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Does Channel Island *Acmispon* (Fabaceae) form cohesive evolutionary groups?

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Abstract.—The California Channel Islands are unique relative to other island chains due to their close proximity to the California mainland and the fact that individual islands, or groups of islands, vary in their distance to the mainland and other islands. This orientation raises questions about whether island taxa with widespread distributions form cohesive evolutionary units, or if they are actually composed of several distinct evolutionary entities, either derived from independent mainland-to-island colonization events or divergence due to prolonged allopatric isolation. The 4 northern islands are clustered in a line (6–8 km separation among islands), while the 4 southern islands are widely spaced (34–45 km separation among islands), which should impact the amount of gene flow and genetic connectivity among islands. We used nuclear microsatellite markers to examine the genetic structure and cohesion of 2 island shrubs, *Acmispon dendroideus* and *A. argophyllus*, which are widely distributed across the California Channel Islands. Both focal species contain varieties with multi-island distributions, with *A. dendroideus* exhibiting a greater distribution on the northern islands and *A. argophyllus* exhibiting a greater distribution on the southern islands. Substantial genetic divergence was observed for 2 single-island endemic varieties, *A. dendroideus* var. *traskiae* and *A. agrophyllus* var. *niveus*, confirming that allopatric isolation can lead to genetic divergence. The widespread *Acmispon dendroideus* var. *dendroideus* and single-island endemic *A. dendroideus* var. *veatchii* formed a cohesive evolutionary group that spans all 4 northern islands and 1 southern island, Santa Catalina, indicating that the northern and southern islands have been genetically linked in the past but do not display evidence of contemporary gene flow. In contrast, widespread *A. argophyllus* var. *argenteus* was composed of moderately distinct genetic groups on each of the 4 southern islands, with no evidence of recent gene flow among islands. These results demonstrate that isolation among islands has led to significant divergence among the southern islands, but that the commonly recognized split between northern and southern islands does not impact all taxa equally.

Resumen.—Las Islas del Canal de California son únicas en comparación a otras cadenas de islas dada su proximidad con California y al hecho de que las islas individuales, o grupos de islas, difieren en cuanto a su distancia con el continente y con otras islas. Tal orientación plantea interrogantes sobre si los taxa insulares de amplia distribución forman unidades evolutivas cohesivas o si se componen, en realidad, de diversas entidades evolutivas, sean derivadas por sucesos independientes de colonización del continente a la isla o por divergencia debido al aislamiento alopátrico prolongado. Las cuatro islas del norte se encuentran agrupadas en una línea (6–8 km de separación entre las islas), mientras que las cuatro islas del sur están espaciadas (34–45 km de separación entre las islas), lo que seguramente repercute en la cantidad de flujo genético y en la conectividad genética entre las islas. Utilizamos marcadores de microsatélites nucleares para examinar la estructura genética y la cohesión de dos arbustos isleños, *Acmispon dendroideus* y *A. argophyllus*, ampliamente distribuidos en las Islas del Canal de California. Ambas especies focales presentan variedades con distribuciones multi-insulares, *A. dendroideus* se encuentra mayoritariamente en las islas del norte, y *A. argophyllus* en las islas del sur. Se observó una divergencia genética sustancial en dos variedades endémicas, *A. dendroideus* var. *traskiae* y *A. agrophyllus* var. *niveus*, confirmando que el aislamiento alopátrico puede generar divergencia genética. Se descubrió que tanto *Acmispon dendroideus* var. *dendroideus* (más generalizado) como el endémico *A. dendroideus* var. *veatchii* forman un grupo evolutivo cohesivo que abarca las cuatro islas del norte y la isla del sur, Santa Catalina, evidenciando que las islas del norte y las del sur estuvieron genéticamente vinculadas en el pasado, aunque no muestren evidencia de flujo genético contemporáneo. Por el contrario, se conoció que el generalizado *A. argophyllus* var. *argenteus* está compuesto por grupos genéticos moderadamente diferenciados en cada una de las cuatro islas del sur, sin evidencia de flujo genético reciente. Tales resultados demuestran que el aislamiento entre las islas produjo divergencias significativas entre las islas del sur, pero que la división comúnmente reconocida entre las islas del norte y las del sur no afecta a todos los taxa por igual.

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A primary goal of evolutionary and conservation biology is to describe biological diversity and determine the processes that have created it. Although biological diversity can be described in many ways (Gaston 2000), most measures of diversity are dependent on numerical approaches summing traditional taxonomic ranks (e.g., species, subspecies, varieties), which are analyzed to determine metrics such as species richness or evenness (Purvis and Hector 2000). Many studies have addressed how biological diversity is impacted by habitat area and isolation (reviewed in Losos et al. 2009), particularly focusing on island taxa due to the discrete nature of islands. Historically, most taxonomic ranks have been described based on morphology, but since the 1980s genetic tools have been applied to dissect both historical evolutionary relationships (Coyne and Orr 2004) and ongoing processes, such as gene flow (Petit and Excoffier 2009), to provide a more accurate representation of units of biological diversity. The importance of accurately describing biological diversity persists, especially in the face of increasing anthropogenic disturbance in many systems. In this work, we examine the California Channel Island plant species *Acmispon argophyllus* (A. Gray) Brouillet (Fabaceae) and *A. dendroideus* (Greene) Brouillet and the varieties they contain, and ask whether they form genetically cohesive entities with distinct contributions to biological diversity.

Many studies of island taxa, particularly sedentary plants, have supported the importance of allopatric isolation among islands as a driver of biodiversity (Price and Wagner 2004, Comes et al. 2008, Ricklefs and Bermingham 2008, Esselstyn et al. 2009). It follows that island-restricted taxa, particularly those with multi-island distributions, may contain more evolutionary diversity than recognized based solely on traditional taxonomy and morphology. Within the Hawaiian Islands, genetic data have documented both multi-island genetic cohesion (McGlaughlin and Friar 2011) and substantial divergence among islands (Baldwin and Friar 2010) within members of the Hawaiian silversword alliance (*Dubautia* [Asteraceae]), and allopatric divergence within a widespread *Schiedea* (Caryophyllaceae; Wallace et al. 2009). Genetic work with Macaronesian plants has documented previously unrecognized subspecific divergence between Azorean and Madeiran-Canarian *Polypodium* (Polypodiaceae; Rumsey et al. 2014), clear genetic differentiation between *Cheirolophus* (Asteraceae) species found on the eastern and western portions of the Canary Islands (Vitales et al. 2014), and *Cistus* (Cistaceae) colonization of the Canary Islands following an east-to-west stepping-stone pattern (Fernández-Mazuecos and Vargas 2011). Collectively, these studies point to the potential importance of isolation among islands in driving divergence both within and among taxa, which has led to an increased appreciation for how gene flow is impacted by spatial scale (Kisel and Barraclough 2010).

The California Channel Islands are composed of 8 oceanic islands located along California's southern coast (Fig. 1). Like many other island chains, the California Channel Islands have never been connected to the mainland (Vedder and Howell 1980), but the islands are much closer to a continental source (minimum distance of 20 km, maximum distance of 98 km; Moody 2009) than most well-studied island chains. The islands are frequently divided into 2 groups: 4 northern islands (San Miguel, Santa Rosa, Santa Cruz, and Anacapa) and 4 southern islands (San Nicolas, Santa Barbara, Santa Catalina, and San Clemente; Fig. 1). The northern islands are closer to the mainland (20–42 km) and to each other (6–8 km) than the southern islands are, and they formed a single large island, Santa Rosae, during the last glacial maximum (Junger and Johnson 1980), whereas the southern islands are more distant from the mainland (32–98 km) and from each other (34–45 km). The 2 island groups also vary in climate, with the northern islands containing more mesic and the southern islands more xeric plant communities (Philbrick and Haller 1977). There is also a clear decrease in annual rainfall from north to south (Junak et al. 1995, Schoenherr et al. 2003).

Few studies have investigated the phylogeography and taxonomy of California Channel Island taxa with multi-island distributions, especially plants. No consistent patterns of colonization or diversification have been validated for the chain, but Riley and McGlaughlin (2016) have suggested that north-to-south colonization, following the dominant dry-season wind direction, best explains the taxonomic diversity of the archipelago-wide flora. In the island animal groups, there is evidence of

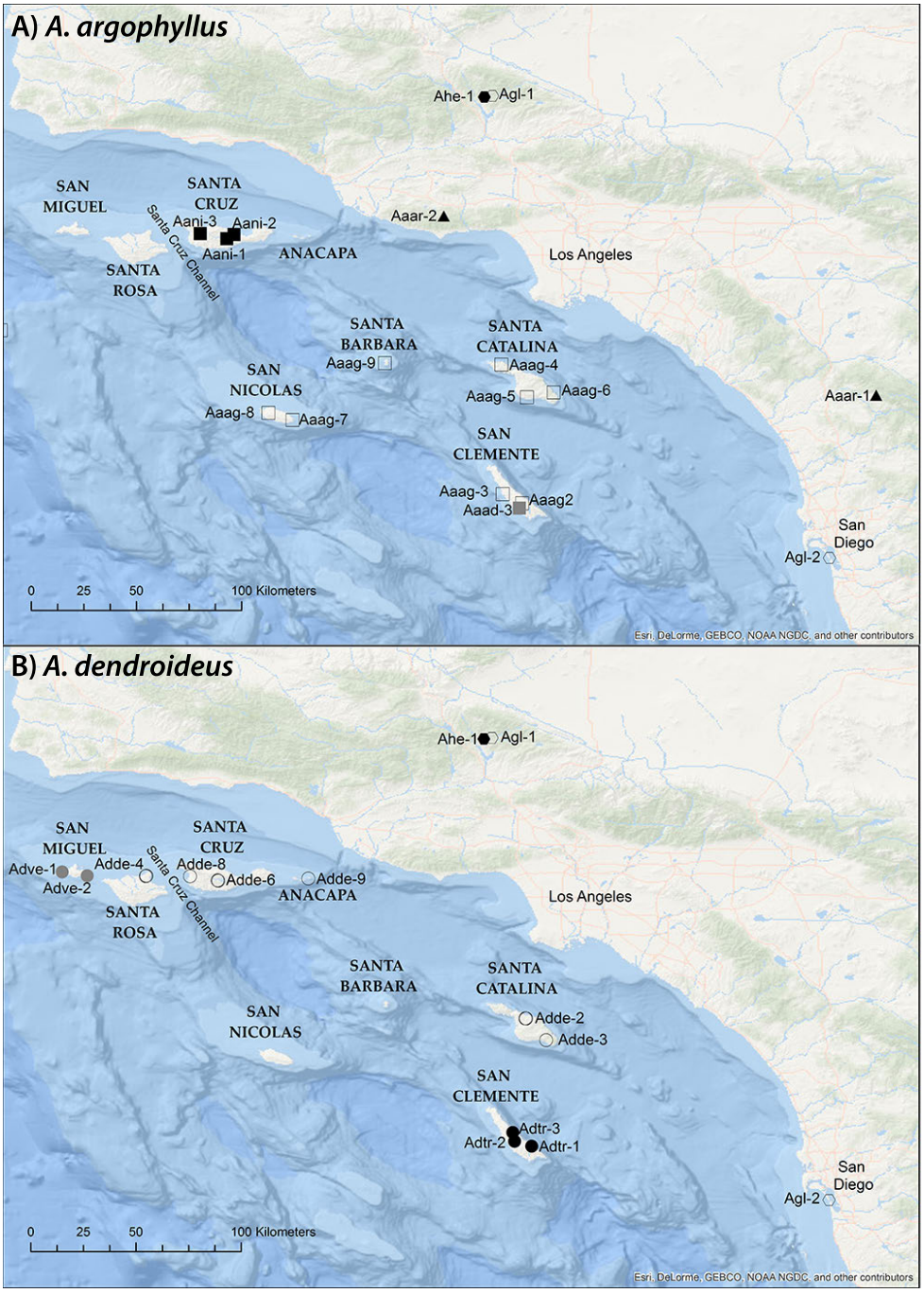

Fig. 1. Locations of sampled populations of *Acmispon* on the California Channel Islands and California Mainland. Population abbreviations are labeled as in Table 1. (**A**) *A. argophyllus* distribution is indicated by squares: *A. a.* var. *niveus* (black), *A. a.* var. *argenteus* (open), and *A. a.* var. *adsurgens* (gray). Mainland populations of *A. argophyllus* var. *argophyllus* are indicated by black triangles. The mainland population of *A. agrophyllus* var. *fremontii* is located approximately 660 km north of Los Angeles (not shown). Mainland *A. glaber* populations are indicated by open hexagons. Mainland *A. heermanii* is indicated by a black hexagon. (**B**) *A. dendroideus* distribution is indicated by circles: *A. d.* var. *traskiae* (black), *A. d.* var. *dendroideus* (open), and *A. d.* var. *veatchii* (gray). Mainland *A. glaber* populations are indicated by open hexagons. Mainland *A. heermanii* is indicated by a black hexagon.

single colonization events followed by dispersal among islands (island foxes, Hofman et al. 2015; spotted skunks, Floyd et al. 2011), multiple independent colonization events (deer mice, Ashley and Wills 1987; Horned Larks, Mason et al. 2014; Song Sparrows, Wilson et al. 2015), and genetic divergence between northern and southern islands (Loggerhead Shrikes, Caballero and Ashley 2011). The limited number of completed studies of plants with multi-island distributions show strong differentiation among southern islands for rare plants (California rockflower, Wallace and Helenurm 2009; Santa Cruz Island rock cress, McGlaughlin et al. 2015) and a clear split between northern and southern populations with a pattern of north-to-south colonization in a widespread species (St. Catherine's lace, Riley et al. 2016).

In this study, we examine patterns of genetic structure within 2 species of *Acmispon* Raf. (Fabceae), *A. argophyllus* and *A. dendroideus*, which each contain 3 varieties that occur on the California Channel Islands. Both focal species were historically placed in *Lotus*, with the same varietal circumscription, but *A. dendroideus* varieties were nested within *L. scoparius* (Torr. & A. Gray) Ottley (now *A. glaber* (Vogel) Brouillet). The 2 focal taxa are closely related but are not sister taxa (Allan and Porter 2000), and are small to medium-sized perennial shrubs. *Acmispon argophyllus* has a decumbent growth form, while *A. dendroideus* has an erect growth form. The 2 taxa are rarely found growing together, with *A. argophyllus* primarily found in more open habitats. Both species are visited by generalist bees (Thorp et al. 1994), but nothing is known about fruit or seed dispersal. *Acmispon argophyllus* is composed of 3 island endemic varieties (found on 1 northern island, all 4 southern islands, and 1 Mexican island) and 2 mainland varieties: *A. argophyllus* var. *adsurgens* (Dunkle) Brouillet on San Clemente Island; *A. argophyllus* var. *argenteus* (Dunkle) Brouillet on San Nicolas Island, Santa Barbara Island, Santa Catalina Island, San Clemente Island, and Guadalupe Island, Mexico; *A. argophyllus* (A. Gray) Brouillet var. *argophyllus* on the mainland in central and southern California; *A. argophyllus* var. *fremontii* (A. Gray) Brouillet on the mainland in the northern Sierra Nevada; and *A. argophyllus* var. *niveus* (Greene) Brouillet on Santa Cruz Island (Fig. 1a). *Acmispon dendroideus* is composed of 3 island varieties, of which 2 are island endemics found on all 4 northern and 2 southern islands: *A. dendroideus* (Greene) Brouillet var. *dendroideus* on Santa Rosa Island, Santa Cruz Island, Anacapa Island, and Santa Catalina Island; *A. dendroideus* var. *traskiae* (Eastw. ex Abrams) Brouillet on San Clemente Island; and *A. dendroideus* var. *veatchii* (Greene) Brouillet on San Miguel Island and northwestern Baja California (Fig. 1b). The exact circumscription of *A. d.* var. *veatchii*, which originally contained specimens from San Miguel Island and northwestern Baja California, has been questioned, but a detailed study verifying the cohesion of the variety has not been conducted (Isley 1981) and specimens from all 4 northern islands have been identified as *A. d.* var. *veatchii*. Three taxa in this group are of conservation concern: *A. a.* var. *adsurgens* and *A. a.* var. *niveus* are California Endangered species (California Department of Fish and Wildlife 2017), and *A. d.* var. *traskiae* is both state listed as endangered and federally listed as Threatened (USFWS 1977, California Department of Fish and Wildlife 2017). Wallace et al. (2017) recently conducted a phylogeographic study of this group utilizing nuclear DNA sequence, chloroplast DNA sequence, and chloroplast microsatellite data to resolve historical patterns of colonization and divergence.

Here, we analyze nuclear microsatellite data to resolve recent evolutionary patterns within *A. argophyllus* and *A. dendroideus*. Our sampling focused on the California Channel Island taxa and close mainland southern California relatives, excluding any occurrences in Mexico. Given the multi-island distribution of the 2 focal species and their varieties, our goal was to address 3 major questions related to evolutionary cohesion in *Acmispon* on the California Channel Islands: (1) Are *A. argophyllus* and *A. dendroideus* genetically cohesive species? (2) Are the *Acmispon* varieties that contain a multi-island distribution genetically cohesive taxa? (3) How has the pattern of island isolation impacted the genetic structure observed in both focal taxa? Answers to these questions will allow a greater understanding of how island placement within the California Channel Islands has impacted diversity and divergence.

Methods

Sampling and Microsatellite Amplification

We sampled a total of 709 individuals of the island taxa throughout their respective ranges: 1 population of *A. a.* var. *adsurgens*, 8 populations of *A. a*. var. *argenteus*, 3 populations of *A. a*. var. *niveus*, 6 populations of *A. d*. var. *dendroideus*, 3 populations of *A. d*. var. *traskiae*, and 2 populations of *A. d*. var. *veatchii* (Table 1, Fig. 1). We also sampled 172 individuals of mainland taxa: 2 populations of *A. a*. var. *argophyllus*, 1 population of *A. a.* var. *fremontii*, 2 populations of *A. glaber*, and 1 population of *A. heermannii* (Durand & Hilg.) Brouillet (Table 1, Fig. 1). The number of individuals sampled from each population ranged from 20 to 40, with an average of 30.4 individuals per population. Detailed collection information and voucher specimens (when available) are shown in Supplementary Material 1. Outgroup taxa were selected to represent all perennial *Acmispon* taxa that occur in southern California adjacent to the California Channel Islands. Due to the clear genetic and morphological differences between *A. dendroideus* and *A. argophyllus*, analyses were conducted on 2 separate data sets containing 1 island taxon and relevant mainland outgroups: (1) all *A. dendroideus* samples and mainland taxa *A. glaber* and *A. heermannii*, and (2) all *A. argophyllus* samples, including mainland varieties, and mainland taxa *A. glaber* and *A. heermannii.*

DNA was extracted from frozen leaf tissue using a modified C-TAB protocol (Friar 2005). Microsatellite data were collected from 15 loci developed for *Acmispon* (McGlaughlin et al. 2011). Amplification was carried out in 12-μL reactions on a MJ Research PTC-200, Eppendorf Mastercycler EP, or BioRad S1000 thermalcycler, following the protocols in McGlaughlin et al. (2011). Amplification products were diluted with water and combined into multiplexes containing 2 to 5 loci each, depending on the population. Each multiplex was electrophoresed with the LIZ-500 size standard on an Avant-3100 Genetic Analyzer (Applied Biosystems, Foster City, CA), following the manufacturer's instructions. Fragments were sized with GeneMapper version 3.7 (Applied Biosystems).

Analysis of Genetic Diversity

Deviations from Hardy–Weinberg equilibrium (HWE) were assessed using GenAlEx version 6.5 (Peakall and Smouse 2012), while the presence of linkage disequilibrium (LD) between loci was assessed with GENEPOP (Rousset 2008). Deviations were considered significant at $P < 0.01$, because the test statistics underestimate the error rate for both multiallelic data and small sample sizes (Lauretto et al. 2009). We used GenAlEx to characterize parameters for each population, including number of alleles per locus (N_A), effective number of alleles per locus (N_E), observed (H_O) and expected (H_E) heterozygosity, and inbreeding coefficient (F_{IS}).

Analysis of Genetic Divergence and Population Structure

GenAlEx 6.5 was used to conduct principal coordinate analyses (PCoA) based on Codem-Genotypic genetic distance calculation with Covariance-Standardized. The software also calculated pairwise F_{ST} and Jost's D (Jost 2008) values among all population pairs. F_{ST} and Jost's D values were averaged within taxon-island groups when multiple populations of a taxon were sampled on a single island. BAYESASS 1.3 (Wilson and Rannala 2003) was used to assess recent migration events among populations with 20,000,000 iterations, a sampling frequency of 2000, a burn-in of 5,000,000, and the following acceptance rate parameters optimized for the 2 data sets: *A. dendroideus* migration rate = 0.30, allele frequencies = 0.35, inbreeding coefficient = 0.35; *A. argophyllus* migration rate = 0.30, allele frequencies = 0.30, inbreeding coefficient = 0.40. BAYESASS reports the fraction of individuals in a population that are migrants derived from another population. Migration values were considered negligible if <2% of genotypes in a population were inferred to be derived from another population.

Population membership based on genotype was estimated using the Bayesian analysis implemented in STRUCTURE 3.2 (Pritchard et al. 2000, Falush et al. 2003, Hubisz et al. 2009). The admixture model with correlated allele frequencies was used, varying the number of inferred populations, K, from 1 to 20. Ten independent runs with a burn-in of 50,000 steps followed by 50,000 steps of data collection were performed for each K. The method of Evanno et al. (2005) was used to determine the "true" K value, as implemented in STRUCTURE HARVESTER (Earl 2012). OBSTRUCT 1.0 was used as an additional

TABLE 1. Microsatellite genetic diversity statistics for *Acmispon* from the California Channel Islands and adjacent mainland. N = number of sampled plants, N_A = number of observed alleles, N_E = effective number of alleles, H_O = observed heterozygosity, H_E = expected heterozygosity, F_{IS} = inbreeding coefficient. Standard deviations are provided in parentheses.

Species	Variety	Population	Island	N	N_A	N_E	H_O	H_E	F_{IS}
A. argophyllus	*adsurgens*	Aaad-3	San Clemente	40	2.667 (0.333)	1.691 (0.154)	0.179 (0.045)	0.330 (0.064)	0.477 (0.061)
	argenteus	Aaag-2	San Clemente	27	4.200 (0.554)	2.755 (0.415)	0.309 (0.048)	0.513 (0.067)	0.397 (0.050)
	argenteus	Aaag-3	San Clemente	30	4.867 (0.710)	2.563 (0.368)	0.269 (0.029)	0.511 (0.057)	0.441 (0.053)
	argenteus	Aaag-4	Santa Catalina	32	6.133 (0.925)	3.279 (0.635)	0.393 (0.060)	0.550 (0.070)	0.261 (0.055)
	argenteus	Aaag-5	Santa Catalina	32	4.533 (0.675)	2.483 (0.406)	0.322 (0.052)	0.469 (0.066)	0.294 (0.079)
	argenteus	Aaag-6	Santa Catalina	32	3.800 (0.626)	2.405 (0.386)	0.343 (0.066)	0.437 (0.077)	0.190 (0.067)
	argenteus	Aaag-7	San Nicolas	31	3.466 (0.568)	1.829 (0.311)	0.179 (0.036)	0.293 (0.071)	0.291 (0.054)
	argenteus	Aaag-8	San Nicolas	31	3.533 (0.487)	1.913 (0.268)	0.187 (0.044)	0.366 (0.063)	0.543 (0.074)
	argenteus	Aaag-9	Santa Barbara	32	1.267 (0.153)	1.018 (0.014)	0.008 (0.005)	0.016 (0.012)	0.202 (0.097)
	niveus	Aani-1	Santa Cruz	32	2.800 (0.393)	1.562 (0.201)	0.067 (0.017)	0.259 (0.060)	0.650 (0.084)
	niveus	Aani-2	Santa Cruz	32	2.933 (0.330)	1.532 (0.178)	0.071 (0.015)	0.263 (0.055)	0.614 (0.100)
	niveus	Aani-3	Santa Cruz	32	2.800 (0.416)	1.406 (0.110)	0.067 (0.021)	0.231 (0.054)	0.703 (0.071)
	argophyllus	Aaar-1	Mainland	32	3.867 (0.710)	1.944 (0.293)	0.212 (0.045)	0.350 (0.072)	0.379 (0.063)
	argophyllus	Aaar-2	Mainland	26	2.600 (0.321)	1.495 (0.110)	0.146 (0.028)	0.277 (0.053)	0.382 (0.080)
	fremontii	Aafr-1	Mainland	27	3.800 (0.470)	2.283 (0.374)	0.385 (0.068)	0.447 (0.056)	0.215 (0.103)
A. dendroideus	*traskiae*	Adtr-1	San Clemente	35	3.867 (0.661)	1.623 (0.168)	0.192 (0.058)	0.303 (0.059)	0.471 (0.080)
	traskiae	Adtr-2	San Clemente	27	3.200 (0.527)	2.013 (0.326)	0.236 (0.059)	0.345 (0.078)	0.301 (0.072)
	traskiae	Adtr-3	San Clemente	30	3.800 (0.732)	2.098 (0.320)	0.249 (0.055)	0.366 (0.080)	0.247 (0.062)
	dendroideus	Adde-2	Santa Catalina	20	3.600 (0.515)	2.196 (0.319)	0.341 (0.074)	0.405 (0.074)	0.230 (0.114)
	dendroideus	Adde-3	Santa Catalina	32	4.400 (0.748)	2.495 (0.389)	0.312 (0.066)	0.441 (0.080)	0.289 (0.085)
	dendroideus	Adde-4	Santa Rosa	31	4.133 (0.515)	2.336 (0.299)	0.293 (0.057)	0.449 (0.074)	0.312 (0.087)
	dendroideus	Adde-6	Santa Cruz	32	6.733 (1.127)	3.145 (0.558)	0.397 (0.063)	0.520 (0.073)	0.229 (0.051)
	dendroideus	Adde-8	Santa Cruz	32	5.600 (0.861)	2.764 (0.382)	0.402 (0.047)	0.541 (0.062)	0.226 (0.049)
	dendroideus	Adde-9	Anacapa	23	3.000 (0.352)	1.811 (0.175)	0.073 (0.023)	0.364 (0.065)	0.730 (0.077)
	veatchii	Adve-1	San Miguel	32	4.467 (0.584)	2.279 (0.380)	0.324 (0.057)	0.422 (0.068)	0.224 (0.045)
	veatchii	Adve-2	San Miguel	32	3.667 (0.630)	2.154 (0.286)	0.341 (0.058)	0.423 (0.069)	0.167 (0.056)
A. heermannii		Ahe-1	Mainland	25	2.933 (0.371)	1.792 (0.168)	0.162 (0.031)	0.366 (0.060)	0.539 (0.072)
A. glaber		Agl-1	Mainland	31	6.334 (0.809)	3.480 (0.512)	0.465 (0.064)	0.574 (0.075)	0.137 (0.053)
		Agl-2	Mainland	31	5.667 (0.599)	2.886 (0.259)	0.457 (0.045)	0.600 (0.046)	0.228 (0.055)

test to determine the number of K genetic clusters based on canonical discriminant analysis (Gayevskiy et al. 2014). Analysis of molecular variance (AMOVA; Excoffier et al. 1992) was estimated in GenAlEx 6.5 for *A. d.* var. *dendroideus* and *A. d.* var. *veatchii*, or *A. a.* var. *argenteus*, grouping populations within islands. POPTREEW (Takezaki et al. 2014) was used to construct neighbor-joining phenograms using the D_A distance (Nei et al. 1983) with 1000 bootstrap replicates.

Results

All loci amplified successfully in all populations except for locus LOAR_216 in *L. a.* var. *argenteus* from San Clemente (18% amplification success) and locus LOAR_21 in *L. d.* var. *dendroideus* and *L. d.* var. *veatchii* (28% amplification success). Principal coordinate analyses were run excluding either locus LOAR_216 or locus LOAR_21 and no changes in the groupings were observed, so both loci were included in the final data sets. Tests for Hardy–Weinberg equilibrium indicated significant disequilibrium for 182 of the 435 locus-by-population comparisons with a corresponding deficiency of heterozygotes (data not shown). Locus LOAR_104 had the greatest amount of disequilibrium, with 22 of 29 sampled populations in violation of Hardy–Weinberg equilibrium. Previous analyses (McGlaughlin et al. 2011) have documented that only locus LOAR_104 exhibited potential null alleles. Given the small and isolated nature of the sampled populations, deviations from Hardy–Weinberg equilibrium are expected based on population-level processes such as inbreeding. Very limited linkage disequilibrium was observed, with only 86 of 3045 locus-by-population comparisons showing significant linkage (data not shown).

Genetic Variation

All measures of genetic variation are shown in Table 1. The number of alleles per locus (N_A) and average number of alleles per locus (N_E) ranged from 1.267 to 6.733 and 1.018 to 3.480, respectively, with the highest values observed in *A. d.* var. *dendroideus* from Santa Cruz (Adde-6, N_A) and *A. glaber* (Agl-1, N_E). Observed (H_O) and expected (H_E) heterozygosity ranged from 0.008 to 0.465 and from 0.016 to 0.600, respectively, with the highest values obtained in *A. glaber* (H_O Agl-1; H_E Agl-2). The lowest values of H_O and H_E were observed in populations on the 2 smallest islands, *A. a.* var. *argenteus* from Santa Barbara and *A. d.* var. *dendroideus* from Anacapa, and taxa restricted to single islands, *A. a.* var. *adsurgens* from San Clemente and *A. a.* var. *niveus* from Santa Cruz (Table 1). Estimates of inbreeding (F_{IS}) varied substantially from a low in *A. glaber* (Agl-1; $F_{IS} = 0.137$) to a high in *A. d.* var. *dendroideus* from Anacapa (Adde-9; $F_{IS} = 0.730$).

Genetic Structure

Analyses of genetic structure were conducted on 2 separate data sets containing one island taxon and relevant mainland outgroups: (1) all *A. dendroideus* samples and mainland taxa *A. glaber* and *A. heermannii*, and (2) all *A. argophyllus* samples, including mainland varieties, and mainland taxa *A. glaber* and *A. heermannii*. The *A. dendroideus* PCoA indicates clear genetic structure with all *A. d.* var. *dendroideus* and *A. d.* var. *veatchii* clustering together, *A. d.* var. *traskiae* clustering together, *A. glaber* clustering together, and *A. heermannii* clustering near to, but distinctive from, *A. glaber* (Fig. 2A). In total, the *A. dendroideus* PCoA explained 66.58% of the observed genetic variability, with axes 1, 2, and 3 (not shown) accounting for 38.69%, 16.24%, and 11.65% of the variability, respectively. PCoA axis 1 supports substantial genetic distinction of *A. d.* var. *dendroideus* and *A. d.* var. *veatchii* relative to *A. d.* var. *traskiae*. The *A. argophyllus* PCoA was less clear with respect to current taxonomic designations and explained only 47.97% of the observed genetic variability, with axes 1, 2, and 3 (not shown) accounting for 19.07%, 15.95%, and 12.95% of the variability, respectively (Fig. 2B). The varieties of *A. argophyllus* were widely scattered, with no clear clustering among samples of *A. a.* var. *argenteus* or *A. a.* var. *argophyllus*. Within the *A. a.* var. *argenteus* samples, populations generally clustered by island: Santa Catalina populations formed a group, and San Nicolas populations were associated with Santa Barbara in close proximity. San Clemente *A. a.* var. *argenteus* had a central position in the PCoA plot, with each sampled population being closely associated with a mainland population of *A. glaber* or *A. heermannii*.

Pairwise measures of genetic differentiation, F_{ST} and Jost's D, are shown for *A. denroideus*

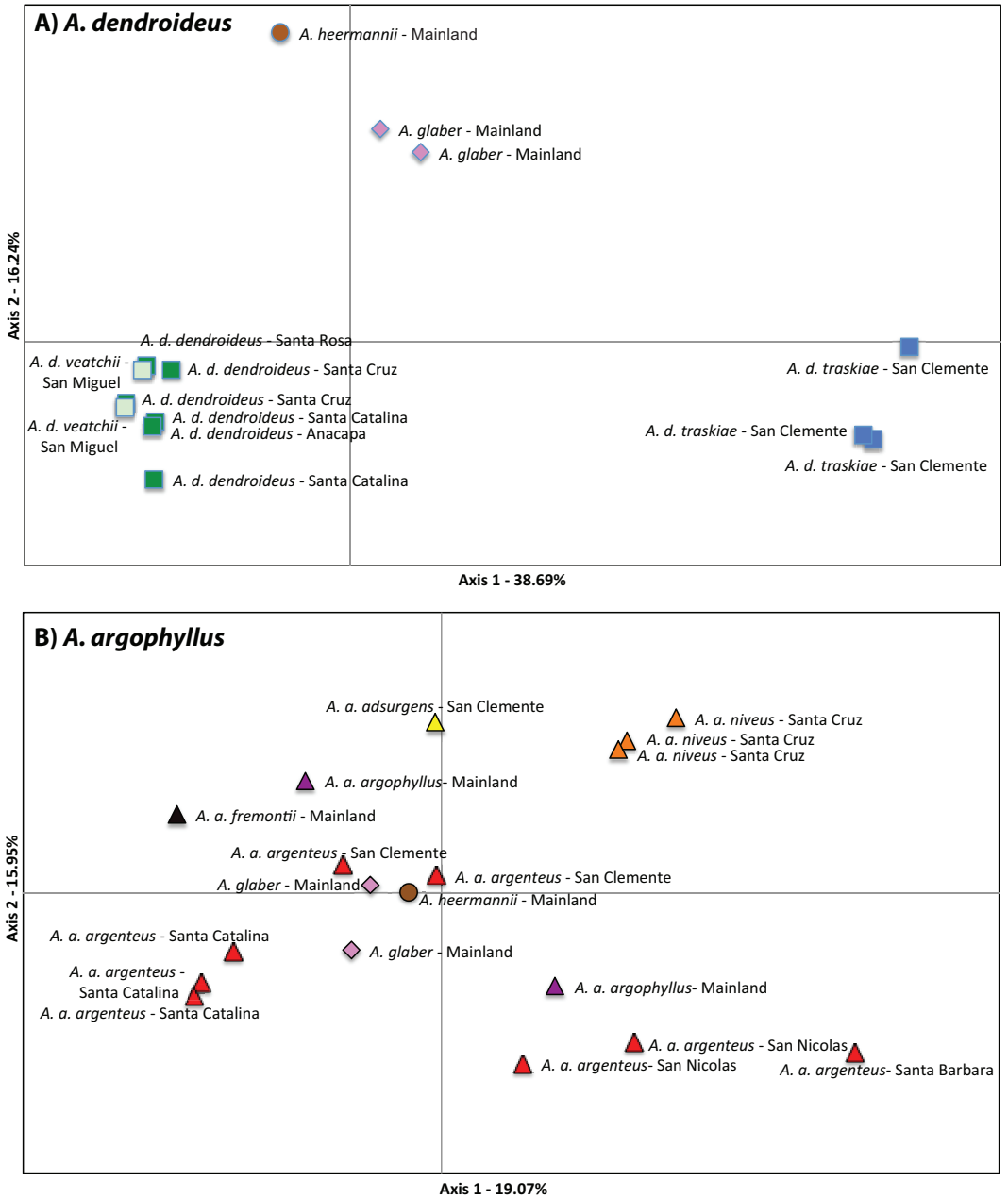

Fig. 2. Principal coordinate analyses (PCoA) based on the Codom-Genotypic genetic distance calculation with Covariance-Standardized as implemented in GenAlEx 6.5 (Peakall and Smouse 2012). (**A**) Comparison of *Acmispon dendroideus* varieties (*A. d.* var. *dendroideus*, *A. d.* var. *traskiae*, and *A. d.* var. *veatchii*) to *A. heermannii* and *A. glaber* with a total of 54.93% of variation represented on axis 1 (38.69%) and axis 2 (16.24%). (**B**) Comparison *Acmispon argophyllus* varieties (*A. a.* var. *adsurgens*, *A. a.* var. *argophyllus*, *A. a.* var. *argenteus*, *A. a.* var. *fremontii*, and *A. a.* var. *niveus*) to *A. heermannii* and *A. glaber*, with a total of 35.02% of the variation represented on axis 1 (19.07%) and axis 2 (15.95%). The collection location is given for each point.

Table 2. Mean pairwise F_{ST} (below diagonal) and Jost's D (above diagonal) values of *Acmispon dendroideus* and mainland *Acmispon* species averaged by island. Values along the diagonal are reported as F_{ST}/Jost's D when multiple populations were sampled within an island. Values <0.2 are shown in bold to indicated limited genetic divergence. Abbreviations: ANA = Anacapa, Main = Mainland California, SCA = Santa Catalina, SCL = San Clemente, SCR = Santa Cruz, SM = San Miguel, SR = Santa Rosa.

	A. d. traskiae–SCL	*A. d. dendroideus*–SCA	*A. d. dendroideus*–ANA	*A. d. dendroideus*–SCR	*A. d. dendroideus*–SR	*A. d. veatchii*–SM	*A. heermannii*–Main	*A. glaber*–Main
A. d. traskiae–SCL	0.203/0.279	0.574	0.678	0.674	0.683	0.674	0.793	0.724
A. d. dendroideus–SCA	0.290	**0.140**/0.234	0.215	**0.186**	**0.186**	0.246	0.625	0.593
A. d. dendroideus–ANA	0.377	**0.149**	na	0.206	0.292	0.251	0.631	0.528
A. d. dendroideus–SCR	0.296	**0.099**	**0.121**	**0.040/0.075**	**0.167**	**0.193**	0.564	0.496
A. d. dendroideus–SR	0.345	**0.099**	**0.191**	**0.090**	na	**0.184**	0.541	0.560
A. d. veatchii–SM	0.345	**0.147**	**0.171**	**0.102**	**0.117**	**0.054/0.069**	0.493	0.601
A. heermannii–Main	0.420	0.331	0.369	0.267	0.302	0.282	na	0.641
A. glaber–Main	0.291	0.228	0.234	**0.169**	0.219	0.230	0.274	**0.155**/0.487

(Table 2) and *A. argophyllus* (Table 3). Low genetic differentiation values indicate limited genetic divergence, with values >0.2 indicative of substantial divergence. Both genetic differentiation measures compared between populations of *A. d.* var. *dendroideus* and *A. d.* var. *veatchii* show limited divergence between Santa Catalina, Santa Cruz, and Santa Rosa, and limited divergence between San Miguel and Santa Rosa, indicative of high historical gene flow (Table 2). The F_{ST} comparisons also support limited differentiation between Anacapa and the other populations of *A. d.* var. *dendroideus* and *A. d.* var. *veatchii*, as well as limited differentiation of *A. d.* var. *dendroideus* Santa Cruz from mainland *A. glaber*. *Acmispon dendroideus* var. *traskiae* shows high levels of differentiation compared to all other sampled taxa. In contrast, within *A. argophyllus*, limited genetic differentiation as measured by F_{ST} and Jost's D is only observed among populations within a taxon and island, or between Santa Catalina *A. a.* var. *argenteus* and mainland *A. glaber* for F_{ST} (Table 3). BAYESASS analysis of recent gene flow documented measurable gene flow within islands for several taxa: *A. d.* var. *traskiae* on San Clemente (Adtr-1 into Adtr-3; 2%), *A. d.* var. *veatchii* (Adve-1 into Adve-2, 24%), *A. a.* var. *argenteus* on San Clemente (Aaag-2 into Aaag-3, 21%), *A. a.* var. *argenteus* on Santa Catalina (Aaag-6 into Aaag-5, 22%), and *A. a.* var. *niveus* (Aani-3 into Aani-1, 22%; Aani-2 into Aani-1, 22%). BAYESASS inferred only within-population mating for all other populations (data not shown).

Analyses using STRUCTURE supported the strong signal of genetic differentiation contained in the *A. dendroideus* data set (Fig. 3) and the conflicting signal in the *A. argophyllus* data set (Fig. 4). STRUCTURE HARVESTER (Supplementary Material 2) and OBSTRUCT (R^2 = 0.97; Supplementary Material 3) both supported grouping the *A. dendroideus* samples into 3 genetic groups, K = 3, containing *A. d.* var. *traskiae* alone, *A. d* var. *dendroideus* and *A. d.* var. *veatchii*, or mainland samples (Fig. 3). Although not strongly supported, dividing the *A. dendroideus* data into 4 genetic groups, K = 4, splits the *A. d* var. *dendroideus* and *A. d.* var. *veatchii* samples into 2 island groups, with San Miguel and Santa Rosa forming one group, and Santa Cruz, Anacapa, and Santa Catalina forming a second group (Fig. 3), as observed in Wallace et al. (2017).

TABLE 3. Mean pairwise F_{ST} (below diagonal) and Jost's D (above diagonal) values of *Acmispon argophyllus* and mainland *Acmispon* species averaged by island. Values along the diagonal are reported as F_{ST}/Jost's D when multiple populations were sampled within an island. Values <0.2 are shown in bold to indicated limited genetic divergence. Abbreviations: Main = Mainland California, SBI = Santa Barbara, SCA = Santa Catalina, SCL = San Clemente, SCR = Santa Cruz, SN = San Nicolas.

	A. a adsurgens–SCL	*A. a. argenteus*–SCL	*A. a. argenteus*–SCA	*A. a. argenteus*–SN	*A. a. argenteus*–SBI	*A. a. niveus*–SCR	*A. a. argophyllus*–Main	*A. a. fremontii*–Main	*A. heermanii*–Main	*A. glaber*–Main
A. a. adsurgens–SCL	na	0.687	0.759	0.764	0.710	0.515	0.650	0.763	0.569	0.632
A. a. argenteus–SCL	0.333	**0.064/0.107**	0.672	0.558	0.642	0.503	0.793	0.701	0.659	0.636
A. a. argenteus–SCA	0.369	0.269	**0.058/0.088**	0.552	0.674	0.613	0.576	0.655	0.506	0.461
A. a. argenteus–SN	0.442	0.289	0.299	**0.129/0.132**	0.312	0.477	0.642	0.838	0.497	0.523
A. a. argenteus–SBI	0.634	0.485	0.516	0.430	na	0.451	0.616	0.861	0.524	0.664
A. a. niveus–SCR	0.386	0.298	0.352	0.366	0.585	**0.119/0.087**	0.565	0.727	0.481	0.537
A. a. argophyllus–Main	0.421	0.379	0.312	0.418	0.628	0.424	**0.464/0.716**	0.773	0.564	0.666
A. a. fremontii–Main	0.378	0.285	0.282	0.398	0.587	0.399	0.395	na	0.743	0.730
A. heermanii–Main	0.351	0.305	0.265	0.320	0.524	0.346	0.364	0.350	na	0.598
A. glaber–Main	0.278	0.218	**0.178**	0.243	0.439	0.272	0.302	0.258	0.251	**0.145/0.453**

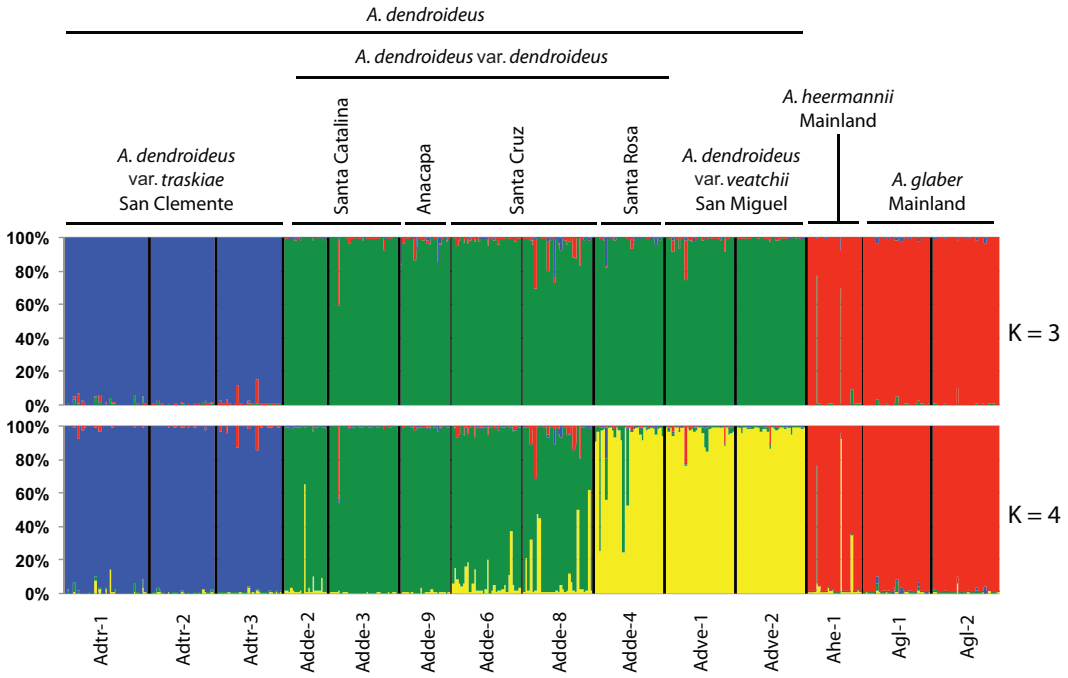

Fig. 3. Bar plot images of STRUCTURE analysis of *Acmispon dendroideus* indicating inferred population assignment of 411 individuals assigned to 3 (K = 3) or 4 (K = 4) genetic groups. The taxon, island, and abbreviation are shown for each population as in Table 1.

The *A. argophyllus* STRUCTURE HARVESTER analyses supported groupings of 2 and 10 genetic clusters (Supplementary Material 4). OBSTRUCT best supported K = 2 (R^2 = 0.96) and K = 3 (R^2 = 0.94), with the population plot for K = 3 and all higher K values clearly segregating into 3 groups (Supplementary Material 5). Based on the STRUCTURE HARVESTER and OBSTRUCT results, K = 2, K = 3, and K = 10 are shown (Fig. 4). The result for 2 genetic clusters, K = 2, infers that the samples of *A. a.* var. *argenteus* San Nicolas and Santa Barbara, all *A. a.* var. *niveus*, and one population of *A. a.* var. *argophyllus* form a group and that all other populations form a second group (Fig. 4). When the number of clusters is increased to three, K = 3 (Fig. 4), *A. a.* var. *adsurgens* (San Clemente Island) clusters with *A. a.* var. *niveus* (Santa Cruz Island) and one population of *A. a.* var. *argophyllus*, while the other 2 clusters are unchanged: *A. a.* var. *argenteus* San Nicolas and Santa Barbara and one population of *A. a.* var. *argophyllus* form one group, and all other populations (*A. a.* var. *argenteus* San Clemente and Santa Catalina, one mainland population of *A. a.* var. *argophyllus*, and the populations of *A. a.* var. *freemontii*, *A. heermanni*, and *A. glaber*) form the other group. The result for 10 genetic clusters, K = 10, infers a division of each taxon or island-taxon group into separate clusters, except for one population of *A. a.* var. *argophyllus* which groups with *A. heermannii* (Fig. 4).

Tree-based clustering analyses support the lack of distinction between *A. d.* var. *dendroideus* and *A. d.* var. *veatchii*, and the uniqueness of *A. d.* var. *traskiae* (Fig. 5A). Additionally, the phenogram joins all varieties of *A. dendroideus* into a single group, but with limited bootstrap support. The *A. argophyllus* phenogram demonstrates the conflict and lack of resolution among taxa in this data set (Fig. 5B). Similar to the STRUCTURE analyses, each island of *A. a.* var. *argenteus* forms a well-supported grouping, but other than *A. a.* var. *argenteus* populations on San Nicolas and Santa Barbara being moderately associated, all other island groups are more closely related to mainland populations, albeit with limited support, than they are to each other. All populations of *A. a.* var. *niveus* also form a

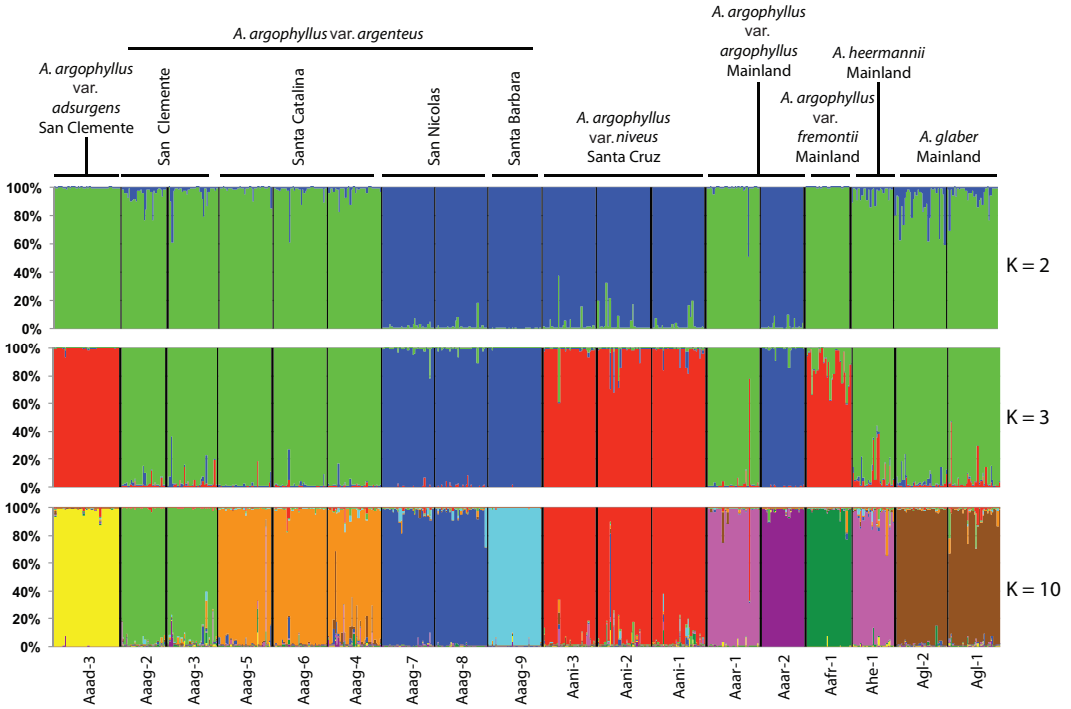

Fig. 4. Bar plot images of STRUCTURE analysis of *Acmispon argophyllus* indicating inferred population assignment of 555 individuals assigned to 2 (K = 2), 3 (K = 3), or 10 (K = 10) genetic groups. The taxon, island, and abbreviation are shown for each population as in Table 1.

well-supported group, which is distinctive from other island *A. argophyllus* populations.

An AMOVA of a subset of populations was utilized to examine genetic cohesion within the island taxa with multi-island distributions. The AMOVA of all populations of *A. d.* var. *dendroideus* and *A. d.* var. *veatchii* supports the lack of distinction among these populations with only 8% of the variability being partitioned among islands, and the vast majority, 81%, contained within populations (Table 4). In contrast, the AMOVA of *A. a.* var. *argenteus* populations found that 33% of the variability is partitioned among islands, 8% among populations within islands, and the remaining 58% within populations (Table 5).

DISCUSSION

The results presented here demonstrate the important role of isolation among islands in shaping genetic divergence and organismal diversity. Although both focal *Acmispon* taxa share many traits and similar distributions, the results show that they have unique evolutionary histories and different levels of connectivity among islands. *Acmispon dendroideus* had substantial genetic cohesion among islands for 2 of its 3 varieties, while *A. argophyllus* exhibited considerable genetic divergence among islands.

Acmispon dendroideus Taxonomy and Genetic Structure

As currently recognized, *A. dendroideus* contains 3 varieties, one of which, *A. d.* var. *dendroideus*, occurs on 3 northern (Anacapa, Santa Cruz, and Santa Rosa) and 1 southern (Santa Catalina) island, while *A. d.* var. *veatchii* and *A. d.* var. *traskiae* are single-island endemics of San Miguel and San Clemente, respectively (Fig. 1b). Under all analyses, *A. d.* var. *traskiae* was substantially diverged from other members of *A. dendroideus* confirming the result of Wallace et al. (2017). Although our mainland sampling is not detailed enough to determine whether *A. d.* var. *traskiae* is descended from an independent mainland-to-island colonization event, *A. d.* var. *traskiae* is equally diverged from both mainland and

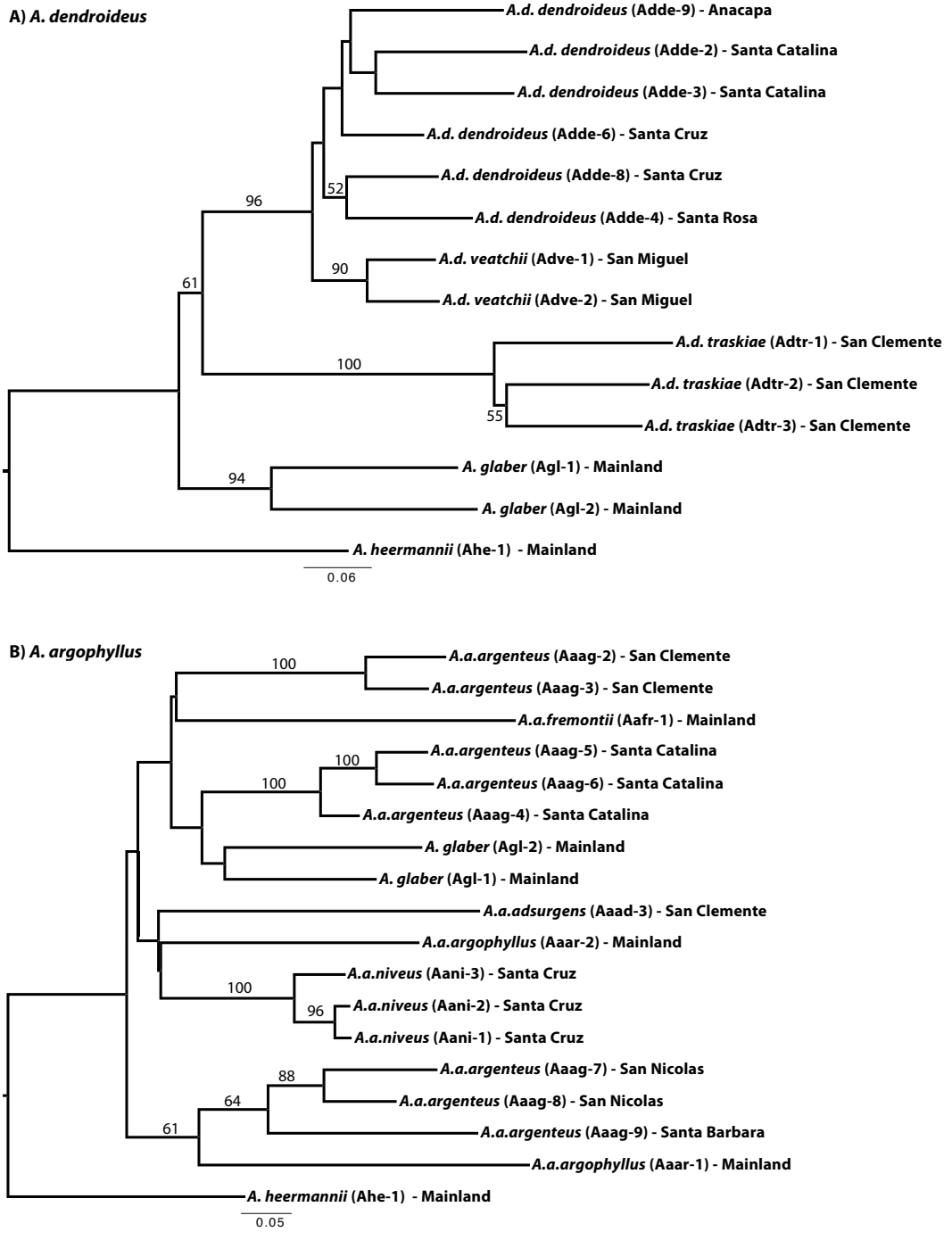

Fig. 5. Neighbor-joining phenogram of (A) *A. dendroideus* and (B) *A. argophyllus,* based on Nei's D. Bootstrap support based on 1000 bootstrap replicates is shown above branches. Population abbreviations are labeled as in Table 1.

Table 4. AMOVA results testing genetic subdivision among islands within *A. dendroideus* var. *dendroideus* and *A. dendroideus* var. *veatchii*.

Source of variation	df	Sum of squares	Variance component	Percentage of variation
Among islands	4	254.353	0.360	8
Among populations/within islands	3	95.023	0.476	11
Within populations	460	1647.541	3.582	81
Total	467	1996.917	4.417	

Table 5. AMOVA results testing genetic subdivision among islands within *A. argophyllus* var. *argenteus*.

Source of variation	df	Sum of squares	Variance component	Percentage of variation
Among islands	3	820.266	2.027	33
Among populations/within islands	4	137.950	0.502	8
Within populations	486	1720.012	3.539	58
Total	493	2678.229	6.067	

island populations (Table 2), the PCoA analysis places it as the most distant grouping of all sampled populations (Fig. 2A), and the phenogram places the *A. d.* var. *traksiae* clade sister to *A. dendroideus* (Fig. 4A) with limited bootstrap support. This leads us to conclude that *A. d.* var. *traskiae* represents a unique genetic entity deserving recognition at the rank of species. In contrast, *A. d.* var. *veatchii* was undistinguishable from *A. d.* var. *dendroideus* in all analyses, which also supports the findings of Wallace et al. (2017). Based on the results of Wallace et al. (2017) and our data presented here, we suggest that additional research should examine the consistency of *A. d.* var. *veatchii* morphological traits in relationship to *A. d.* var. *dendroideus* populations on Santa Rosa to determine whether multiple taxa should be recognized. Our examination of the taxonomy of the entirety of *A. dendroideus* shows that there is 1 substantially diverged lineage, *A. d.* var. *traskiae*, and 2 very similar varieties, *A. d.* var. *dendroideus* and *A. d.* var. *veatchii.* This result leaves the group composed of only 2 genetic entities throughout the California Channel Islands.

The genetic structure contained within *A. dendroideus* greatly expands our understanding of genetic connectivity among populations and islands. The lack of distinction among *A. d.* var. *dendroideus* and *A. d.* var. *veatchii* suggests that there has been considerable genetic connectivity among populations and islands during this group's evolutionary history. The historical island Santa Rosae, the large northern island that existed during the last glacial maximum (Junger and Johnson 1980), may have facilitated dispersal among populations that are now located on distinct islands. The STRUCTURE analysis dividing genotypes into 4 groups (K = 4, Fig. 3), although suboptimal, does show a distinction between populations of *A. d.* var. *veatchii* on San Miguel and *A. d.* var. *dendroideus* on Santa Rosa from *A. d.* var. *dendroideus* on Santa Cruz and Anacapa as observed in the phylogenetic analyses of Wallace et al. (2017). This genetic structure within the northern islands may be attributable to the Santa Cruz Channel, which separates Santa Rosa and Santa Cruz and served as the first major split of Santa Rosae when sea levels rose after the last glacial maximum. Our results also confirm the findings of Wallace et al. (2017) documenting genetic cohesion of *A. d.* var. *denroideus* and *A. d.* var. *veatchii* across both northern and southern islands. Although there is significant genetic structure based on AMOVA ($P = 0.001$), only 8% of the genetic variability is explained among islands (Table 4), while the remaining 92% is within islands and populations. This is the first example of genetic cohesion within plants distributed on northern and southern islands, where previous studies of widespread taxa have found clear distinctions among island groups (Riley et al. 2016, Helenurm unpublished data). Interestingly, we did not find evidence of recent gene flow between the northern islands and Santa Catalina (Table 2) or data to infer the pattern of colonization of the California Channel Islands and subsequent dispersal. The genetic distinction of *A. d.* var. *traskiae* lends support

to the long-held view that San Clemente is the most distinct of the California Channel Islands based on its isolation and dry climate (Raven 1967, Philbrick and Haller 1977, Moody 2000).

Acmispon argophyllus Taxonomy and Genetic Structure

In contrast to *A. dendroideus*, *A. argophyllus* shows a less clear signal when the genetic units contained within the California Channel Islands are considered. Of the 3 varieties, *A. a.* var. *argenteus* is the most widespread, occurring on all 4 southern islands, while *A. a.* var. *adsurgens* and *A. a.* var. *niveus* are single-island endemics found on San Clemente and Santa Cruz, respectively. The phylogenetic data of Wallace et al. (2017) clearly placed *A. a.* var. *niveus* as a monophyletic group, distinctive from all other varieties of *A. argophyllus* and worthy of elevation to the rank of species. Although our current data support the conclusion that *A. a.* var. *niveus* is a distinctive genetic entity that is not breeding with other *A. argophyllus* varieties, the degree of genetic divergence is equal to that of the other varieties, not offering clear support for recognition as a distinct species. Our data also support recognition of *A. a.* var. *adsurgens* as a distinctive genetic entity, although the relationship to other taxa is unclear. Phylogenetic analyses by Wallace et al. (2017) nested *A. a.* var. *adsurgens* within *A. a.* var. *argenteus*, and based on their findings, taxonomic changes are not currently warranted. Finally, *A. a.* var. *argenteus* is the most perplexing of the sampled taxa. STRUCTURE analyses indicated that *A. a.* var. *argenteus* splits into 2 genetic groups, San Nicolas and Santa Barbara or Santa Catalina and San Clemente (Fig. 4), while the PCoA and phenogram further split Santa Catalina and San Clemente into separate groups (Fig. 2B, Fig. 5B). What is more interesting is that depending on the analysis, individuals of *A. a.* var. *argenteus* from different islands have an affinity for different mainland relatives. This is best seen in the STRUCTURE analysis dividing all samples into 2 or 3 groups (K = 2 or 3, Fig. 5), where the 2 major *A. a.* var. *argenteus* groups have an affinity for different mainland taxa. Although we did not investigate the timing of divergence within groups, the lack of genetic structure within *A. a.* var. *argenteus* could be indicative of a fairly recent colonization of the California Channel Islands. An examination of the taxonomy of the entirety of *A. argophyllus* makes it clear that there are 2 cohesive varieties composed of the single-island endemic taxa *A. a.* var. *niveus* and *A. a.* var. *adsurgens*, and a final widespread variety, *A. a.* var. *argenteus*, with divergence among islands, leaving a minimum of 3 genetic entities, but possibly more depending on how *A. a.* var. *argenteus* is parsed.

Despite the lack of strong signal in the *A. argophyllus* data set, our data do inform our thinking of evolutionary patterns on the California Channel Islands, particularly the southern islands. The genetic distinction of *A. a.* var. *niveus* indicates that there was one colonization event of the northern islands, but that there was limited further gene flow between the northern and southern islands as documented in other phylogeographic studies (Caballero and Ashley 2011, Riley et al. 2016). The strong distinction among islands for *A. a.* var. *argenteus* based on the STRUCTURE (K = 10, Fig. 5) and phenogram analyses (Fig. 4B) confirms that connectivity among southern islands is limited. This conclusion is supported by the fact that 33% of *A. a.* var. *argenteus* genetic variation is partitioned among islands (Table 5), and there is no evidence of recent gene flow among islands (Table 3). What remains unclear is whether all *A. a.* var. *argenteus* populations are derived from a single colonization event followed by interisland dispersal, which is the conclusion reached by Wallace et al. (2017) based on phylogenetic analyses, or whether *A. a.* var. *argenteus* is derived from multiple colonization events. In studies of Aegean *Nigella* (Ranunculaceae), Comes et al. (2008) concluded that genetic drift is the primary mechanism leading to divergence among islands due to evidence of genetic structure and a lack of gene flow. In *A. a.* var. *argenteus*, we observed structure among islands, a lack of gene flow, and low diversity on the smallest islands San Nicolas and Santa Barbara (see below; Table 1), all of which would be expected results associated with high rates of genetic drift. Future studies with *A. a.* var. *argenteus* should examine breeding system, particularly whether selfing is possible, and the timing of divergence among islands based on a molecular clock to better understand the observed genetic structure.

Conservation Implications

California Channel Island *Acmispon* has faced historical and ongoing threats due to small population sizes, isolation, nonnative taxa, and human-induced habitat modification, all of which can reduce genetic diversity and impact evolutionary potential. Previous work (McGlaughlin et al. 2014) indicated that mean levels of microsatellite diversity within *A. argophyllus* and *A. dendroideus* are similar to levels observed in other Channel Island endemic taxa (e.g., Furches et al. 2009, Wallace and Helenurm 2009, Riley et al. 2016) and levels generally observed in other endemic plant taxa (Nybom 2004). However, there is considerable variability in levels of diversity among taxa and islands (Table 1), with several groups that are of concern. In particular, the lowest levels of diversity are seen in populations on the smallest islands, *A. a.* var. *argenteus* on Santa Barbara and *A. d.* var. *dendroideus* on Anacapa, which have substantial constraints on total population size due to limited available habitat. These populations should be monitored to avoid unnecessary disturbance which could further reduce population size and genetic diversity. Furthermore, *A. d.* var. *dendroideus* on Anacapa (East Anacapa) exhibited high levels of inbreeding (F_{IS}), suggesting that this island may warrant active management to boost the total population size. Given the lack of genetic distinction among *A. d.* var. *dendroideus* populations, introduction of plants from nearby Santa Cruz could help bolster genetic diversity. Of the 3 taxa of conservation concern, *A. a.* var. *niveus* and *A. a.* var. *adsurgens* show low levels of genetic diversity, while *A. d.* var. *traskiae* is moderately diverse (Table 1). Inbreeding is also a concern for *A. a.* var. *niveus*, but given the recent removal of large nonnative mammals from Santa Cruz, recent population size increases (McGlaughlin personal observation), and measureable gene flow among populations, no current management is recommended. Genetic management for *A. a.* var. *adsurgens* is more challenging given its limited distribution, predominantly in the Shore Bombardment Area (SHOBA) on San Clemente, and low diversity observed in other populations (McGlaughlin unpublished data). Management of *A. a.* var. *adsurgens* populations should focus on reducing disturbance and maintaining the largest populations possible. Although the remaining *A. argophyllus* and *A. dendroideus* populations are not highly diverse, limited evidence suggests that they are in need of active management for the maintenance of genetic diversity. Interestingly, we found very limited evidence for contemporary gene flow among populations within islands, despite strong evidence of historical genetic connectivity within *A. dendroideus*. This result raises questions about whether rates of gene flow have been impacted by anthropogenic disturbance, which could impact the long-term survival of these taxa.

Conclusions

The research presented here adds to our understanding of connectivity and isolation among islands, and taxonomic accuracy within endemic California Channel Island *Acmispon* species. Contrary to previous research with California Channel Island plants (Riley et al. 2016, Helenurm unpublished data), it is clear that there was historical genetic connectivity within *A. d.* var. *dendroideus* between the northern islands and Santa Catalina, although the directionality of that connectivity is unclear and no evidence of contemporary gene flow is evident. Within *A. a.* var. *argenteus*, genetic isolation among the southern islands was observed, which fits with the geography of the southern islands and studies that have examined taxa on San Clemente and Santa Catalina (Wallace and Helenurm 2009, McGlaughlin et al. 2015). Collectively, these results demonstrate that closely related taxa may undergo very different evolutionary trajectories, even when they are functioning in the same system. When considering *Acmispon* taxonomy, we identified a single taxon, *A. d.* var. *veatchii,* that was not supported genetically. However, when considering the species rank, we identified one taxon, *A. d.* var. *traskiae*, that is substantially differentiated and worthy of recognition as a distinct species. Overall, these data add to a growing understanding of how allopatric isolation among islands and limited gene flow have impacted diversity in the California Channel Islands.

Supplementary Material

Five online-only supplementary files accompany this article (scholarsarchive.byu.edu/wnan/vol78/iss4/24).

Supplementary Material 1. Locations and voucher references for populations of *Acmispon* sampled for this study.

Supplementary Material 2. *Acmispon dendroideus* STRUCTURE HARVESTER results showing the rate of change of delta K for K = 1–20.

Supplementary Material 3. OBSTRUCT canonical discriminant analysis plots of the median and 50% ellipse for each taxon by island: 1, *Acmispon dendroideus* var. *traskiae* San Clemente; 2, *A. d.* var. *dendroideus* Santa Catalina; 3, *A. d.* var. *dendroideus* Anacapa; 4, *A. d.* var. *dendroideus* Santa Cruz; 5, *A. d.* var. *dendroideus* Santa Rosa; 6, *A. d.* var. *veatchii* San Miguel; 7, *A. glaber* Mainland 1; 8, *A. glaber* Mainland 2; 9, *A. heermannii* Mainland.

Supplementary Material 4. *Acmispon argophyllus* STRUCTURE HARVESTER results showing the rate of change of delta K for K = 1–20.

Supplementary Material 5. OBSTRUCT canonical discriminant analysis plots of the median and 50% ellipse for each taxon by island: 1, *Acmispon argophyllus* var. *adsurgens* San Clemente; 2, *A. a.* var. *argenteus* San Clemente; 3, *A. a.* var. *argenteus* Santa Catalina; 4, *A. a.* var. *argenteus* San Nicolas; 5, *A. a.* var. *argenteus* Santa Barbara; 6, *A. a.* var. *niveus* Santa Cruz; 7, *A. a.* var. *argophyllus* Mainland 1; 8, *A. a.* var. *argophyllus* Mainland 2; 9, *A. a.* var. *fremontii* Mainland; 10, *A. heermannii* Mainland; 11, *A. glaber* Mainland 1; 12, *A. glaber* Mainland 2.

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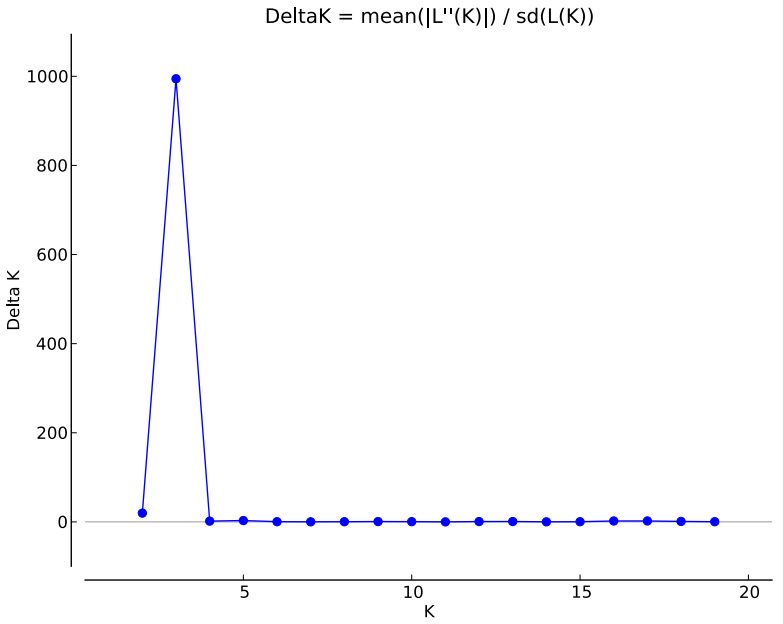
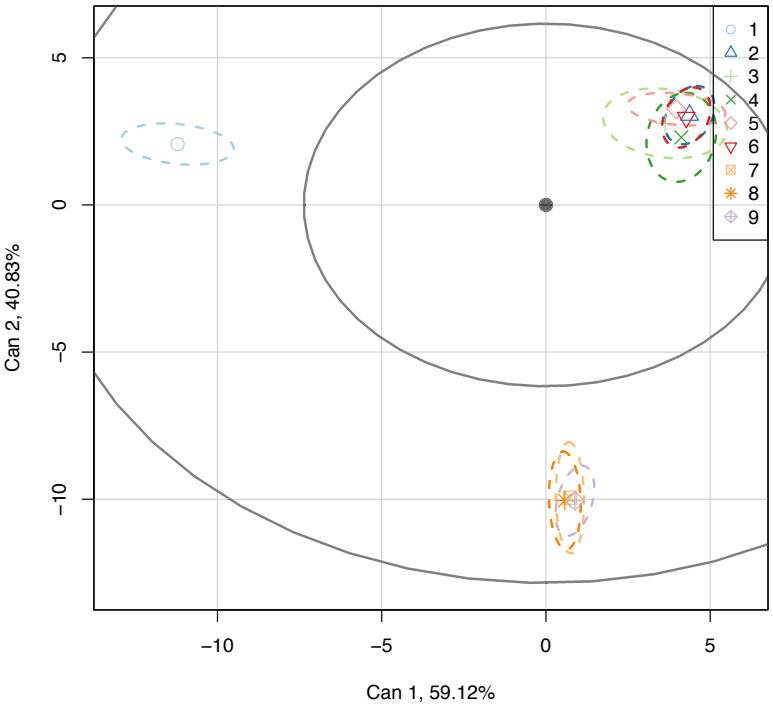
SUPPLEMENTARY MATERIAL 1. Locations and voucher references for populations of *Acmispon* sampled for this study. All available vouchers are deposited at the University of Northern Colorado (GREE), Greeley, Colorado, and are shown with their collector number (MEM = M.E. McLaughlin) and accession number. NA = not available.

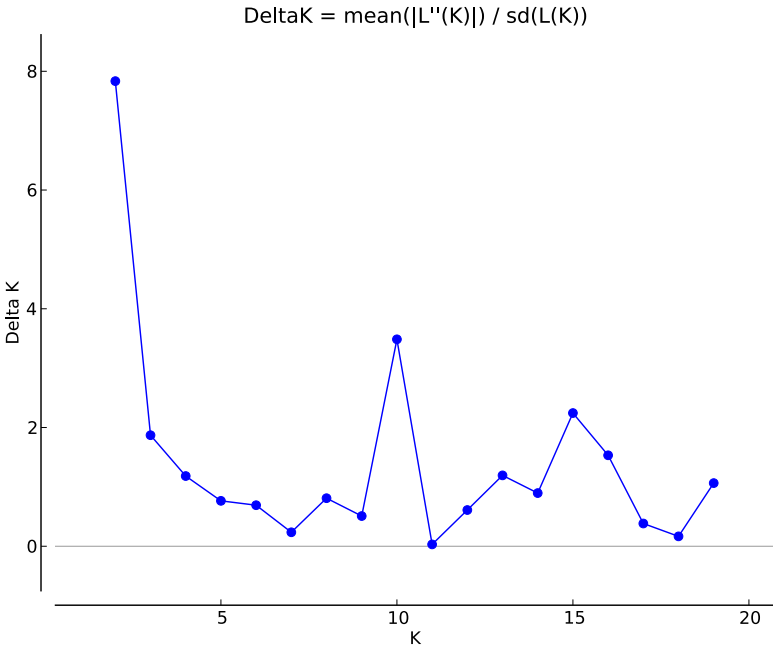
Population	Island	Longitude	Latitude	Voucher
Acmispon argophyllus var. *adsurgens*				
Aaad-3	San Clemente	−118.467	32.852	NA
Acmispon argophyllus var. *argenteus*				
Aaag-2	San Clemente	−118.546	33.000	NA
Aaag-3	San Clemente	−118.539	32.914	NA
Aaag-4	Santa Catalina	−118.524	33.460	MEM 189, GREE9190
Aaag-5	Santa Catalina	−118.435	33.327	NA
Aaag-6	Santa Catalina	−118.345	33.322	MEM 192, GREE9188
Aaag-7	San Nicolas	−119.437	33.230	NA
Aaag-8	San Nicolas	−119.456	33.258	NA
Aaag-9	Santa Barbara	−119.043	33.473	MEM 314, GREE22106
Acmispon argophyllus var. *argophyllus*				
Aaar-1	Mainland	−116.944	33.335	MEM 229, GREE9184
Aaar-2	Mainland	−118.792	34.101	MEM 265, GREE22105
Acmispon argophyllus var. *fremontii*				
Aafr-1	Mainland	−121.302	39.554	MEM 261, GREE9195
Acmispon argophyllus var. *niveus*				
Aani-1	Santa Cruz	−119.730	33.998	MEM 204, GREE9178
Aani-2	Santa Cruz	−119.857	34.021	MEM 212, GREE9175
Aani-3	Santa Cruz	−119.703	34.006	MEM 182, GREE9185
Acmispon dendroideus var. *dendroideus*				
Adde-2	Santa Catalina	−118.440	33.416	NA
Adde-3	Santa Catalina	−118.343	33.323	MEM 191, GREE9189
Adde-4	Santa Rosa	−120.053	34.016	NA
Adde-6	Santa Cruz	−119.682	33.981	MEM 216, GREE9180
Adde-8	Santa Cruz	−119.873	34.024	MEM 211, GREE9176
Adde-9	Anacapa	−119.368	34.014	NA
Acmispon dendroideus var. *traskiae*				
Adtr-1	San Clemente	−118.493	32.925	NA
Adtr-2	San Clemente	−118.412	32.866	NA
Adtr-3	San Clemente	−118.485	32.887	NA
Acmispon dendroideus var. *veatchii*				
Adve-1	San Miguel	−120.420	34.038	NA
Adve-2	San Miguel	−120.312	34.022	NA
Acmispon glaber var. *glaber*				
Agl-1	Mainland	−118.582	34.614	NA
Agl-2	Mainland	−117.141	32.640	MEM 220, GREE21280
Acmispon heermannii var. *heermannii*				
Ahe-1	Mainland	−118.581	34.614	MEM 238, GREE9183



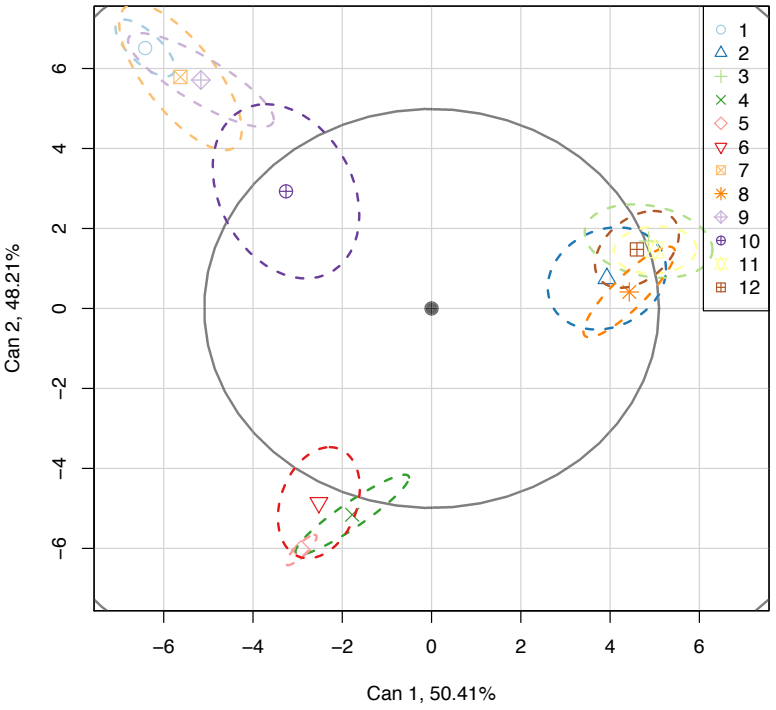
SUPPLEMENTARY MATERIAL 2. *Acmispon dendroideus* STRUCTURE HARVESTER results showing the rate of change of delta K for K = 1–20.



SUPPLEMENTARY MATERIAL 3. OBSTRUCT canonical discriminant analysis plots of the median and 50% ellipse for each taxon by island: **1,** *Acmispon dendroideus* var. *traskiae* San Clemente; **2,** *A. d.* var. *dendroideus* Santa Catalina; **3,** *A. d.* var. *dendroideus* Anacapa; **4,** *A. d.* var. *dendroideus* Santa Cruz; **5,** *A. d.* var. *dendroideus* Santa Rosa; **6,** *A. d.* var. *veatchii* San Miguel; **7,** *A. glaber* Mainland 1; **8,** *A. glaber* Mainland 2; **9,** *A. heermannii* Mainland.



SUPPLEMENTARY MATERIAL 4. *Acmispon argophyllus* STRUCTURE HARVESTER results showing the rate of change of delta K for K = 1–20.



SUPPLEMENTARY MATERIAL 5. OBSTRUCT canonical discriminant analysis plots of the median and 50% ellipse for each taxon by island: **1,** *Acmispon argophyllus* var. *adsurgens* San Clemente; **2,** *A. a.* var. *argenteus* San Clemente; **3,** *A. a.* var. *argenteus* Santa Catalina; **4,** *A. a.* var. *argenteus* San Nicolas; **5,** *A. a.* var. *argenteus* Santa Barbara; **6,** *A. a.* var. *niveus* Santa Cruz; **7,** *A. a.* var. *argophyllus* Mainland 1; **8,** *A. a.* var. *argophyllus* Mainland 2; **9,** *A. a.* var. *fremontii* Mainland; **10,** *A. heermannii* Mainland; **11,** *A. glaber* Mainland 1; **12,** *A. glaber* Mainland 2.