Evolutionary origins of a latitudinal plant height gradient

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Abstract

Latitudinal gradients are among the most pervasive patterns in biodiversity, yet their evolutionary origins remain poorly understood, especially in plants. Here we identify a latitudinal gradient in plant height within cacti, an iconic and rapidly evolving plant family spanning the Americas, with taller species inhabiting warmer, lower latitudes. To uncover how this gradient arose, we reconstructed repeated directional changes towards taller and shorter descendants across the cactus phylogeny, distinguishing these from shifts in evolutionary variance and growth-form differences. We combined continuous-space reconstructions of ancestral geographical dispersal with palaeoclimate to recover the warming and cooling experienced by species throughout their diversification. We find that species that dispersed through warmer places increased in height, whereas those that dispersed through colder places shrank. These opposing associations reveal how historical climate change was linked to directional height evolution, providing an evolutionary explanation for the emergence of the latitudinal height gradient. By reconstructing complex phenotypic, geographical and climatic histories together in high resolution, we trace the origins of an extant latitudinal gradient through the evolutionary changes from which it emerged. Our framework provides a method to reveal the evolutionary origins of geographical gradients and trait-environment relationships across the Tree of Life.

Introduction

Latitudinal gradients are among the most pervasive patterns describing the distribution of life on Earth [1–5]. In mammals and birds, body size often increases

34 towards colder, higher latitudes, a pattern known as Bergmann's rule [6,7]. However, in
35 plants the opposite pattern is found, with mean plant height declining almost 30-fold
36 from the equator to the poles [8]. Several ecological and evolutionary explanations
37 may account for this gradient, but their relative contribution remains poorly
38 understood. This is because, unlike contemporary hypotheses, historical
39 explanations require reconstructing how height evolved as well as the environmental
40 changes experienced by dispersing ancestors [9]. Models of trait evolution and
41 historical biogeography are well established. However, many make assumptions that
42 limit our ability to distinguish different processes underlying trait evolution or
43 reconstruct how ancestral lineages moved through space [10–14]. Recent advances
44 now allow directional evolution to be distinguished from changes in evolutionary rate
45 (evolutionary variance) and trait-dependent effects [15,16], such as different growth
46 forms, and to estimate ancestral dispersal through continuous geographical space
47 and palaeoclimates [17–21]. Combining these trait-evolution and biogeographic
48 approaches offers a unique opportunity to uncover the evolutionary origins of the
49 inverse latitudinal plant height gradient.
50
51 Turning to the hypotheses themselves, may reflect geographical turnover among
52 growth forms, particularly the greater prevalence of trees towards the tropics [8,22,23],
53 although the gradient also occurs within growth forms and can differ in strength or
54 even reverse among lineages [8,24–27]. Secondly, precipitation and temperature
55 constraints on plant height through water availability, hydraulic limitations and
56 growing conditions [8,22,28–33]. Indeed, global variation in plant height is strongly
57 associated with contemporary climate, particularly precipitation but also temperature
58 [834–38]. Finally, the gradient may reflect the cumulative evolutionary consequences of
59 historical environmental change, as ancestors evolved into taller or shorter
60 descendants while dispersing through changing climates throughout their
61 evolutionary history.
62
63 The cactus family (Cactaceae) is a relatively young, species-rich radiation with
64 exceptional variation in height and growth form [38–45]. Since originating around 30
65 million years ago, probably in the Andes, the ~1,850 cactus species have spread
66 across much of the Americas, from southern Canada to Patagonia [43,46]. They have
67 adapted to environments ranging from tropical forests and hyper-arid deserts to high-

elevation grasslands by evolving exceptional ecomorphological diversity [42–44,47–50]. During a relatively short timeframe, cacti span more than three orders of magnitude in height, from tiny plants only centimetres tall (e.g. *Blossfeldia liliputana*, ~1.2 cm) to towering columnars approaching 20 m (e.g. *Brasiliopuntia brasiliensis*) [42], and this is latitudinally structured, with smaller cacti found further from the equator (Figs. 1 and 2). Alongside this, cacti have evolved striking diversity in growth forms, flowers, and pollination systems [42–44,47,48]. This combination of exceptional height variation, an 80-degree latitudinal distribution and latitudinal height gradient allows us to ask how this size gradient evolved through time.

Reconstructing the evolutionary origins of a latitudinal height gradient requires distinguishing the different processes through which height variation arises. Along evolutionary lineages, height can change through (1) directional evolution towards taller or shorter descendants [15,51,52], (2) changes in evolutionary variance that generate more or less variation in height among descendants [52–55], and (3) differences associated with transitions between growth forms, such as from small globose plants to larger shrubby forms [44,56,57]. These processes can be difficult to distinguish [16], because a transition between growth forms may produce height differences that resemble directional evolutionary changes. Disentangling them allows us to investigate whether historical environmental change was associated specifically with directional height evolution. If historical temperature change contributed to the emergence of the latitudinal height gradient, we predict that lineages experiencing greater warming accumulated greater directional increases in height, whereas those experiencing greater cooling accumulated greater directional decreases. Testing this prediction requires reconstructing the changing environments experienced by ancestral lineages rather than relying on the climates occupied by species today.

As cacti dispersed across the Americas, ancestral lineages encountered different environments while climates within individual regions also changed through time. The temperature changes experienced by a lineage therefore depended on both its geographical movements and the changing climates of the regions it occupied. Recent advances in Bayesian continuous-space biogeographical models allow ancestral geographical locations to be reconstructed through deep time, while

accounting for variation in dispersal rates among lineages [17–19]. Combining these geographical reconstructions [21] with spatially and temporally explicit palaeoclimate models [58,59] allows us to estimate the cumulative warming and cooling experienced along each lineage's evolutionary history. These reconstructed climatic histories can then be associated with directional changes in height, allowing us to investigate how historical environmental change relates to the evolution of the latitudinal height gradient.

Here we examine the evolutionary origins of a latitudinal gradient in cactus height. We first establish the gradient and test for its associations with growth-forms, contemporary precipitation and temperature. We then reconstruct directional changes in height and changes in evolutionary variance across the cactus phylogeny, while accounting for growth-form differences. By combining these evolutionary reconstructions with ancestral geographical movements and palaeoclimate, we test whether historical warming and cooling are associated with directional height evolution.

Results and discussion

A latitudinal gradient in cactus height

Cactus height is strongly phylogenetically structured (Pagel's $\lambda = 0.943$, likelihood-ratio test, $P = 1.65 \times 10^{-259}$), indicating that closely related species tend to be similar in height [60,61] (Figure 3). Nevertheless, substantial height variation occurs across the cactus family. Among subfamilies, mean height ranged from 23 cm in Maihuenioideae to 627 cm among the early-diverging leafy cacti (*Leuenbergeria* and *Pereskia*), while Cactoideae alone spanned almost the entire family-wide range, from 1.2 to 1,800 cm. In smaller taxonomic groups, mean height ranged from 5 cm in tribe Fraileeae to 729 cm in *Leuenbergeria*. Thus, substantial variation in height exists alongside strong phylogenetic signal.

Growth form is an important contributor to global variation in plant height, including latitudinal height gradients [8]. In cacti, eight major growth forms are recognised in CactEcoDB across the 922 sampled species [42], of which 683 (74.1%) are assigned to a single growth-form category and the remainder are polymorphic. Globose solitary is the most common growth form (344 species), followed by globose

caespitose (234), arborescent (127), epiphytic (73), columnar (41), shrubby, cushion-forming (25) and geophytic (14). We found that growth-form variation explains 37.4% of the variation in cactus height (median Bayesian $R^2 = 0.374$ across six independent MCMC runs; Fig. 1). Of the 28 pairwise comparisons among growth forms, 15 showed statistically supported differences. Arborescent and columnar species were generally taller than the shorter species, which were predominantly globose, geophytic and shrubby forms, while epiphytic species were intermediate. The remaining 13 pairwise comparisons were not statistically significant, including comparisons among several taller growth forms (e.g. arborescent and columnar, and columnar and epiphytic; Fig. 1). Growth form is therefore an important contributor to differences in cactus height, although substantial variation remains within growth forms.

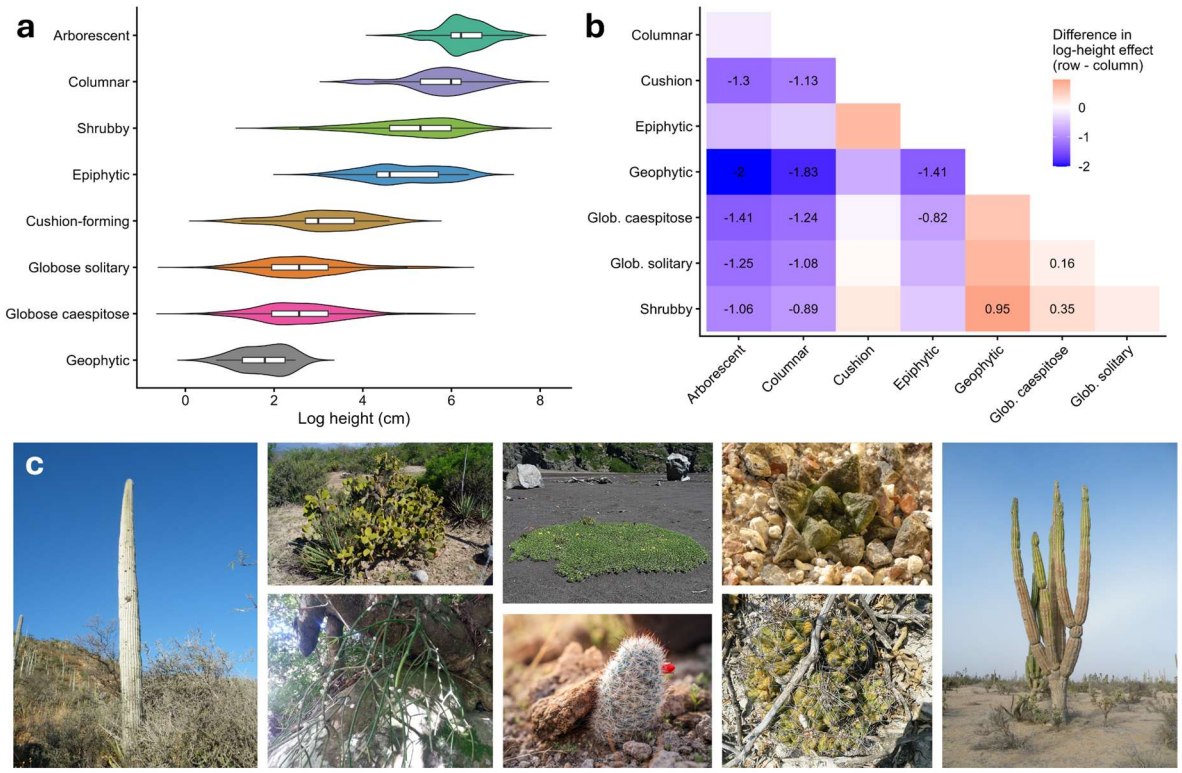


Figure 1: Cactus height varies strongly among growth forms. *(a) Distribution of cactus height (log cm) between eight cactus growth forms. Violin plots show the distribution of species heights and boxplots show the median and interquartile range. (b) Pairwise differences in height among growth forms estimated in a Bayesian phylogenetic regression. Cells show the posterior mean difference in log height between the growth form in each column and row (row − column). Numerical values*

are shown for pairwise comparisons which are significantly different. Cell colour indicates the magnitude and direction of the estimated difference. (c) Representative cactus species illustrating different growth forms. Images are arranged clockwise from the large left-hand image: Cephalocereus columna-trajani *(columnar),* Opuntia microdasys *(shrubby),* Maihuenia poeppigii *(cushion),* Ariocarpus fissuratus *(geophytic), and* Pachycereus pringlei *(arborescent). The bottom row continues from right to left with* Thelocactus leucacanthus *(globose caespitose),* Mammillaria tetrancistra *(globose solitary), and* Rhipsalis baccifera *(epiphytic). Images are photographs available under Creative Commons licenses or in the public domain. The photographs of* Cephalocereus columna-trajani*,* Pachycereus pringlei*, and* Thelocactus leucacanthus *were taken by Amante Darmanin and are licensed under a Creative Commons Attribution 2.0 License (https://creativecommons.org/licenses/by/2.0/). The photograph of* Opuntia microdasys *was taken by Sergio Niebla, and that of* Ariocarpus fissuratus *by Dr. Hans-Günter Wagner; both are licensed under a Creative Commons Attribution-ShareAlike 2.0 License (https://creativecommons.org/licenses/by-sa/2.0/). The photograph of* Maihuenia poeppigii *was taken by Krzysztof Ziarnek (Kenraiz) and is licensed under a Creative Commons Attribution 4.0 International License (https://creativecommons.org/licenses/by/4.0/). The photograph of* Mammillaria tetrancistra *was taken by Jesse Pluim (BLM) and is marked as being in the public domain using the Public Domain Mark 1.0 (https://creativecommons.org/publicdomain/mark/1.0/). The photograph of* Rhipsalis baccifera *was taken by Maria Vorontsova and released under the Creative Commons CC0 1.0 Universal Public Domain Dedication (https://creativecommons.org/publicdomain/zero/1.0/).*

Given the importance of growth-form turnover in explaining global latitudinal height gradients [8], we accounted for growth form when examining geographical variation in cactus height. We identified a negative latitudinal gradient in cactus height, with taller species occurring closer to the equator. This relationship was supported by a Bayesian phylogenetic model accounting for differences among growth forms (posterior mean $\beta = -0.018$, 95% credible interval [CrI] = −0.028 to −0.007, pMCMC = 0.004; median marginal $R^2 = 0.17$, median conditional $R^2 = 0.91$; Fig. 2) [62]. The 10% of cactus species closest to the equator were 7.3-fold taller on average than

those in the 10% furthest from the equator (259 versus 35.3 cm, respectively). A model excluding growth form resulted in nearly identical latitudinal association (β = −0.019, 95% CrI = −0.032 to −0.007, pMCMC = 0.002; median marginal R^2 = 0.004, median conditional R^2 = 0.90). The similarity of these relationships indicates that differences in growth-form composition alone do not account for the latitudinal gradient in cactus height.

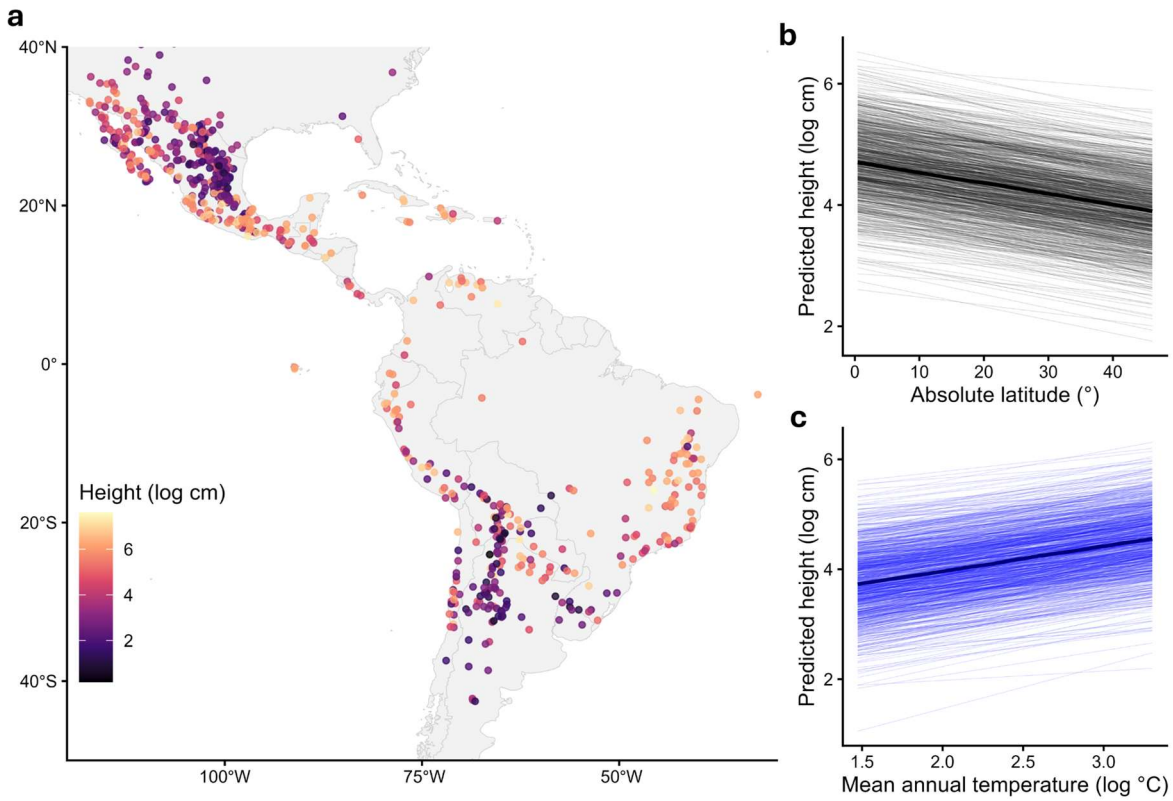


Figure 2: A latitudinal gradient in cactus height across the Americas. *(a) Geographical centroid of the 753 cactus species with both height and spatial distribution data, coloured from shorter (dark purple) to taller (yellow). (b) Posterior predictions of cactus height (log cm) across absolute latitude from a Bayesian phylogenetic regression model, showing the negative latitudinal height gradient. (c) Posterior predictions of cactus height (log cm) across mean annual temperature (log °C), showing the positive association between height and temperature. In (b) and (c), the median trend is plotted as a thicker line.*

The latitudinal plant height gradient has been associated more strongly with precipitation than temperature in global analyses [8], but we found a different pattern

among cacti. Cactus height was positively associated with mean annual temperature (MAT) after accounting for growth form (posterior mean β = 0.430, 95% CrI = 0.150–0.730, pMCMC = 0.004; median marginal R^2 = 0.16, median conditional R^2 = 0.90), whereas precipitation showed no clear association (pMCMC = 0.906). These results were consistent with models excluding growth form, in which temperature was positively associated with height (β = 0.508, 95% CrI = 0.183–0.835, pMCMC = 0.008; median marginal R^2 = 0.004, median conditional R^2 = 0.94), whereas precipitation was not (β = 0.082, 95% CrI = −0.031–0.196, pMCMC = 0.142). Across all three environmental predictors (latitude, temperature and precipitation), additive models were better supported than interaction models allowing relationships to differ among growth forms (ΔDIC = 10.0–13.5). Thus, we found little evidence that the associations between environmental conditions and cactus height varied substantially among growth forms.

Our results reveal a latitudinal gradient in cactus height that resembles patterns observed across plants more broadly, but with potentially different underlying environmental and morphological associations [8]. Taller cactus species tend to occur in warmer, lower-latitude environments, and these relationships cannot be explained solely by differences in growth form. The occurrence of such a gradient within cacti, a relatively young plant lineage with a crown age of approximately 29 million years [39–43], suggests that broad-scale geographical patterns in plant height can emerge within relatively short evolutionary timescales. However, most studies of latitudinal size gradients in plants and animals focus on variation among extant species, leaving their evolutionary origins largely unexplored. In particular, we know little about how these gradients originate and change through time, or how historical environmental change may have shaped their evolution. Understanding these processes requires examining how plant height has changed across the cactus phylogeny and through evolutionary time.

Disentangling different types of phenotypic change in cactus evolution

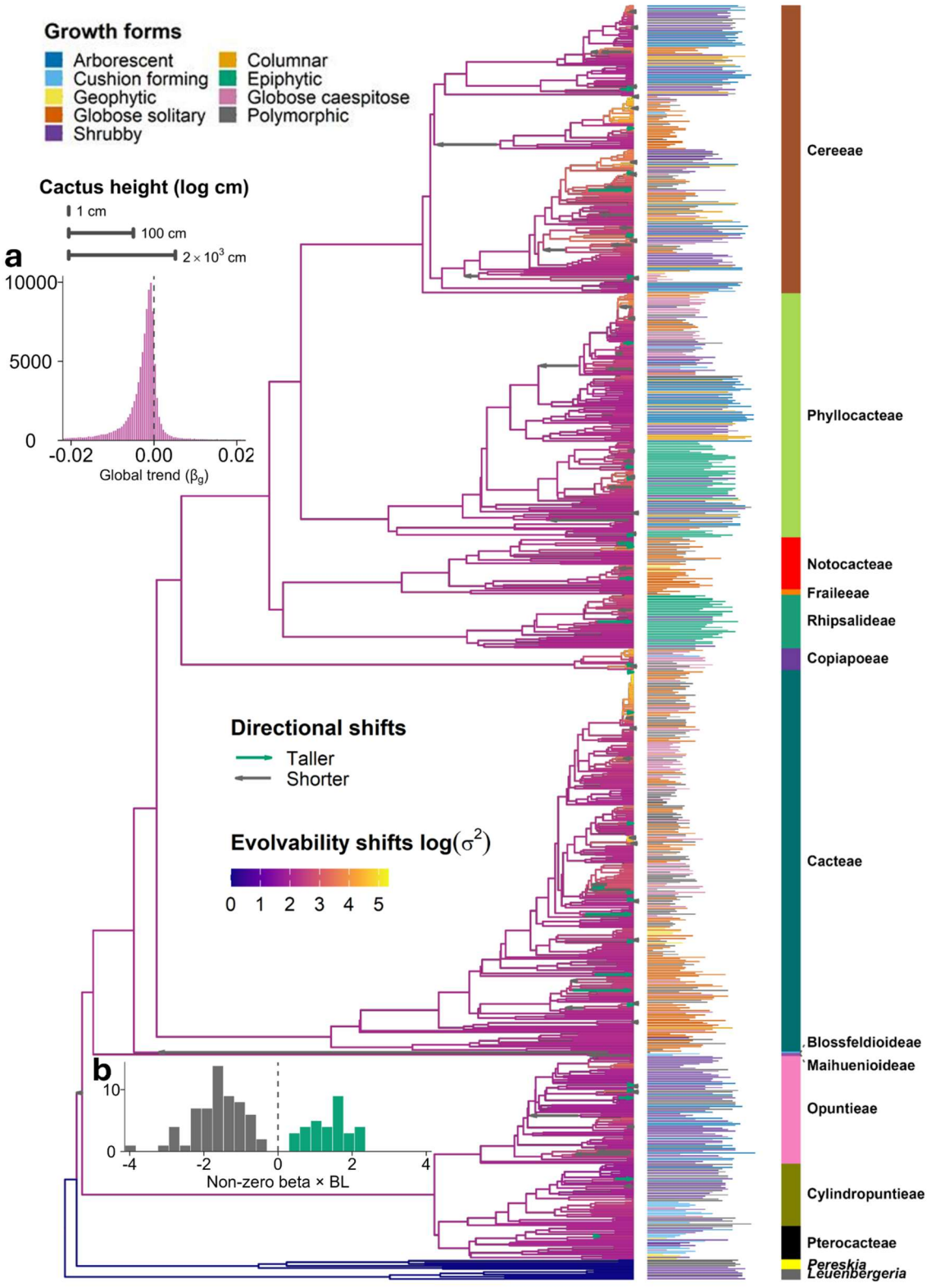
Important evolutionary processes likely to shape cactus height variation include long term macroevolutionary trends [63], changes in the rate of phenotypic divergence through time (evolvability) [53,55,64], and lineage-specific directional shifts towards taller or shorter species [15,52]. Cacti have also repeatedly evolved into different growth

forms, from small globose species to larger columnars [42,44]. We therefore reconstructed height evolution using models that account for growth-form differences while testing for these temporal and lineage-specific patterns of variance and directionality [16]. We compared Bayesian models with different combinations of these four components of height evolution. Evolvability and directional shifts were inferred across the phylogeny using reversible-jump MCMC, allowing their number and phylogenetic locations to be estimated from the data. These models were implemented using the Fabric [15], Fabric regression [16], variable rates [53], and variable rates regression models [52], available in BayesTraits V5 (https://www.evolution.reading.ac.uk/BayesTraitsV5.0.0/BayesTraitsV5.0.0.html).

The best-supported model included all four components (median log marginal likelihood across six independent runs = −1,149) and explained 54.3% of variation in cactus height (median marginal R^2 = 0.543). Support declined when the global trend was removed (Bayes factor = 13), and substantially more when directional shifts or growth-form effects were excluded (Bayes factors = 58-119). Cactus height therefore reflects the combined effects of growth-form effects and continuous evolutionary changes in both the direction and variance of height evolution. Furthermore, these changes occur within a weak family-wide background directional trend towards decreasing height (median trend = −0.00186, with 78% of the posterior below zero) (Figure 3).

We found 200 strongly supported shifts in evolvability and 92 strongly supported directional shifts in height (Figure 3). All supported evolvability shifts increased variance, whereas 60 directional shifts decreased and 32 increased height. The earliest strongly supported shift in evolvability occurred in the ancestor of all genera except *Leuenbergeria*, resulting in a ~7.43-fold increase in the accumulation of variance in height (mean non-1 scalar = 7.43) over a relatively short period of ~28.49 million years. A subsequent directional decrease in height occurred at the subsequent ancestor, from which the succulent cacti descend (~28.13 Mya), encompassing all species except the leafy *Leuenbergeria* and *Pereskia* [65,66]. This has a mean non-0 $\beta \times t$ of −2.75, corresponding to a decrease of about 15.6 cm over about 0.28 million years [15]. Thus, increased evolutionary variance preceded, rather than coincided with, a directional shift towards reduced height during early cactus

evolution. The temporal separation of these types of shifts illustrates the importance of distinguishing changes in evolutionary variance from directional changes, as the diversification of height began to accelerate before the subsequent reduction. Approaches that model phenotypic change primarily through shifts between evolutionary regimes or selective optima may struggle to distinguish these sequential changes in variance and direction [10,67–69]. More broadly, by identifying the episodes of directional changes, we provide an opportunity to ask which ecological forces are associated with different directions of morphological evolutionary change.



284

285 ***Figure 3: Directional changes and shifts in evolvability throughout cactus***

286 ***height evolution.*** *A phylogenetic reconstruction of height evolution from the Fabric*

287 *regression model including growth form. Branch colour indicates evolvability, with*

warmer colours indicating greater evolvability. Grey and green branch arrows indicate strongly supported directional shifts towards shorter and taller descendants, respectively. Bars at the tips indicate species height (log cm) and are coloured by growth form. A labelled taxonomy bar indicates tribes and in some cases, genera and subfamiles, following a recently-published phylogenomic classification [70]. Inset (a) shows the distribution of the global trend parameter, truncated for visual interpretation. Inset (b) shows the distribution of magnitudes of significant directional shifts.

Directional shifts are distributed throughout the family. Decreases outnumbered increases in both Cactoideae (51 decreases and 27 increases) and Opuntioideae (seven versus five), and there is a single decrease in the monogeneric Maihuenioideae. One directional shift, the most powerful (mean non-0 $\beta \times t = -3.83$) occurs leading to *Blossfeldia liliputana*, the smallest cactus and the sister to the remainder of Cactoideae [70]. In the eleven tribes in which we found directional changes, decreases outnumber increases in six, there are equal numbers in four, and more increases than decreases in only one. Decreases were particularly prevalent in Cereeae (20 decreases versus 7 increases), Phyllocacteae (12 versus 4), Opuntieae (6 versus 3) and Cacteae (14 versus 11). Notocacteae also contained fewer decreases than increases (2 versus 3), while *Maihuenia* contained a single decrease and no increases. Rhipsalideae, Cylindropuntieae and Copiapoeae each contained equal numbers of increases and decreases (one of each), while Blossfeldieae contained a single decrease. Pterocacteae was the only tribe in which increases occurred without any decreases, with a single directional increase.

Repeated directional changes in height appear to be a common feature in cactus evolution through time, and are not restricted to a single lineage. Of these, decreases are both the most common and have the largest magnitudes (mean negative non-0 $\beta \times t = -1.65$, mean positive non-0 $\beta \times t = 1.38$), which may indicate a general tendency for selection towards smaller cacti. This is also supported by the weakly negative global directional trend. However, directional shifts with opposite effects do occur within the same tribes, indicating there are different evolutionary trajectories and selective pressures experienced even by close relatives (Figure 3). Notably, there are many cases of nested directional changes, indicating that

selective pressures have changed through time as lineages speciated. These nested changes are found both over long timescales, given the early decrease in the ancestor of all succulent non-leafy cacti, and within the same tribes, notably in Cereeae and Phyllocacteae (Figure 4).

Dispersal through palaeoclimate shaped the latitudinal gradient through directional evolution

How the extant latitudinal gradient in cactus height emerged over deep time remains unclear. Its association with current temperature raises the possibility that the gradient reflects repeated evolutionary responses to climate as cacti dispersed and diversified across the Americas. If so, lineages experiencing greater cumulative cooling should show greater directional decreases in height, whereas those experiencing warming should show increases. We therefore reconstructed the directional history of height leading to each extant species. From the common ancestor of cacti to each species, we separately summed the magnitude of all directional increases and decreases inferred by the Fabric model, providing pathwise measures of total directional height evolution through time for each species.

Similarly, we reconstructed corresponding pathwise directional measures of palaeotemperature change by combining historical biogeography [17–19,21] with palaeoclimate reconstructions [59,71]. We used a recent reconstruction of cactus dispersal through time in continuous space [21], in which constant rates of dispersal were relaxed and lineage-specific dispersal rates were estimated [17] (Figure 4). From the posterior sample of geographical locations of each phylogenetic ancestor, we extracted temperatures from palaeoclimate reconstructions [59,71]. We then calculated the change in median temperature between the ancestral and descendant nodes of each phylogenetic branch, distinguishing positive changes (warming) from negative changes (cooling). Finally, for each extant species (n = 753), we summed the positive temperature changes and the absolute magnitudes of negative changes separately along the phylogenetic path connecting the root to each species. This provided two measures representing the cumulative warming and cooling experienced over each species' evolutionary history, rather than the net temperature difference between the root and the present [17,18] (Figure 4).

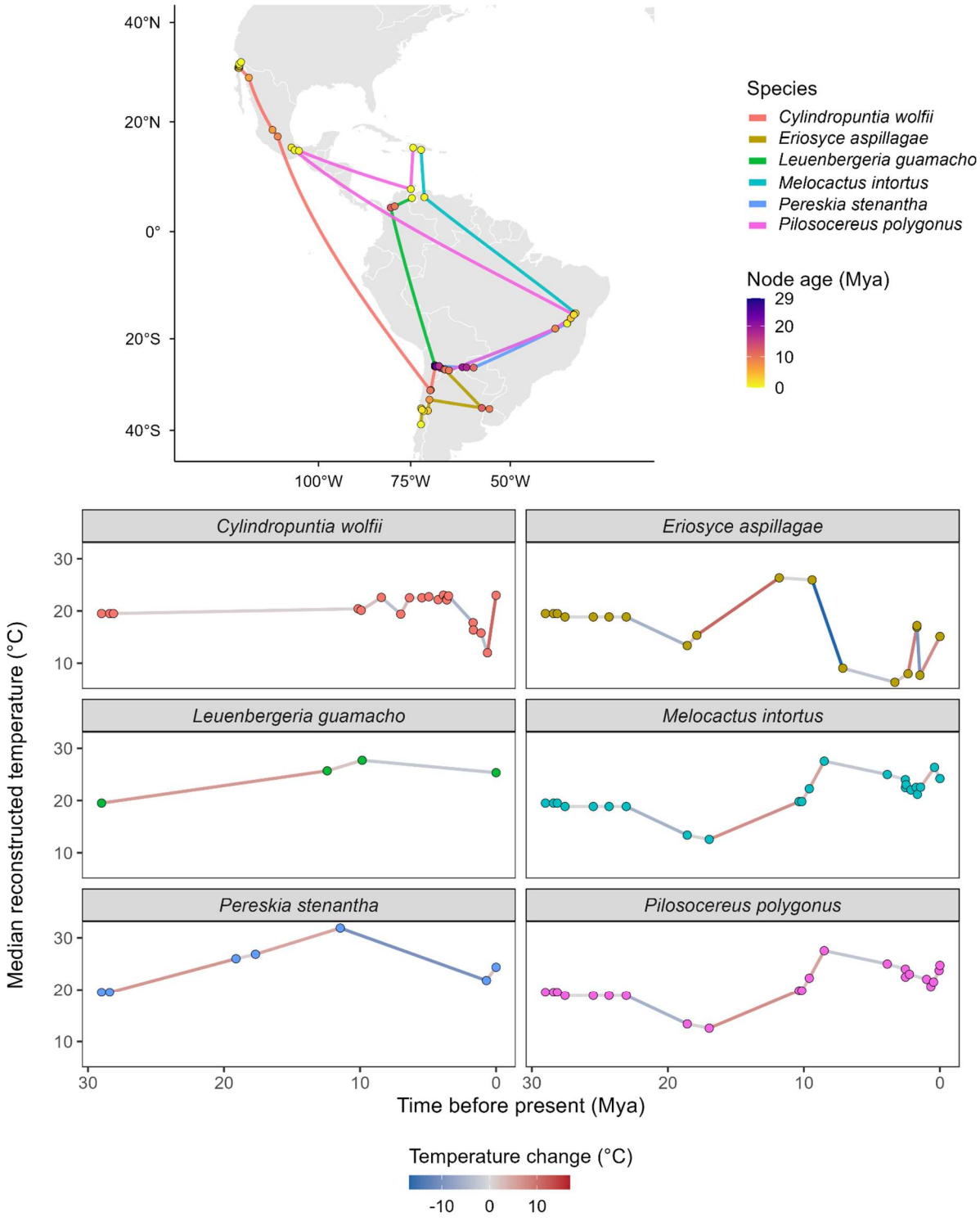


Figure 4: Dispersal of cacti through changing environments over 29 million years in continuous space. *Top, reconstructed geographic trajectories for six exemplar species* [21]*. Points show median posterior reconstructed ancestral locations from root-to-tips and are coloured by node age. Coloured lines connect the ancestor-descendent node locations in a hypothetical dispersal path. Several points and lines*

are shared by different species due to shared ancestry. Bottom, median reconstructed palaeotemperature at the ancestral nodes through time from root-to-tip. Points are coloured by species matching the geographic trajectories above. Line segments show the change in reconstructed temperature between successive nodes, with blue indicating cooling and red indicating warming towards the present.

We jointly modelled the cumulative evolutionary increases and decreases in height using a multi-response phylogenetic MCMCglmm [62,72], in which associations of both historical warming and cooling with both directions of cumulative height evolution were estimated simultaneously. We included current temperature to distinguish associations with historical temperature change from those attributable to temperatures currently inhabited by species. Greater historical warming was associated with greater evolutionary increases in height (posterior mean $\beta = 0.566$, 95% CrI = 0.417–0.699, $p_{MCMC} < 0.001$), but not decreases (posterior mean $\beta = -0.063$, 95% CrI = −0.136–0.007, $p_{MCMC} = 0.088$). Conversely, greater historical cooling was associated with greater evolutionary decreases in height (posterior mean $\beta = 0.216$, 95% CrI = 0.139–0.287, $p_{MCMC} < 0.001$), but not increases (posterior mean $\beta = -0.121$, 95% CrI = −0.280–0.026, $p_{MCMC} = 0.112$) (Figure 5). To assess what was gained by reconstructing directional height evolution rather than analysing extant height alone, we also tested whether historical temperature change was associated with extant variation in cactus height. Cumulative historical warming was positively associated with extant height ($\beta = 1.154$, 95% CrI = 0.566–1.741, pMCMC < 0.001), whereas cumulative cooling was negatively associated with height ($\beta = -1.278$, 95% CrI = −1.892 to −0.658, pMCMC < 0.001). In contrast, the total magnitude of historical temperature change, regardless of direction, was not associated with extant height ($\beta = -0.139$, 95% CrI = −0.538–0.274, pMCMC = 0.534). Therefore, historical temperature change is associated not only with reconstructed directional height evolution but also with extant height variation. However, associations with extant height alone cannot reveal which evolutionary changes gave rise to these differences. By distinguishing directional evolution from other components of height evolution [16], the Fabric regression model allows us to connect historical temperature change to the historical evolutionary changes through which extant diversity evolves.

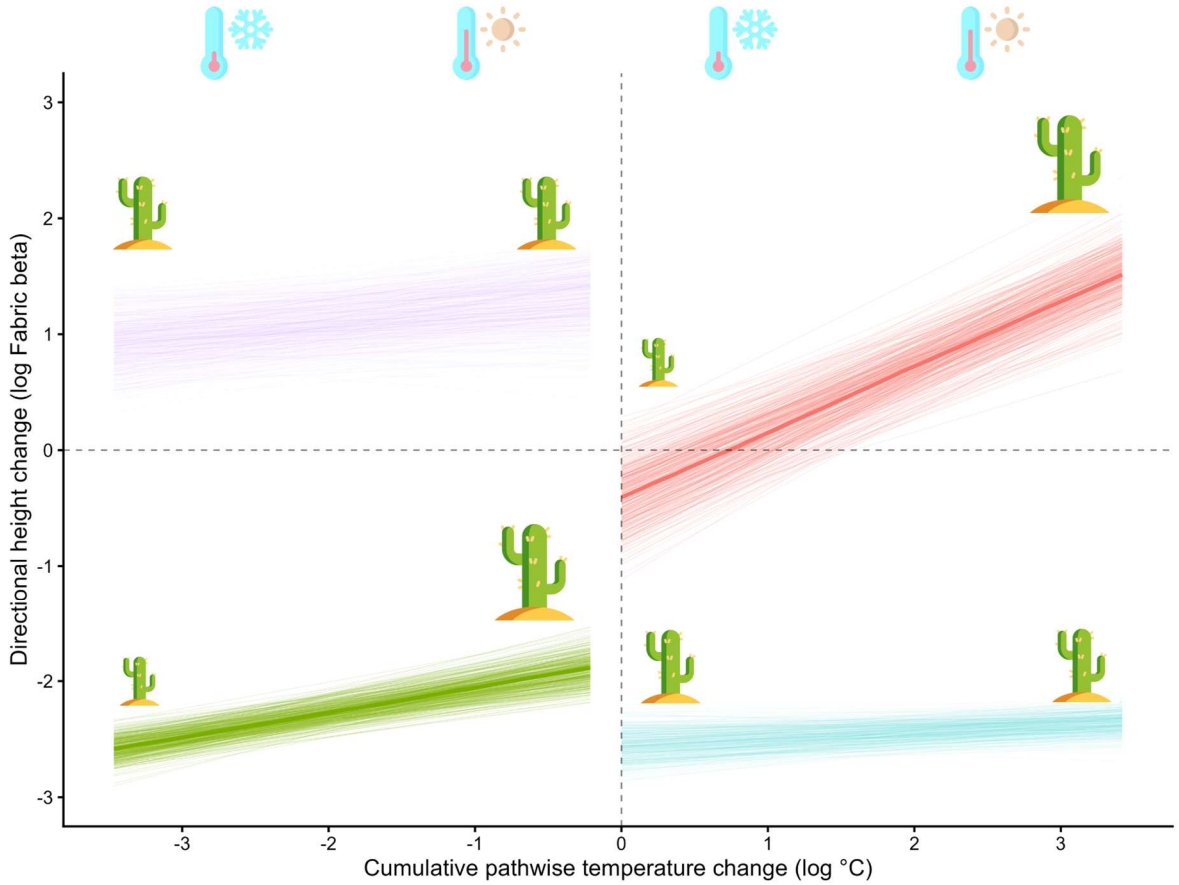


Figure 5: Historical temperature change is associated with the direction of cactus height evolution. *Relationships between cumulative palaeotemperature change and cumulative directional height evolution from root-to-tip across the cactus phylogeny (n = 753). Historical warming and cooling are shown on the positive and negative x-axes, respectively, and directional increases and decreases in height on the positive and negative y-axes. Lines show posterior predictions from the multi-response phylogenetic model, accounting for contemporary temperature. Median fitted relationships are presented as thicker lines in the two significant comparisons. Historical warming was significantly associated with evolutionary increases in height (red), but not decreases (blue), whereas historical cooling was significantly associated with evolutionary decreases in height (green), but not increases (purple). Asterisks denote significant associations. Dashed lines indicate zero temperature change and zero directional height change.*

Historical warming and cooling were associated with opposing directions of cactus height evolution. Lineages that accumulated greater warming experienced greater directional increases in height, whereas those that accumulated greater cooling

experienced greater directional decreases. This directional relationship provides a historical mechanistic origin of the contemporary cactus height gradient, in which taller cacti occur in warmer, lower-latitude environments. Our framework demonstrates how the geographical distribution of extant height carries the signature of climatic changes experienced by their ancestors.

More broadly, our results show that latitudinal gradients can emerge not only through adaptation to contemporary environments, but through repeated directional evolution as lineages experience changing environments through time. Similar geographical patterns therefore need not have the same evolutionary origins. The latitudinal gradient in cactus height resembles that observed across plants globally, persists after accounting for growth form, and is associated with temperature rather than precipitation. Similar macroecological patterns can therefore hide different combinations of morphological evolution and climatic history. Contemporary trait-environment relationships represent only the endpoints of these histories. Reconstructing how both traits and environments changed through time moves our explanations from describing latitudinal gradients to understanding how they evolved.

Conclusion

Over only around 30 million years, cacti have diversified across the Americas into one of the most remarkable radiations of flowering plants, spanning more than three orders of magnitude in height, from centimetre-scale plants to towering plants around 20 m [41–44,46]. From this extraordinary variation has emerged a latitudinal gradient in height, a strong example of one of the most pervasive geographical patterns in plant life worldwide [6,7,73,74]. Here, we traced its origins through the evolutionary and environmental histories from which it arose. Cactus height resulted from a complex history of repeated directional changes towards both larger and smaller species, changes in evolutionary variance, and growth-form transitions across the phylogeny. Yet underlying this complexity is a simple historical story. Species that experienced greater warming in their ancestors increased in size, whereas those that experienced greater cooling decreased. By reconstructing height evolution in context of the geographical and climatic histories experienced by lineages, we reveal how an

extant geographical gradient can emerge from repeated evolutionary change through deep time.

Crucially, the resemblance between the cactus height gradient and the broader pattern across plants does not tell us how either pattern arose [8]. Contemporary latitudinal height gradients are the endpoints of evolutionary trajectories in which species have changed in height, moved through space and experienced environments that themselves changed through time. Here, by reconstructing these histories together, we were able to trace a latitudinal gradient among living species back to the directional evolutionary changes accumulated by their ancestors, and to the climatic histories associated with those changes. This provides a general framework for moving beyond describing gradients to reconstructing their evolutionary and environmental origins. Applying it across other radiations could test whether some of the most pervasive geographical patterns in nature share common evolutionary origins, or whether similar patterns have repeatedly emerged through different histories.

Materials and Methods

Trait and spatial data, and phylogenetic framework

We used cactus height data (in centimetres), growth forms, and spatial data, from the Cactus Ecological Database (CactEcoDB) [42] which we recently published and have cleaned further here. We identified height outliers within each growth form using the 1.5 × interquartile range (IQR) criterion and additionally examined the tallest and shortest 10% of species within each growth form. This identified 203 species for manual review, including 21 outliers. Several suspicious measurements appeared to represent individual stem segment lengths rather than total plant height. Following inspection, we corrected 12 height measurements and excluded three species. We subsequently screened log-transformed height for outliers within genera using the same IQR criterion. 25 were identified as outliers, of which two were corrected and 23 retained following inspection. Manual review also resulted in two additional height corrections and the exclusion of one species, yielding a final dataset of 922 species. CactEcoDB records nine growth forms (arborescent, barrel, columnar, cushion-forming, epiphytic, geophytic, globose caespitose, globose solitary and shrubby) for 1,061 species. We revised 36 growth-form classifications

and combined barrel with globose solitary, as these forms are distinguished primarily by a height threshold of 50 cm [44]. CactEcoDB provides geographic range data in coordinates, most of which are sampled from IUCN expert range polygons, and a small number from curated GBIF coordinates. From these, we calculated median latitudes for every species. Although we investigated climate impacts (mean annual temperature and precipitation), we did not use those provided by CactEcoDB, which are derived from CHELSA [75], because we needed consistency with the spatial palaeoclimatic data. Instead, we used the coordinates from CactEcoDB and sampled from PALEO-PGEM [71,76]. The phylogenetic framework underlying the comparative analyses is a time-calibrated molecular phylogeny of 1,052 species built from a supermatrix sampling up to 15 plastid and three nuclear loci [41,42]. The topology was estimated with a recent phylogenomic backbone for the family [70] (CactEcoDB phylogeny V2), and calibrated against geological time using a secondary calibration on the crown age. In the absence of an informative cactus fossil record, estimates of the crown age remain uncertain, although several molecular-clock studies place the early diversification of living cacti at approximately 27–37 Ma [41–43,48,77].

Characterising the latitudinal height gradient

We tested for a latitudinal gradient in cactus height using Bayesian phylogenetic mixed models implemented in MCMCglmm [62]. Species height in centimetres was log-transformed and modelled as a Gaussian response, and the absolute value of median species latitude as the geographical predictor. We used absolute latitude because cacti occur in both hemispheres and our hypothesis concerns distance from the equator rather than hemisphere. Absolute latitude was log-transformed before analysis, and phylogenetic relationships were controlled for. To determine whether the latitudinal gradient can be explained by turnover among cactus growth forms, we compared four models: (1) an intercept-only model, (2) a model containing latitude alone, (3) an additive model containing latitude and the eight growth forms, (4) and a model including interactions between latitude and growth form [62]. Growth forms were analysed as binary variables due to polymorphism within the data [42]. We repeated these analyses using log-transformed mean annual temperature (MAT) and precipitation in place of latitude to assess the contemporary climatic correlates of cactus height. MAT and precipitation were taken from [71]-PGEM [71]. Analyses were restricted to species with complete height, growth-form, latitude, MAT and

514 precipitation data, ensuring comparisons used the same set of species. MCMCglmm
515 models used weak inverse-Wishart priors for phylogenetic and residual variance and
516 were run for 5.5 million Markov chain Monte Carlo (MCMC) iterations, with a burn-in
517 of 500,000 iterations and sampling every 5,000. We compared models using the
518 deviance information criterion (DIC) and summarised fixed effects using posterior
519 means, 95% credible intervals and pMCMC values. We assessed MCMC
520 convergence using effective sample sizes.
521
522 **Models of cactus height evolution**
523 We tested a series of Bayesian models of log-transformed height evolution with
524 increasing complexity, and compared their fits. These were all estimated in
525 BayesTraits V5
526 (https://www.evolution.reading.ac.uk/BayesTraitsV5.0.0/BayesTraitsV5.0.0.html).
527 Five models were univariate, including only cactus height and not growth forms. We
528 compared (1) a homogenous Brownian Motion model in which only an overall rate of
529 height evolution is estimated [60,78], (2) a variable rates model in which lineages
530 undergoing height evolvability changes are proposed in a reversible-jump framework
531 [53], (3) a variable rates model with a global trend from root to tips, (4) a Fabric model
532 [15], in which directional shifts are proposed by reversible-jump in conjunction with
533 variable rates, (5) a Fabric model with a global trend. Five models included growth
534 forms, which were one-hot encoded, as predictors in a regression framework [16].
535 Under the models, evolvability and directional changes in height are estimated
536 simultaneously alongside accounting for the effects of growth forms on height. We
537 compared (1) a regression model with a homogeneous rate of height evolution, (2) a
538 variable rates regression model [52], (3) a variable rates regression model with a
539 global trend, (4) a Fabric regression model [16], (5) a Fabric regression model with a
540 global trend. All models were run six times to ensure stability and were run to
541 convergence. We sampled 240 million reversible-jump (rj) MCMC generations per
542 model every 10,000, and discarded the first 40 million as burn-in. Marginal
543 likelihoods for every model run were estimated using stepping stone sampling,
544 sampling 1,000 stones for 20,000 generations each [79]. We provided a broad
545 exponential prior on variable rates, and a prior on the ancestral height with a normal
546 distribution centred around 4.6 (natural log of 100 cm) and a standard deviation of
547 one, based on preliminary variable rates runs. In models with a global trend, this was

given a relatively uninformative prior, with a normal distribution centred around zero and a standard deviation of one. In regression models, growth form betas were given relatively uninformative normal priors centred around zero with a standard deviation of five. To set priors on directional changes, for Fabric models, we estimated the expected amount of directional change for every branch in the phylogeny (n = 1,826) using the FabricUniversalPrior.py script (https://github.com/AndrewPMeade/FabricTools), and assessed the fit of Gamma, Lognormal and Weibull distributions. We then calculated the threshold cost for directional shifts, to ensure that they enter the model with a minimum cost of -2 natural log units, based on the branchwise distribution of prior probabilities. In all models, we removed clades consisting of a unique height value, which are known to detrimentally affect the estimation of variable rates, and are far more likely to originate from trait measurement inaccuracy than there exist cacti with exactly the same heights. To do this, we used the script PruneToUniqueData (https://github.com/AndrewPMeade/FabricTools). Variable rates and Fabric models were summarised with the variable rates and Fabric post-processors, respectively, and then combined (https://www.evolution.reading.ac.uk/BayesTraitsV5.0.3/BayesTraitsV5.0.3.html).

Quantifying rates of palaeoclimate change along phylogenetic branches

To investigate whether historical temperature change was associated with directional evolution in cactus height, we reconstructed the temperature histories experienced by ancestral cactus lineages as they dispersed across the Americas. We obtained ancestral geographical coordinates from a separate biogeographical reconstruction of 897 cactus species, based on 397,172 geographical coordinates from expert-validated species ranges and occurrence records from CactEcoDB [21,42]. Ancestral locations were estimated using the continuous-space Geo model [17,19] implemented in BayesTraits v5 (https://www.evolution.reading.ac.uk/BayesTraitsV5.0.3/BayesTraitsV5.0.3.html). This model reconstructs ancestral geographical coordinates on a spherical Earth using a three-dimensional Cartesian coordinate system, with a variable-rates Brownian motion model allowing dispersal rates to vary among lineages and through time [53]. To account for continental movement and the terrestrial distribution of cacti, ancestral locations were constrained using palaeogeographical maps at 1-Myr

intervals, with proposed locations falling within palaeo-oceans rejected [80]. These analyses provided posterior estimates of ancestral geographical locations throughout cactus evolutionary history. We combined these geographical reconstructions with spatially and temporally explicit estimates of mean annual temperature (MAT). For ancestral nodes older than 5 Ma, we used HadCM3 palaeoclimate reconstructions at 10, 14, 19, 25 and 31 Ma [59], selecting the temporally nearest reconstruction for each node [17,18]. For nodes within the last 5 Myr, we used the PALEO-PGEM-Series, which provides global palaeoclimate reconstructions at 1-kyr temporal resolution and 1° × 1° spatial resolution [71]. Each ancestral geographical location was matched to the geographically nearest climate grid cell in the temporally closest reconstruction, providing estimates of historical MAT along the evolutionary paths leading to extant species [17,18]. We quantified the cumulative warming and cooling experienced along each root-to-tip evolutionary path by calculating temperature differences between successive reconstructed ancestral locations. Positive temperature changes were summed to estimate cumulative historical warming, while the absolute magnitudes of negative changes were summed separately to estimate cumulative historical cooling. We applied the same approach to the directional changes in height inferred by Fabric regression [16], separately accumulating directional changes towards taller descendants and the absolute magnitudes of changes towards shorter descendants along each root-to-tip path. This produced four species-level measures: cumulative warming, cumulative cooling, cumulative directional increases in height and cumulative directional decreases in height. All four measures were log1p-transformed before analysis.

Impacts of palaeoclimate change on directional height evolution

We tested the associations between these reconstructed histories using Bayesian multivariate phylogenetic mixed models implemented in MCMCglmm [62]. Log-transformed cumulative directional increases and decreases in height were modelled jointly as two Gaussian response variables, with log-transformed cumulative warming and cooling included as response-specific fixed effects. Our preferred model additionally included log-transformed contemporary MAT, allowing associations with historical temperature change to be estimated while accounting for the temperatures currently experienced by species. Unstructured covariance matrices were fitted between the two responses for both phylogenetic and residual

effects. We also fitted a model excluding contemporary MAT and a model including an interaction between cumulative warming and cooling, without contemporary MAT, to assess sensitivity to model specification. All three models were run for 5.5 million MCMC iterations, discarding the first 500,000 as burn-in and sampling every 5,000 iterations. Convergence was assessed using effective sample sizes. We used inverse-Wishart priors with $V = I_2$ and $v = 2$ for both phylogenetic and residual covariance matrices. To assess sensitivity to prior specification, we refitted all three models using alternative priors ($V = \frac{2}{3}I_2$, $v = 3$), which specify uniform marginal priors on correlations rather than favouring stronger correlations between responses.

As a complementary analysis, we tested whether reconstructed historical temperature changes were associated directly with the heights of extant species, rather than their reconstructed directional height evolution. We fitted two Gaussian phylogenetic MCMCglmm models with log-transformed extant height as the response. The first included cumulative historical warming and cooling as separate predictors, while the second included the total absolute magnitude of historical temperature change, irrespective of direction. Both models accounted for log-transformed contemporary MAT and used the same MCMC settings described above, with inverse-Wishart priors of $V = 1$ and $v = 0.002$ for phylogenetic and residual variances.

Data availability

All data analysed and outputs from this study will be made available at GitHub (https://github.com/jamie-thompson/cactus_height) upon acceptance.

Code availability

BayesTraits version 5 is available at https://www.evolution.reading.ac.uk/BayesTraitsV5.0.3/BayesTraitsV5.0.3.html.

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