al. 2002). Since we were interested in the value of habitat preferences, growth form, and functional traits for the prediction of plant longevity, we divided our predictors into three categories: 1) habitat preferences of plants (habitat type, moisture, stability, elevation optima, elevation range), 2) plant size and growth form (plant height and growth form) and 3) functional traits (the proportion of mechanical and storage tissue, LNC, LPC, LCC, RNC, RPC,  $\delta^{15}\text{N}$ ,  $\delta^{13}\text{C}$ , radial growth and bark-xylem ratio measured on the root collar) (Fig. 2). The proportion of conductive tissue was not included as a predictor since it is the remaining proportion after considering mechanical and storage tissues. From the original dataset of 355 species, we excluded 49 species due to missing values, resulting in 306 species used in all the analyses. We used model selection based on the Akaike information criterion (AIC) with longevity as a response variable and three categories of predictors sequentially. We selected predictors from the first category and used these as covariates for the second one, and finally added selected predictors from the second category and used these with selected predictors from the first category as covariates when selecting predictors

from the third category. By this sequential process, we determined which predictors from later categories improve the prediction of longevity. Model selection within each category of predictors was always done from a set of models with all possible combinations of predictors (including the null model without any predictors; e.g. all possible 2048 models for the third category of predictors were fitted). We explored not only the selected model but also models with similar performance to check the robustness of our results (models with  $\Delta\text{AIC} < 2$  are considered as having similar performance) and models without selected predictors to explore the importance of these predictors.

We transformed longevity, LNC, LPC, LCC, RNC, RPC, and radial growth using natural logarithm and plant height, bark-xylem ratio, and proportion of mechanical tissue using square root before the analyses to satisfy the model assumption of approx. normal distribution of residuals. The analyses were done in R ver. 4.1.1 ([www.r-project.org](http://www.r-project.org)) using package 'phylolm' (function *phylolm* ver. 2.6.2; Ho and Ané 2014) for phylogenetic linear models. We estimated the strength of the phylogenetic signal (Pagel's lambda; Pagel 1999 where  $\lambda = 0$  corresponds to no phylogenetic signal and  $\lambda = 1$  to the Brownian motion model of evolution) using maximum likelihood each time we fitted the model. To explore relationships among continuous predictors, we performed a phylogenetic principal component analysis (Supporting information).

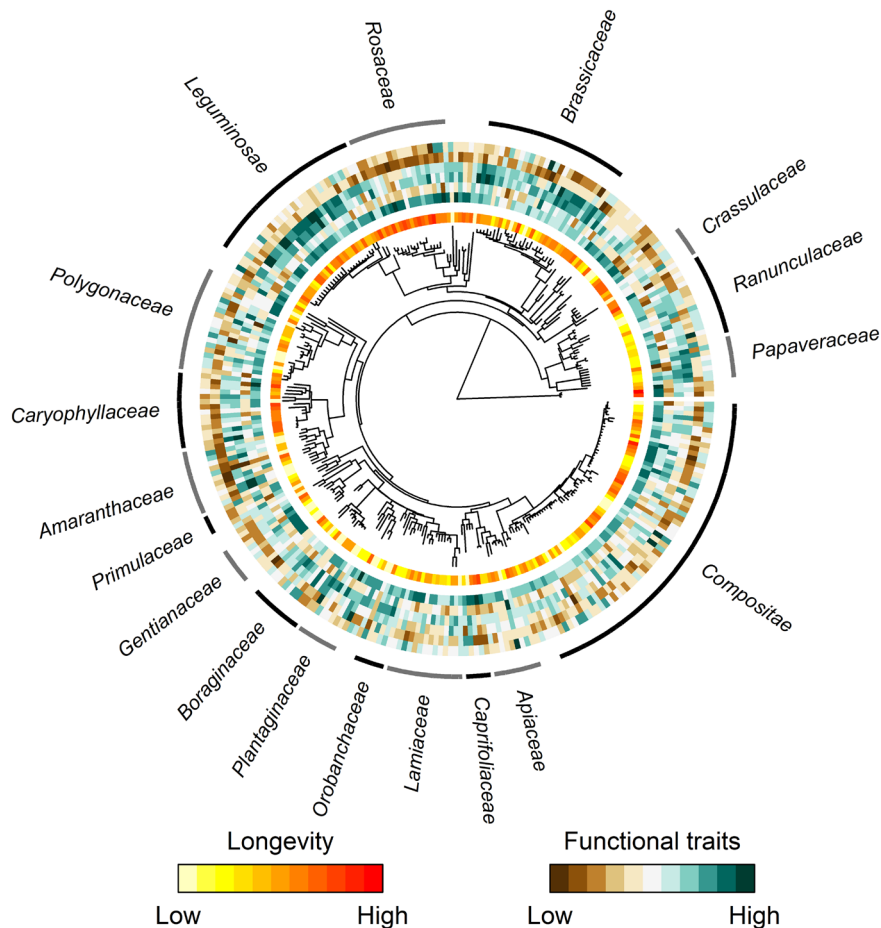


Figure 4. Phylogenetic tree with visualization of longevity and selected functional traits. The order of functional traits corresponds to high-lighted traits in Fig. 2. From inside: leaf carbon, leaf phosphorus, root phosphorus, root nitrogen, bark–xylem ratio, and radial growth.

## Results

### Habitat preferences

The longevity of our species was best predicted by each species' preferred habitat type. This model explained 12.1% variability in the data (adj- $R^2$ ; Fig. 2, 5, Supporting information). The best model without habitat type as a predictor had a much lower performance ( $\Delta AIC=22.8$ ). Plants with a lifespan of 10–15 years are most abundant in steppes and screes, while species reaching regularly 20–30 years old are found mainly in the high-elevation alpine (mean= 11.3 years, SD =13.24) and subnival (mean=15 years, SD=11.62 years) zones above 5200 m. Short-lived species account for 70% of species at the lowest elevation, primarily in ruderal vegetation (mean= 6.66 years, SD=10.18 years), and their percentage decreases with elevation, reaching 15 to 30% at 5500 m. The screes and subnival zone have the least short-lived plants, although alpine pastures can include up to 30% (a high number of Gentianaceae).

### Size and growth form

The growth form was selected as the only predictor that improved longevity prediction made by habitat type (which

was selected in the previous step). The resulting model explained 31.5% of the variability (adj- $R^2$ ) and the best model without growth form as a predictor had a much worse performance ( $\Delta AIC=69.5$ , Supporting information). High longevity was achieved by alpine cushion plants with taproots (mean=16.08 years, SD=14.30), clonal plants with longer rhizomes (mean= 12.20 years, SD=11.38), or forbs with pleiocorms (mean=14.52 years, SD=10.36) and woody shrubs (mean=24.63 years, SD=21.26 years) (Fig. 5, Supporting information). Among the oldest forbs are *Artemisia stracheyi* and *Potentilla pamirica* (up to 70 years) and *Arenaria bryophylla* (60 years) in the alpine area above 5200 m. The oldest plant individual at the highest place with angiosperms at an elevation of 6150 m was the 22-year-old endemic *Ladakiella klimesii*.

### Functional traits

By including functional traits, the prediction of longevity improved to 51.5% variability (adj- $R^2$ ) with selected predictors being leaf carbon content (LCC), leaf phosphorus content (LPC), root phosphorus content (RPC), root nitrogen content (RNC), bark–xylem ratio, and radial growth. The best model which did not have LCC ( $\Delta AIC=1.29$ ), LPC ( $\Delta AIC=1.41$ ), RPC ( $\Delta AIC=1.77$ ), had a very similar fit,



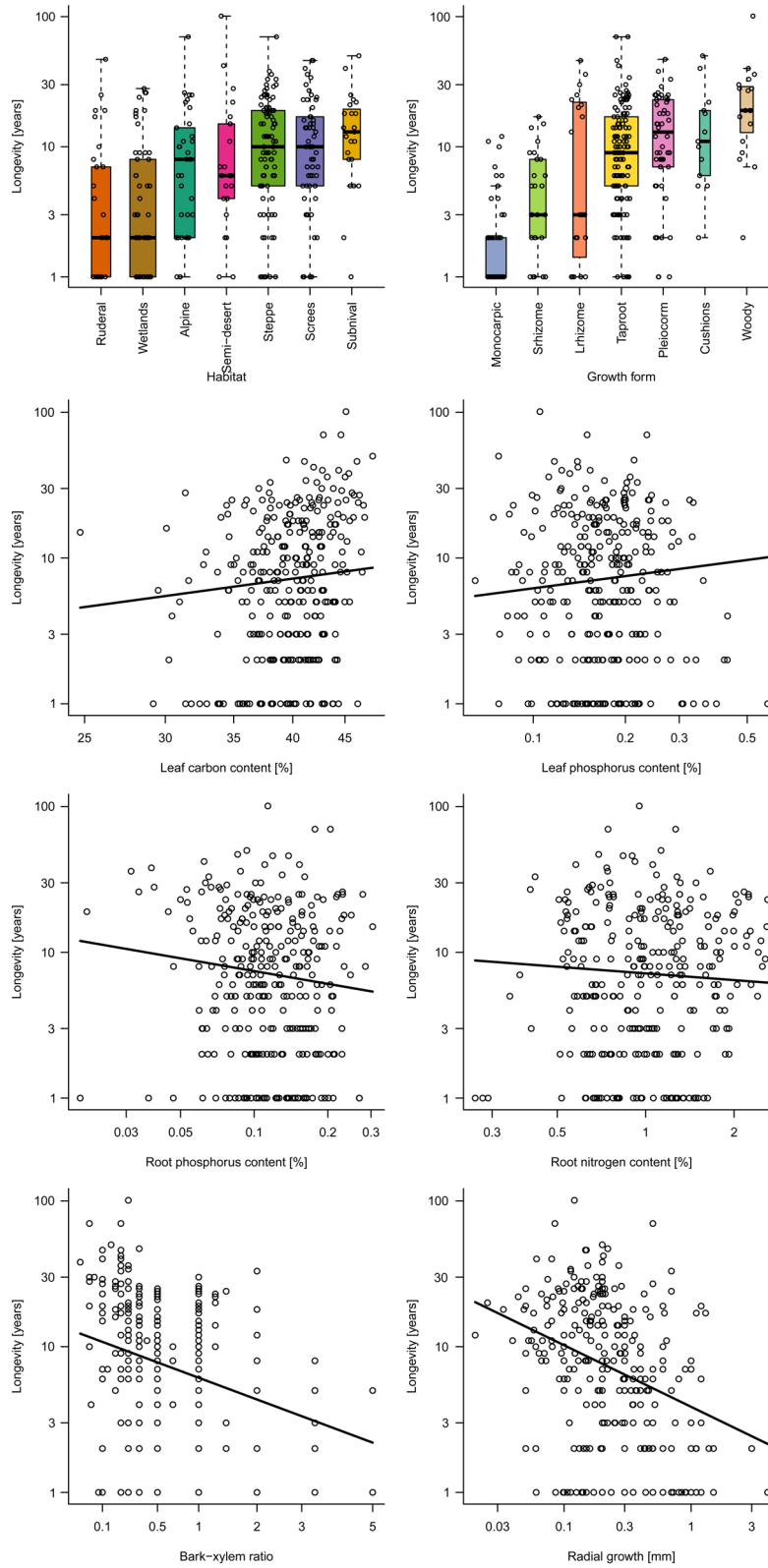


Figure 5. Relationships between selected predictors and longevity of plants. The width of boxplots corresponds to the square root of the number of observations in each category, the thick middle line is median, boxes range from first to third quartile, and whiskers up to  $3/2$  of the interquartile range from the box. Boxplots are ordered according to mean values. Relationships are estimates from phylogenetic linear regression.

while models without bark-xylem ratio ( $\Delta\text{AIC}=42.26$ ), and radial growth ( $\Delta\text{AIC}=47.19$ ) had much worse fit, marking bark-xylem ratio and radial growth as the main predictors among functional traits (model with only covariates from previous analyses and these two predictors ( $\Delta\text{AIC}=8.46$ , Supporting information). Longevity increased with LCC, and LPC while decreased with RPC, RNC, bark-xylem ratio, and radial growth (Fig. 5).

### Phylogenetic signal

Phylogenetic signal ( $\lambda$ ) in individual predictors ranged from very high (for  $\Delta^{13}\text{C}$  and bark-xylem ratio with  $\lambda$  0.91 and 0.84 respectively) to very low (for leaf phosphorus with  $\lambda$  0.00; Supporting information), suggests that traits with high  $\lambda$  values are strongly conserved among related taxa and that the evolutionary history of the plants is an important factor influencing the trait, while trait with low  $\lambda$  is not strongly conserved across related taxa and might reflect environmental conditions. The phylogenetic signal in the model using only the first category of predictors was 0.72, with the second category it was 0.65, and with the third it was 0.22 (Supporting information).

### Discussion

Our research demonstrates the combined influences of various factors on plant longevity among over 300 vascular dicot plant species in the western Himalayas across 2800–6150 m elevation gradient. The study shows that plant longevity is closely associated with species' habitat preferences. Species inhabiting ecologically stressful environments, such as alpine and subnival habitats, tend to be long-lived. On the other hand, habitats with higher productivity, such as ruderal and wetland areas, contain a larger proportion of short-lived species. The research also indicates that growth form is a significant factor in determining plant longevity. Small-stature cushion plants, characterized by compact canopies and deep roots, are more likely to achieve longevity. These plants are commonly found in cold and infertile alpine and subnival soils. Furthermore, plant traits that reflect adaptations to stress and disturbance play a role in interspecific differences in longevity. These traits likely enable plants to cope with challenging environmental conditions. It is worth noting that a negative correlation between longevity and growth rate is observed. Slow-growing plants have a higher likelihood of reaching high age. Finally, changes in carbon, nitrogen, and phosphorus content in root and leaf tissues are associated with variations in longevity. These nutrient dynamics are likely linked to the physiological functioning and resource allocation strategies of plants, which can affect their overall longevity. These findings provide valuable insights into the relationships between environmental factors, morphological traits, physiological dynamics, and anatomical constraints, and their combined influence on plant longevity. Understanding these relationships contributes to our

understanding of the stability, productivity, and resilience of Himalayan ecosystems under increasing environmental pressure.

### Habitats shaping plant longevity patterns

We discovered that interspecific differences in plant longevity were largely driven by species habitat preferences. Consistent with our first hypothesis, we found that long-lived species were more common in stable and unproductive habitats, such as alpine and subnival habitats, whereas short-lived species mainly occurred in productive and unstable habitats, such as ruderal sites and grazed wetlands. Hence, our study illustrates how longevity variation across habitats emerges locally from the complex and interconnected effects of climatic, edaphic, and disturbance factors (Lehmann and Parr 2016). Habitats with limited resources but stable over long periods, such as upper alpine and subnival zones promoted long-lived species with slow growth and resource-conservative strategies characterized by low tissue nutrient contents (Adler et al. 2014, Büntgen et al. 2015). Conversely, fertile environments with frequent or intense disturbances such as animal resting places selected for short-lived species with fast growth and resource-acquisitive strategies (Grime 2006, Herben et al. 2018).

Most of the species in lower elevation ruderal sites and wetlands (less harsh environmental conditions) are younger, with a few outliers, followed by semideserts, while the plants of the highest subnival zones have reached a much older age (Körner and Hiltbrunner 2021). This is consistent with our previous investigation of intraspecific variation in *P. pamirica* plants from the western Himalayas, in which the plants in the mesic alpine zone were relatively younger than subnival and steppe plants, however, there was no significant difference in maximum longevity between habitats (Doležal et al. 2021). Furthermore, there are more short-lived species at lower elevations, presumably due to higher soil nitrogen and the intensity of disturbances (Dvorský et al. 2011), related to pasturing activities (Schweingruber and Poschlod 2005), or possibly due to more active microbial activity and nutrient cycling, supporting the prevalence of short-lived monocarpic species (Řeháková et al. 2011).

### Longevity variation is related to growth form

Longevity in studied plants is also well related to their growth forms. Consistent with previous studies (Schweingruber and Poschlod 2005), we found that the non-clonal cushion plants are older than clonal or monocarpic plants (Supporting information), probably due to their diminutive nature of life form, which allows them to thrive in harsh conditions (Liancourt et al. 2020) and establish in low-fertility, dry substrates with little developed soils (Doležal et al. 2018). Cushion plants can reach very old age due to a slow expansion rate of only a few millimeters per year (McCarthy 1992), long-term space occupancy (Boucher et al. 2016), and specific factors related to plant nutrition and soil stability (Körner 2021).

Plants with taproots were generally older than clonal plants with rhizomes, with the tendency of taproots to persist in generally cold and dry soils (Klimešová et al. 2011). A possible limitation of this comparison is that clonal plants tend to discard older tissues and allocate resources in favor of new growth (Roeder et al. 2021). This would make the age of existing active (live) tissue in clonal plants younger than the potential age of vegetatively reproducing genet (age based on the time elapsed since zygote formation), which may be hundreds of years in clonal plants. Yet, some clonal plants at altitudes above 5000 m reached great age (retaining old living tissue for long periods), suggesting that investment in vegetative asexual reproduction in harsh conditions may promote longevity (Körner and Hiltbrunner 2021), while preferences for sexual reproduction observed in monocarpic plants reduces longevity (Adler et al. 2014, Salguero-Gómez et al. 2016). Clonal plants comprised about < 20% of the total species number. About 16–20% of plant species were clonal from the lowest elevations up to 5000 m a.s.l., from where the proportion of clonal plants gradually declined down to zero at the highest elevation (Supporting information). Short monocarpic plants are also increasingly rare at high altitudes above 5000 m due to low temperatures and short growing seasons preventing rapid growth and completion of the life cycle (Klimeš 2003), there are a few exceptions such as *Hedinia tibetica* which ascends to 5800 m where it is bound to nutrient-rich ruderal sites formed by colonies of the Himalayan pika *Ochotona himalayana*. Fewer woody shrubs were also older than herbs, which agrees with Ehlén and Lehtilä (2002) who found that woody plants had four times longer lifespans than non-woody plants (Supporting information).

### Longevity is linked to plant strategies

Plant longevity was largely driven by external factors reflected in species habitat preferences but internal factors such as slower growth (smaller mean annual radial increments) and specific leaf and root physiological parameters played also a significant role. This is consistent with previous studies documenting the plant's ability to achieve longevity through slow metabolism and growth (Thomas 2013, Büntgen et al. 2019), prolonged growth dormancy (Peñuelas and Munné-Bosch 2010), and limited nutrient uptake typically due to temperature and soil moisture limitation (Macek et al. 2012). Our results demonstrate that long-lived herbs grow in conditions where the short growing season and low temperatures (Doležal et al. 2016), or soil water and mineral nutrient deficiencies limit the meristematic growth processes (Nobis and Schweingruber 2013) and nutrient uptake (Rosbakh and Poschlod 2018), as is evident from the negative relationship between longevity, radial increments, and phosphorus content in the roots. Phosphorus is one of the most important macronutrients for plants that are acquired from the soil primarily as inorganic phosphate. Long-lived alpine and subnival forbs have a larger root allocation (Doležal et al. 2021), which is much higher at low phosphorus content, reflecting

low phosphorus levels in the soil (Heydari et al. 2019). Plants tend to store more nutrients essential for photosynthesis such as phosphorus in the active tissues like leaves and less in the supporting organs like roots (Körner 2021).

Slow radial growth was a defining feature of old age. Slow-growing plants are thought to gain a selective advantage over fast-growing plants by allocating resources from growth towards the production of non-structural carbohydrates, as suggested by Chlumská et al. (2022). This shift in resource allocation can lead to the synthesis of compounds that improve plant defenses against environmental stressors and increase plant resistance (Hartmann and Trumbore 2016), but at the expense of reduced growth (Wiley and Helliker 2012). On the other hand, fast-growing plants increase in size earlier but rarely reach the same age as slow-growing plants because the initial advantage of large size (i.e. increased availability of light and nutrients) and rapid growth and reproduction is often offset by reduced resistance to pests and pathogens and reduced investments in defense capability and biomechanical stability in an environment with a higher probability of random extreme stress and disturbance events such as summer frost, solifluction, etc. (Doležal et al. 2016).

The result of increasing longevity in plants with high LPC is probably to maximize light capture and photosynthetic activity (Körner 2021) while low RPC in extreme environmental conditions of subnival zones is consistent with low levels of available phosphorus, but also ammonium and nitrates in mineral soils between 5500 and 6000 m (Devetter et al. 2017). This supports the hypothesis of 'death by starvation' (Molisch 1938, Thomas 2003) which assumes that soil and plant tissue mineral nutrient deficiencies should prolong plant life span by slowing vegetative growth and reducing the production of sink tissues such as flowers and seeds (Rosbakh and Poschlod 2018). Our recent studies from the Himalayas (Doležal et al. 2021) support this notion because plants growing in a stressful environment tended to spend less on sexual reproduction and more on vegetative development to escape resource depletion.

The high LCC and root bark–xylem ratio supported plant longevity probably due to a large amount of plant tissue rich in carbon in older plants (Li et al. 2009, Chlumská et al. 2022). The high carbon content is usually found in long-lived leaves of perennial plants (Reich et al. 1992), while the large bark–xylem ratio is typical of massive taproots without internal tissue decomposition (Doležal et al. 2018). Long-persisting leaves and roots in long-lived plants are a significant source of carbon, which is utilized for a variety of metabolic functions such as respiration, development, and defense, as well as to withstand and survive stress or disturbances such as herbivory (Körner 2021). This is in line with Genet et al. (2010), who found that the relative carbon storage allocation increased with age in diffuse-porous species. Most of our perennial plants studied have diffuse-porous or semi-ring porous xylem (Doležal et al. 2018) with vessel conduits formed preferentially in earlywood, while latewood is made up of massive axial and radial bands of living parenchyma cells with high carbon storage capacity to protect plants from

frost or drought damage and thus secure long live. There is more information available about the allocation of carbon with respect to age in trees (Piispanen and Saranpää 2001, Barbaroux and Breda 2002, Genet et al. 2010), but there is a lack of studies linking nutrients with growth and longevity in herbaceous plants (Mooney and Billings 1960). According to a study by Dee et al. (2018), plant age is positively associated with the percentage of carbon in the root area. However, more research is required to determine the link between plant-tissue carbon concentration and longevity.

### Implications for persistence, biodiversity conservation, and future research potentials

Understanding interspecific variation in plant longevity is increasingly important for better understanding plant strategies and responses to ongoing environmental changes (Roeder et al. 2021). Plant age and longevity have mostly been studied on a limited set of species in selected plant communities (Dietz and von Arx 2005, von Arx et al. 2016, Rosbakh and Poschlod 2018), while some attempts have been made to understand variation in lifespan across environmental gradients both between (Nobis and Schweingruber 2013) and within species (Doležal et al. 2021). Fewer studies have attempted to understand the effect of various extrinsic and intrinsic factors (Rosbakh and Poschlod 2018). The current study is one of the first to take a new look at plant longevity by examining how different factors affect differences among several hundred taxa.

Maximum plant lifespan is an indicator of population persistence and as such may be more important for population survival than seed production or seedling establishment, especially in harsh environmental conditions that limit seedling regeneration more than adult plant survival (Graae et al. 2011, Salguero-Gómez et al. 2016). The importance of longevity for persistence and biodiversity conservation is becoming relevant with the recent accelerated warming in the mountains, which exposes cold-adapted plants to challenging conditions. Alpine plants inhabiting the highest elevations are considered potentially sensitive to climatic warming and endangered because of more intense competition from stronger plants migrating from lower elevations (Steinbauer et al. 2018). Although these negative trends may be mitigated by the enormous microhabitat heterogeneity of mountain ecosystems rich in thermal and edaphic refuges (Scherrer and Körner 2011), making alpine populations highly resistant to climatic change (Körner and Hiltbrunner 2021), the evolutionary adaptation of persistence and longevity may be another important but rarely studied maintenance mechanism in alpine plants. Certainly, further research can be conducted to investigate the effects of climate change on plant longevity in the Himalayas, as this region is particularly vulnerable to dynamic climate impacts. Additionally, the role of symbiotic associations with mycorrhizal fungi in contributing to plant longevity in this region warrants further study. Moreover, exploring the influence of anthropogenic factors, such as overgrazing and tourism, on plant longevity in the

Himalayas is also crucial. Understanding these factors and their impacts on the longevity of Himalayan plant species can help inform conservation efforts and strategies for mitigating the negative effects of climate change and human activities on the region's biodiversity.

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**Thinles Chondol:** Investigation (equal); Writing – original draft (lead); Writing – review and editing (equal). **Adam Klimeš:** Formal analysis (lead); Investigation (supporting); Writing – review and editing (equal). **Jan Altman:** Investigation (supporting); Writing – review and editing (supporting). **Kateřina Čapková:** Investigation (supporting). **Miroslav Dvorský:** Investigation (supporting). **Inga Hiiesalu:** Investigation (supporting). **Veronika Jandová:** Investigation (supporting). **Martin Kopecký:** Investigation (supporting). **Martin Macek:** Investigation (supporting). **Klára Reháková:** Investigation (supporting). **Pierre Liancourt:** Conceptualization (supporting); Investigation (supporting); Writing – review and editing (supporting). **Jiří Doležal:** Conceptualization (lead); Data curation (lead); Formal analysis (equal); Funding acquisition (lead); Investigation (lead); Methodology (lead); Project administration (lead); Writing – original draft (equal); Writing – review and editing (lead).

### Data availability statement

Data are available from the Dryad Digital Repository: <https://doi.org/10.5061/dryad.2bvq83bvx> (Chondol et al. 2023).

### Supporting information

The Supporting information associated with this article is available with the online version.

### References

- Adler, P. B., Salguero-Gómez, R., Compagnoni, A., Hsu, J. S., Ray-Mukherjee, J., Mbeau-Ache, C. and Franco, M.. 2014. Functional traits explain variation in plant life history strategies. – *Proc. Natl Acad. Sci. USA* 111: 740–745.
- Barbaroux, C. and Breda, N. 2002. Contrasting distribution and seasonal dynamics of carbohydrate reserves in stem wood of adult ring-porous sessile oak and diffuse-porous beech trees. – *Tree Physiol.* 22: 1201–1210.
- Boucher, F. C., Lavergne, S., Basile, M., Choler, P. and Aubert, S. 2016. Evolution and biogeography of the cushion life form in angiosperms. – *Perspect. Plant Ecol. Evol. Syst.* 20: 22–31.

- Büntgen, U., Hellmann, L., Tegel, W., Normand, S., Myers-Smith, I., Kirilyanov, A. V., Nievergelt, D. and Schweingruber, F. H. 2015. Temperature-induced recruitment pulses of Arctic dwarf shrub communities. – *J. Ecol.* 103: 489–501.
- Büntgen, U., Krusic, P. J., Piermattei, A., Coomes, D. A., Esper, J., Myglan, V. S., Kirilyanov, A. V., Camarero, J. J., Crivellaro, A. and Körner, C. 2019. Limited capacity of tree growth to mitigate the global greenhouse effect under predicted warming. – *Nat. Commun.* 10: 2171.
- Chlumská, Z., Liancourt, P., Hartmann, H., Bartoš, M., Altman, J., Dvorský, M., Hubáček, T., Borovec, J., Čapková, K., Kotlínek, M. and Doležal, J. 2022. Species- and compound-specific dynamics of nonstructural carbohydrates toward the world's upper distribution of vascular plants. – *Environ. Exp. Bot.* 201: 104985.
- Chondol, T., Klimeš, A., Altman, J., Čapková, K., Dvorský, M., Hiiesalu, I., Jandová, V., Kopecký, M., Macek, M., Řeháková, K., Liancourt, P. and Doležal, J. 2023. Data from: Habitat preferences and functional traits drive longevity in Himalayan high-mountain plants. – Dryad Digital Repository, <https://doi.org/10.5061/dryad.2bvq83bvz>.
- Craine, J. M., Elmore, A. J., Aidar, M. P. M., Bustamante, M., Dawson, T. E., Hobbie, E. A., Kahmen, A., Mack, M. C., McLaughlan, K. K., Michelsen, A., Nardoto, G. B., Pardo, L. H., Peñuelas, J., Reich, P. B., Schuur, E. A. G., Stock, W. D., Templer, P. H., Virginia, R. A., Welker, J. M. and Wright, I. J. 2009. Global patterns of foliar nitrogen isotopes and their relationships with climate, mycorrhizal fungi, foliar nutrient concentrations, and nitrogen availability. – *New Phytol.* 183: 980–992.
- de Pinto, M. C., Locato, V. and de Gara, L. 2012. Redox regulation in plant programmed cell death: redox regulation in plant PCD. – *Plant Cell Environ.* 35: 234–244.
- de Witte, L. C. and Stöcklin, J. 2010. Longevity of clonal plants: why it matters and how to measure it. – *Ann. Bot.* 106: 859–870.
- de Witte, L. C., Armbruster, G. F. J., Gielly, L., Taberlet, P. and Stöcklin, J. 2012. Aflp markers reveal high clonal diversity and extreme longevity in four key arctic-alpine species: longevity of arctic-alpine clonal plants. – *Mol. Ecol.* 21: 1081–1097.
- Dee, J. R., Adams, H. D. and Palmer, M. W. 2018. Belowground annual ring growth coordinates with aboveground phenology and timing of carbon storage in two tallgrass prairie forb species. – *Am. J. Bot.* 105: 1975–1985.
- Devetter, M., Háněl, L., Řeháková, K. and Doležal, J. 2017. Diversity and feeding strategies of soil microfauna along elevation gradients in Himalayan cold deserts. – *PLoS One* 12: e0187646.
- DeWoody, J., Rowe, C. A., Hipkins, V. D. and Mock, K. E. 2008. “Pando” lives: molecular genetic evidence of a giant aspen clone in central Utah. – *West. N. Am. Nat.* 68: 493–497.
- Díaz, S. et al. 2016. The global spectrum of plant form and function. – *Nature* 529: 167–171.
- Dietz, H. and von Arx, G. 2005. Climatic fluctuation causes large-scale synchronous variation in radial root increments of perennial forbs. – *Ecology* 86: 327–333.
- Doležal, J., Dvorsky, M., Kopecky, M., Liancourt, P., Hiiesalu, I., Macek, M., Altman, J., Chlumská, Z., Rehakova, K., Capkova, K., Borovec, J., Mudrak, O., Wild, J. and Schweingruber, F. 2016. Vegetation dynamics at the upper elevational limit of vascular plants in Himalaya. – *Sci. Rep.* 6: 24881.
- Doležal, J., Dvorský, M., Börner, A., Wild, J. and Schweingruber, F. H. 2018. Anatomy, age and ecology of high mountain plants in Ladakh, the Western Himalaya. – Springer.
- Doležal, J., Jandova, V., Macek, M. and Liancourt, P. 2021. Contrasting biomass allocation responses across ontogeny and stress gradients reveal plant adaptations to drought and cold. – *Funct. Ecol.* 35: 32–42.
- Doležal, J., Jandova, V., Macek, M., Mudrak, O., Altman, J., Schweingruber, F. H. and Liancourt, P. 2021. Climate warming drives Himalayan alpine plant growth and recruitment dynamics. – *J. Ecol.* 109: 179–190.
- Dvorský, M., Altman, J., Kopecký, M., Chlumská, Z., Řeháková, K., Janatková, K. and Doležal, J. 2015. Vascular plants at extreme elevations in eastern Ladakh, northwest Himalayas. – *Plant Ecol. Divers.* 8: 571–584.
- Dvorský, M., Doležal, J., de Bello, F., Klimešová, J. and Klimeš, L. 2011. Vegetation types of East Ladakh: species and growth form composition along main environmental gradients: vegetation of east Ladakh. – *Appl. Veg. Sci.* 14: 132–147.
- Dvorský, M., Macek, M., Kopecký, M., Wild, J. and Doležal, J. 2017. Niche asymmetry of vascular plants increases with elevation. – *J. Biogeogr.* 44: 1418–1425.
- Ehrlén, J. and Lehtilä, K. 2002. How perennial are perennial plants? – *Oikos* 98: 308–322.
- Farquhar, G. D., Ehleringer, J. R. and Hubick, K. T. 1989. Carbon isotope discrimination and photosynthesis. – *Annu. Rev. Plant Biol.* 40: 503–537.
- Freckleton, R. P., Harvey, P. H. and Pagel, M. 2002. Phylogenetic analysis and comparative data: a test and review of evidence. – *Am. Nat.* 160: 712–726.
- Gärtner, H. and Schweingruber, F. H. 2013. Microscopic preparation techniques for plant stem analysis. – Originalausg. Kessel, Remagen-Oberwinter.
- Genet, H., Breda, N. and Dufrene, E. 2010. Age-related variation in carbon allocation at tree and stand scales in beech (*Fagus sylvatica* L.) and sessile oak (*Quercus petraea* (Matt.) Liebl.) using a chronosequence approach. – *Tree Physiol.* 30: 177–192.
- Graae, B. J., Ejrnæs, R., Lang, S. I., Meineri, E., Ibarra, P. T. and Bruun, H. H. 2011. Strong microsite control of seedling recruitment in tundra. – *Oecologia* 166: 565–576.
- Grime, J. P. 1974. Vegetation classification by reference to strategies. – *Nature* 250: 26–31.
- Grime, P. J. 2006. Plant strategies, vegetation processes, and ecosystem properties, 2nd edn. – Wiley.
- Hartmann, H. and Trumbore, S. 2016. Understanding the roles of nonstructural carbohydrates in forest trees – from what we can measure to what we want to know. – *New Phytol.* 211: 386–403.
- Herben, T., Klimešová, J. and Chytrý, M. 2018. Effects of disturbance frequency and severity on plant traits: an assessment across a temperate flora. – *Funct. Ecol.* 32: 799–808.
- Heydari, M. M., Brook, R. M. and Jones, D. L. 2019. The role of phosphorus sources on root diameter, root length and root dry matter of barley (*Hordeum vulgare* L.). – *J. Plant Nutr.* 42: 1–15.
- Ho, L. S. T. and Ané, C. 2014. A linear-time algorithm for gaussian and non-gaussian trait evolution models. – *Syst. Biol.* 63: 397–408.
- Kipkeev, A. M., Onipchenko, V. G., Tekeev, D. K., Erkenova, M. A. and Salpagarova, F. S. 2015. Age of maturity in alpine herbaceous perennials in the northwest Caucasus. – *Biol. Bull. Rev.* 5: 505–511.
- Kirkwood, T. B. L. 1977. Evolution of ageing. – *Nature* 270: 301–304.
- Kirkwood, T. B. L. and Austad, S. N. 2000. Why do we age?. – *Nature* 408: 233–238.
- Klimeš, L. 2003. Life-forms and clonality of vascular plants along an altitudinal gradient in E Ladakh (NW Himalayas). – *Basic Appl. Ecol.* 4: 317–328.

- Klimeš, L. and Klimešová, J. 1999. CLO-PLA2 – A database of clonal plants in central Europe. – *Plant Ecol.* 141: 9–19.
- Klimešová, J., Doležal, J., Dvorský, M., de Bello, F. and Klimeš, L. 2011. Clonal growth forms in Eastern Ladakh, Western Himalayas: classification and habitat preferences. – *Folia Geobot.* 46: 191–217.
- Klimešová, J., Martínková, J., Pausas, J. G., de Moraes, M. G., Herben, T., Yu, F.-H., Puntieri, J., Vesk, P. A., de Bello, F., Janeček, Š., Altman, J., Appezzato-da-Glória, B., Bartušková, A., Crivellaro, A., Doležal, J., Ott, J. P., Paula, S., Schnablová, R., Schweingruber, F. H. and Ottaviani, G. 2019. Handbook of standardized protocols for collecting plant modularity traits. – *Perspect. Plant Ecol. Evol. Syst.* 40: 125485.
- Körner, C. 2021. Alpine plant life: functional plant ecology of high mountain ecosystems. – Springer International Publishing.
- Körner, C. and Hiltbrunner, E. 2021. Why is the alpine flora comparatively robust against climatic warming? – *Diversity* 13: 383.
- Larson, D. W. 2001. The paradox of great longevity in a short-lived tree species. – *Exp. Gerontol.* 36: 651–673.
- Lehmann, C. E. R. and Parr, C. L. 2016. Tropical grassy biomes: linking ecology, human use and conservation. – *Phil. Trans. R. Soc. B* 371: 20160329.
- Li, C., Wu, C., Duan, B., Korpelainen, H. and Luukkanen, O. 2009. Age-related nutrient content and carbon isotope composition in the leaves and branches of *Quercus aquifolioides* along an altitudinal gradient. – *Trees* 23: 1109–1121.
- Liancourt, P., Song, X., Macek, M., Santrucek, J. and Doležal, J. 2020. Plant's-eye view of temperature governs elevational distributions. – *Global Change Biol.* 26: 4094–4103.
- Lütz, C. (ed.). 2012. Plants in alpine regions: cell physiology of adaption and survival strategies. – Springer.
- Macek, M., Dvorský, M., Klimeš, A., Wild, J., Doležal, J. and Kopecký, M. 2021. Midpoint attractor models resolve the mid-elevation peak in Himalayan plant species richness. – *Ecography* 44: 1665–1677.
- Macek, P., Klimeš, L., Adamec, L., Doležal, J., Chlumská, Z., de Bello, F., Dvorský, M. and Řeháková, K. 2012. Plant nutrient content does not simply increase with elevation under the extreme environmental conditions of Ladakh, NW Himalaya. – *Arct. Antarct. Alp. Res.* 44: 62–66.
- McCarthy, D. P. 1992. Dating with cushion plants: establishment of a silene acaulis growth curve in the Canadian rockies. – *Arct. Alp. Res.* 24: 50.
- Molisch, H. 1938. The longevity of plants. – Published by the Translator, Science Press Printing Co.
- Mooney, H. A. and Billings, W. D. 1960. The annual carbohydrate cycle of alpine plants as related to growth. – *Am. J. Bot.* 47: 594–598.
- Morris, W. F., Pfister, C. A., Tuljapurkar, S., Haridas, C. V., Boggs, C. L., Boyce, M. S., Bruna, E. M., Church, D. R., Coulson, T., Doak, D. F., Forsyth, S., Gaillard, J.-M., Horvitz, C. C., Kalisz, S., Kendall, B. E., Knight, T. M., Lee, C. T. and Menges, E. S. 2008. Longevity can buffer plant and animal populations against changing climatic variability. – *Ecology* 89: 19–25.
- Munné-Bosch, S. 2008. Do perennials really senesce? – *Trends Plant Sci.* 13: 216–220.
- Munné-Bosch, S. 2018. Limits to tree growth and longevity. – *Trends Plant Sci.* 23: 985–993.
- Nobis, M. P. and Schweingruber, F. H. 2013. Adult age of vascular plant species along an elevational land-use and climate gradient. – *Ecography* 36: 1076–1085.
- Obeso, J. R. 2002. The costs of reproduction in plants. – *New Phytol.* 155: 321–348.
- Pagel, M. 1999. Inferring the historical patterns of biological evolution. – *Nature* 401: 877–884.
- Peñuelas, J. and Munné-Bosch, S. 2010. Potentially immortal? – *New Phytol.* 187: 564–567.
- Piispänen, R. and Saranpää, P. 2001. Variation of non-structural carbohydrates in silver birch (*Betula pendula* Roth) wood. – *Trees* 15: 444–451.
- Řeháková, K., Chlumská, Z. and Doležal, J. 2011. Soil cyanobacterial and microalgal diversity in dry mountains of Ladakh, NW Himalaya, as related to site, altitude, and vegetation. – *Microb. Ecol.* 62: 337–346.
- Reich, P. B., Walters, M. B. and Ellsworth, D. S. 1992. Leaf lifespan in relation to leaf, plant, and stand characteristics among diverse ecosystems. – *Ecol. Monogr.* 62: 365–392.
- Robinson, D. 2001.  $\delta^{15}\text{N}$  as an integrator of the nitrogen cycle. – *Trends Ecol. Evol.* 16: 153–162.
- Roeder, A., Schweingruber, F. H., Ebeling, A., Eisenhauer, N., Fischer, M. and Roscher, C. 2021. Plant diversity effects on plant longevity and their relationships to population stability in experimental grasslands. – *J. Ecol.* 109: 2566–2579.
- Ronquist, F. and Huelsenbeck, J. P. 2003. MrBayes 3: Bayesian phylogenetic inference under mixed models. – *Bioinformatics* 19: 1572–1574.
- Rosbakh, S. and Poschlod, P. 2018. Killing me slowly: harsh environment extends plant maximum life span. – *Basic Appl. Ecol.* 28: 17–26.
- Salguero-Gómez, R., Jones, O. R., Jongejans, E., Blomberg, S. P., Hodgson, D. J., Mbeau-Ache, C., Zuidema, P. A., de Kroon, H. and Buckley, Y. M. 2016. Fast–slow continuum and reproductive strategies structure plant life-history variation worldwide. – *Proc. Natl Acad. Sci. USA* 113: 230–235.
- Scherrer, D. and Körner, C. 2011. Topographically controlled thermal-habitat differentiation buffers alpine plant diversity against climate warming: topographical control of thermal-habitat differentiation buffers alpine plant diversity. – *J. Biogeogr.* 38: 406–416.
- Schweingruber, F. H., Börner, A. and Schulze, E.-D. 2013. Anatomical adaptations to environmental conditions. – In: Atlas of stem anatomy in herbs, shrubs and trees. Springer, pp. 355–370.
- Schweingruber, F. H., Kučerová, A., Adamec, L. and Doležal, J. 2020. Anatomic atlas of aquatic and wetland plant stems. – Springer Nature Switzerland AG.
- Schweingruber, F. H. and Wirth, C. 2009. Old trees and the meaning of 'old.' – In: Wirth, C., Gleixner, G. and Heimann, M. (eds), Old-growth forests. Springer, pp. 35–54.
- Schweingruber, F. H. and Poschlod, P. 2005. Growth rings in herbs and shrubs: life span, age determination and stem anatomy. – *For. Snow Lands. Res.* 79:195–415.
- Silvertown, J. 2013. The long and the short of it: the science of life span and aging. – Univ. of Chicago Press.
- Steinbauer, M. J. et al. 2018. Accelerated increase in plant species richness on mountain summits is linked to warming. – *Nature* 556: 231–234.
- Thomas, H. 2003. Defining senescence and death. – *J. Exp. Bot.* 54: 1127–1132.
- Thomas, H. 2013. Senescence, ageing and death of the whole plant. – *New Phytol.* 197: 696–711.

- Violle, C., Navas, M.-L., Vile, D., Kazakou, E., Fortunel, C., Hummel, I. and Garnier, E. 2007. Let the concept of trait be functional! – *Oikos* 116: 882–892.
- von Arx, G., Crivellaro, A., Prendin, A. L., Čufar, K. and Carrer, M. 2016. Quantitative wood anatomy – practical guidelines. – *Front. Plant Sci.* 7: 781.
- Wiley, E. and Helliker, B. 2012. A re-evaluation of carbon storage in trees lends greater support for carbon limitation to growth. – *New Phytol.* 195: 285–289.
- Wright, I. J. et al. 2004. The worldwide leaf economics spectrum. – *Nature* 428: 821–827.
- Yang, Y., Siegwolf, R. T. W. and Korner, C.. 2015. Species specific and environment induced variation of  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  in alpine plants. – *Front. Plant Sci.* 6: 423.
- Zheng, X., Chen, J., Sun, J. and Ren, R. 2022. Association plant leaf longevity to the increasing warm and arid environment. – *IOP Conf. Ser. Earth Environ. Sci.* 983: 012084.