A fully resolved backbone phylogeny reveals numerous dispersals and explosive diversifications throughout the history of Asteraceae

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The sunflower family, Asteraceae, comprises 10% of all flowering plant species and displays an incredible diversity of form. Asteraceae are clearly monophyletic, yet resolving phylogenetic relationships within the family has proven difficult, hindering our ability to understand its origin and diversification. Recent molecular clock dating has suggested a Cretaceous origin, but the lack of deep sampling of many genes and representative taxa from across the family has impeded the resolution of migration routes and diversifications that led to its global distribution and tremendous diversity. Here we use genomic data from 256 terminals to estimate evolutionary relationships, timing of diversification(s), and biogeographic patterns. Our study places the origin of Asteraceae at ~83 MYA in the late Cretaceous and reveals that the family underwent a series of explosive radiations during the Eocene which were accompanied by accelerations in diversification rates. The lineages that gave rise to nearly 95% of extant species originated and began diversifying during the middle Eocene, coincident with the ensuing marked cooling during this period. Phylogenetic and biogeographic analyses support a South American origin of the family with subsequent dispersals into North America and then to Asia and Africa, later followed by multiple worldwide dispersals in many directions. The rapid mid-Eocene diversification is aligned with the biogeographic range shift to Africa where many of the modern-day tribes appear to have originated. Our robust phylogeny provides a framework for future studies aimed at understanding the role of the macroevolutionary patterns and processes that generated the enormous species diversity of Asteraceae.

Significance

Flowering plant species represent at least 95% of all vascular plants on Earth, and members of the sunflower family comprise roughly 10% of this diversity. The family is often considered taxonomically difficult primarily because it is enormous in size and cosmopolitan in distribution. Using phylogenomics, we were able to fully resolve the backbone of the sunflower family tree. We provide evidence for a late Cretaceous origin followed by explosive diversifications and dispersals during the middle Eocene—ultimately resulting in the family’s 25,000+ extant species. Our results provide a framework to interpret the spatiotemporal patterns of migration out of South America and the family’s explosive diversifications out of Africa that led to its global evolutionary and ecological success.

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Data deposition: Raw sequencing reads were deposited in the National Center for Biotechnology Information (NCBI) Sequence Read Archive and Genbank under BioProject PRJNA54287. The resulting alignments and phylogenetic trees were deposited in FigShare at https://doi.org/10.6084/m9.figshare.7697834 and https://doi.org/10.6084/m9.figshare.7695929.

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some topological differences compared with the concatenated phylogenies, but in nearly every case, the ASTRAL tree nodes had lower support (SI Appendix, Fig. S4). We found a few major differences from previous phylogenies in tribal placements. For example, in our ML tree, Hyalideae was placed sister to Stiffiteae, and Perteaee diverged just before the Carduoideae subfamily (thistles) and immediately after the monotypic Hecatoeideae. Two subfamilies previously circumscribed (17, 25) are not supported as monophyletic: Carduoideae [here recovered as three clades: (i) Cardueae, (ii) Oldenburgeae + Tarchonantheae, and (iii) Diocoeae, brown shading in Fig. 2], and Cichorioideae [dandelions, here recovered as two clades: (i) tribe Cichorieae and (ii) six remaining tribes referred to herein as Vernonioideae (26) blue shading in Fig. 2]. In the enormous subfamily Asteroideae (17,000+ species), relationships among the five tribes that comprise nearly 10,000 species, including Anthemideae (chrysanthemums), Astereae (asters), Gnaphalieae (strawflowers), Calenduleae (pot marigolds), and Senecioneae (ragworts), were resolved with high support (Fig. 2). The exception was the sister group relationship between Anthemideae and Senecioneae with bootstrap support for this relationship lower than at most other nodes at 75%. The Bayesian analysis yielded the same topology for these five tribes with posterior probabilities of 1.0 (SI Appendix, Fig. S3). The ASTRAL analysis resulted in high support for a different topology for these five tribes with Senecioneae as the sister group to a clade of the four remaining tribes (SI Appendix, Fig. S4). Within the large Heliantheae alliance, which includes sunflowers and coneflowers (supertribe Helianthodae, 13 tribes), maximal support was recovered for most intertribal relationships (Fig. 2 and SI Appendix, Figs. S2–S4).

Dating Analysis. The ML phylogeny, calibrated in two analyses by constraining the minimum ages of nodes with either seven or eight fossils, provided similar estimates that indicate Asteraceae likely originated during the Late Cretaceous (~53 MYA) (95% CI, 91–64 MYA, Fig. 3 and SI Appendix, Figs. S5 and S6 for all CIs, and SI Appendix, Table S1 for all scenarios tested). The earliest diversifying lineage (64 MYA) is tribe Barnadesiaeae followed by the monospecific Famaatinantheae, which diverged from the rest of the family roughly 62 MYA near the Cretaceous–Paleogene (K–Pg) boundary. Roughly 5–10 MYA passed before extant members of tribes Stiffiteae, Nassauvieae, Mutisieae, Wunderlichieae, and Gochnatieae began to diverge during the early Eocene (~53 MYA). Divergence of the large clade that includes monotypic Hecatoeideae and the rest of the family occurred in the early Eocene around 50 MYA. Explosive diversification occurred within the remaining three subfamilies (Cichorieae, Anthemideae, and Asteroideae) in the middle to late Eocene (42–37 MYA) that gave rise to most of the present-day tribes that harbor 95% of Asteraceae’s extant species. Lastly, the radiation of the Heliantheae alliance began during the late Oligocene, roughly 25 MYA, including the origins of its tribes (SI Appendix, Figs. S5 and S6). Our age estimates are in general agreement with three recent publications which also used the newer fossil data (SI Appendix, Table S1) (6, 23, and 24).

Historical Biogeography. The AIC model selection supports the BAYAREALIKE model, including the jump (j) speciation parameter (Fig. 3 and SI Appendix, Figs. S7 and Table S2). Our ancestral range estimates support an origin of Asteraceae in southern South America with subsequent range extensions into the north and central Andes region and later into the Guiana Shield region and Brazil. From South America, Asteraceae likely migrated to North America but a possible dispersal to southern central Africa or to Asia is also estimated (Fig. 3, see stem node leading to Hecatoeideae and SI Appendix, Fig. S7). Next, the family dispersed either to southern and central Africa or to Asia. Beginning around 42 MYA, all major nodes along the backbone of the Asteraceae tree have an ancestral range of south and central Africa. The only exception is the large New World Heliantheae alliance with a broad ancestral range estimate that includes north and central Andes + Mesoamerica–Caribbean and the north and central Andes region alone. The Biogeographic Stochastic Mapping (BSM) results indicate the presence of numerous dispersal events, some requiring long-distance
events occurred out of the north and central Andes region, south and central Africa, and North America, respectively. The highest number of anagenetic events was to North America and the north and central Andes region, respectively, with the fewest events to south and central Africa. By far, the greatest number of founder events occurred out of south and central Africa and coincide with the explosive diversifications during the middle Eocene. Founder events were highest to North America and mostly resulted from events out of the north and central Andes region and Mesoamerica–Caribbean regions coinciding with dispersal events during the Miocene (20–5 MYA) (Fig. 3 and SI Appendix, Fig. S7 and Table S3).

Diversification Analyses. The diversification analyses revealed five instances of rate acceleration (shown in red numbered boxes) and three of deceleration (shown in blue numbered boxes) (Fig. 3 and SI Appendix, Fig. S8 and Table S4). The rate decreases (nos. 7, 8, and 9) occur at nodes leading to tribes with only one or two species. The five accelerations coincide with major diversifications, including the core of the family following divergence from tribes Barnadesiaceae and Famatinanthaceae (no. 5), the explosive diversifications in Africa during the middle Eocene (nos. 2 and 6), and on the stems leading to the Vernonieae (no. 4) and the Heliantheae alliance (no. 3).

Discussion

Asteraceae Phylogeny Backbone. With at least 25,000 named species and more than 1,700 genera, the backbone phylogeny of the sunflower family has been notoriously difficult to resolve. However, with increasing access to next generation sequencing technologies, solving the phylogenies of megafamilies has become a reality. Our phylogenomics approach resulted in a highly resolved and well-supported nuclear phylogeny that shares similarities with the previously published plastid (23, 24, 27) and phylotranscriptomic (6) phylogenies despite the substantial differences in sampling of genomes, loci, and taxa. It is also important to note there is little discordance among our different analyses (ML, Bayesian, ASTRAL; Fig. 2 and SI Appendix, Figs. S2–S4), which may be attributed to our use of the conservative pipeline, PHYLUCE, for assigning orthology (retaining only loci recovered as single copy for each taxon). Our deep sampling of genes and broad, dense sampling of select taxa representing each major lineage resulted in a fully resolved tree for each of the stem nodes along the backbone, which was previously intractable. In addition, we sampled a number of genera that have been historically difficult to place, including Stiffita, Hylasia, Hecastocleis, Cavea, Platycarpha, Gundelia, Cyclolepis, and Cynarinum. The inclusion of anomalous and difficult to place genera (which also tend to be species poor) is important for biogeographic analyses of the family as previously noted (17), their placement nearly always anchors a large radiation.

While recent progress has been made to better understand the evolution of the Asteraceae through molecular phylogenetics using plastid markers (23, 27) and phylotranscriptomics (6), the highly resolved backbone of our nuclear phylogeny presented here further alters our perception of evolution in the family and will necessitate several changes in its classification, particularly for subfamilies Stiffitioideae, Carduoideae (thistles), and Cichorioideae (dandelions). First, a few taxa (Hyalocephalas, Gongylolepatis, and Leucomeris), previously placed in Wunderlichioideae are now in Stiffitioideae, which except for Leucomeris, are placements also supported by Panero and colleagues (23, 28). This makes more sense biogeographically and adds an interesting potential for long-distance dispersal events from South America to Asia for Leucomeris and its sister genus, Nouelia (not sampled here). Second, our data do not support the monophyly of subfamily Carduoideae (sensu 17). The southern African tribes Oldenburgiae + Tarchonanthaceae and the widespread African Dicomeae no longer share a most recent common ancestor (MRCA) with Carduoideae (core thistles) and instead form a grade paraphyletic to Carduoideae. Thus, the classification of Carduoideae needs revision to render it monophyletic.

Third, the Asian Pertyeae is now shown to diverge immediately after the north American Hecastoleiaceae. This makes the case for an earlier arrival of the family into Asia than previously considered (see section below). Finally, our data do not support the monophyly of subfamily Cichorioideae, which necessitates a more narrowly defined

Fig. 2. Maximum likelihood tree for the Asteraceae family. Tree is color coded by subfamily with most tribes indicated in text to the right of each. Diamonds at nodes indicate bootstrap support of 94% or higher. All nodes along the backbone have maximal bootstrap support of 100%.

dispersals throughout the entirety of the Asteraceae’s evolutionary history (59% of total events, SI Appendix, Table S3). Among these dispersals, anagenetic events (i.e., range expansions) were more prevalent than founder events (73%). The highest number of anagenetic
subfamily consisting of only tribe Cichorieae. The six remaining tribes of subfamily Cichorioideae are monophyletic and need to be recognized as a separate subfamily named Vernonioideae. These vernonioid members of the currently defined Cichorioideae are atypical for Cichorieae, and understanding this new relationship may help identify the defining traits that support a newly defined Vernonioideae (already described in ref. 29).

Evolution of Asteraceae in Space and Time. Our data provide evidence that Asteraceae most likely originated during the late Cretaceous (~83 MYA, 95% CI 91–64) in southern South America and underwent several range extensions, dispersal events, and diversifications (Fig. 3). During the Cretaceous, the global climate was warmer than today with tropical forest vegetation at the poles (30). Cooling began at the end of the Cretaceous, resulting in increased seasonality (31) with subdeserts beginning to form in central South America. The oldest lineage is the enigmatic Barnadesieae which is a small South American tribe of about 90 species with a strikingly different morphology than the rest of the family, which includes the frequent presence of prominent, paired axillary spines. Our analyses indicate that additional extant lineages of Asteraceae did not diverge until after the K–Pg. Alternatively, earlier lineages from other southern hemisphere continents may have been lost during this mass extinction event. Given the sparse fossil record for Asteraceae, and the general difficulty in assigning late Cretaceous/early Eocene pollen to extant angiosperm genera (23, 31), there are few data to more fully explore this since the only Cretaceous Asteraceae fossil evidence (Tubuliflorides lilliei type A, putatively attributed to Barnadesioideae) is from Antarctica and New Zealand (76–66 MYA; refs. 24 and 32), whereas the earliest diverging extant lineage is restricted to South America.
Alliance diverged from the rest of the family and its descendants arrived in the New World coincident with the end of the Oligocene and another brief warming period and major climatic shift (Late Oligocene Warming Event). About 21 MYA, diversifications occurred that resulted in the segregate tribes of the Heliantheae alliance (representing more than 5,600 species), and the highest rate acceleration measured in our study was along this stem (Fig. 3, red box no. 3). Previous studies also support a WGD event at the crown node of the Heliantheae alliance (4, 6), that, along with long-distance dispersal into new habitats, likely played a major role in its diversification.

While we broadly sampled across Asteraceae, our study still represents a small portion of the total number of genera in the family (207 of the ~1,700 genera). Furthermore, extinctions that took place during the history of Asteraceae are difficult to account for in historical biogeographic analyses with a sparse pollen record. Nonetheless, our primary goal was to estimate the ancestral geographic ranges along the backbone of the tree, i.e., deep biogeographical events, thus we carried out this analysis at the level of genus to represent as much geographic diversity as possible. We coded geographic range by genus sampled, as opposed to species because species-level sampling is not practical given the size of the family and wide distribution of some sampled taxa. However, genus-level, rather than species-level coding could bias the results toward more widespread ancestral range estimates and/or lower estimates of found species. It is important to note that we did not see this bias in our results along the backbone of the tree, and our estimates are in general agreement with other biogeographic studies of the family (25, 43). Still we note that some interpretations of the biogeographic estimates, especially of lower taxonomic scales, will require more extensive sampling in future studies. For example, our sampling of the two very large tribes, Heliantheae and Eupatorieae, represents less than 0.005% of the ~5,600 species within the Heliantheae alliance, and therefore our limitations in sampling and/or coding could explain why the ancestral range for this lineage is estimated as more widespread than previous studies which hypothesize a North American origin (25, 43). Given that our Asteraceae-specific probe set has been successful in resolving phylogenetic relationships within both tribes and genera (ref. 44; Fig. 2), future studies testing biogeographical hypotheses at these levels may yield further insight into the migrations and dispersals of the family.

Phylogenomics coupled with biogeographic and diversification rate analyses have revealed that a series of explosive diversifications that began during the Eocene resulted in the tremendous diversity of this family. These diversifications were rapid, occurring in a few million years, and were associated with extensive dispersal events and significant climatic changes since the Cretaceous. The impact of these diversifications and dispersals on angiosperm biodiversity is substantial, with members of Asteraceae found on every continent and comprising 10% of flowering plants thus encompassing a major component of angiosperms, the world’s most dominant group of terrestrial plants.

Materials and Methods

Taxon Sampling, Hyb-Seq, and Data Processing. We carried out sequence capture for 238 samples and incorporated 18 transcriptomes representing 13 subfamilies, ~45 tribes, 207 genera, and three outgroup taxa (Dataset S2). Sequence capture was performed and data were processed following the bioinformatic workflow and methods of ref. 45. Raw sequencing reads were deposited in the National Center for Biotechnology Information (NCBI) Sequence Read Archive under BioProject PRJNA540287.

Phylogenomic and Divergence Time Analyses. We generated phylogenetic trees based on a concatenated data matrix of all loci using a ML approach, Bayesian estimation, and a pseudo-coalescence method to compute a consensus “species” tree based on individual gene trees. We generated a time-calibrated ML phylogeny and used fossil calibration points (SI Appendix, Table S1) to constrain nodes in the ML phylogeny. The resulting alignments and phylogenetic trees were deposited in FigShare DOIs 10.6084/m9.figshare.7697834 and 10.6084/m9.figshare.7695299 (46, 47).

Historical Biogeography and Diversification Analyses. Ancestral ranges were estimated using ML implemented on the dated phylogeny pruned to include only one species per genus (SI Appendix, Table S5). Diversification rates were estimated using the dated tree described above and pruned to the level of
tribe (I Appendix, Table S6). Additional details of the methods are available in S1 Appendix.

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