The evolutionary history of Senna ser. Aphyllae (Leguminosae–Caesalpinioideae), an endemic clade of southern South America
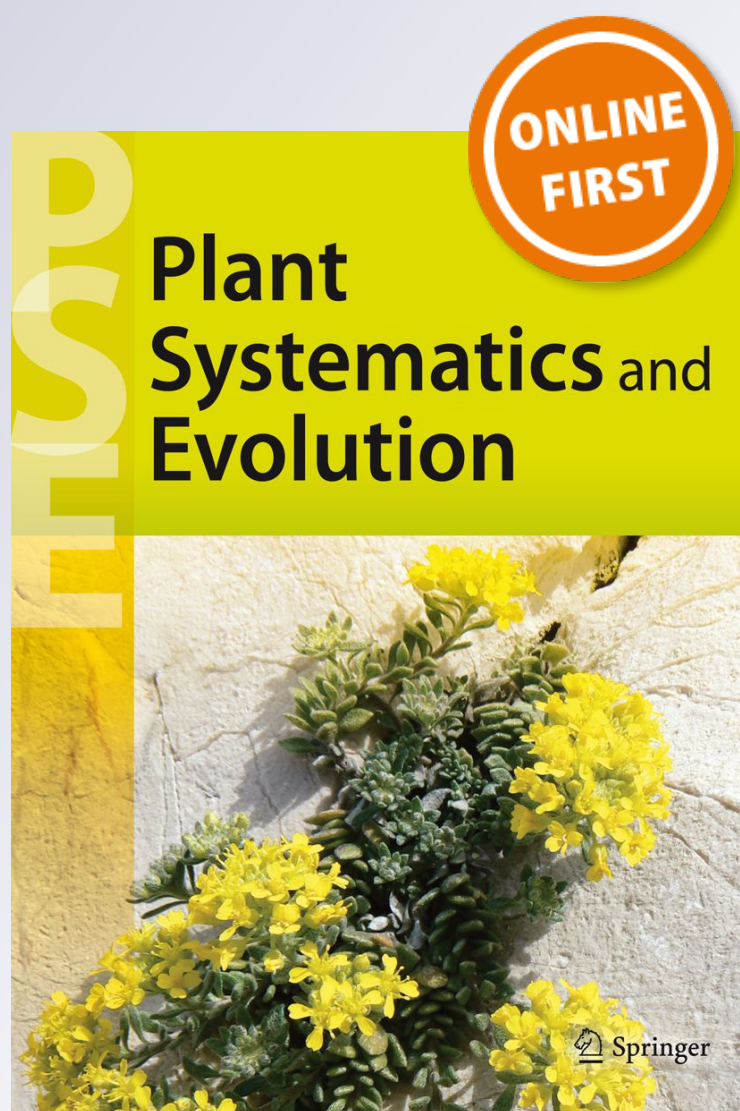
Federico O. Robbiati, Ana Anton, Brigitte Marazzi, Marilyn Vásquez-Cruz & Renée H. Fortunato

ONLINE FIRST

PSE

Plant Systematics and Evolution

Springer

Springer

Springer

CrossMark

ORIGINAL ARTICLE

The evolutionary history of *Senna* ser. *Aphyllae* (Leguminosae–Caesalpinioideae), an endemic clade of southern South America

Federico O. Robbiati[1,2] · Ana Anton[2,3] · Brigitte Marazzi[4] · Marilyn Vásquez-Cruz[5] · Renée H. Fortunato[2,6,7]

Abstract In the legume genus *Senna*, series *Aphyllae* includes seven species of leafless shrubs and subshrubs from arid, semiarid and xerophilous areas of three different biogeographic subregions in southern South America. In this study, we investigated the evolutionary history of *Aphyllae* in a molecular phylogenetic framework. We reconstructed phylogenetic relationships among *Aphyllae* species based on DNA sequence data of four plastid (*rpS16, rpL16, mat*K, *trn*L-F) and one nuclear (ITS) region from 23 accessions, analyzed with parsimony, Bayesian and maximum likelihood methods. We inferred the evolutionary and biogeographic history estimating divergence times and reconstructing ancestral character states and ancestral areas of distribution. Series *Aphyllae* was found to be monophyletic, and the taxa formed two main clades: Clade A gathering *S. aphylla* var. *aphylla, S. crassiramea, S. rigidicaulis* and *S. spiniflora*; and Clade B grouping *S. acanthoclada*, *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*. The morphologically complex *S. aphylla* appears thus polyphyletic. Molecular dating and ancestral area reconstructions suggest that the *Aphyllae* clade started to diversify in the South American Transition Zone in the Late Pliocene. Based on these results, we hypothesize that this diversification occurred during the last period of Andean uplift with the aridification in South America. The ancestral character state reconstructions suggest that, in addition to the loss of leaves in adult plants, series *Aphyllae* evolved various morphological features, such as fastigiate, thickened or decumbent–subdecumbent branches during the colonization and establishment in different arid and semiarid lands in South America.

Handling Editor: Michal Ronikier.

✉ Federico O. Robbiati
federico.robbiati@conicet.gov.ar

Ana Anton
anton@imbiv.unc.edu.ar

Brigitte Marazzi
marazzibrigitte@gmail.com

Marilyn Vásquez-Cruz
marilynvas07@gmail.com

Renée H. Fortunato
fortunato.renee@inta.gob.ar;
rhfortunato2002@yahoo.com.ar

1 Estación Experimental Agropecuaria, INTA, Ruta Nac. 68 – Km 172, 4403 Cerrillos, Prov. de Salta, Argentina

2 Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Ciudad de Buenos Aires, Argentina

3 Instituto Multidisciplinario de Biología Vegetal (IMBIV), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) – Universidad Nacional de Córdoba, Vélez Sarsfield 1611–X5016GCA, 5000 Córdoba, Prov. de Córdoba, Argentina

4 Natural History Museum of Cantone Ticino, Viale Cattaneo 4, 6900 Lugano, Switzerland

5 Biología Evolutiva, Instituto de Ecología, A.C, Carretera antigua a Coatepec 351, El Haya, 91070 Xalapa, Veracruz, Mexico

6 Instituto de Recursos Biológicos, INTA, Nicolás Repetto y de Los Reseros s/n°, 1686 Hurlingham, Prov. de Buenos Aires, Argentina

7 Facultad de Agronomía y Ciencias Agroalimentarias, Universidad de Morón, Cabildo 134, 6° piso, 1708 Morón, Prov. de Buenos Aires, Argentina

🖄 Springer

Introduction

Series *Aphyllae* (Benth.) H.S.Irwin & Barneby is a taxonomically complex group of xeromorphic shrubs and subshrubs of the caesalpinioid legume *Senna* Mill., from arid, semiarid and xerophilous areas of southern South America (Bravo 1978a, b; Irwin and Barneby 1982). This group of seven species is morphologically unique among *Senna* species, as adult plants lack leaves, their stems are junciform, green and photosynthetic, and their roots deep and woody. These xerophytic features allow them to grow and survive in extreme habitats with limited precipitation, high temperatures and rocky substrate (Bravo 1978a; Burkart 1952; Robbiati et al. 2014a, b, 2017).

Characterized by a complex taxonomic history, series *Aphyllae* was formerly part of *Cassia* L. and consisted of five species (Burkart 1952). The first comprehensive taxonomic treatment identified 11 species, including two subspecies and two varieties (Bravo 1978a, b, 1982; Bravo et al. 1986), which were then transferred to *Senna* (Irwin and Barneby 1982). Recently, a series of taxonomic studies (Robbiati et al. 2013, 2014a, b) based on phenetic and seed protein profiles reinterpreted *Aphyllae*'s taxonomy and finally proposed seven species and three varieties: *S. acanthoclada* (Griseb.) H.S.Irwin & Barneby (Fig. 1a), *S. aphylla* (Cav.) H.S.Irwin & Barneby var. *aphylla* (Fig. 1b), var. *divaricata* (Hieron.) Robbiati & Fortunato (Fig. 1c), and var. *pendula* Robbiati & Fortunato (Fig. 1d), *S. crassiramea* (Benth.) H.S.Irwin & Barneby (Fig. 1e), *S. nudicaulis* (Burkart) H.S.Irwin & Barneby, *S. pachyrrhiza* (L.Bravo) H.S.Irwin & Barneby (Fig. 1f), *S. rigidicaulis* (Burkart ex L.Bravo) H.S.Irwin & Barneby (Fig. 1g), and *S. spiniflora* (Burkart) H.S.Irwin & Barneby (Fig. 1h).

Aphyllae's evolutionary history is strictly related to the history of the arid lands that its species currently inhabit. They occur in deserts and xerophilous areas of South America in southern Bolivia, central and northwestern Argentina and southeastern Paraguay. Their biogeographic distributions cover the South American Transition Zone (SATZ) including Monte and Prepuna province, the Chaco province in neotropical region, and the northern part of Central Patagonian province in Andean region (Fig. 2; Morrone 2006). Therefore, as one of the most characteristic groups of these arid regions (Bravo 1978a), series *Aphyllae* is an ideal study group to increase our understanding of the influence of aridization and Andean uplift in southern South America on the evolution of these biota.

The aridification of South America has been attributed to several geological and climatic events such as the uplift of the Andes (Roig et al. 2009; Hoorn et al. 2010; Baranzelli et al. 2014), a phase of global cooling and the upwelling northflowing of the cold Humboldt Current (Hartley and Chong 2002). The Andes were formed in discrete periods, progressing from south to north and from west to east with regional differences in timing and type of uplift (Taylor 1991; Hoorn 1993; Hoorn et al. 1995; Gregory-Wodzicki 2000; Garzione et al. 2008). The Patagonian and Principal Cordilleras were uplifted during middle Miocene and reached their current altitude with Pliocene and Pleistocene uplift events (van der Hammen and Cleef 1986; Gregory-Wodzicki 2000; Ramos and Ghiglione 2008). As the peaks arose, they formed a physical barrier to atmospheric circulation in the southern hemisphere which produced a slow decrease in precipitation originating the Arid Diagonal (Cabrera and Willink 1980; Gregory-Wodzicki 2000). The Arid Diagonal is a narrow region with limited precipitation (<300 mm/year). It comprises the Altiplano andino, the western coast desert, Atacama Desert and the Monte and Patagonia phytogeographical provinces (Cabrera and Willink 1980).

However, palynological findings have shown the existence of small regions with semidesertic conditions in central-western Argentina during Eocene (Volkheimer 1971), while the main aridization process began during the Pliocene (Hartley and Chong 2002; Roig et al. 2009). The present semiarid conditions were established in southern South America by the end of the Pliocene, and hyperaridity did not begin until the late Pliocene, i.e., 2 million years ago (My) (Ari et al. 2011). New evidence provided by fossil pollen records indicates that Patagonian desertification was not exclusively a consequence of Andean uplift, but also a result of a much widespread phase of aridity that affected other regions of Earth (for example, central Australia and northern Chile) during the Pliocene–Pleistocene interval (Palazzesi et al. 2014). Ultimately, the climatic fluctuations during the Pleistocene also contributed to the expansion of South America arid and semiarid regions (Palma et al. 2005; Viruel et al. 2012).

The origin of arid lands in South America had indeed significant consequences on biota, favoring biotic diversification, the formation and increase in xeric shrubland landscapes, such as the Monte, Prepuna and Patagonian province (López 2003; Palma et al. 2005; Antonelli et al. 2009; Roig et al. 2009; Hoorn et al. 2010; Ari et al. 2011). Along with the Andean uplift new ecological opportunities arose as an evolutionary force promoting diversification, for example, via allopatric speciation, habitat fragmentation and/or ecological displacement (von Hagen and Kadereit 2003; Bell and Donoghue

Fig. 1 Habit of the study taxa from *Senna* ser. *Aphyllae*. **a** *Senna acanthoclada*, **b** *S. aphylla* var. *aphylla*, **c** *S. aphylla* var. *divaricata* **d** *S. aphylla* var. *pendula*, **e** *S. crassiramea*, **f** *S. pachyrrhiza*, **g** *S. rigidicaulis* and **h** *S. spiniflora*

2005; Palma et al. 2005; Hughes and Eastwood 2006; Pirie et al. 2006; Ribas et al. 2007; Antonelli et al. 2009; Guerrero et al. 2013).

Molecular phylogenetic studies of *Senna* (Marazzi et al. 2006; Marazzi and Sanderson 2010) showed that this monophyletic series originated almost 12 million
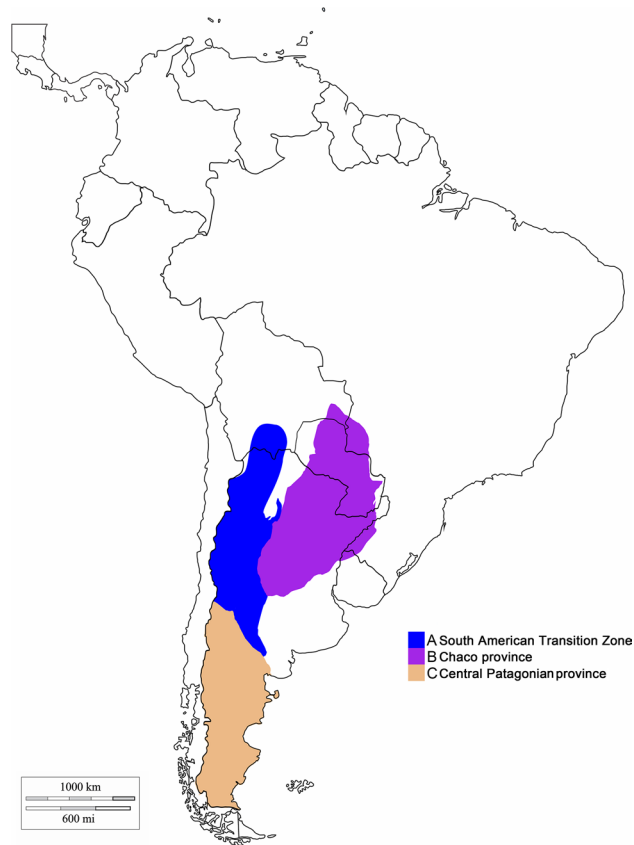
Fig. 2 Biogeographic subregions in southern South America in which *Senna* ser. *Aphyllae* occurs. Subregion *A* corresponds to the South American Transition Zone, *B* to Chaco province and *C* to Central Patagonian province

years ago, but included only few species. This means that the *Aphyllae* lineage most probably occurred in this region already before/during aridization and the Andean uplift, but phylogenetic relationships of its extant species and its evolutionary history are still unresolved.

This study aims to resolve the *Aphyllae* puzzle as a representative case study to understand how past geoclimatic events are reflected on the series' evolutionary history. We want to test the current taxonomic classification proposal by Robbiati et al. (2014a, 2017). Moreover, we propose the following hypotheses: (1) the evolutionary history of *Aphyllae* was influenced by geological and climatic changes that occurred during Pliocene and Pleistocene (2) reduction in shrub size, change in pattern of branch thickening and branching pattern favored colonization of new habitats. The reduction in shrub size, the thickening of the branches and the existence of different branching pattern were interpreted as adaptations to extreme environmental condition and thus treated as "xerophytic traits" (Bravo 1978a; Robbiati et al. 2017).

To explore these hypotheses, we carried out molecular phylogenetic analyses to infer relationships within series *Aphyllae* and to provide a framework for subsequent biogeographic and character evolution analyses. These analyses reflect the evolutionary history of series *Aphyllae*, including possible scenarios for its lineage diversification and morphological evolution. Finally, we compare our results with other studies that attempted to unravel the link between aridification paleoevents and the evolutionary history of plants in extant arid lands.

Materials and method

Taxonomic sampling

A total of 23 accessions (15 taxa, one to six accessions per taxon) were used in this study. The ingroup contained 22 accessions representing the most recent taxonomic proposal of seven species and three varieties belonging to the *Aphyllae* series (Robbiati et al. 2013, 2014a, b, 2017). The outgroup included *Senna chloroclada* (Harms) H.S.Irwin & Barneby, which was selected based on previous phylogenetic analyses (Marazzi et al. 2006; Marazzi and Sanderson 2010). Voucher information and GenBank accession numbers are provided in Online Resource 1 (Electronic Supplementary Material).

DNA extraction, amplification and sequencing

Total genomic DNA was extracted either from silica-dried field-collected material or from herbarium material using cetyltrimethylammonium bromide (CTAB) (Doyle and Doyle 1987). Four plastid DNA regions, including the first three originally used by Marazzi et al. (2006) and Marazzi and Sanderson (2010), *rpL16*, *rpS16*, *mat*K and *trn*L-F; and one nuclear region (ITS) were selected and PCR-amplified in 50 µL reaction solutions with Gotaq GreenMasterMix (3 mM MgCl$_2$), following the manufacturer's instructions. The following primers were used: F71 and R1516 to amplify a partial sequence of the rpL16 intron; primers rpsF and rpsR2 to amplify the complete rpS16 intron, primers matK3R and matK3F to amplify the central portion of the matK gene, primers B49873 and A50272 to amplify the intergenic spacer between the trnL (UAA) 3′ exon and trnF (GAA) 5′, primer ITS4 and ITS5 to amplify the ITS region. Primer sequences are listed in Online Resource 2. Sequencing was performed for two DNA strands. The PCR products were visualized under UV light after electrophoretic separation on a 1% agarose Tris/Borate/EDTA gel and staining with SYBR Safe gel stain (Invitrogen, Carlsbad, California, USA). Amplified products were sent to Macrogen Inc. (Seoul, South Korea) for purification and sequencing.

Phylogenetic reconstructions

Sequences were assembled and edited using BioEdit ver. 7.0.9. The sequences generated and GenBank sequences were pre-aligned in Muscle (Edgar 2004), implemented in MEGA5 (Tamura et al. 2011) and manually adjusted. To infer phylogenetic relationships we used three different approaches: Bayesian inference, parsimony and maximum likelihood. Nucleotide substitution models for Bayesian analyses were evaluated for each DNA regions separately, combined plastid and total evidence matrix (outgroup + ingroup) in jModelTest ver. 2.1.3 (Posada 2008), using the Akaike information criterion (AIC; Akaike 1974) and default search values. The best-fit models of evolution obtained are shown in Table 1. To evaluate congruence between the plastid regions and ITS data, two Bayesian analyses were run in MrBayes ver. 3.1.2 (Huelsenbeck and Ronquist 2001), one with plastid combined regions and other with ITS data. With the resulting trees we calculated the Incongruence Index, computed based on the size of their maximum agreement sub-tree and comparing its value to a null-hypothesis distribution obtained by simulation of random trees (de Vienne et al. 2007). Further, the trees also were visually evaluated to detect incongruence. For Bayesian inference (BI) three partitioning schemes were evaluated using the Bayes factor: ITS + combined plastid DNA using two molecular evolution models, ITS + partitioned plastid DNA using a molecular evolution model for each region, and all combined regions using a single molecular evolution model. The Bayes factor assesses the relative support for models using information theoretic criteria (Kass and Raftery 1995). Bayes factors can be estimated as a twice the difference in the harmonic mean of marginal lnL scores from Bayesian MCMC sampling (Newton and Raftery 1994). Interpretation of Bayes factors was based on the following guidelines, for $2\ln B_{ab}$: <0 support for M_b; 0–2 difference barely worth mentioning; 2–5 positive support for M_a; 5–10 strong support for M_a; >10 very strong support for M_a (Raftery 1995). The harmonic means were calculated using the 'sump' command in MrBayes. The Bayesian phylogenetic inference consisted of 2 MCMC runs and a Metropolis coupling of four chains (one cold and three heated), sampling every 1000 generations. Total number of generations was 10 million with a burn-in of the first 25% generations. Chain convergence was evaluated with Tracer ver. 1.5 (Rambaut and Drummond 2009). The posterior probabilities (PP) of clades were determined by a 50% majority rule consensus of the trees retained. parsimony analyses were performed in TNT ver. 1.5 (Goloboff et al. 2008), using the following specifications (all combined regions): 5000 replicates with random sequence addition, tree bisection–reconnection (TBR) branch swapping, keeping all trees found during branch swapping and other parameters using the default options. Branch support was estimated using bootstrapping with 1000 replicates, ten random addition sequence replicates with a maximum of ten trees saved per replicate, and all other settings used as in the heuristic search. For maximum likelihood (ML) analysis, the program randomized axelerated maximum likelihood (RAxML) ver. 8.1.11 (Stamatakis 2014) was used, which implements a rapid hill-climbing algorithm (Stamatakis 2006). Analyses were run for the best-scoring ML tree inferences under the GTR-GAMMA model. Rapid bootstrapping was performed with 1000 replications using the GTRCAT estimation to assess branch support (Stamatakis 2006). This analysis was run in the Cipres Gates using RAxML-HPC ver. 8 on XSEDE (8.2.4) tool.

Divergence times estimation

The Bayesian inference approach was used to estimate divergence times with combined chloroplast and nuclear data that was implemented in the package BEAST ver. 1.8.2 (Drummond et al. 2012). The substitution and clock model were unlinked, while the tree model was linked. To run this analysis selected representatives of all *Senna* clades of the matrix constructed by Marazzi and Sanderson (2010; see Online Resource 1) were chosen. *Cassia fistula* L. was selected as outgroup to root the trees. We used an uncorrelated relaxed molecular clock with a log-normal distribution of rates, and Yule speciation model (Yule 1925; Gernhard 2008), and the respective prior. Calibration points were obtained by age constraints derived from one fossil (calibration *N* in Bruneau et al. 2008) and the age estimates from *Senna* stem node by Marazzi and Sanderson (2010). The stem node of the *Senna* clade was constrained

Table 1 Summary of four plastid regions and nuclear ribosomal DNA (ITS) used in this study

	rpL16	*rpS16*	*mat*K	*trn*L-F	ITS	Combined plastid + ITS
Number of taxa	23	23	23	21	20	23
Number of base pairs	867	828	620	442	691	3448
Selected model of evolution, Akaike information criterion (AIC)	TPM3 + G	TPM1 + I	TPM1uf	TPM3uf	GTR + G	TIM3 + G

using a normal prior distribution (mean = 47.62, standard deviation = 2, Million years = My), and the *Senna* crown node was constrained using a exponential prior distribution (mean = 45, standard deviation = 2 My). Moreover, the *Aphyllae* series was constrained as monophyletic, based on previous results with a comprehensive taxonomic sampling (Marazzi et al. 2006; Marazzi and Sanderson 2010). Analyses consisted of two independent MCMC runs of 20 million generations, sampling every 2000 generations. To assess the ESSs (200 >), convergence and confidence intervals of each run, the program Tracer ver. 1.5 (Rambaut and Drummond 2009) was used, and then runs were combined after excluding burn-in of the initial twenty-five percent of trees (2500 trees) in Logcombiner ver. 1.8.2. To summarize probable trees and obtain a maximum clade credibility (MCC) tree, we used Tree Annotator ver. 1.8.2 included in the BEAST package (Drummond et al. 2012), with the posterior probability (PP) limit set to 0 and including mean node heights. Consensus trees (i.e., maximum clade credibility tree, MCC), PP and 95% highest probability density (HPD) intervals derived from the combined post-burn-in trees from independent runs were analyzed with TreeAnnotator ver. 1.8.2 and visualized with FigTree ver. 1.4.2.

Area reconstruction

To investigate the historical biogeography of series *Aphyllae* we reconstructed the ancestral area on phylogenetic trees using the statistical dispersal–vicariance analysis (S-DIVA) and Dispersal–Extinction–Cladogenesis (DEC). Both analyses were implemented in the Reconstruct Ancestral State in Phylogenies (RASP) program ver. 3.2 (Yu et al. 2015). We coded 9 areas based on the current distribution of *Aphyllae* and its outgroup, in the biogeographic regions proposed by Morrone (2006) for America: (A) the South American Transition Zone; (B) the Chaco province (C) the Central Patagonian province, (D) the Chiapas province, (E) Caatinga province, (F) Cerrado provinces, (G) Brazilian Atlantic Forest province, (H) Parana forest province, (I) Yungas province. For the taxa that inhabit non-American lands we used a biogeographic classification made by Cabrera and Willink (1980): (J) Eastern and (K) Australian region. To account for phylogenetic uncertainty in the statistical analyses S-DIVA and DEC ancestral areas were optimized on each 15,000 sampled post-burn-in tree from the BEAST runs. Results were plotted in the consensus tree of the dating analysis.

Ancestral character state reconstructions

To explore the evolution of xerophytic traits we analyzed the phylogenetic distribution of three morphological features selected according to taxonomic and ecological importance (Bravo 1978a; Robbiati et al. 2014a, b). Traits for ancestral character state reconstruction were coded, considering that the pattern of branch thickening is not homologous with branching pattern, because the two characters are independent from each other. We coded them into the following discrete, multistate characters: (A) shrub size: 0 = tall shrubs (up to 3 m height), 1 = median shrubs (up to 2 m height), 2 = subshrubs (up to 0.50 m height); (B) pattern of branch thickening: 0 = not thickened, 1 = thickened in the central part, 2 = uniformly thickened; (C) branching pattern: 0 = straight, 1 = decumbent–subdecumbent, 2 = fastigiate, 3 = pendulous, 4 = intricate-ramose. Character states were scored based on herbarium specimens. These analyses were developed for the combined matrix of 11 taxa with *Senna* members of clade VI of this study (i.e., *S. kurtzii* (Harms) H.S.Irwin & Barneby, *S. holwayana* (Rose) H.S.Irwin & Barneby, *S. tonduzii* (Standl.) H.S.Irwin & Barneby and all species of series *Aphyllae*). To reconstruct ancestral state we used Mesquite ver. 2.7.5 (Maddison and Maddison 2011), using Likelihood ancestral state as a reconstruction method with ML criterion (Mk1 model) to trace the character history over the majority rule tree and over 1000 random bayesian trees obtained of 8061 post-burn-in trees from the Bayesian inference run in MrBayes (GTR + G model) to account for phylogenetic uncertainty.

Results

Phylogenetic analyses

Despite repeated efforts to optimize PCR conditions, we were unable to generate sequences for *S. nudicaulis*. The Incongruence Index (Icong = 1.53, *P* value = 0.0006) suggested that the plastid DNA and nrDNA are more congruent than expected by chance. Support for the partition scheme (ITS + partitioned plastid DNA (harmonic mean lnL = −7347.54) provided the best fit in combined analyses, compared to total evidence (harmonic mean lnL = −7577.17, B_{21} = 459.26) or second partition scheme (ITS + combined plastid DNA: harmonic mean lnL = −7443.05, B_{23} = 191.02). Based on these results we chose to use the ITS + partitioned plastid DNA partition scheme to run the Bayesian analysis. The alignment of the 23 accession dataset consisted of 3448 aligned positions (Table 1 and Online Resource 3). Bayesian and ML inferences recovered the series *Aphyllae* as monophyletic (BS = 94, PP = 0.99; Fig. 3). In this series two main clades were resolved (BS = 100, PP = 1), Clade A formed by *Senna aphylla* var. *aphylla*, *S. crassiramea*, *S. rigidicaulis* and *S. spiniflora* and Clade B formed by *S. aphylla* var.
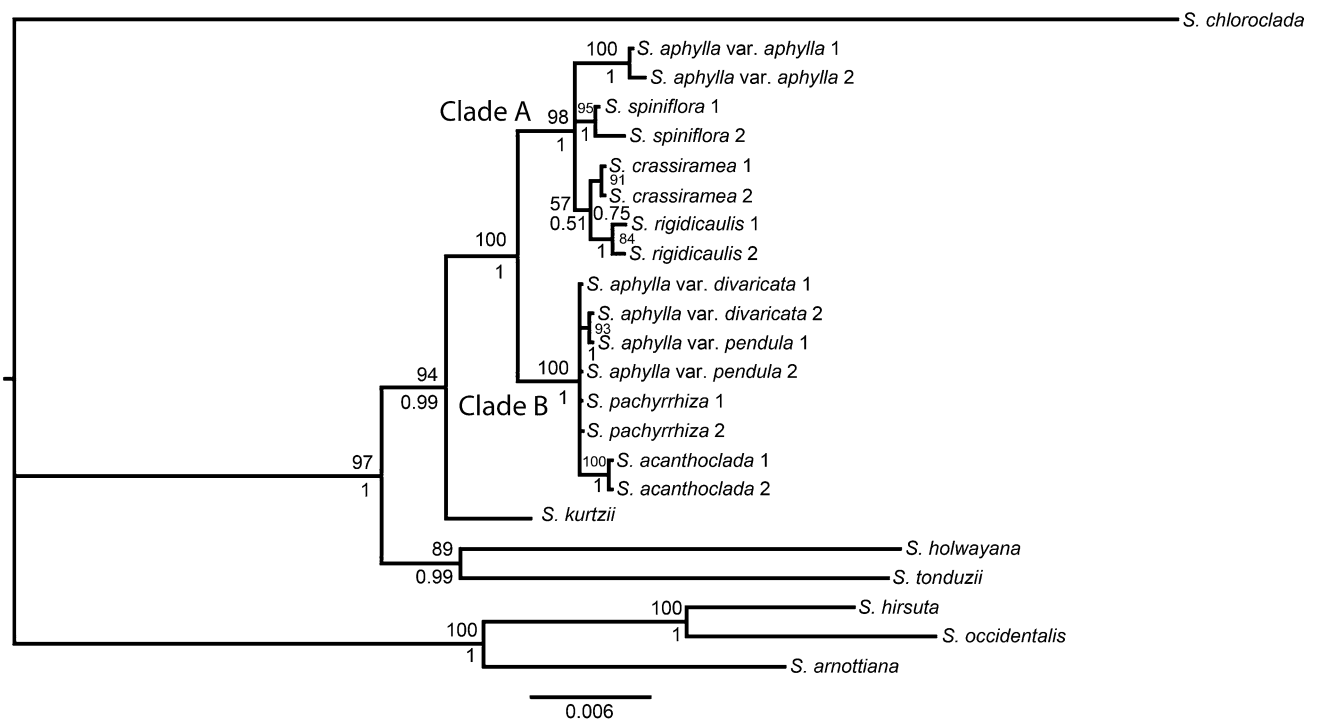
Fig. 3 Fifty percent majority rule Bayesian consensus of the molecular phylogenetic analyses of combined ITS and plastid DNA sequences showing relationships of *Senna* ser. *Aphyllae*. Posterior probabilities are indicated below the branches. Bootstrap supports are indicated above the branches. Clade A includes *S. aphylla* var. *aphylla*, *S. spiniflora*, *S. crassiramea* and *S. rigidicaulis*, and Clade B includes *S. acanthoclada*, *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*

divaricata, *S. aphylla* var. *pendula*, *S. pachyrrhiza* and *S. acanthoclada*. The main diagnostic characteristic of species in the Clade A is the thickened branches. These analyses showed *S. aphylla* as non-monophyletic taxon. In the Clade B *S. acanthoclada* appears to be more closely related to *aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*, and these three taxa formed a polytomy. Representatives of clade are characterized by the presence of not thickened branch. *Senna crassiramea* and *S. rigidicaulis* were retrieved as a monophyletic group but with low support (BS = 57; PP = 0.51). The strict consensus tree resulting from parsimony analyses shows the similar topology as the 50% majority rule consensus tree resulting from BI and ML, nodes of Clade A and B were strongly supported (>85% BS), but *S. crassiramea* and *S. rigidicaulis* were not retrieved together (Online Resource 4).

Divergence times estimation

The results of the *Aphyllae* divergence time estimation are presented in Fig. 4. Given the polytomy obtained in Clade B for *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*, we selected two representative sequences of this group. This analysis shows that *Aphyllae* diverged from its sister taxon in the middle Miocene, approximately

10.81 My (95% HPD 6.58–15.7 My), and the diversification of the two clades began in the early Pliocene, approximately 4.55 My (95% HPD 2.59–6.78 My). The divergence between *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. acanthoclada* began in the mid-Pleistocene, approximately 1.26 My (95% HPD 0.49–3.65 My). The divergence of Clade A began 3.48 My (95% HPD 1.68–4.72 My).

Ancestral area reconstruction

The reconstructions of S-DIVA are shown in Fig. 5 and Online Resource 5. The S-DIVA and DEC analyses displayed current distribution of series *Aphyllae* was formed by three dispersal, one vicariance and five dispersals, one vicariance events, respectively. The only vicariance event reported by both analyses occurred in the node of ancestor of *S. spiniflora* and *S. crassiramea-S. rigidicaulis* clade. The ancestral geographic ranges obtained by S-DIVA analysis suggest that *Aphyllae* originated in SATZ (A 100; PP = 0.79). This analysis showed that the putative ancestor of Clade A occurred in both SATZ and the Chaco province (AB 100; PP = 0.79) and the putative ancestor of Clade B occurred in SATZ (A 100; PP = 100). The ancestral area reconstruction obtained by DEC analysis suggests that *Aphyllae* probably inhabited
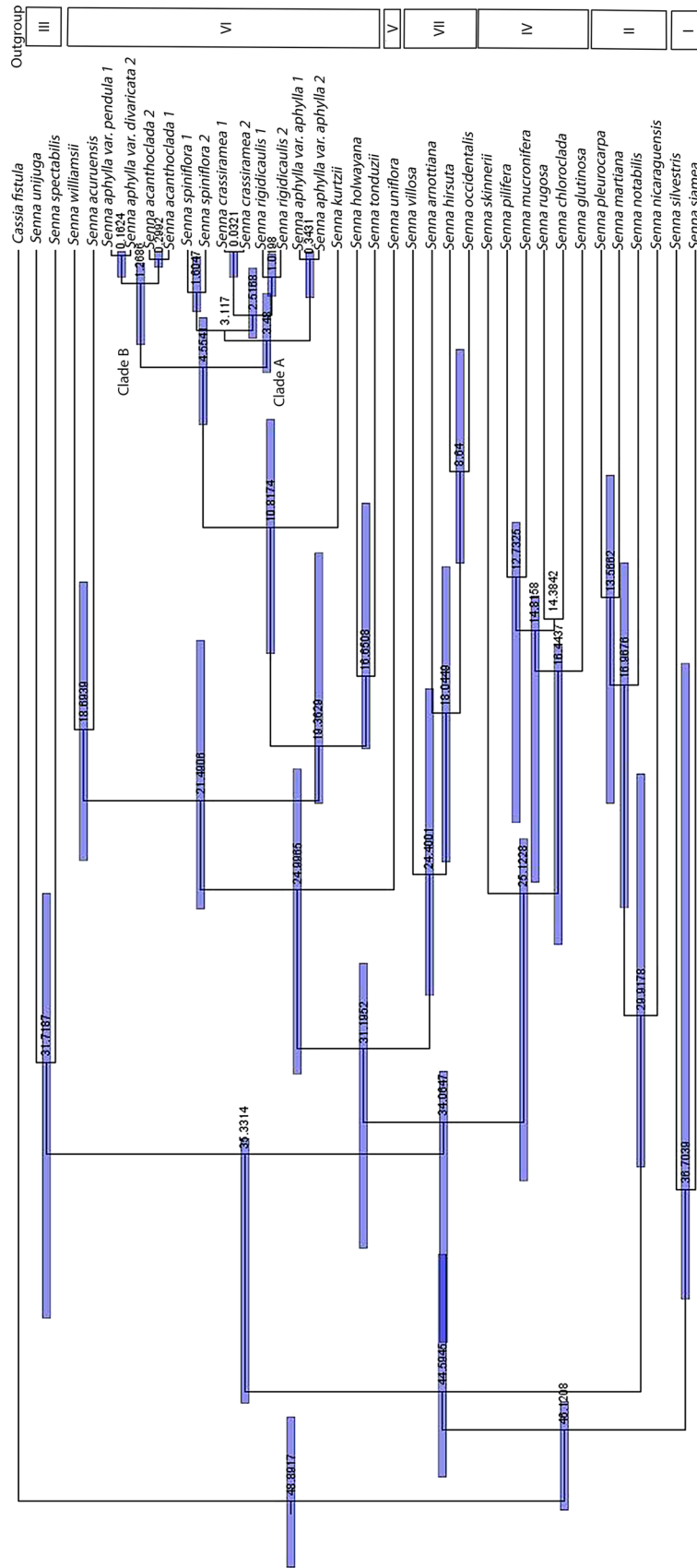
Fig. 4 BEAST chronogram of the combined dataset of representatives of *Senna*. Estimated ages are presented in millions of years (My), and 95% confidence intervals are given at the nodes. Clades *I–VII* represent the big clades retrieved by Marazzi and Sanderson (2010) for genus *Senna*. Clade A includes *S. aphylla* var. *aphylla*, *S. spiniflora*, *S. crassiramea* and *S. rigidicaulis*, and Clade B includes *S. acanthoclada*, *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*

Fig. 5 Statistical dispersal–vicariance analysis (S-DIVA) reconstruction of ancestral distributions for *Senna* ser. *Aphyllae*, indicated by the pies with *different colors* and proportions. Current distributions are indicated before the species names
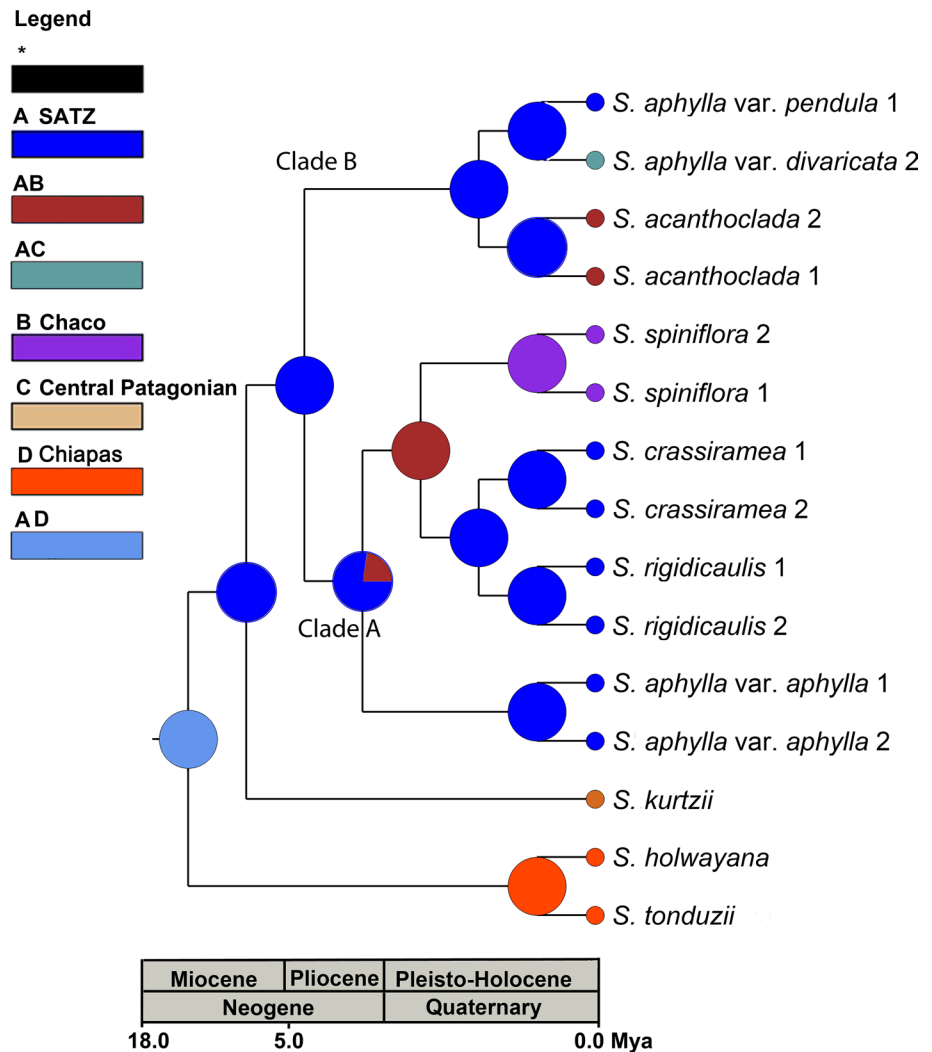
SATZ or SATZ and the Chaco province, with low probability (A 53.25, AB 46.75; PP < 0.5). According to this analysis the putative ancestors of Clade A probably occurred in SATZ and in Chaco province or in SATZ but with low probability (AB 54.21, A 45.49; PP < 0.5). The putative ancestor of Clade B probably inhabited SATZ or SATZ and Chaco or SATZ and Central Patagonian province, with low probability (A 47.77, AB 35.35, AC 16.88; PP < 0.5, respectively, DEC results were not shown in the tree).

Ancestral character state reconstructions

The putative *Aphyllae* ancestor was inferred as a tall or median shrubs (PP = 0.46, PP = 0.39; in the reconstruction over 1000 trees no states reported for this node), with no thickened branches (PP = 0.99; in reconstruction over 1000 all trees estimated this state) and straight branches (P = 0.68; in reconstruction over 1000 only 182 trees estimated this state). The putative ancestor of *S.*

aphylla var. *aphylla*, *S. crassiramea*, *S. rigidicaulis* and *S. spiniflora* is inferred as a tall shrub form (PP = 0.90; in reconstruction over 1000 trees no states reported for this node), with no thickened branches (PP = 0.92; in reconstruction over 1000 all trees estimated this state) and straight branches (PP = 0.64; in reconstruction over 1000 only 316 trees estimated this state). The ancestor of *S. crassiramea* and *S. rigidicaulis* is inferred as a tall shrubs form (PP = 0.77; in reconstruction over 1000 trees no states reported for this node), with branches thickened in the central part (PP = 0.96; in reconstruction over 1000 all trees estimated this state) and fastigiated branches (PP = 0.91; in reconstruction over 1000 all trees estimated this state). The putative ancestor of *S. acanthoclada*, *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza* is inferred as a median shrub and subshrub forms (PP = 0.59, PP = 0.40; in reconstruction over 1000 all trees estimated this state), not thickened branch (PP = 1; in reconstruction over 1000 all trees estimated this state) and decumbent–subdecumbent
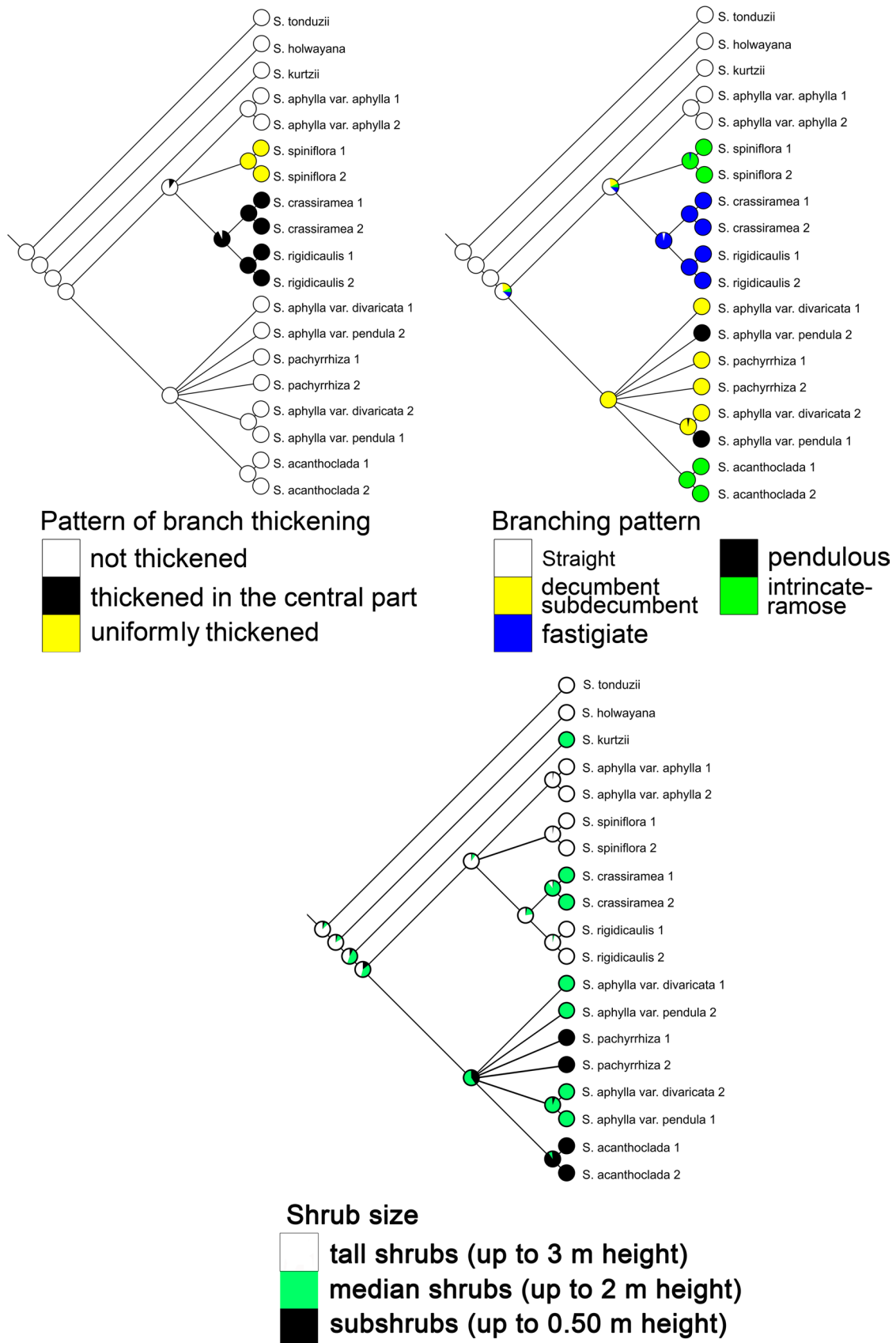
Fig. 6 Maximum likelihood ancestral character state reconstructions of xerophytic traits in *Senna* ser. *Aphyllae*; *pie diagrams* indicate the probability of each ancestral character state

branch (PP = 0.99; in reconstruction over 1000 only 306 trees estimated this state; Fig. 6).

Discussion

Phylogenetic relationships within series *Aphyllae*

The present phylogenetic analysis including all species of the series *Aphyllae* (except *Senna nudicaulis*) is the first one to elucidate phylogenetic relationships in this clade of highly xerophytic plants from the arid and semiarid regions of southern South America. Our analysis confirmed the monophyly of the series (Marazzi et al. 2006; Marazzi and Sanderson 2010) and inferred two main clades: Clade A, including *S. aphylla* var. *aphylla*, *S. spiniflora*, *S. crassiramea* and *S. rigidicaulis*, and Clade B including *S. acanthoclada*, *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*.

One unexpected result is that *S. aphylla* is paraphyletic. Although *S. aphylla* var. *aphylla* (in Clade A) is similar to the other varieties (all in Clade B), this taxon differs because of the presence of straight and armed branches and a glabrous to densely pubescent calyx (all others have pendulous or decumbent unarmed branches, and the calyx is mainly glabrous). Because *S. aphylla* s.l. does not appear as an independent single evolutionary unit, the morphological similarity among varieties could be an example of morphological convergence or a cryptic speciation that have confused taxonomists (Bickford et al. 2007). Non-monophyly of *S. aphylla* could reflect some underlying evolutionary mechanism shaping the relationships within *Aphyllae*, such as incomplete lineage sorting (Pamilo and Nei 1988; Maddison 1997), reticulate evolution (hybridization and introgression; Wendel and Doyle 1998) and/or peripatric speciation (Mayr 1982). Nevertheless, a broad taxonomic and geographic sampling is required to confirm the monophyly of *S. aphylla*. Relationships between *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza* remain unresolved. The lack of resolution is also observed phenotypically, since morphological and multivariate analysis (Robbiati et al. 2017) reveals no significant phenotypic differences among *S. aphylla* var. *divaricata* and *S. pachyrrhiza*, suggesting that *S. pachyrrhiza* may be part of the *S. aphylla* lineage in Clade B or at least it would confirm its close relatedness (both taxa have non-thickened branches, up to 4 mm in diameter). Close morphological similarity can be attributed to either early stages of lineage differentiation or to gene flow between the entities in question (Popp et al. 2005; Schmidt-Lebuhn et al. 2012; Nylinder et al. 2013). Interestingly, *S. pachyrrhiza* was in fact most likely recognized as a separated taxon because of its peculiar geographic distribution. It was described as endemic

of the "Campos del Arenal" located in the highlands of the Monte province in SATZ (Bravo 1978a). This area is an extensive plain of about 2000 km², delimited by a group of mountains belonging to the "Sierras Pampeanas" system. Nores (1986) has suggested that the isolation of this area could function as an ecological island. Given the geographic isolation of *S. pachyrrhiza* in "Campos del Arenal," analysis of any gene flow between *S. aphylla* and *S. pachyrrhiza* would thus be necessary to verify if *S. pachyrrhiza* represents a recent endemism since it may be an example of peripatric speciation. Several studies in Andean South American plants have suggested that the predominant speciation mode is allopatric (Berry 1982; Molau 1988; Norman 2000).

Another unexpected result is the close relationship of *S. acanthoclada* with *S. aphylla* var. *divaricata*, *S. aphylla* var. *pendula* and *S. pachyrrhiza*. *Senna acanthoclada* is morphologically different, due to petals pubescence, and intricate–branches with spiny apices. This species inhabits salty soils mainly in southern–western Chaco and Monte province; in this last biogeographic region distribution of *S. acanthoclada* and *S. aphylla* overlap. Nevertheless, no intermediate individuals were observed either in herbarium or in the field (Robbiati et al. 2017). This means that it is a good species; however, it is a difficult task to find sufficient molecular variation which usually requires large amounts of data (Hughes et al. 2006).

Relationships among the other species also deserve a closer look. On the one hand, *S. crassiramea* and *S. rigidicaulis* appear as sister taxa in some of our analyses (BI and ML, but not MP), but with little support. On the other hand, this sister relationship is actually supported by morphological similarities, especially because both species have fastigiated and thickened branches. In addition, both species share a similar geographic distribution in SATZ, mainly occur in the Prepuna province, with partly overlapping distribution (Robbiati et al. 2017). Based on morphological analysis and geographic distribution, Robbiati et al. (2017) suggested that these entities could represent a single species, finding several populations with intermediate morphological characteristics. Given this intermediacy both taxa could represent a potential hybrid complex; however, further studies will be needed to test these hypotheses before making any taxonomic decisions about their status. On the other hand, *S. spiniflora* appears a close relative of *S. aphylla* var. *aphylla*, *S. crassiramea* and *S. rigidicaulis*, but S. *spiniflora* is morphologically distinct because of its uniformly thickened and intricate–branchy branches (instead of non-thickened or thickened in the central part and straight or fastigiated branches as *S. aphylla* var. *aphylla*, *S. crassiramea* and *S. rigidicaulis*). The distribution patterns of *S. spiniflora* in the xerophilous forest in Chaco province

suggested that it might have originated through parapatric speciation.

Evolutionary history of series *Aphyllae*

Our results suggest that the series *Aphyllae* lineage originated during the diversification of the major clades of *Senna*, which appeared during mid-Eocene to early Oligocene and started diversifying during the early Oligocene to early Miocene (Marazzi and Sanderson 2010).

Our findings indicate that the crown clade of series *Aphyllae* probably originated in SATZ during the early Pliocene with a mean age 4.5 My, while Clades A and B diverged during middle Pliocene-early Pleistocene around 3.4–1.8 My in the same geographic zone. In this scenario, *Aphyllae* appeared and diverged when the Andean peaks were already formed, the eastern orographic systems of Argentina (e.g., Puna, Pampean Mountain Range) were settled (Ortiz-Jaureguizar and Cladera 2006), and present arid and semiarid conditions were established (Kleinert and Strecker 2001). This meant a drastic reduction in precipitation levels and landscape change in the eastern side of the Andes that had important ecological consequences promoting habitat fragmentation, inter- and intraspecific differentiation and forced many tropical and subtropical lineages to adapt to the new conditions (Pascual et al. 1996; Alberdi et al. 1997; Richardson et al. 2001; López 2003; Hughes and Eastwood 2006; Antonelli et al. 2009; Pennington and Dick 2010; Baranzelli et al. 2014; Amarilla et al. 2015). Moreover, during Pleistocene, the cyclical advance and retreat of glaciers produced notable dimension changes of arid and semiarid landscapes permitting opportunities for plants living in these habitats (Marshall and Cifelli 1990).

Several studies about the evolutionary history of different taxa that inhabit in arid land of southern South America suggest that aridization processes in South America are strongly associated with their diversification and evolutionary history (Jakob et al. 2009; Schmidt-Jabaily and Sytsma 2010; Viruel et al. 2012; Ossa et al. 2013; Baranzelli et al. 2014; Amarilla et al. 2015; Sanín et al. 2016).

Our results suggest that the arid and semiarid habitats available in southern South America when *Aphyllae* diverged could have favoured diversification and geographic and ecological expansion of series *Aphyllae*. Several studies support the hypothesis that dry-adapted taxa diverged concomitantly with the expansion of arid environments in different arid regions of the world (Richardson et al. 2001; Verboom et al. 2003; Good-Avila et al. 2006; Moore and Jansen 2007; Catalano et al. 2008).

Conversely, the fact that *S. spiniflora* occurs exclusively in the xerophilous forest of the Chaco province supports the hypothesis that the Chaco flora is a Tertiary or early Pleistocene relict established in salty soils (Iriondo 1993; Spichiger et al. 2004; Caetano et al. 2008; Werneck 2011).

Evolution of xerophytic traits as adaptations to arid habitats

Plants that inhabit semiarid and arid regions developed special morphological traits in the process of adaptation to the environmental stress (Ward 2009). The most important morphological xerophytic adaptation in the series *Aphyllae* is the loss of leaves in adult plants (Burkart 1952; Bravo 1978a; Irwin and Barneby 1982; Robbiati et al. 2017). Our results suggest that the putative ancestor of series *Aphyllae* was distributed over SATZ and might have been a tall or medium-sized shrub, with non-thickened, straight branches, similarly to three extant taxa of the series *Aphyllae* that retained these features: *S. aphylla* var. *aphylla*, *S. rigidicaulis* and *S. spiniflora*. Instead, the remaining taxa became smaller. Nevertheless, the adaptive value of plant size in *Aphyllae* is unclear (if relevant at all), since we observed no correlation with other xeromorphic traits and an uncertain position in the phylogenetic analysis.

The putative ancestor of Clade A was distributed in SATZ and Chaco and had non-thickened branches. *Senna crassiramea* and *S. rigidicaulis* mainly occur in the Prepuna province in SATZ, growing in mounted slopes with rocky soils and *S. spiniflora* occurs in Chaco province growing mainly in dry and salty soils and the three taxa have thickened branches (Robbiati et al. 2014b, 2017). Our results further suggest that the acquisition of thickened branches may have favored the establishment of *S. crassiramea*, *S. rigidicaulis* and *S. spiniflora* in drier and saltier soils that those occupied by the other species. This phenomenon of branch thickening has been observed as adaptive response to arid and salty environments in other plants (Eamus and Palmer 2007).

Our results also revealed the fastigiated condition as an acquired phenotype in *S. crassiramea* and *S. rigidicaulis*. Their branch features may have played an important role in order to colonize and establish in the arid environment of Prepuna province in SATZ. Changes in the angle of plant canopy and branching architecture have been reported as adaptive responses to water stress in other plants (Neufeld et al. 1988; Hesp 1991; De Soyza et al. 1997).

The putative ancestor of Clade B was a medium-sized shrub or subshrub, with non-thickened, decumbent to sub-decumbent branches. It is possible that it occurred in sandbanks patches in arid regions, given the fact that extant taxa that inherited these habit characteristics (*S. aphylla* var. *divaricata* and *S. pachyrrhiza* grow in sandy soils).

In conclusion, our analyses found that the series *Aphyllae* consists of two main clades, but phylogenetic

relationships within these clades remain unresolved. Future cytogenetic, phylogeographic and molecular population studies will be required to clarify the systematics of this group. Our study provides a solid example illustrating the impact the Andean uplift and aridization had in the evolution of South American floras. It remains unclear as to whether the orogenetic events and aridization of southern South America could have provided new opportunities for the geographic and ecological expansion of series *Aphyllae*. Moreover, other evolutionary forces such as polyploidy and hybridization processes could have driven *Aphyllae* evolution; the existence of different levels of ploidy could have conferred an adaptive advantage to colonize or stabilize in unfavorable environments. Nevertheless, the loss of leaves and adaptations in branching patterns and branch morphology are the most conspicuous xerophytic features that evolved in this group as a response to the different arid and semiarid conditions in southern South America.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Information on Electronic Supplementary Material

Online Resource 1. Biogeographic distribution, voucher information and Genbank accessions of the samples used in this study. Bold code represents generated sequences in this work. Bold taxa are those used in phylogenetic analyses.

Online Resource 2. Primer sequences for amplification and sequencing of ITS and plastid DNA regions used in this study.

Online Resource 3. Alignment used to produce phylogeny.

Online Resource 4. Phylogenetic tree obtained from parsimony analysis of combined ITS and plastid DNA sequences showing relationships of *Senna* ser. *Aphyllae*.

Online Resource 5. Graphical representation of ancestral distributions for genus *Senna* obtained by S-DIVA analysis, indicated by the pies with different colors and proportions.

References

Akaike H (1974) A new look at the statistical model identification. IEEE Trans Automat Contr 19:716–723. doi:10.1109/TAC.1974.1100705

Alberdi MT, Bonadonna FP, Ortiz-Jaureguizar E (1997) Chronological correlation, paleoecology, and paleobiogeography of the late Cenozoic South American Rionegran land-mammal fauna: a review. Rev Esp Paleontol 12:249–255

Amarilla LD, Anton AM, Chiapella JO, Manifiesto MM, Angulo DF, Victoria S (2015) *Munroa argentina*, a grass of the South American Transition Zone, survived the Andean uplift, aridification and glaciations of the quaternary. PLoS ONE 10:e0128559. doi:10.1371/journal.pone.0128559

Antonelli A, Nylander JAA, Persson C, Sanmartín I (2009) Tracing the impact of the Andean uplift on neotropical plant evolution. Proc Natl Acad Sci USA 106:9749–9754. doi:10.1073/pnas.0811421106

Ari I, Artabe AE, Morel EM (2011) The evolution of Patagonian climate and vegetation from the Mesozoic to the present. Bot J Linn Soc 103:409–422. doi:10.1111/j.1095-8312.2011.01657.x

Baranzelli MC, Johnson LA, Cosacov A, Sérsic AN (2014) Historical and ecological divergence among populations of *Monttea chilensis* (Plantaginaceae), an endemic endangered shrub bordering the Atacama Desert, Chile. Evol Ecol 28:751–774. doi:10.1007/s10682-014-9694-y

Bell CD, Donoghue MJ (2005) Phylogeny and biogeography of Valerianaceae (Dipsacales) with special reference to the South American valerians. Organisms Diversity Evol 5:147–159

Berry PE (1982) The systematics and evolution of *Fuchsia* sect. *Fuchsia* (Onagraceae). Ann Missouri Bot Gard 69:1–198

Bickford D, Lohman DJ, Sodhi NS, Ng PK, Meier R, Winker K, Ingram KK, Das I (2007) Cryptic species as a window on diversity and conservation. Trends Ecol Evol 22:148–155. doi:10.1016/j.tree.2006.11.004

Bravo LD (1978a) El género *Cassia* en la Argentina, I. Serie Aphyllae. Darwiniana 21:43–391

Bravo LD (1978b) Estudio comparativo de las plántulas de las subespecies de *Cassia aphylla* Cav. Darwiniana 21:393–399

Bravo LD (1982) Estudio de las semillas del genero *Cassia,* Serie Aphyllae. Darwiniana 24:455–468

Bravo LD, Agulló M, Palacios R (1986) Additional notes on *Senna crassiramea*, *S. rigidicaulis*, and seeds of series *Aphyllae* (Caesalpiniaceae). Brittonia 38:269–272. doi:10.2307/2807354

Bruneau A, Mercure M, Lewis GP, Herendeen PS (2008) Phylogenetic patterns and diversification in the caesalpinioid legumes. Botany 86:697–718. doi:10.1111/j.1558-5646.2010.01086.x

Burkart A (1952) *Cassia*. In: Burkart A (ed) Las Leguminosas argentinas silvestres y cultivadas II. Acme Agency, Buenos Aires, pp 162–169

Cabrera AL, Willink A (1980) Biogeografía de América Latina. Secretaría General de la Organización de los Estados Americanos (OEA), Washington DC

Caetano S, Prado D, Pennington RT, Beck S, Oliveira-Filho A, Spichiger R, Naciri Y (2008) The history of seasonally dry tropical forests in eastern South America: inferences from the genetic structure of the tree *Astronium urundeuva* (Anacardiaceae). Molec Ecol 17:3147–3159. doi:10.1111/j.1365-294X.2008.03817.x

Catalano SA, Vilardi JC, Tosto D, Saidman BO (2008) Molecular phylogeny and diversification history of *Prosopis* (Fabaceae: mimosoideae). Biol J Linn Soc 93:621–640. doi:10.1111/j.1095-8312.2007.00907.x

De Soyza G, Whitford WG, Martinez-Meza E, van Zee JW (1997) Variation in creosote bush (*Larrea tridentata*) canopy morphology in relation to habitat, soil fertility and associated annual plant communities. Amer Midl Naturalist 137:13–26

de Vienne DM, Giraud T, Martin OC (2007) A congruence index for testing topological similarity between trees. Bioinformatics 23:3119–3124. doi:10.1093/bioinformatics/btm500

Doyle JJ, Doyle JA (1987) A rapid DNA isolation procedure for small quantities of fresh leaf tissue. Phytochem Bull 19:11–15. doi:10.1007/BF00986191

Drummond AJ, Suchard MA, Xie D, Rambaut A (2012) Bayesian phylogenetics with BEAUti and the BEAST 1.7. Molec Biol Evol 29:1969–1973. doi:10.1093/molbev/mss075

Eamus D, Palmer AR (2007) Is climate change a possible explanation for woody thickening in arid and semi-arid regions? Int J Ecol 2007:37364. doi:10.1155/2007/37364

Edgar RC (2004) MUSCLE: a multiple sequence alignment method with reduced time and space complexity. BMC Bioinform 5:113. doi:10.1186/1471-2105-5-113

Garzione CN, Hoke GD, Libarkin JC, Withers S, MacFadden B, Eiler J, Ghosh P, Mulch A (2008) Rise of the Andes. Science 320:1304–1307. doi:10.1126/science.1148615

Gernhard T (2008) The conditioned reconstructed process. J Theor Biol 253:769–778. doi:10.1016/j.jtbi.2008.04.005

Goloboff PA, Farris JS, Nixon KC (2008) TNT, a free program for phylogenetic analysis. Cladistics 24:774–786. doi:10.1111/j.1096-0031.2008.00217.x

Good-Avila SV, Souza V, Gaut BS, Eguiarte LE (2006) Timing and rate of speciation in *Agave* (Agavaceae). Proc Natl Acad Sci USA 103:9124–9129. doi:10.1073/pnas.0603312103

Gregory-Wodzicki KM (2000) Uplift history of the Central and Northern Andes: a review. Geol Soc Amer Bull 112:1091–1105. doi:10.1130/0016-7606(2000)112

Guerrero PC, Rosas M, Arroyo MTK, Wiens JJ (2013) Evolutionary lag times and recent origin of the biota of an ancient desert (Atacama–Sechura). Proc Natl Acad Sci USA 110:11469–11474. doi:10.1073/pnas.1308721110

Hartley AJ, Chong G (2002) Late Pliocene age for the Atacama Desert: implications for the desertification of western South America. Geology 30:43–46. doi:10.1130/0091-7613(2002)

Hesp PA (1991) Ecological processes and plant adaptations on coastal dunes. J Arid Environm 2:165–191

Hoorn C (1993) Marine incursions and the influence of Andean tectonics on the Miocene depositional history of northwestern Amazonia: results of a palynostratigraphic study. Palaeogeogr Palaeoclimatol Palaeoecol 105:267–309

Hoorn C, Guerrero J, Sarmiento GA, Lorente MA (1995) Andean tectonics as a cause of changing drainage patterns in Miocene northern South America. Geology 23:237–240

Hoorn C, Wesselingh FP, TerSteege H, Bermudez MA, Mora A, Sevink J, Sanmartín I, Sanchez-Meseguer A, Anderson CL, Figueiredo JP, Jaramillo C (2010) Amazonia through time: Andean uplift, climate change, landscape evolution and biodiversity. Science 330:927–993. doi:10.1126/science.1194585

Huelsenbeck JP, Ronquist F (2001) MRBAYES: bayesian inference of phylogenetic trees. Bioinformatics 17:754–755

Hughes C, Eastwood R (2006) Island radiation on a continental scale: exceptional rates of plant diversification after uplift of the Andes. Proc Natl Acad Sci USA 10:10334–10339. doi:10.1073/pnas.0601928103

Hughes CE, Eastwood RJ, Bailey DC (2006) From famine to feast? Selecting nuclear DNA sequence loci for plant species-level phylogeny reconstruction. Philos Trans Roy Soc London B Biol Sci 361:211–225. doi:10.1098/rstb.2005.1735

Iriondo M (1993) Geomorphology and late quaternary of the Chaco (South America). Geomorphology 7:289–303. doi:10.1016/0169-555X(93)90059-B

Irwin HS, Barneby RC (1982) The American Cassiinae: a synoptical revision of Leguminosae, tribe Cassiae subtribe Cassiinae in the New World. Mem New York Bot Gard 35:567–570

Jakob SS, Martinez-Meyer E, Blattner FR (2009) Phylogeographic analyses and paleodistribution modeling indicate Pleistocene in situ survival of *Hordeum* species (Poaceae) in southern Patagonia without genetic or spatial restriction. Molec Biol Evol 26:907–923. doi:10.1093/molbev/msp012

Kass RE, Raftery AE (1995) Bayes factors. J Amer Statist Assoc 90:773–795

Kleinert K, Strecker MR (2001) Climate change in response to orographic barrier uplift: paleosol and stable isotope evidence from the late Neogene Santa Maria basin, northwestern Argentina. Geol Soc Amer Bull 113:728–742. doi:10.1130/0016-7606

López RP (2003) Phytogeographical relations of the Andean dry valleys of Bolivia. J Biogeogr 30:1659–1668. doi:10.1046/j.1365-2699.2003.00919.x

Maddison WP (1997) Gene trees in species trees. Syst Biol 46:523–536. doi:10.1093/sysbio/46.3.523

Maddison WP, Maddison DR (2011) Mesquite: a modular system for evolutionary analysis, version 2.75. Available at: http://mesquite-project.org

Marazzi B, Sanderson MJ (2010) Large-scale patterns of diversification in the widespread legume genus *Senna* and the evolutionary role of extrafloral nectaries. Evolution 64:3570–3592. doi:10.1111/j.1558-5646.2010.01086.x

Marazzi B, Endress PK, de Queiroz LP, Conti E (2006) Phylogenetic relationships within *Senna* (Leguminosae, Cassiinae) based on three chloroplast DNA regions: patterns in the evolution of floral symmetry and extrafloral nectaries. Amer J Bot 93:288–303. doi:10.3732/ajb.93.2.288

Marshall LG, Cifelli RL (1990) Analysis of changing diversity patterns in Cenozoic land mammal age faunas South America. Palaeovertebrata 19:169–210

Mayr E (1982) Speciation and macroevolution. Evolution 36:119–1132

Molau U (1988) Scrophulariaceae, I: Calceolarieae. The New York Botanical Garden, Bronx

Moore MJ, Jansen RK (2007) Origins and biogeography of gypsophily in the Chihuahuan desert plant group *Tiquilia* subg. *Eddya* (Boraginaceae). Syst Bot 32:392–414. doi:10.1600/036364407781179680

Morrone JJ (2006) Biogeographic areas and the transition zones of Latin America and the Caribbean Islands based on panbiogeographic and cladistics analyses of the entomofauna. Annual Rev Entomol 5:467–494. doi:10.1146/annurev.ento.50.071803.130447

Neufeld HS, Meinzer FC, Wisdom CS, Sharifi MR, Rundel PW, Neufeld MS, Goldring Y, Cunningham GL (1988) Canopy architecture of *Larrea tridentate* (DC.) Cov., a desert shrub: foliage orientation and direct beam radiation interception. Oecologia 75:54–60

Newton MA, Raftery AE (1994) Approximate bayesian inference with the weighted likelihood bootstrap. J R Stat Soc B 56:3–48

Nores M (1986) Diez nuevas subespecies de aves provenientes de islas ecológicas argentinas. Hornero 12:262–273

Norman ME (2000) Buddlejaceae. Flora neotropica monograph 81. The New York Botanical Garden, Bronx

Nylinder S, Cronholm B, de Lange PJ, Walsh N, Anderberg AA (2013) Species tree phylogeny and character evolution in the genus *Centipeda* (Asteraceae): evidence from DNA sequences from coding and non-coding loci from the plastid and nuclear

genomes. Molec Phylogen Evol 68:239–250. doi:10.1016/j.ympev.2013.03.020

Ortiz-Jaureguizar E, Cladera GA (2006) Paleoenvironmental evolution of southern South America during the Cenozoic. J Arid Environm 66:498–532. doi:10.1016/j.jaridenv.2006.01.007

Ossa PG, Pérez F, Armesto JJ (2013) Phylogeography of two closely related species of Nolana from the coastal Atacama Desert of Chile: post-glacial population expansions in response to climate fluctuations. J Biogeogr 40:2191–2203. doi:10.1111/jbi.12152

Palazzesi L, Barreda VD, Cuitiño JL, Guler MV, Tellería MC, Ventura Santos R (2014) Fossil pollen records indicate that Patagonian desertification was not solely a consequence of Andean uplift. Nat Commun 5:3558. doi:10.1038/ncomms4558

Palma RE, Marquet PA, Boric-Bargetto D (2005) Inter- and intraspecific phylogeography of small mammals in the Atacama Desert and adjacent areas of northern Chile. J Biogeogr 32:1931–1941. doi:10.1111/j.1365-2699.2005.01349.x

Pamilo P, Nei M (1988) Relationships between gene trees and species trees. Molec Biol Evol 5:568–583

Pascual R, Ortiz-Jaureguizar E, Prado JL (1996) Land mammals: paradigm for Cenozoic South American geobiotic evolution. Münchner Geowiss Abh A 30:265–319

Pennington RT, Dick CW (2010) Diversification of the Amazonian flora and its relation to key geological and environmental events: a molecular perspective. In: Hoorn C, Wesselingh FP (eds) Amazonia, landscape and species evolution: a look into the past. Wiley-Blackwell, Oxford, pp 373–385

Pirie MD, Chatrou LW, Mols JB, Erkens RHJ, Oosterhof J (2006) Andean-centred genera in the short branch clade of Annonaceae: testing biogeographical hypothesis using phylogeny reconstruction and molecular dating. J Biogeogr 33:31–46. doi:10.1111/j.1365-2699.2005.01388.x

Popp M, Erixon P, Eggens F, Oxelman B (2005) Origin and evolution of a circumpolar polyploid species complex in *Silene* (Caryophyllaceae) inferred from low copy nuclear RNA polymerase introns, rDNA, and chloroplast DNA. Syst Bot 30:302–313. doi:10.1600/0363644054223648

Posada D (2008) jModelTest: phylogenetic model averaging. Molec Biol Evol 25:1253–1256. doi:10.1093/molbev/msn083

Raftery AE (1995) Bayesian model selection in social research. Sociol Methodol 25:111–163

Rambaut A, Drummond AJ (2009) Tracer, version 1.5. Available at: http://beast.bio.ed.ac.uk/Tracer

Ramos VA, Ghiglione MC (2008) Tectonic evolution of the Patagonian Andes. In: Rabassa J (ed) The late Cenozoic of Patagonia and Tierra del Fuego. Elsevier, Oxford, pp 205–226. doi:10.1016/S1571-0866(07)10004-X

Ribas CC, Moyle RG, Miyaki CY, Cracraft J (2007) The assembly of montane biotas: linking Andean tectonics and climatic oscillations to independent regimes of diversification in Pionus parrots. Proc Roy Soc London B Biol Sci 274:2399–2408. doi:10.1098/rspb.2007.0613

Richardson JE, Pennington RT, Pennington TD, Hollingsworth PM (2001) Rapid diversification of a species-rich genus of neotropical rain forest trees. Science 293:2242–2245. doi:10.1126/science.1061421

Robbiati FO, Lamarque A, Anton AM, Fortunato RH (2013) Phenetic analysis of the complex *Senna fabrisii—S. trichosepala* (Leguminosae, Caesalpiniodeae, *Aphyllae*) based on morphological characters and seed protein electrophoretic profiles. Phytotaxa 145:1–14. doi:10.11646/phytotaxa.145.1.1

Robbiati FO, Ariza Espinar L, Anton AM, Fortunato RH (2014a) Lectotypification, synonymy, and a new name in *Senna* series *Aphyllae* (Leguminosae: caesalpinioideae). Phytotaxa 162:84–90. doi:10.11646/phytotaxa.162.2.2

Robbiati FO, Anton AM, Fortunato RH (2014b) A new synonym in *Senna*, Series *Aphyllae* (Leguminosae, Caesalpinioideae). Syst Bot 39:1120–1126. doi:10.1600/036364414X683840

Robbiati FO, Amarilla LD, Anton AM, Fortunato RH (2017) Phenotypic variation in arid and semiarid zones of southern South American: the case of *Senna* Series *Aphyllae* (Fabaceae, Caesalpinioideae). Bot J Linn Soc 183:454–473. doi:10.1093/botlinnean/bow012

Roig FA, Roig-Juñent S, Corbalán V (2009) Biogeography of the Monte Desert. J Arid Environm 73:164–172. doi:10.1016/j.jaridenv.2008.07.016

Sanín MJ, Kissling WD, Bacon CD, Borchsenius F, Galeano G, Svenning JC, Olivera J, Ramírez R, Trénel P, Pintaud JC (2016) The Neogene rise of the tropical Andes facilitated diversification of wax palms (*Ceroxylon*: arecaceae) through geographical colonization and climatic niche separation. Bot J Linn Soc 182:303–317. doi:10.1111/boj.12419

Schmidt-Jabaily R, Sytsma KJ (2010) Phylogenetics of *Puya* (Bromeliaceae): placement, major lineages, and evolution of Chilean species. Amer J Bot 97:337–356. doi:10.3732/ajb.0900107

Schmidt-Lebuhn AN, de Vos JM, Keller B, Conti E (2012) Phylogenetic analysis of *Primula* section *Primula* reveals rampant non-monophyly among morphologically distinct species. Molec Phylogen Evol 65:23–34. doi:10.1016/j.ympev.2012.05.015

Spichiger R, Calenge C, Bise B (2004) Geographical zonation in the Neotropics of tree species characteristic of the Paraguay-Paraná Basin. J Biogeogr 31:1489–1501. doi:10.1111/j.1365-2699.2004.01071.x

Stamatakis A (2006) RAxML-VI-HPC: maximum likelihood-based phylogenetic analyses with thousands of taxa and mixed models. Bioinformatics 22:2688–2690. doi:10.1093/bioinformatics/btl446

Stamatakis A (2014) RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics 30:1312–1313. doi:10.1093/bioinformatics/btu033

Tamura K, Peterson D, Peterson N, Stecher G, Nei M, Kumar S (2011) MEGA5: molecular evolutionary genetics analysis using maximum likelihood, evolutionary distance, and maximum parsimony methods. Molec Biol Evo 28:2731–2739. doi:10.1093/molbev/msr121

Taylor DW (1991) Paleobiogeographic relationships of Andean angiosperms of Cretaceous to Plioceneage. Palaeogeogr Palaeoclimatol Palaeoecol 88:69–84

van der Hammen T, Clef AM (1986) Development of the high Andean páramo flora and vegetation. In: Vuilleumier F, Monasterio M (eds) High tropical altitude biogeography. Oxford University Press, New York, pp 153–201

Verboom GA, Linder HP, Stock WD (2003) Phylogenetics of the grass genus *Ehrharta*: evidence for radiation in the summer-arid zone of the South African Cape. Evolution 57:1008–1021. doi:10.1554/00143820(2003)057[1008:POTGGE]2.0.CO;2

Viruel J, Catalán P, Segarra-Moragues JG (2012) Disrupted phylogeographical microsatellite and chloroplast DNA patterns indicate a vicariance rather than long-distance dispersal origin for the disjunct distribution of the Chilean endemic *Dioscorea biloba* (Dioscoreaceae) around the Atacama Desert. J Biogeogr 39:1073–1085. doi:10.1111/j.1365-2699.2011.02658.x

Volkheimer W (1971) Aspectos paleoclimatológicos del terciario argentino. Revista Mus Argent Ci Nat 18:172–190

von Hagen KB, Kadereit JW (2003) The diversification of *Halenia* (Gentianaceae): ecological opportunity versus key innovation. Evolution 57:2507–2518

Ward D (2009) The biology of desert. University Press, Oxford

Wendel JF, Doyle JJ (1998) Phylogenetic incongruence: window into genome history and molecular evolution. In: Soltis DE, Soltis PS, Doyle JJ (eds) Molecular systematics of plants II:

DNA sequencing. Kluwer Academic, Boston, pp 265–296. doi:10.1007/978-1-4615-5419-6

Werneck FP (2011) The diversification of eastern South American open vegetation biomes: historical biogeography and perspectives. Quatern Sci Rev 30:1630–1648. doi:10.1016/j.quascirev.2011.03.009

Yu Y, Harris AJ, Blair C, He XJ (2015) RASP (Reconstruct Ancestral State in Phylogenies): a tool for historical biogeography. Molec Phylogen Evol 87:46–49. doi:10.1016/j.ympev.2015.03.008

Yule GU (1925) A mathematical theory of evolution, based on the conclusions of Dr. JC Willis, FRS. Philos Trans Roy Soc London B Biol Sci 213:21–87