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Phylogeny of Polemoniaceae Based on Nuclear Ribosomal Internal Transcribed Spacer DNA Sequences

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PHYLOGENY OF POLEMONIACEAE BASED ON NUCLEAR RIBOSOMAL INTERNAL TRANSCRIBED SPACER DNA SEQUENCES

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ABSTRACT

Nuclear ribosomal internal transcribed spacer (ITS) DNA sequences are used to estimate the phylogeny of 53 members of Polemoniaceae, representing all but two genera of the family. Fitch parsimony analysis of equal-weighted nucleotide sites result in 1080 minimal-length trees. However, when alignment-ambiguous positions are removed and an 11:10 transition to transversion weighting is imposed only eight trees are found. These data are used to address two issues: 1) patterns of diversification in Polemoniaceae, and 2) the circumscription and monophyly of the genus *Gilia*. Although the monophyly of Polemoniaceae is well supported, relationships inferred among the earliest diverging lineages are altered by character weighting, treatment of indels, and taxon inclusion. In spite of the lack of reliable resolution at the basal nodes, ITS data provide evidence that *Gilia*, as currently interpreted, is polyphyletic and comprises at least five independent lineages.

Key words: DNA sequences, *Gilia*, Polemoniaceae, phylogeny.

INTRODUCTION

The Polemoniaceae are a small, mostly New World, group of between 300 and 340 species (Grant 1959; Willis 1966; Cronquist 1981; Thorne 1992; Patterson 1993). Evolutionary relationships and classification within this family have long been controversial (Bentham 1845; Gray 1870; Kuntze 1891; Brand 1907; Rydberg 1913; Dawson 1936; Wherry 1940; Mason 1945; Mason and Grant 1948; Grant 1959; Cronquist 1984; Day and Moran 1986). Although generally considered a "natural" group by most authors, generic and species relationships within the family have remained obscure: the circumscription of species, genera and higher taxonomic groups differing radically from author to author. The most recent family-wide classification of Polemoniaceae is that of Grant (1959), who recognizes 18 genera, placed into five tribes (Table 1). Over the past 30 years, a broad spectrum of data has accumulated bearing on relationships within the family. These include family- and generic-level surveys of palynological (Stuchlik 1967; Loeblich 1964; Taylor and Levin 1975; Chuang et al. 1978; Timbrook 1986; Lord and Eckard 1986; Day and Moran 1986), histological (Carlquist et al. 1984), chemosystematic (Smith et al. 1977; Dean et al. 1978; Harborne and Smith 1978; Harborne 1980; Smith et al. 1982; Wilken et al. 1982; Kiredjiad and Smith 1985), and reproductive data (Grant and Grant 1965; Plitmann and Levin 1983; 1990). Although neither phenetic nor phylogenetic analyses have been conducted on the family using this wide array of morphological and chemical data (but

see Wilken and Hartman 1991), recent phylogenetic analyses of nucleotide sequences of the chloroplast gene *mat*K (Steele 1991, 1992; Steele and Vilgalys 1994; Johnson and Soltis 1995; Johnson et al. 1996) suggest these tribes are not monophyletic. In light of these data, realignment of Grant's (1959) tribes appears inevitable. This study provides additional insight into classification and evolutionary history of Polemoniaceae from an independent source of evidence. Presented is a molecular phylogenetic study using sequences from the nuclear genes coding for ribosomal RNA, the internal transcribed spacers (ITS1 and ITS2).

Taxonomic Background

The contrast between various classifications of Polemoniaceae has been reviewed by Grant (1959: 4). There is little consensus among historical classifications. The few general similarities (particularly for classifications since 1900) include: 1) the tendency to place the tropical genera *Cantua*, *Cobaea* and *Bonplandia* in separate tribes apart from the remainder of the so-called "temperate genera" (Brand 1907; 1908; Grant 1959); and 2) the inclusion of *Linanthus* and *Leptodactylon* in the same tribe with *Gilia* (Brand 1907; 1908; Wherry 1940; 1945; Grant 1959) while placing *Phlox* in a different tribe (Wherry 1940; 1945; Grant 1959). A particularly striking disparity among historical classifications in the family is the circumscription of the genus *Gilia*. These differences range from broad circumscriptions that include nearly all of

the "temperate genera" except *Polemonium, Phlox,* and *Collomia* (e.g., Bentham and Hooker 1876) to the present treatment by Grant (1959) that segregates *Allophyllum, Ipomopsis, Eriastrum, Langloisia, Loeseliastrum,* and *Navarretia* from *Gilia.*

Grant's currently accepted tribal classification has not found support from recent molecular studies. Steele and Vilgalys (1994) provided evidence from cpDNA *mat*K sequences that some genera included in tribe Polemonieae share more recent common ancestry with genera placed in tribe Gilieae. For example *Collomia* and *Allophyllum* (tribe Polemonieae) are more closely related to *Gilia* and *Navarretia* (tribe Gilieae) than to either *Phlox* or *Polemonium* (tribe Polemonieae). Indeed, Steele and Vilgalys suggest that there is no support for two temperate tribes, rather Polemoniaceae possess a single temperate lineage. These results were not only corroborated by Johnson and Soltis (1995) and Johnson et al. (1996) but due to the increased taxon sampling, evidence was presented that the genus *Gilia,* as currently circumscribed, is a polyphyletic assemblage. Some members of *Gilia* (e.g., *G. filiformis*) are more closely related to genera never associated with tribe Gilieae (e.g., *Phlox*) than to other members of *Gilia.*

This controversy over relationships within Polemoniaceae is problematic because Polemoniaceae have been used as a model system for the study of many aspects of character evolution and reproductive biology. The family as well as individual species have been used as primary examples for the pattern and modes of evolution of pollination mechanisms (Grant and Grant 1965; Galen and Kevan 1983; Paige and Whitham 1985, 1987a; Galen and Stanton 1989), evolution of plant sexual systems (Grant 1959; Schoen 1982a, b, c, 1983; Plitmann and Levin 1983, 1990), hybridization and its evolutionary consequences (Grant 1950, 1954a, b, 1957, 1979; Grant and Grant 1960; Day 1965; Levin 1979; Coyne 1974; Grant and Wilken 1987), natural selection on floral traits (Ellstrand 1983; Paige and Whitham 1985; Wolf et al. 1986; Ellstrand and Mitchell 1988; Campbell 1989; 1991), and evolution life history (Paige and Whitham 1987a, b). An accurate phylogenetic backdrop is necessary to interpret evolutionary changes in these traits (Donoghue 1989). The chloroplast phylogenies provide an important inroad into understanding the phylogeny of Polemoniaceae. However, a single gene phylogeny may not correspond with an organismal phylogeny (Dwyer et al. 1991; Doyle 1992; Neigel and Avise 1986; Sanderson and Doyle 1992). A phylogeny based on an independent source of data is both desirable and necessary to either corroborate or refute phylogenetic inferences from the chloroplast genome.

In this paper I provide a preliminary assessment of phylogenetic relationships of Polemoniaceae using DNA sequences of the internal transcribed spacers (ITS1 and ITS2) of the nuclear ribosomal cistron. Because these spacer sequences are encoded in the nuclear genome, they represent an independent source of phylogenetic information from chloroplast sequences.

METHODS

Sampled Taxa

An attempt was made to include representatives of all of the 19 currently recognized genera of Polemoniaceae (Table 1). Because of the controversy over the limits of the genus *Gilia* and its relation to *Ipomopsis,* 13 species of *Gilia* (members of sections *Arachnion, Gilia, Giliandra, Giliastrum, Gilmania,* and *Saltugilia*) and five species of *Ipomopsis* (members of sections *Ipomopsis, Microgilia, Phloganthea* [including the *Giliopsis* group]). In addition, a largely unknown woody member of Polemoniaceae, *Gilia scabra* (treated by Grant 1959 as a synonym of *Linanthus nuttallii*), is also included. Only two genera are not represented. *Gymnosteris,* with two species, is not included because the DNAs did not sequence well enough for reasonable interpretation (nearly all of the nucleotide sites appeared polymorphic). In addition, the DNA sequence from *Huthia coerulea* is not included because it is unalignable with other Polemoniaceae sequences.

Selection of an outgroup for Polemoniaceae is problematical. In spite of most present classifications (Cronquist 1981; Takhtajan 1980; Thorne 1968, 1992), molecular data from the chloroplast gene *rbc*L (Olmstead et al. 1992; Chase et al. 1993) provide evidence that Polemoniaceae are more closely related to Ericalean groups, usually placed in the Dilleniidae, than to the Asteridae families with which they are traditionally placed: i.e., Convolvulaceae, Hydrophyllaceae, and Solanaceae (but see Anderberg 1992; Martin and Dowd 1991). In particular, chloroplast sequence data from *rbc*L (Olmstead et al. 1992) and *mat*K (Johnson et al. 1996) place Fouquieriaceae as the sister group to Polemoniaceae. By contrast, the Chase et al. study of *rbc*L supports a close relationship between Polemoniaceae and Diapensiaceae. Close relationships between Polemoniaceae and both Diapensiaceae (Linnaeus 1753; Don 1822) and Fouquieriaceae (Humbolt et al. 1823; Nash 1903; Engler and Gilg 1924; Abrams 1951; Thorne 1977) have been suggested based on morphological evidence. Based on this evidence, I have selected Diapensiaceae and Fouquieriaceae as outgroups. Attempts were made to also include sequences from Solanales, e.g., *Nicotiana rustica* L. (Venkateswarlu and Nazar 1991); however, these sequences are unalignable.

Collection and DNA Extraction

Material collected in the field was placed on ice for transport to storage at −80°C. Total genomic DNA extracts from leaf material, powdered under liquid nitrogen, used a 2X CTAB protocol, modified from Doyle and Doyle (1987). In some cases a 4X CTAB (with 0.1% PVP-40) extraction buffer was used for species with high polysaccharide content. The DNAs were further purified using cesium chloride/ethidium bromide gradients (Maniatis et al. 1982). The DNAs of *Acanthogilia gloriosa* and *Gilia scabra* were extracted from leaf material desiccated using silica gel (Liston and Rieseberg 1990; Chase and Hillis 1991). The DNAs of six taxa were obtained from leaf fragments taken from herbarium collections (Table 1). Subsequently, all DNA samples obtained from herbarium specimens have been verified using freshly collected material, and in no case did the DNA sequences of ITS differ at more than a single, uninformative nucleotide site (Porter unpubl.).

PCR Amplification and Sequencing

Sequencing was performed on 39 samples using the method of Sanger et al. (1977). Single-stranded DNAs for both strands of the two internal transcribed spacers of the nuclear ribosomal cistron, ITS1 and ITS2, were amplified directly by asymmetric polymerase chain reaction (PCR), using a 1:20 ratio of the primers "ITS5" (5′-GGA AGT AAA AGT CGT AAC AAG G-3′) and "ITS2" (5′-GCT GCG TTC TTC ATC GAT GC-3′) for the ITS1 spacer, and the primers "ITS3" (5′-GCA TCG ATG AAG AAC GCA GC-3′) and "ITS4" (5′-TCC TCC GCT TAT TGA TAT GC-3′) for the ITS2 spacer (White et al. 1990). PCR amplifications, PCR product purification and direct single-stranded DNA sequencing followed the procedures described by Baldwin (1992), except for the following modifications. The amplification of DNA from herbarium material employed a modified 10× *Taq* polymerase reaction buffer consisting of 67mM Tris-HCl (pH 8.8), 2 mM MgCl$_2$, and 2 mg/ml bovine serum albumin (Pääbo et al. 1989; Pääbo 1990). The PCR products were electrophoresed using a 1.5% agarose gel (prestained with ethidium bromide) in a 1× TBE (pH 8.3) buffer, to confirm the single-stranded product and purified using differential filtration in Millipore Ultra-Free-MC microfuge tubes (Millipore UFC-3 THK00). The purified DNAs were sequenced using the dideoxy chain termination reaction, following the procedures outlined in the TAQuence® kit (U. S. Biochemical Co.). Sequencing products were labeled using α-^{35}SdATP. To reduce base compressions, 7-deaza-dGTP was substituted for dGTP in all of the sequencing reactions. The gels were fixed in 5% acetic acid/5% methanol for 45 min and vacuum dried at 80°C for 1 hr. The autoradiographs were exposed for a minimum of 24 hr. All sequences were checked by sequencing the opposite strand.

Sequence data for 14 species were obtained using an Applied Biosystems Model 373A Automated DNA Sequencing System (Perkin Elmer). Template DNAs were prepared by symmetric PCR of the entire ITS region using a 1:1 ratio of primers ITS5 and ITS4. Direct cycle-sequencing of the ITS region followed manufacturers specifications, using the PRISM® DyeDeoxy® Terminator Kit (Perkin Elmer) and employed primers ITS2, ITS3, "ITS5I" (5′-AGG TG ACC TGC GGA AGG ATC ATT-3′) and "ITS4I" (5′-GGT AGT CCC GCC TGA CCT GG-3′). Primers ITSP5I and ITSP4I are internal to and flanking ITS5 and ITS4, respectively. The four primers provide sequences for overlapping fragments that collectively cover both strands of the entire ITS region (ITS1, 5.8S and ITS2).

Sequence Alignment

The 56 sequences were truncated to include only ITS1 and ITS2. Identification of the terminal ends of ITS1 and ITS2 was based on comparisons with published sequences for *Daucus carota* L., *Nicotiana rustica*, *Vicia faba* L., species of *Astragalus,* and representatives of *Madiinae* (Baldwin 1992, 1993; Venkateswarlu and Nazar 1991; Wojciechowski et al. 1993; Yokota et al. 1989). The two spacer regions were aligned separately, using PILEUP (Feng and Doolittle 1987; Higgins and Sharp 1987; see also Needleman and Wunch 1987), a multiple sequence alignment program of the University of Wisconsin Genetics Computer Group (UWGCG) software package. The alignment parameters were varied from GapWeight= 1.0 and GaplengthWeight= 0.1 to GapWeight= 5.0 and GaplengthWeight= 3.0. With higher gap and length penalty values, fairly large blocks of aligned sequences are produced, but single taxa are sometimes grossly misaligned. Low gap and length penalties result in alignments lacking grossly misaligned taxa, but with a greater frequency of gaps. Evenso, alternative alignments were evident. The resulting alignments were both analyzed directly and manually realigned and reanalyzed. The PILEUP aligned sequences were manually realigned to maximize the number of invariant nucleotide positions. The results presented refer to sequences which were realigned manually; however, the conclusions apply to both alignments.

Phylogenetic Analysis

Estimations of phylogenetic relationships, based on combined ITS1 and ITS2 sequences, were obtained using Fitch parsimony as implemented in PAUP (version 3.0; Swofford 1991). A variety of heuristic approaches

Table 1.   Classification of Polemoniceae, sensu Grant (1959) and collections utilized in the phylogenetic analysis of nrDNA internal transcribed spacer sequences. The cross (†) indicates those taxa for which herbarium collections were used for DNA extractions. The asterisk (*) indicates those species for which cycle sequencing and automated sequencing procedures were used. Vouchers are cited and housed as indicated. The taxa included to serve as the outgrup are designated as such parenthetically.

Tribe Cantuaeae (2: ca. 9)

Acanthogilia (1 species)	$n = 9$	A. gloriosa (Brandeg.) Day & Moran. 2.4 km west of Punta Prieta, Baja California, Mexico. [J. M. Porter & K. D. Heil 7987; SJNM]
Cantua (ca. 12 species)	$n = 27$	C. quercifolia Cav. Cult. San Francisco State University, San Francisco, California. [R. Patterson sn; RSA]
Huthia (2 species)	$n = ?$	†* H. coerulea Brand. Arequipa, Peru. [O. Tovar 3543; MO]

Tribe Bonplandieae (2: ca. 17)

Bonplandia (2 species)	$n = 15$	B. geminiflora Cav. Cult. San Francisco State University, San Francisco, California. [R. Patterson sn; RSA]
Loeselia (ca. 15 species)	$n = 9$	L. glandulosa (Cav.) Don. Patagonia Mountains, Santa Cruz County, Arizona. [J. M. Porter & C. Campbell 9231; AZ, SJNM]

Tribe Cobaeeae (1: 19)

Cobaea (19 species)	$n = 27$	C. scandens Cav. Cult. San Francisco State University, San Francisco, California. [R. Patterson sn; RSA]
	$n = ?$	† C. baiurita Standl. Puerto Vienda, Chiapas, Mexico. 20 Aug. 1967. [O. Clarke 293; AZ]

Tribe Polemonieae (5: ca. 102)

Allophyllum (5 species)	$n = 9$	* A. divaricatum (Nutt.) A. Grant & V. Grant. Mt. Konocti, Lake County, CA. [J. M. Porter & L. Machen 10819; RSA]
	$n = 9$	A. gilioides (Benth.) Grant & Grant. 5 miles south of Oracle, Santa Catalina Mtns., Pinal County, Arizona. [J. M. Porter & L. Machen 8751; AZ, SJNM]
Collomia (15 species)	$n = 8$	* C. grandiflora Lindl. Libre Mtns., Los Angeles County, California. [T. Ross & S. Boyd 8142; RSA]
	$n = 8$	C. linearis Nutt. 10 miles northeast of Pogosa Springs, Archuleta County, Colorado. [J. M. Porter 8565; AZ]
Gymnosteris (2 species)	$n = 6$	—
Phlox (ca. 60 species)	$n = 7$	P. stanburyi (Torr.) Heller. 4 miles south of the junction of Nevada Highway 160 and US Highway 95, Nye County Nevada. [J. M. Porter 8841; SJNM]
	$n = 7$	* P. (Microsteris) gracilis E. Greene. Mt. Emma, Los Angeles County, California [J. M. Porter 10566; RSA]
Polemonium (ca. 20 species)	$n = 9$	P. foliosissimum Gray. Lizard Head Pass, along Colorado Highway 145, Dolores County, Colorado. [J. M. Porter 7526; SJNM]

Tribe Gilieae (8: ca. 200)

Eriastrum (14 species)	$n = 7$	E. diffusum (Gray) Mason. Tucson Mountains, 4 miles west of Tucson, Pima County, Arizona. [J. M. Porter 8754; AZ]
Gilia (ca. 70 species)	$n = 8$	G. caespitosa Gray. Capitol Reef National Park, Wayne County, Utah [J. M. Porter & K. Heil 7352; SJNM]
	$n = 9$	†* G. campanulata Gray. 5 mi W Independence, Inyo County, CA [Kerr 638; RSA]
	$n = 9$	† G. capitata Sims. E of Washougal, Skamania County, Washington. 14 July 1984. [R. S. Halse 2900.; AZ]
	$n = 72$	†* G. crassifolia Benth. Rio Molles, Endesa, Coquimbo, Chile. 5 Nov. 1970. [J. Simon 161.; RSA]
	$n = 9$	* G. filiformis Gray. 12 mi E. Independence, Inyo County, CA [J. M. Porter & L. Machen 10849; RSA]
	$n = 18$	G. flavocincta A. Nels subsp. australis A. & V. Grant. Ca. 6 miles east of Roosevelt Reservoir, roadside, Pinal County, Arizona. [J. M. Porter 7060; SJNM]
	$n = 9$	† G. foetida Gil. ex Benth. Las Heras camino a Yalguaraz, Provincia de Mendoza, Argentina. 27 Dec. 1947. [R. Leal, 11,113; AZ]
	$n = 9$	G. hutchinsifolia Rydb. 5 mi S of Lathrop Wells, Nye County, Nevada. [J. M. Porter 8209; AZ, SJNM]
	$n = ?$	* G. inyoensis I. M. Johnston. Anchorite Pass, Mineral County, NV. [J. M. Porter & L. Machen 10842; RSA]
	$n = 18$	G. latifolia Wats. Cataract Canyon, Canyonlands National Park, San Juan County, Utah. [J. M. Porter & M. Heil 8804; AZ, SJNM]
	$n = 9$	* G. maculata Parish. Whitewater Canyon, Riverside County, CA. [J. M. Porter & L. Machen 10560; RSA]
	$n = 8$	G. mcvickerae M. E. Jones. Ca. 4 mi S of Panguitch on UT89, Garfield County, Utah. [J. M. Porter and L. Machen 7184; SJNM]

Table 1.   Continued.

	$n = 8$	*G. nyensis* J. Reveal. Red001 burned area, Red Rock Canyon, Nevada Nuclear Test Site, Nye County, Nevada. [*J. M. Porter 8202*; AZ]
	$n = 9$	*G. ochroleuca* M. E. Jones. Ca. 4 mi. S of Big Pine, Inyo County, California. [*J. M. Porter 8860*; AZ, SJNM]
	$n = 8$	*G. pentstemonoides* M. E. Jones. N side of the Gunnison River, Cimarron, Montrose County, Colorado. [*J. M. Porter 7192*; AZ, SJNM]
	$n = 9$	*G. rigidula* Benth. 7 miles southeast of Sonoita, Santa Cruz County, Arizona. [*J. M. Porter 8723*; RSA, SJNM]
	$n = 9$	*G. scabra* Brandegee. Cerro Prietto, near San Hipalito, Viscaino Desert, Baja California Sur, Mexico. [*J. M. Porter and K. Heil 7991*; SJNM]
	$n = 9$	*G. scopulorum* M. E. Jones. Hualapai Indian Reservation, Mohave County, Arizona. 21 Apr. 1973. [*P. S. Martin, sn*; AZ]
	$n = 9$	* *G. splendens* Dougl. ex Lindl. Pacific Crest Trail, San Bernardino County, CA. [*J. M. Porter 10142*; RSA]
	$n = 9$	*G. stellata* Heller. Roadside along AZ85, 9 mi N of Why, Pima County, AZ. [*J. M. Porter 9492*; SJNM]
	$n = 8$	*G. subnuda* Gray. W of Steamboat Canyon, Apache County, AZ. [*J. M. Porter & K. Heil 7355*; SJNM]
	$n = 18$	† *G. tenerrima* A. Gray. 70 mi NW of Elko, Elko County, Nevada. 26 June 1937. [*N. Nichols & L. Lund 206*; AZ]
	$n = 9$	*G. tricolor* Benth. Cult. University of Arizona, seed from retail seed source. [*J. M. Porter sn*; AZ]
Ipomopsis (30 species)	$n = 7$	*I. gunnisonii* (Torr. and Gray) V. Grant. Navajo Mine, 12 miles south of Waterflow, San Juan County, New Mexico. [*J. M. Porter 9295*; AZ, SJNM]
	$n = 7$	*I. multiflora* (Nutt.) V. Grant. Molino Basin, Santa Catalina Mountains, Pima County, Arizona. [*J. M. Porter 8052*; AZ, SJNM]
	$n = 7$	*I. longiflora* (Torr.) V. Grant. 8 mi south of Safford, Greenlee County, Arizona. [*J. M. Porter 7142*; SJNM]
	$n = ?$	†* *I. sonorae* (Rose) A. Grant. Sonora, Mexico. [*T. R. VanDevender 93-20*; AZ, RSA]
	$n = 7$	† *I. tenuifolia* (Gray) V. Grant. 11 May 1980. Sierra San Pedro Martir, Mexico. [*S. P. McLaughlin 2465*; AZ]
Langloisia (1 species)	$n = 7$	*L. setosissima* (Torr. & Gray) Greene subsp. *setosissima.* 4 miles south of the junction of Nevada Highway 160 and US Highway 95, Nye County Nevada. [*J. M. Porter 8561*; SJNM]
	$n = 7$	*L. setosissima* (Torr. & Gray) Greene subsp. *punctata* (Cov.) Timbr. 4 miles south of the junction of Nevada Highway 160 and US Highway 95, Nye County Nevada. [*J. M. Porter 8840*; SJNM]
Leptodactylon (7 species)	$n = 9$	* *L. pungens* (Torr.) Rydb. 3 mi S Natural Bridges Nat. Mon., Kane County, Utah. [*J. M. Porter 8561*; SJNM]
	$n = 9$	*L. watsonii* (Gray) Rydb. Waterpocket Fold, Capitol Reef National Park, Wayne County, Utah. [*J. M. Porter 8571*; SJNM]
Linanthus (41 species)	$n = 9$	*L. aureus* (Nutt.) Greene. 2 miles north of Searchlight, along US Highway 95, Clark County, Nevada. [*J. M. Porter 8822*; AZ, SJNM]
	$n = 9$	* *L. bicolor* (Nutt.) Greene. Hayfork, Trinity County, CA. [*J. M. Porter & L. Machen 10821*; RSA]
	$n = 9$	*L. nuttallii* (Gray) Greene ex Mlkn. 8 miles north of Granville, near Grey Peak, White Mountains, Greenlee County, Arizona. [*J. M. Porter & L. Machen 9004*; RSA, SJNM]
	$n = 9$	* *L. parryae* (Gray) Greene. Valyermo, Los Angeles County, CA. [*J. M. Porter 10553* RSA]
Loeseliastrum (2 species)	$n = 7$	*L. matthewsii* (Gray) Timbr. San Bernardino County, California. [*J. M. Porter 8869*; SJNM]
	$n = 7$	*L. schottii* (Torr.) Timbr. 14 mi S of Parker, along AZ95, Yuma County, Arizona. [*J. M. Porter 8856*; RSA]
Navarretia (32 species)	$n = 9$	*N. breweri* (Gray) Greene, Columbine Pass, Uncompahgre Plateau, Montrose County, Colorado. [*J. M. Porter 8599*; RSA]
Diapensiaceae (Outgroup)	$n = 6$	*† *D. laponica* L. Mt. McKinley, Alaska. [*M. Fay 325*; ARIZ]
Fouquieriaceae (Outgroup)	$n = ?$	*F. columnaris* (Kell.) Hendrickson. Cult., The University Mall, University of Arizona Campus, Tucson, Arizona. [*J. M. Porter sn*; SJNM]
	$n = 6$	*F. splendens* Engelm. Cult., Old Main, University of Arizona Campus, Tucson, Arizona. [*J. M. Porter sn*; SJNM]

was employed to help assure that all of the most parsimonious solutions were obtained. Initially, CLOSEST addition and TBR (tree bisection-reconnection) swapping was performed. This was followed by 100 replicates of RANDOM addition and TBR branch swapping. Finally, 500 replicates of RANDOM addition were carried out with no swapping, followed by TBR swapping on the resulting set of trees (Maddison 1991). This should insure that all minimal length tree are found, even if multiple "islands" of equally parsimonious trees exist. The data matrix was analyzed with equal weights at all nucleotide positions and using an 1.1 to 1 transition/transversion bias (based on estimates from Polemoniaceae sequences). Insertions and/or deletions (indels) were coded initially as missing data. Subsequent analyses treated indels as a "fifth nucleotide" (GAPMODE=FIFTHSTATE option in PAUP) and as additional binary characters.

Robustness and Sampling

An assessment of the relative support for clades was performed using the decay index (Bremer 1988; Donoghue et al. 1992) and the bootstrap (Felsenstein 1985). Decay values were calculated for one, two and three steps longer than the shortest trees. My purpose is to identify only the weakly supported nodes. Bootstrap values were calculated from 100 replicate Fitch parsimony analyses, as implemented by PAUP 3.0, using heuristic searches based on CLOSEST addition and TBR branch swapping.

The distribution of tree-lengths, based on 10,000 randomly generated trees was evaluated for skewness (g1). A strongly left-skewed tree length distribution has been suggested to indicate that a data set is highly structured and likely contains strong phylogenetic signal (Goodman et al. 1979; Fitch 1984; Werman 1986; Hillis and de Sa 1988; Hillis and Dixon 1989; Hillis and Huelsenbeck 1992; Hillis 1991; Huelsenbeck 1991). However, this test actually assesses whether the g1 of a given data set deviates from the expected g1 of random data (see also Kallersjo et al. 1992).

To investigate the sensitivity of these data to taxon sampling, an analysis based on resampling terminal taxa was conducted. Lanyon's jackknife is a heuristic tool to explore the effect of taxon sampling on internal branch support and tree structure (Lanyon 1985a, b; Felsenstein 1988; Siddall 1995). Lanyon (1985a) points out that trees generated during this jackknife procedure simulate possible outcomes had the investigator failed to sample a taxon or if extinction had occurred. Clearly, taxon sampling (inclusion) can have profound consequences on assessment of monophyly (Doyle and Donoghue 1987; Gauthier et al. 1988; Donoghue et al. 1989; Novacek 1991). The manually realigned data matrix was analyzed by sequentially re-

moving each member of the ingroup, one at a time, and performing a parsimony analysis (CLOSEST addition, HOLD= 2, TBR branch swapping) on the resulting data sets (pseudoreplicates). In all cases, only the strict-consensus tree from analyses of each pseudoreplicate was retained. The trees were compared and pseudoreplicate-consensus trees were generated. This was done by first returning the excluded taxon from each pseudoreplicate run back into the pseudoreplicate strict-consensus tree. The excluded taxon was placed in the same position it is found in the strict-consensus tree from the analysis of the entire data set. This is based on the assumption that the analysis of the full data set provides evidence for the taxon's placement. After the excluded taxon is returned to the tree, a consensus of the entire set of trees was obtained, using strict and majority rule consensus procedures.

RESULTS

ITS Sequence Variation

The ITS1 regions of all representatives of Polemoniaceae, as well as the outgroups, were consistently larger than the ITS2 regions. Within Polemoniaceae the ITS1 region is generally 249–253 bases. Extremes beyond this range include the sequences of *Gilia rigidula* at 242 base pairs (bp) and *Gilia scabra* at 262 bp. The outgroups possessed the largest ITS1 sequence of this analysis, 267 bp. In Polemoniaceae, percent G+C content of ITS1 ranged from 42.2% in *Cobaea baiurita* to 65.0% in *Acanthogilia gloriosa*. ITS2 sequences were mostly between 188 and 193 bases, but ranged from 187 bp in *Phlox standsburyi* and *Linanthus nuttallii* to 195 in *Gilia foetida*. The ITS2 region of the outgroups was slightly larger, 196 bp. Percent G+C content of ITS2 in Polemoniaceae varied from 48.3% in *Cobaea baiurita* to 61.8% in *Acanthogilia gloriosa*.

Alignment

Both ITS1 and ITS2 possess several highly conserved regions, flanked by more variable regions (Baldwin et al. 1995). The conserved regions are easily and nearly unambiguously aligned; however, the variable regions become increasingly difficult to align as pairwise distance increases. As a result, alignment of some taxa (i.e., outgroup taxa) should be viewed with skepticism. The variable regions of ITS in Polemoniaceae are frequently associated with short indels of one to a few nucleotides. Sequences of ITS1 and ITS2, aligned using variable weights in PILEUP produce similar alignments; however, the more variable regions seemed better aligned (i.e., there is a higher frequency of invariant sites) using GapWeight= 1.0 and GaplengthWeight= 0.1 (the aligned sequences us-

ing PILEUP are not shown). The manually realigned sequences are used in all of the following comparisons. Even so, the manually aligned sequence data set is not without alternative alignments and alignment in some regions remains ambiguous (Appendix I).

The aligned, combined sequences of ITS1 and ITS2 required, on average, gaps at 14.39% of the sites of any given sequence. The inclusion of the outgroup was responsible for gaps at 4.53% of these nucleotide positions. The combined data matrix, possesses 529 characters, of which 120 are invariant and 346 (65.28%) are potentially informative. The pair-wise levels of divergence range from 0.18% (between *Langloisia setosissima* subsp. *setosissima* and *Langloisia setosissima* subsp. *punctata*) to 42.4% (between *Diapensia lapponica* and *Cantua quercifolia*).

Phylogenetic Analysis

All of the parsimony analyses of the full, equal-weighted data set, with indels treated as missing, resulted in recovery of the same 1080 minimum-length trees. For the data set including only potentially informative sites, the most parsimonious trees were of 1552 steps (tree length including all sites is 1646), with a consistency index (C.I.) of 0.423, retention index (R.I.) of 0.622, and rescaled consistency index (R.C.) of 0.263. The strict consensus tree (Fig. 1) shows that the primary areas of disagreement in the set of minimal length trees involves 1) relationships involving *Ipomopsis sonorae* and *Loeseliastrum matthewsii* in the Loeselieae Clade, 2) relationship of *Gilia campanulata* within the Linanthieae Clade, and 3) relationships between three clades of *Gilia* species as well as *Gilia splendens,* within the Gilieae Clade. Aside from the basal nodes, the phylogenetic estimate is not highly sensitive to coding of the indel regions or transition-transversion weighting. Figure 2 shows the strict consensus trees from analyses that 1) treat each indel position as a fifth character state while all other sites are equally weighted, and 2) remove all indels and alignment-ambiguous positions while imposing a 1.0:1.1 transition to transversion weighting. In both analyses many fewer trees are recovered, 12 and 8 respectively. The topologies differ primarily in the inferred relationships of *Cobaea, Cantua, Bonplandia* and *Acanthogilia* to the remainder of Polemoniaceae. Both of these trees are very similar in structure to the full, equal-weighted analysis.

Tree Support

Bootstrap.—The bootstrap 50% majority rule tree (not shown) is structurally identical to the strict consensus insofar as the majority rule tree is resolved. Many of the clades are supported with high bootstrap percentage values (see Fig. 1). Ten clades are supported by

values of 95% or greater. Hillis and Bull (1993), suggest that 70% bootstrap percentage values or greater might be a better estimate of a "95% confidence interval" for estimating the correct phylogeny. If this can be generalized to all data (but see Felsenstein and Kashino 1993), another 14 clades fall within the confidence limit.

Some portions of the trees, show little bootstrap support. In general the highest values are associated with the more terminal portions of the phylogeny. The values tend to decrease toward the more basal nodes. The only notable exception is the ancestral node of Polemoniaceae, that possesses a significant value of 99%.

Decay index.—The decay index is here employed to identify those clades that are weakly supported or may be prone to errors in phylogenetic estimation. Decay values shown in Fig. 1 range from one to three. In general, the most weakly supported nodes are the more basal nodes within Polemoniaceae. Again, the only exception to this trend is the ancestral node of Polemoniaceae.

Lanyon's jackknife.—The jackknife procedure required 53 analyses, the consensus of these analyses is shown in Fig. 3. The regions of the phylogeny that are affected by the inclusion of particular taxa are represented as unresolved polytomies. The lack of resolution indicates that a major portion of the phylogenetic estimate can change based solely on which terminal taxa are included in the analysis. However, it is the removal of *Acanthogilia* and the placement of *Cantua* that are responsible for much of the lack of resolution (i.e., the Loeselieae Clade). Resolved portions of the consensus tree represent relationships not effected by the current sampling. This includes the Gilieae Clade, the *Giliandra* clade, the *Gilia foetida-G. rigidula* clade, the Linanthieae Clade, the *Ipomopsis longiflora-I. multiflora-I. gunnisonii* clade, and the *Cobaea scanned-C. biaurita* clade.

Skewness of the distribution of random trees.—One of the primary limitations to evaluation of the skewness of the distribution of sets of random trees for a data set of this size is that no 95% confidence limit for skewness values (g1) has been determined for data sets in excess of 25 terminal taxa (Hillis 1991; Hillis and Huelsenbeck 1992; Hulsenbeck 1991). However, as a conservative test, I have used the 95% confidence limit of the 25-taxon tree for those trees with 25 or more taxa (Hillis and Huelsenbeck 1992).

The distribution of tree length for a set of 10,000 trees based on all 56 sequences is significantly skewed, with a g1 statistic of −0.6622. Following Hillis (1991), I removed well-supported clades (as measured by bootstrap values of 95% or greater), except for one
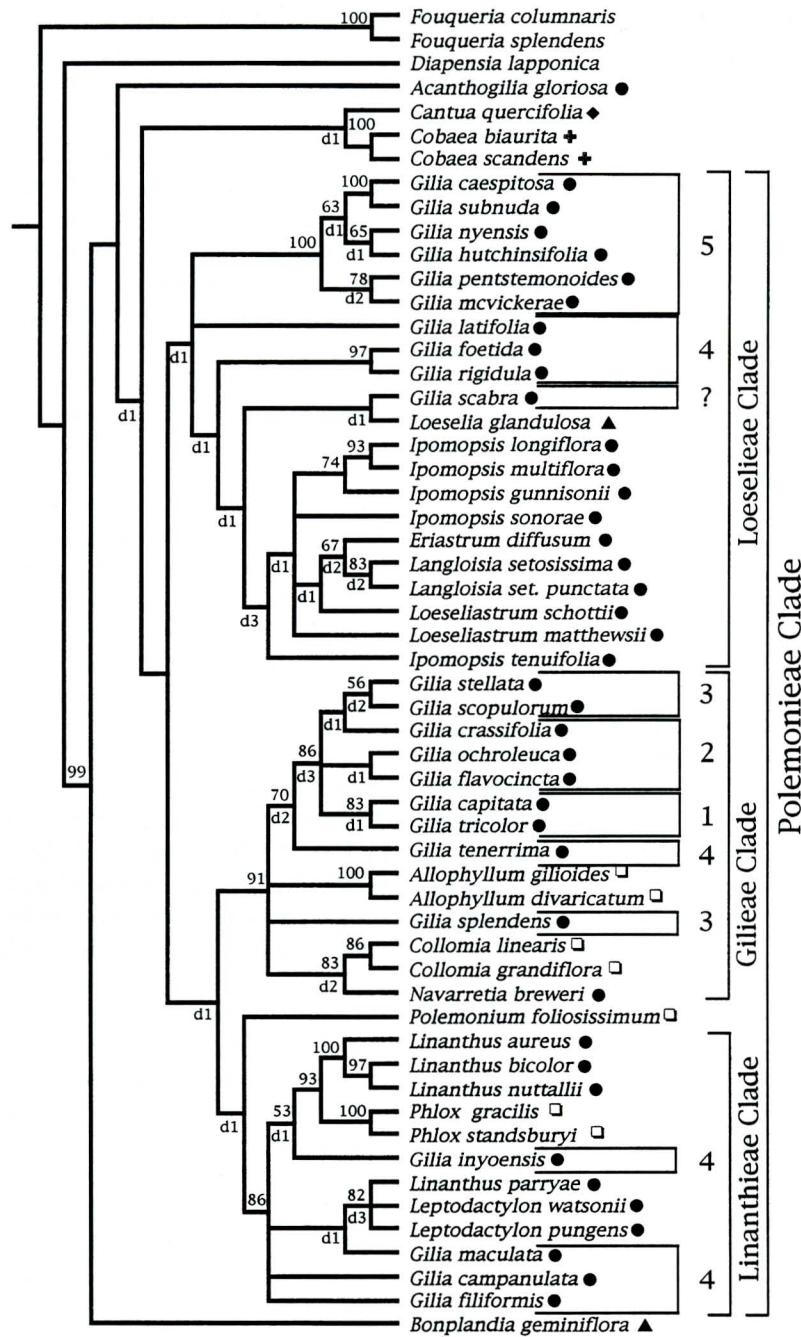
Fig. 1. The strict-consensus of 1080 trees of 1646 steps (CI = 0.423, RC = 0.263), resulting from the PAUP analysis of the manually realigned PILEUP alignment of 53 members of Polemoniaceae and three outgroup members. The number below each internode is the decay index for that branch, ranging from d1 to d3. The frequency at which clades occur in 100 bootstrap replications is shown as a percentage, above each internode. Tribal classification of Grant (1959) is indicated by the symbol following each ingroup taxon name (✚ = Cobaeeae; ◆ = Cantuaeae; ▲ = Bonplandieae; □ = Polemonieae; ● = Gilieae). Intrageneric classification of *Gilia* is indicated by the numbered brackets (1 = sect. Gilia; 2 = sect. Arachnion; 3 = sect. Saltugilia; 4 = sect. Giliastrum; 5 = sect. Giliandra). *Gilia scabra* is labeled "?" because Grant erroneously considered it synonymous with *Linanthus nuttallii*.

randomly selected representative, leaving 46 taxa. Another set of 10,000 random trees was generated. Again, the distribution is significantly skewed: $g1 = -0.4441$. This was followed by removal of clades with bootstrap values of 90% or greater, save one representative, leaving 31 taxa, and skewness was determined. Similarly, the distribution remains significantly skewed ($g1 = -0.4881$). This process continued, using 21, 15, 12, 11, 10, nine, and eight taxa, each time removing members of the most well supported clades. In all cases, the $g1$ value significantly differs from the expected value of random data. Similar results (Porter unpublished) are obtained from permutation tail probability tests (Faith and Cranston 1991).
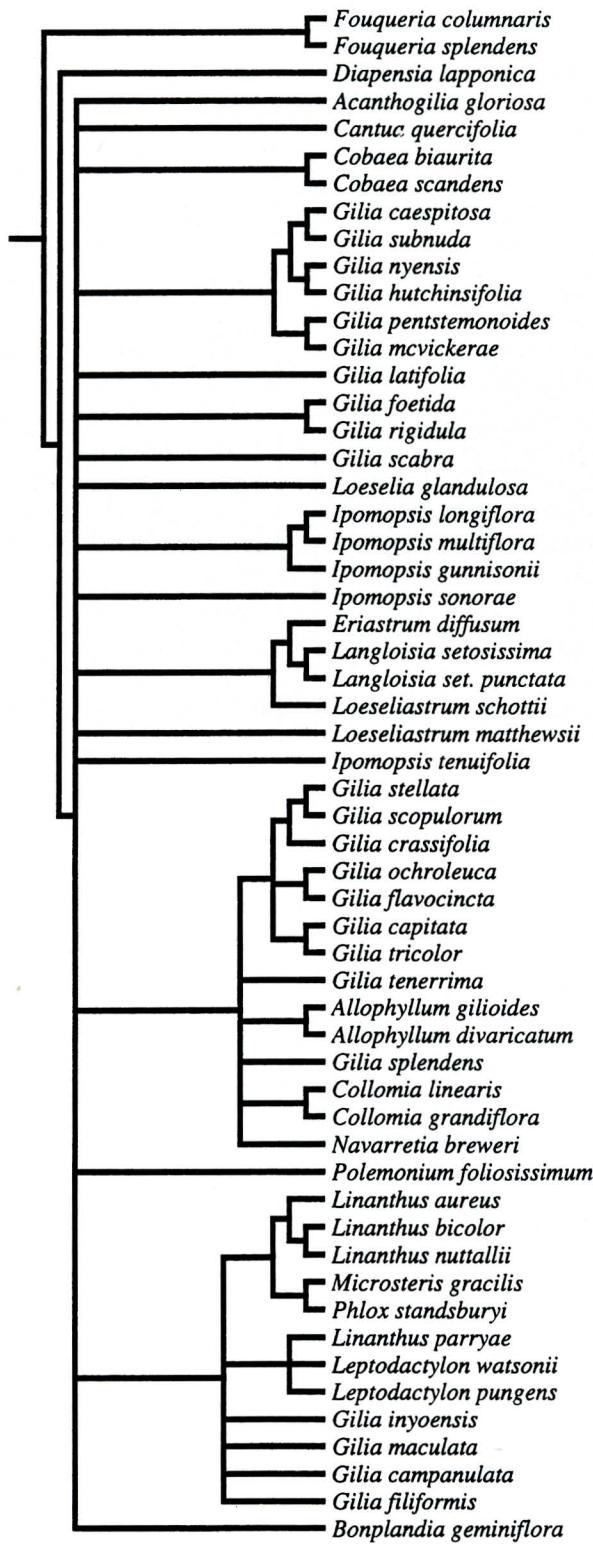
Fig. 2. Lanyon's jackknife strict-consensus tree, identifying those regions of the phylogeny affected by taxon inclusion. Resolved portions of the tree indicate those nodes not influenced single taxon removal. Unresolved portions (compare with Fig 1) change based on the removal of a single terminal taxon.

The tree includes the following taxa:

Fouqueria columnaris
Fouqueria splendens
Diapensia lapponica
Acanthogilia gloriosa
Cantua quercifolia
Cobaea biaurita
Cobaea scandens
Gilia caespitosa
Gilia subnuda
Gilia nyensis
Gilia hutchinsifolia
Gilia pentstemonoides
Gilia mcvickerae
Gilia latifolia
Gilia foetida
Gilia rigidula
Gilia scabra
Loeselia glandulosa
Ipomopsis longiflora
Ipomopsis multiflora
Ipomopsis gunnisonii
Ipomopsis sonorae
Eriastrum diffusum
Langloisia setosissima
Langloisia set. punctata
Loeseliastrum schottii
Loeseliastrum matthewsii
Ipomopsis tenuifolia
Gilia stellata
Gilia scopulorum
Gilia crassifolia
Gilia ochroleuca
Gilia flavocincta
Gilia capitata
Gilia tricolor
Gilia tenerrima
Allophyllum gilioides
Allophyllum divaricatum
Gilia splendens
Collomia linearis
Collomia grandiflora
Navarretia breweri
Polemonium foliosissimum
Linanthus aureus
Linanthus bicolor
Linanthus nuttallii
Microsteris gracilis
Phlox standsburyi
Linanthus parryae
Leptodactylon watsonii
Leptodactylon pungens
Gilia inyoensis
Gilia maculata
Gilia campanulata
Gilia filiformis
Bonplandia geminiflora

DISCUSSION

Sequence data from the two intergenic ITS spacers provide a compelling insight into some aspects of phylogeny. However, the phylogenetic inferences from ITS sequences contains both well-supported conclusions and highly questionable ones. In general, the weakly supported portions of the phylogeny are nodes associated with the earliest diverging lineages within the family. The lack of well-supported relationships at the base of the tree could be due to biological reasons or artifact. Biological reasons could include rapid diversification, low nucleotide substitution rate, or spurious long-branch attraction. Artifacts also could result from the difficulty in aligning the more divergent lineages and the treatment of the large blocks of question marks in the data matrix that result from indels. Fortunately, even with the ambiguity of relationships associated with the earliest diverging lineages, the robust portions of the phylogeny provides an understanding of relationships in Polemoniaceae that directly conflicts with current classification. This insight includes the pattern of diversification of the phlox family as well as issues of monophyly of the genera *Gilia*, *Ipomopsis* and *Linanthus*.

Monophyly of Polemoniaceae

The only well-supported node (based on bootstrap, decay and Lanyon's jackknife) in the basal region of the ITS phylogeny is the Polemoniaceae clade. Regardless of differential weighting of indels and/or transitions versus transversions, ITS sequences support the monophyly of Polemoniaceae. There is no support for the treatment of *Cobaea* as a separate family, Cobaeaceae (Don 1824). However, this study provides at best a weak test of the monophyly of Polemoniaceae. If the phylogeny is treated as unrooted, it can be recognized that a monophyletic Polemoniaceae is the result of the two longest branches (*Diapensia* and the *Fouqueria* clade) being united. These two lineages are characterized by nucleotide changes at nearly 30% of all potentially informative sites and between 37% and 50% of potentially informative sites not involving indels, raising the potential for long-branch attraction. This caution is tempered by the concordant support for a monophyletic Polemoniaceae from both chloroplast genes *mat*K (Steele and Vilgalys 1994; Johnson et al. 1996) and *ndh*F (A. Prather, pers. comm.) and nuclear 18S sequences (as cited in Johnson et al. 1996).

Diversification of Polemoniaceae

The evolution and diversification of Polemoniaceae have been characterized by recent authors (e.g., Grant 1959) as a lineage of tropical origin that has radiated into the temperate regions. Grant postulates the inde-
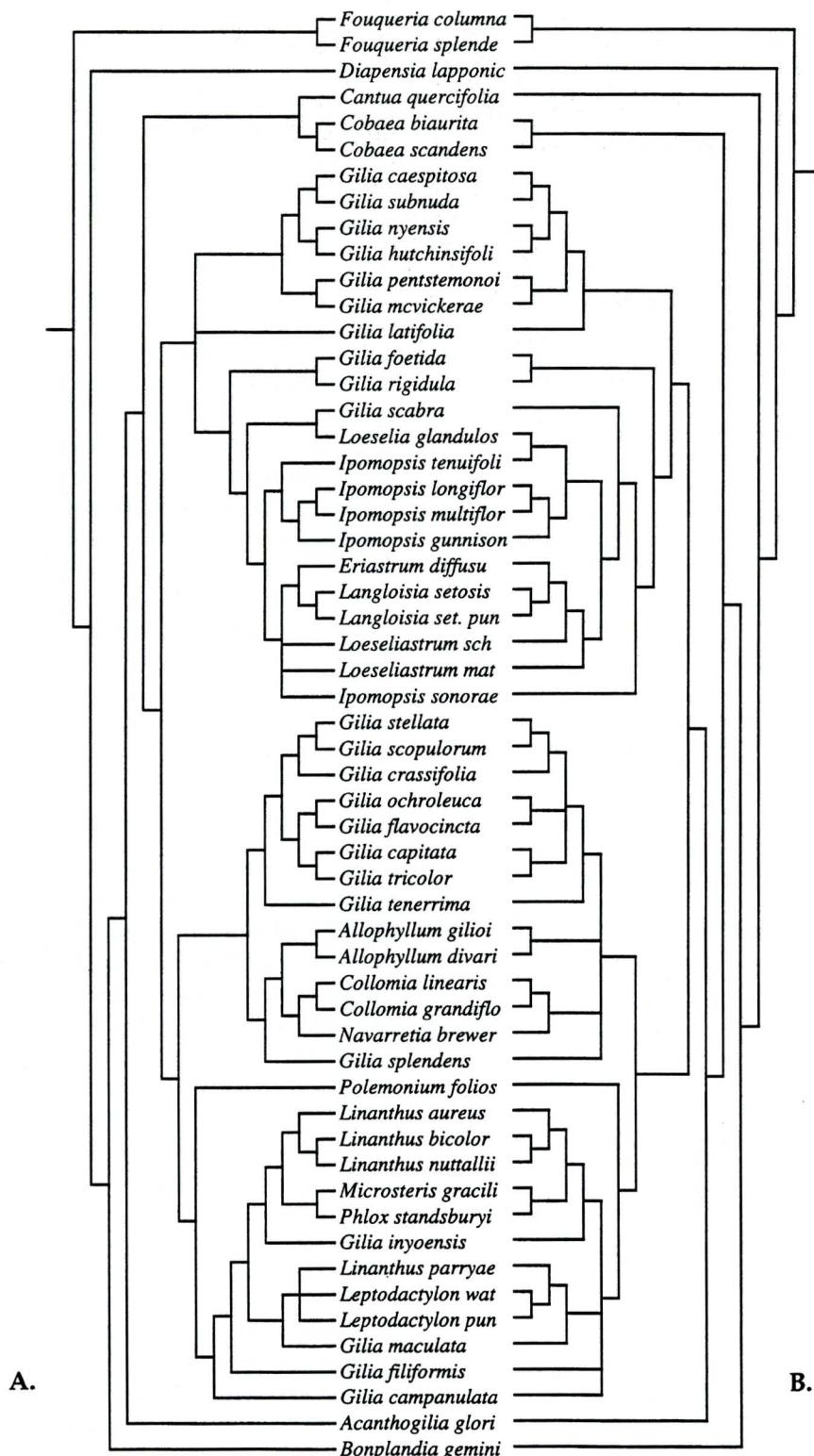
Fig. 3. Comparison of two parsimony analyses differing in the weighting of gaps and transitions vs. transversions. Tree A is the strict consensus of 12 trees, resulting from the analysis in which no transition/transversion bias is imposed but all gap positions were treated as a fifth nucleotide state (GAPMODE = FIFTHSTATE option in PAUP). The analysis for tree B utilized a 1.1 to 1.0 transition to transversion bias but all gap and alignment-ambiguous positions were removed. Tree B is the strict consensus of the resulting eight trees.

pendent origin of two temperate lineages, tribes Polemonieae and Gilieae, from different tropical groups (tribes Cobaeeae and Bonplandieae, respectively). This tropical-temperate distinction has persisted. The phylogenetic inference based in *matK*, presented by Steele and Vilgalys (1994), is suggested to support a tropical origin although the strict consensus tree shows ambiguous relationships between the included genera of tropical distribution. Steele and Vilgalys also propose a single "temperate" lineage, rather than two . Johnson et al. (1996) present a more complete *matK* phylogeny in which *Cantua, Cobaea,* and *Bonplandia* form a "tropical clade." Aside from *Acanthogilia,* the remaining genera are included in a large "temperate clade." Given that *Acanthogilia* is not tropical in distribution, the ancestral condition, if reconstructed using parsimony, is dependent on the distribution of the outgroups. Because Diapensiaceae are temperate in distribution and Fouquieriaceae are subtropical to tropical the ancestral character state assignment is ambiguous.

The phylogeny based on nucleotide sequences from ITS 1 and 2 show *Acanthogilia, Cantua, Cobaea,* and *Bonplandia* as a paraphyletic group, positioned as the earliest diverging lineages of the family. This could be interpreted as support for a tropical origin of Polemoniaceae. However, the relationships between these four genera, as inferred by ITS, have been shown to change based on taxon inclusion, character weighting (indels and transition-transversion) and how alignment-ambiguous sites are treated. It is difficult to argue that the ITS phylogeny is error free in this region. Given the uncertainty of relationships at the basal nodes the ancestral character state assignment is also uncertain. The question of a tropical origin for the family remains unanswered. Such determination again requires a more rigorous assessment of sister and outgroup relationships for Polemoniaceae than is presented here or in previous studies (e.g., Chase et al. 1993; Steele and Vilgalys 1994; Johnson et al. 1996).

The ITS spacer sequences support the remaining genera of Polemoniaceae (i.e., *Gilia, Ipomopsis, Eriastrum, Langloisia, Loeseliastrum, Loeselia, Allophyllum, Collomia, Navarretia, Phlox, Linanthus, Leptodactylon* and *Polemonium*) as a monophyletic group. This lineage has been referred to as the "Temperate Clade" (Steele and Vilgalys 1994; Johnson et al. 1996). While it is true that the majority of species in this clade occur in temperate regions, such a characterization may obscure the geographic patterns associated with diversification and dispersal within the lineage. This lineage includes several clades that are either wholly or partly tropical in distribution. For example, *Loeselia* ranges from southernmost Arizona, USA, through Mexico and Central America to Columbia, but all species occur in the tropics of Mexico.

Similarly, *Gilia* section *Giliastrum* (exclusive of *Gilia tenerrima, G. filiformis, G. campanulata, G. inyoensis,* and *G. maculata*) occurs not only in the temperate southwestern United States, but also the subtropical and tropical latitudes of both Mexico and South America (Peru, Bolivia, and Argentina). These examples serve to point out this clade includes tropical, subtropical, temperate, and even boreal lineages. It is not evident where the early diversification took place. Indeed, Grant's (1959) hypothesis of several independent lineages dispersing to and radiating within the temperate regions cannot be ruled out. For convenience I will refer to this clade as the Polemonieae Clade (Fig. 1).

The Polemonieae Clade is comprised of members from three of Grant's (1959) tribes (tribes Gilieae, Polemonieae, and Bonplandieae). None of these three tribes is supported as monophyletic (Fig. 1). Rather, three primary lineages, in addition to the genus Polemonium, make up this clade in all of the most parsimonious trees. The support for relationships between these clades is weak, but the membership of the three lineages is consistent regardless of weighting of transitions vs. transversions or indels and it is corroboration from other molecular phylogenetic studies (Steele and Vilgalys 1994; Prather 1995; Johnson et al. 1996). I refer to these clades as the Loeselieae, Gilieae, and Linanthieae Clades (Fig. 1). Note that the endings are used to distinguish the clades from generic names and are not intended to imply a particular hierarchic rank.

The Loeselieae Clade, which includes *Gilia* section *Giliandra, Gilia* section *Giliastrum* (exclusive of *Gilia campanulata, G. inyoensis, G. maculata* and *G. filiformis*), *Gilia scabra, Loeselia, Ipomopsis, Langloisia, Loeseliastrum,* and *Eriastrum,* is the sister group to the remaining members of the Polemonieae Clade. The members of this clade have traditionally been included within tribe Gilieae, with the exception of *Loeselia,* which has been placed in Bonplandieae (Grant 1959). The potential relationship between *Loeselia* and certain members of tribe Gilieae was recognized by Grant (1959), given his hypothesis that tribe Gilieae has its origin within or near *Loeselia.* Support, as measured by bootstrap, jackknife and decay, is low for many of the nodes within the Loeselieae Clade. Even so, the phylogenetic inference is highly consistent with that of *matK* (Johnson et al. 1996). Several noteworthy points can be made concerning the phylogenetic inferences based on ITS sequences. First, the two subspecies of *Langloisia setosissima* are supported as the sister group of *Eriastrum.* The sister group of this (the Eriastrum-Langloisia Clade) is *Loeseliastrum schottii,* supporting Timbrook's (1986) assertion that *Langloisia s.s.* and *Loeseliastrum* do not represent a monophyletic group. This represents a direct conflict between ITS and phylogenetic inferences from chloro-

plast *mat*K (Johnson et al. 1996) and *trn*L intron (Porter, unpubl.) sequences, that support *Langloisia* and *Loeseliastrum* as sharing most recent common ancestry. Second, note that *Ipomopsis* is not monophyletic. The placement of *Ipomopsis tenuifolia* as sister to *Ipomopsis, Eriastrum, Langloisia,* and *Loeseliastrum* is not surprising, given that this member of the Giliopsis group was formerly included in *Loeselia* (Gray 1876; 1886). *Ipomopsis sonorae* is also not unambiguously more closely related to other *Ipomopsis* species than to other genera. There may be three lineages, representing a grade, included in the current circumscription of *Ipomopsis*. The paraphyly of *Ipomopsis* is corroborated by chloroplast DNA sequence data (Johnson et al. 1996; Porter, unpubl.) and a more detailed ITS analysis of the Loeselieae Clade (Porter, in prep.). Third, the members of *Gilia* within this clade are neither monophyletic (although, *Gilia* section *Giliandra* is monophyletic) nor closely related to those *Gilia* associated with the type (i.e., *Gilia* section *Gilia*, type = *G. laciniata* R. & P.). In fact, *Gilia* section *Giliastrum*, as circumscribed by Grant (1959) is polyphyletic. In addition, *Gilia scabra*, considered by Grant (1959: 140) synonymous with *Linanthus nuttallii*, is suggested to share common ancestry with *Loeselia*.

The Gilieae Clade includes *Allophyllum, Collomia, Navarretia, Gilia* sections *Saltugilia, Arachnion, Gilia,* and one member of section *Giliastrum*. Historically, *Allophyllum* has been variously treated within *Collomia* (Bentham 1845; Gray 1870), close to *Collomia* in tribe Polemonieae (Grant and Grant 1955; Grant 1959),or included within *Gilia* (Nuttall 1848; Brand 1907; Mason and Grant 1948; Cronquist 1984), in tribe Gilieae. *Navarretia* has been of equally uncertain affiliation: included within *Gilia* by Gray (1870); allied with *Collomia, Cantua* and *Gymnosteris* in the *Collomia* tribe (Wherry 1945); or within Tribe *Gilieae*, near *Langloisia* and *Leptodactylon* (Grant 1959). It is perhaps not surprising that ITS sequences support a close relationship between *Navarretia, Collomia, Allophyllum* and a portion of what is now considered *Gilia*. Even so, this is a striking noncorrespondance with traditional taxonomy because it not only segregates only a portion of *Gilia* and allies it with *Navarretia* (both members of Grant's tribe Gilieae), but also strongly supports the relationships between this group and two members of tribe Polemonieae, *Allophyllum* and *Collomia*. The monophyly of the Gilieae Clade, supported here by ITS sequences, is both well supported in terms of bootstrap values and Lanyon's jackknife analysis and also corroborated by sequence data from the chloroplast gene *mat*K (Steele 1992; Steele and Vilgalys 1994; Johnson and Soltis 1995). The correspondence between ITS and *mat*K gene phylogenies and the relative robustness of this portion of the ITS

phylogeny strongly supports the monophyly of this group.

Perhaps the most intriguing of the hypothesized relationships based on ITS sequence data involves the Linanthieae Clade. This clade is comprised of the genera *Linanthus, Leptodactylon* and *Phlox,* as well as four species of *Gilia* currently included in section *Giliastrum*. The Linanthieae Clade is well supported but very inconsistent with current classification. Although there is a long history associating *Linanthus* and *Leptodactylon* together (Gray 1870; Brand 1907; Wherry 1940; 1945; Grant, 1959) and they are currently both included in tribe Gilieae, *Phlox* (currently in tribe Polemonieae) has not been considered closely related. While floral morphology of *Leptodactylon* has been compared with that of *Phlox* (Gray 1870), no direct relationship was espoused. It is noteworthy that *Linanthus, Phlox* and *Leptodactylon* all possess pantoporate pollen grains (Marticorena 1961; Stuchlik 1967; Taylor and Levin 1975), very similar leaf flavonoid and phenolic chemistry (Smith et al. 1977; Dean et al. 1978; Smith et al. 1982), and possess opposite leaves, an unusual trait in Polemoniaceae.

The Linanthieae Clade is also intriguing because four species of *Gilia* (*G. campanulata, G. inyoensis, G. filiformis,* and *G. maculata*) are also members. These species have historically been closely aligned (see for example Brand 1907); however, most treatments prior to 1989 maintained *G. maculata* in *Linanthus,* while the remaining species were retained in *Gilia* sect. *Giliastrum*. Patterson (1989) has cautiously argued that, in spite of the unique traits possessed by *G. maculata*, there is sufficient morphological similarity between *Gilia campanulata, G. inyoensis, G. filiformis* and *G. maculata* to include them in the same genus, i.e., *Gilia*. ITS sequence data, presented here, supports in essence Patterson's proposition; however, this group of species is related not to members of *Gilia*, but to *Linanthus, Leptodactylon* and *Phlox.* It is also important to note that ITS sequences do not support the monophyly of these species.

Gilia is not monophyletic

The genus *Gilia* has remained, over the last 100 years, a recurring taxonomic problem, the circumscription of which has changed radically. Mason and Grant (1948) correctly point out that all of the herbaceous genera of the Polemoniaceae, with the exception of *Polemonium* and *Phlox,* have historically been placed in *Gilia*. One of the most broad interpretations of the genus was that of Asa Gray (1886). While recognizing that *Gilia* was "certainly a polymorphous . . . genus" (Gray 1870: 262), he included the currently recognized genera *Langloisia, Loeseliastrum, Gymnosteris, Leptodactylon, Linanthus, Navarretia, Ipomopsis* and *Er-*

iastrum within *Gilia.* The most recent classification of the family (Grant 1959) has maintained all these segregate genera. However, as noted above, Cronquist (Cronquist 1959; Cronquist 1984) has included *Ipomopsis* and *Allophyllum* within *Gilia.*

ITS sequence data provide insight into this conflict. The monophyly of representatives of *Gilia* sects. *Gilia* (here represented by *G. capitata* and *G. tricolor*), *Arachnion* (represented by *G. crassifolia, G. ochroleuca* and *G. flavocincta*), *Saltugilia* (represented by *G. stellata* and *G. scopulorum*), and *Giliastrum*, sensu Grant 1959 (represented by *G. tenerrima*) is supported (Fig. 2). Conspicuously absent from this clade are *Gilia scabra* and members of *Gilia* sects. *Giliastrum* (represented by *G. foetida, G. rigidula, G. latifolia, G. campanulata, G. inyoensis, G. filiformis,* and *G. maculata*), and *Giliandra* (represented by *G. caespitosa, G. subnuda, G. pentstemonoides, G. mcvickerae, G. hutchinsifolia* and *G. nyensis*). All of these samples, except *G. campanulata, G. inyoensis, G. filiformis,* and *G. maculata* (in the Linanthieae Clade), are within the Loeselieae Clade. The most parsimonious placement of any one of these species into the Gilieae Clade requires between 27 and 51 steps (ACCTRANS reconstruction). Moreover, if representatives of *Gilia* sensu Grant (1959) are constrained to be monophyletic, three minimal-length trees are found of tree length 1693, 47 steps longer than the unconstrained analysis. The inclusion of representatives of *Ipomopsis* into *Gilia* (as proposed by Cronquist) does not ameliorate this situation. If *Ipomopsis* is constrained to be included in *Gilia,* sensu Cronquist, the eight trees found are 59 steps longer than the unconstrained analyses. Including additional taxa to make the representatives of *Gilia* monophyletic—if one considers the minimal-length ITS trees—will result in a group as morphologically diverse as Gray's circumscription of *Gilia.* If this is considered an unacceptable treatment (i.e., considering the Polemonieae Clade as a single genus, *Polemonium*), then the genus *Gilia* is must be recognized in the more strict sense (i.e., *G. tenerrima* and its the sister group), it will be necessary to remove those clades that do not share common ancestry.

Conclusions

Sequences of the two internal transcribed spacers, ITS1 and ITS2, have been demonstrated to provide reliable phylogenetic estimates for portions of Polemoniaceae. The bootstrap, decay index, Lanyon's jackknife, and distribution of the tree length of random trees provide support for much of the phylogenetic inference. These include the *Giliandra* clade, *Gilia foetida-G. rigidula* clade, the *Allophyllum-Gilia* clade, the *Linanthus-Phlox-Leptodactylon* clade, and the *Ipomopsis gunnisonii-I. longiflora-I. multiflora* clade. Un-

fortunately, resolution of relationships among most of the major lineages, representing the basal radiation of the family, is suspect. Even given this limitation, ITS data unambiguously fail to support the current classification of Polemoniaceae that recognizes two temperate tribes, Polemonieae and Gilieae (Table 1). Rather, all of the genera, exclusive of *Bonplandia, Cantua* (and presumably *Huthia*), *Cobaea,* and *Acanthogilia,* are within a single clade, with members of Grant's tribes, Polemonieae and Gilieae variously related. Further, it has been shown that ITS sequence data does not support *Gilia,* as currently circumscribed, as a monophyletic group. The taxa now treated as *Gilia* represent at least five independent lineages, and potentially more (Fig. 1).

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Appendix I

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                              10        20        30        40        50        60        70        80        90        100       110
                              .         .         .         .         .         .         .         .         .         .         .

Fouqueria columnaris          TCGATACCTGCCTAGCAGAACGACCTGTGAACTCGT.CTCCAAAATGAGGTGAT.TCGC.GGGTGGGC.CTCC..GGTCCCCCCGTGGGGAA..CCTTCTCGGGGAAGCG
Fouqueria splendens           TCGATACCTGCCTAGCAGAACGACCTGTGAACTCGT.CTCCAAAATGAGGTGAT.TCAY.GGGTGGGC.CTCT..GGTCCTCCCGTGCGGAA..CCTTATCGGGGAAACG
Diapensia lapponica           TCGATATCTGCACAGCAGAACGACCCGCGAACTAGTATTTCACTTTG.GGAGAT.GCATTGGGCGCTC.TAAT..CCCCCT.CCTTTGTGCT..CTCCCAC......GCC
Acanthogilia gloriosa         TCGAAACCTGCCTAGCAGAACGACCCGCGAACCTGTATTCCAAACTCGGGT.ACG.GGCGTTGGC.CTC...GGTCCT.CGCCCGTTCC..CCCT....CCGGA...
Cantua quercifolia            TCGAAA.CTGTCCCGAAACACGACCCGTGAACTTGTATTCTCAYATCGGGT.GC.ATG.GTCGATCGG.CTTC..GGTCCT.CGGCCATTGCTACCCCGTTGGTGGT.GT
Bonplandia geminiflora        TCGAAACCTGCCTAGCAGAACAACCCGCGAACTCGT.TTAACATATCGGG..GC.AC...TCGGC....CTTC..GGA.CT.CCGGTCCTGG.GTCGTTT.GCCCCTCGT
Cobaea biaurita               TCGAAACCTGCATRGCAGAAAGACCYGTGAACTTAT.TTTTAAACTCAG.TGGC.ATG.ATTGGGGA..CTYC..GGTCCT.CCTTTGTTGY.CCCTTT.RCTTGTTG.
Cobaea scandens               TCGAAACCTGCATGGCAGAAAGACCTGTGAACTTAT.TTTTAAACTCAG.TGGC.ATG.ATTGGGGA..CTTC..GGTCCT.CATTTGTTGC.CCCTTT.GCT.GTTG.
Gilia caespitosa              TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAAACTAAA.TCGGGTGGC.ACG.AGCATGGTC.CTTT..GGTCCA.AACTCGTTGC.CCCCA..GCTGGGT..
Gilia subnuda                 TCGAAACCTGCCTAGCAGAACGACCC.CCAACTTGTAAACTAAA.TCGGGTGGC.ACG.AGCATGGTC.CTTT..GGTCCA.AACTCGTTGC.CCCCA..GCTGGGT..
Gilia pentstemonoides         TCGAAACCTGCCTAGCAGAMCGACCCGCGAACTTGTAAACTAAA.TCGGCTGGC.ACG.GGCATGGSS.CTTT..GGTCCT.AGCCCGTTGC.CCCCA..GCTGGGT..
Gilia mcvickerae              TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAAACTAAA.TCGGGTGGC.ACG.GRCATGGTC.CTTT..GGTCCT.AGCTCGTTGC.CCCCA..GCTGGGT..
Gilia nyensis                 TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAAACTAAA.TCGGGTGGC.ACG.AGCATGGTC..CTTC..GGTCCT.AGCTCGTTGC.CCCCA..GCTGGGT.T
Gilia hutchinsifolia          TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAAACTAAA.TTGGGTGGC.ACG.AGCATGGTC.CTTT..GGTCCT.ACCTCGTTGC.CCCCA..GCTGGGC..
Gilia latifolia               TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAAACTACA.TCGGGTGGC.ACG.GGCGTCGTA.CTCC..GGTCCT.CGTTCGTTGC.CCCCT..GCTGGGTGT
Gilia foetida                 TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TCGGGA.GC.ACG.GTCGTGGAC.CTTT..GGTCCT.TGCTCG.TGC.CCC.A..GCTGGGA..
Gilia rigidula                TCGAAACCTGCCTASCAGCACGACCCGCGAACTTGTAATCAAAA.TCGGGT.GC.ACG.GTCGTGGAC.CTTC..GGTTCA.CGATCGATGC.CCC.A..GCTGGGA..
Gilia scabra                  TCGAAACCTGTCTAGCAGAACGACCCGCGAACTTGTAATCACA.CCGGGT.GC.ACG.GACGTGGAC.CTCC..GGTCCT.TGGCCGTTGC.ACCCA..GCCGGAT..
Loeselia glandulosa           TCGAAACCTGCCTAGCAGAACGACCCGCGAACATGTTATCAAA.CTCGGGTGGC.GCG.GGCGTGGGC.CTTTT.GGTCCT.CGCTCGTTCC.CCCCA..GCCGACA..
Ipomopsis longiflora          TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAACA.TCGGGTGGC.ACG.GGTGTGGTC.CTAT..GGTCCT.CCCCCGTTGC.CACTA..GCTGACA..
Ipomopsis gunnisonii          TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAACA.TCGGGTGGC.ACG.GGTGTGGTC.CTAT..GGTCCT.CACCCATTGC.CCCTA..GCTGACG..
Ipomopsis multiflora          TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAACA.TCGGGTGGC.ACG.GGTGTBGTC.CTAT..GGTCCT.CCCCCGTTGC.CCCTA..GCTGCCA..
Ipomopsis tenuifolia          TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TCGGGTGGC.AC..GGCGTGGTC.CTTTTTGGTCCT.CGCCCGTTGC.CCCTA..GCTGACT..
Ipomopsis sonorae             TCGAAGCCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TCAGGT.GCTGCG.GGTGGGGTC.CTTC..GGTCCT.CGCTCGTTGC.CCCTA..GCCGACA..
Eriastrum diffusum            TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TTGGGCGGC.ACG.GGTGTSGCC.CTCTATGGTCCT.CGCTCGTTGC.CCCTA..GCCGACA..
Loeseliastrum matthewsii      TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCWAAA.TCGGGTGGC.ACG.GGTGTGGTC.CTWT..GGTCCY.CGCTCGTTGC.CCCTA..GCTGACA..
Loeseliastrum schottii        TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TTGGGTGGC.ACG.GGTGTGGTC.CTTT..GGTCCC.CGCTCGTTGC.CCCTA..GCTGACA..
Langloisia setosissima        TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TTGGGTGGC.ACG.GGCTTGGTC.CTCT..GGTCCT.CGCTCGTTGC.CCCTA..GCCGGCA..
Langloisia set. punctata      TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAA.TTGGGTGGC.ACG.GGCTTGGTC.CTCC..GGTCCT.CGCTCGTTGC.CCCTA..GCCGGCA..
Gilia stellata                TCGAAACCTGCCTAGCAGCACGACCCGCGAACTTGTATTCAAAACTAGGGTGGT.GCG.GGCSTTGTATCTC...GGTACG.CGCTCGTTCC.CCCAAC.GCTAGGA..
Gilia crassifolia             TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTAATCAAAACTAGGGTGG..GCG.GGCGTTGTATCTC...GGTACT.CGCTCGTTCC.CCCAAC.GCTAGGA..
Gilia scopulorum              TCGAAACCTGCCTAGCAGCACGACCCGCGAACTTGTATTCAAAACTAGGGTGGT.GCG.GGCGTTGTATCTC...GGTACT.CGCTCGTTCC.CCC.AC.GCTAGGA..
Gilia ochroleuca              TCGAAACCCGCCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTAGGGTGG..GCG.GGTGTTGTATCTC...GGTACT.CGCTCGCTC..CCCCAC.GCTAGGA..
Gilia flavocincta             TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATTCTAAACTAGGGTGGGCACG.GGTGTTGTATCTT...GGTACTACGCTCGTTC..CCCCAC.GCTAGGA..
Gilia tenerrima               TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTWGGGTGGT.GYG.GGCGTTGTATCTC...GGTACT.TGCHCGTTC..CCCCAC.GCTAGGA..
Gilia capitata                TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTAGGGTGGT.GCG.GGCSTTGTATCTC...GGTACT.CGCCCGTTC..CACCTC.GCTAGGA..
Gilia tricolor                TCGAAACCTGCCTAGCAGAACGACCCGCGAACATGTATTCAAAACTTGGGTGGT.RCG.GGTGTTGTATCTC...GGTACT.CGCTCGTTC..CCCCAC.GCTAGGA..
Allophyllum gilioides         TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTCGGGTGGT.ACG.GGCGTTGTATCTC...GGTACT.CGCTCGTTTC.CCCCAC.GCTAGGA..
Allophyllum divaricatum       TCGAAACCTGCCTAGCAGAACGACCCGTGAACTTGTATTCAAAACTCGGGTGGT.ACG.GGCGTTGTATCTC...GGTGCT.CGCTCGTTTC.CCCCAC.GCTAGGA..
Gilia splendens               TCGAGACCTGTCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTCGGGTGGC.TCG.GGCGTCGTA.CTTC..GGTGCT.CGCGCGTTGC.CCCCAC.GCTAGGA..
Collomia linearis             TCGACAC.TGCATAGCAGAACGACCCGTGAACTTGTATTCAAAACTTGGGTGGT.GCG.GGTGTTGTATCT.C..GGTACT.CGCTCGTTAC.CCCCAC.GCTAGGA..
Collomia grandiflora          TCGACACCTGCCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTCGGGTGGT.GCG.GGTGTAGTACT..C..GGTACT.CGCTCGTTAC.CCCCAC.GCTAGGA..
Navarretia breweri            TCGAAACCTGCCTAGCAGAACGACCCGTGAACTTGTATTCAAAACTTGGGTGGT.GCG.GGCATTGTATCATC..GGTACT.CGCTCGTTAC.CCCCAC.GCTAGGA..
Polemonium foliosissima       TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATTCAAAACTCGGGTGGC.ACG.ARCGTAGTA.CTTC..GGTACT.CGCCCGTYGC.CCCCTC.GCTGTGA..
Linanthus aureus              TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATTACAAACTTGGGGGGTGACG..AC.TTGTG.CTTC..GGTACT...CACGTTGC.CCCCTT.GCTGGTG..
Linanthus parryae             TCGAAACCTGCCTAGCAGAACGACCCGCGAACTCGTATTCAAAACTCGGGCGGC.ACA.TGC.TTGTG.CTTC..GGTACT.CGCATGTTGC.CCCCTTTGCTGGTG..
Gilia maculata                TCGAAACCTGCCTAGCAGAACGACCCGTGAACTCGTATTCAAAACTCGGGCGGT.GCG.GAC.ATGTG.CTTC..GGTACT.TGTCCGTTGC.CCCCTT.GCTGGTG..
Gilia campanulata             TCGAAACCTGCCTAGCAGAACGACCCGTGAACTCGTATTCAAAACTCGGGTGGC.ATG.GGC.TTGTG.CTTC..GGTGCT.CGCCTGTTGC.CCCCTT.GCTGGTG..
Gilia inyoensis               TCGAAACCTGCCTAGCAGAACGACCCGTGAACTCGTATTCAAAACCTGGG.GGT.GCG.GGC.TTGTG.CTTC..GGTACT.CGCCCGTTGC.CCCCTC.GCTGGTG..
Gilia filiformis              TCGAAACCTGCTTAGCAGAACGACCCGTGAACTCGTATTCAAAACTCGGGTGGC.GCG.GGC.TTGTG.CTTC..GGTACT.CGTCCGTTGC.CCCCTC.GCTGGTG..
Phlox gracilis                TCGACACCTGCCTAGCAGCAKAACGACCCGCGAACTTGTATTCAAAACTTGGGTTGT.GG.TGC.TTCGG.CTTC..GGCCGT.TGCTCG.TGCATTGCCCTCGCTGGTG..
Phlox standsburyi             TCGAAACCTGCCTAGCATAACGACCCGCGAACTTGTATTCAAAACTTGGGTTGT.GCG.TGC.TTCT..CTTC.CGGCCGT.TGCTCG.TGT..AACCWC.GCTGGTG..
Leptodactylon watsonii        TCGAAACCTGCCTAGCAGAACGACCCGCGAACTCGTATCCAAAACTCGGGTGGY.GCA.GGC.TTGTG.CTTC..GGTACT.CGCCTGTTGC.CCCCTTTGCTGGTG..
Leptodactylon pungens         TCGAAACCTGCCTAGCAGAACGACCCGCGAACTCGTATTCAAAACTCGGGTGGC.GCA.TGC.TTGTG.CTTC..GGTACT.CGCCTGTTGC.CCCCTTTGCTGGTG..
Linanthus bicolor             TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATCACAAACTTGGGGGGT.GAT.GAC.TTGTG.CTTT..GGTACT.TGTATGTTGC.CCCCTT.GCTGGTG..
Linanthus nuttallii           TCGAAACCTGCCTAGCAGAACGACCCGCGAACTTGTATCACAAACTTGGGTGGT.GAC.GAC.TTGTG.CTTC..GGTACT.TGCACGTTGC.CCCCTT.GCTGGTG..
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Appendix 1. Aligned nrDNA ITS1 and ITS2 nucleotide sequences from 53 representatives of Polemoniaceae and three outgroup members. Nucleotide sites are numbered 5′ to 3′, beginning with the first base of ITS1. Position 278 is the final base of ITS1 and position 279 is the first base of ITS2 (the 5.8S region has been removed). Nucleotide coding follows standard IUB-IUPAC codes. Gaps at nucleotide positions are denoted by dots.

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                               120       130       140       150       160       170       180       190       200       210       220
                                .         .         .         .         .         .         .         .         .         .         .

Fouqueria columnaris        TGCTCCGAGCAATGCACGCAGCTGCACGGCGAA.GGGTGCGCTTCTCTGGAAAACAACGAA.CCCCGGCGCG.AACTGCGCCAAGGAAAC.TTAACAAAGAGATC..TCG
Fouqueria splendens         TGCTCTGAGCAATGCACGCAGTTGCACGGCAGG.GGGTGCGCTTCTCCGAAAACAACGAA.CCCCGGCGCG.AGCCGCGCCAAGGAAAC.TTAACAAAGAGATC..TCG
Diapensia lapponica         TGGAGTGTGC.TCGTATCCCTTTCGGGGGCCAC.GGGTGCAC..CTC..GAGGACAAC.AA.CCCCGGCGCG.AATAGTGCCAAGGAATT.TAAACATTGAGTGCCCATA
Acanthogilia gloriosa       GTGCA....TC.CGCGGCC.....CCGCGCCGC.GCTCGCATTTCCCGGCCGACAAACCAACGAA.CCCCGGCGCGG.CATGCG.CAAGGAACT.GTAACACAGAGAGCC.T.G
Cantua quercifolia          GACCACTCGCCCAGCGGCGTGAGCTTCTGGTGATGGTTGCGCTTCCCGGCACAACCAACGAA.CCCCGGCACG..CATGTG.CAAGGAATT.GTAACGAAGA......TGG
Bonplandia geminiflora      CGGGATGCACGCACCGCGCGGCGAAASTCGCGAGGCGTGTATTTCTCGGCAACCGAACAAACCCCCGGCGCGG.CA....CCAAGGAACTTG.AACAAAGAGAGC..TTG
Cobaea biaurita             ATGCARTTGTTTCATAGCGAGAGCTCTGTCAACTAC.TGTATTTTTCAGCACATTAACGAA.CTCCGGCGCGG.TATGTGCCAAGGAAAW.RWAATAAAGAGTGC..TTG
Cobaea scandens             ATGCAGTTGTTTCATAGCGAGAGCTCTGTCAACTAC.TGTATTTTTCAGCACATTAACGAA.CTCCGGCGCGGATGTGTGCCAAGGAAAA.GAAATAAAGAGTGC..TTG
Gilia caespitosa            GTGCA....TT.CATGGCC.....TATGGCAA.TGGTTGCACTTCCTGGCACAKTAACGAA.CCCCGACGCAG.TGTGCGTCAAGGAATT.GTAACAAAGAGAGC..TTG
Gilia subnuda               GTGCA....TT.CATGGCC.....TATGGCAA.TGGTTGCACTTCCTGGCACATTAACGAA.CCCCGACGCAG.TGTGCGTCAAGGAATT.GTAACAAAGAGAGC..TTG
Gilia pentstemonoides       GTGCA....WT.CAAGGCC.....TATGGCAA.TGATTGCACTTCCCGGCACATTAACGAA.CCCCGGCGCAG.TATGCGTCAAGGAATTCGTAACAAAGAGAGC..TTG
Gilia mcvickerae            GTGCA....TT.CAAGGCC.....TATGGCAA.TGATTGCACTTCCCGGCACATTAACGAA.CCCCGGCGCAG.TATGCGTCAAGGAATT.GTAACAAAGAGAGC..TTG
Gilia nyensis               GTGCA....TT.CATGGCC.....TTTGGCAA.TGTTTTCACTTCTCGGCACATTAACGAA.CCCCGACGCAG.TTTGCGTCAAGGAAAT.GTAACAAAGACAGC..TTG
Gilia hutchinsifolia        GTGCA....TC.CATGGCC.....TATGGCAA.TGGTTGCACTTCCCGGCACATTAACGAA.CCCCGGCGCAG.TGTGCGTCAAGGAATT.GTAACAAAGAGAGC..TTG
Gilia latifolia             GTGCA....TT.CACGGCC.....TACGGTTTGTGGATGCACTTCCCGGCACATTAACGAA.CCCCGGCGCAG.TATGCGTCAAGGAATT.GTAACAAAGAGAGC..TTG
Gilia foetida               GTGCA....AT.CACGACC.....TATGGCACG.GGTTGTGCTTCCTGGCACATAAACGAACCCCCGGCGCAG.TATGCG.CAAGGAATT.GTAACAAAGAGAGC..TTG
Gilia rigidula              GTGCA....AT.CACGGCC.....TACGGCACG.GGTTGTGCTTATTAGCAAAT.AACTAACCCCCGGCGCAG.TACGCGTCAAGGAATT.ATAACAAATACAGC..TT.
Gilia scabra                GTGCT....CT.CACGGCC.....TTGCGGTAG.TGTTTGCACTC..CGGCAAACTAACGAA.CCCCGGCGCAG.TACGCGCCAAGGAATT.GTAACGAAGAGAGC..TTG
Loeselia glandulosa         GTGCT....AT.CATGGCC.....TACGGCGA.TGGTTGCACTT..CGGCGTTTTAACTAA.CCCCGGCGCAG.TAAGCG.CAA....TT.GTAACAAAGAGAGC..TTR
Ipomopsis longiflora        GTGCA....CT.CACGGCC.....TCTGGCAA.TGTTTGCACTT..CGGCAAACTAACGAA.CCCCGGCGCAG.TGTGCG.CAAGGAATT.GTAACAAAGAGAGC..TTG
Ipomopsis gunnisonii        GTGCA....CT.CATGGCC.....TATGGCAA.TGTTTGCACTG.AACGCAAATTAACGAA.CCCCGGCGCAG.TGTGCGCCAAGGAATT.GTAACAAAGAGAGC..TTG
Ipomopsis multiflora        GTGCA....CT.CACGGCC.....TCTGGCAA.TGTTTGCACTT..CGGCAAATTAACGAA.CCCC.GCGCAG.TGTGCG.CAAGGAATT.GTAACAAAGAGAGC..TTG
Ipomopsis tenuifolia        GTGCA....CT.CRTGGCC.....TATGGCAA.TGTTTGCACTT.TTGGCACATTAACGAA.CCCC.GCGCAG.TGTGCGCCAAGGAATT.GTAACAAAGAGAGC..TTG
Ipomopsis sonorae           GTGCA....CT.CGCGCCT.....TAGGGCAC.GGCCCGCACTT..CGGCACATTAACGAACCCCCGGCGCAG.TGTGCGCCAAGGAATT.GTAACAAAGAGAGC..TTG
Eriastrum diffusum          GTGCA....CT.CA...........GCAA.TGTTT.CACCT..AGGCACCCTAACGAA.CCCCGGCGCAG.TGTGCG.CAAGGAATT.GTAACAAAGACAGC..TTG
Loeseliastrum matthewsii    GTGCA....CT.CACGGCC.....TATGGCWATTGTTTGCACTT..CGGCACATTAACGAA.CCCCGGCGCAG.TGTGCG.CAAGGAATT.GTAACAAAGASAGC..T..
Loeseliastrum schottii      GTGCA....CT.CACGGCC.....TATGGCTG.TGTTTGCACTT..CGGCACATTAACGAA.CCCCGGCGCAG.TGTGCG.CAAGGAATT.GTAACAAAGACAGC..TTG
Langloisia setosissima      GTGCA....CT.CACGGCC.....TATGGCAA.TGTTTGCACTT..CGGCACATCAACGAA.CCCCGGCGCAG.TGTGCGCCAAGGAATT.GTAACAAAGACAGC..TTG
Langloisia set. punctata    GTGCA....CT.CACGGCC.....TATGGCAA.TGTTTGCACTT..CGGCACACTAACGAA.CCCCGGCGCAG.TGTGCG.CAAGGAATT.GTAACAAAGACAGC..TTG
Gilia stellata              GTGTA....AT.CACGGCC.....TCGGCAA.TGCTTGCACTTCCCGGCACATCAACGAA.CCCCGGCGCGG.TCTGCG.CAAGGAAAT.ATAACAAAGAGAGCT.TTG
Gilia crassifolia           GTGGA....AG.CACGGCC......TCGGCAA.TGCTTGCACTTCCCGGAACATCAACGAA.CCCCGGCGCGG.TCTGCGCCAAGGAAAT.GTAACAAAGAGAGCA.TTG
Gilia scopulorum            GTGTA....AT.CATGGCC.....TCGGCAA.TGGTTGTACTTCCCGGTACATCAACGAA.CCCCGGCGCGG.TCTGCGCCAAGGAAAT.GTAACAAAGAGAGCG.TTG
Gilia ochroleuca            GTTTA...AG.CACGGCC.....TCGGCAA.TGCTTGCACTTCCCTGCACATCAACGAA.CCCCGGCGCGG.TCTGCG.CAAGGAATT.GTAACAAAGAGAGCG.TTG
Gilia flavocincta           GTGTCA...AG.CACGGCC.....WCGGCAA.TGCCTGCACTTCTCGGCACATCAACGAA.CCCCGGCGCGG.TCTGCG.CAAGGAAAT.GTAACAAAGAGAGGG.TTG
Gilia tenerrima             GTGTA....AT.CAYGGCY......TCGGCAA.TGCTAGCACTTCCCGGCACATAAACGAA.CCCCGACGCAG.TATGCG.CAAGGAAAT.GTAACAAAGAGAGCG.TTG
Gilia capitata              ATGTA....AT.CACGGCC.....TCGGCAA.TGCTTGCACTTCCTGGCACATCAACGAA.CCCCGGCGCGG.TCTGCG.CAAGGAAAT.GTAACAAAGAGACCG.TTG
Gilia tricolor              GTGTA....AT.CACGGCC.....TCGGCAA.TGCTTGCACTTCCCGGCACATCAACGAA.CCCCGGCGCGG.TTTGCGCCAAGGAAAT.GTAACAAAGAGAGCG.TTG
Allophyllum gilioides       GTGTA....AT.CATTGCC.....TCGGCAA.TACTTGCACTTCCCTAG.CATTAACGAA.CCCCGGCGCGG.TATGCG.CAAGGAAAT.GTAACAAAGAGAGCA.TTG
Allophyllum divaricatum     GTGTG....AT.CATTGCC.....TCGGCAA.TACTTGCACTTCCCTAG.CATTAACGAA.CCCCGGCGCGG.TATGCGCCAAGGAAAT.GTAACAAAGAGAGCA.TTG
Gilia splendens             GTGTA....AT.CACGGCC.....TCGGCAA.TGCTTGCACTTCCCGGCACATCAACGAA.CCCCGGCGCGG.TATGCGCCAAGGAAAT.GTAACAAAGACAGCT.TTG
Collomia linearis           GTGTA....AT.CAAGGCC.....TCGGCAA.TGCTTGCACTTCCCGGCACATCAACGAA.CCCCGGCGCGG.TATGCGCCAAGGAAAT.GTAACAAAGAGAGCT.TTG
Collomia grandiflora        GTGTA....AT.CAAGGCC.....TTGGCAA.TGCTTGCACTTCCCGGCACATCAACGAA.CCCCGGCGCAG.TACGCGCCAAGGAATT.GTAACAAAGAGAGCC.TTG
Navarretia breweri          GTGTA....AT.CAAGGCC.....TTGGCAA.TGCTTGCACTTCCTCGCAAATCAACGAA.CCCCGGCGCGG.TAATGCG.CAAGGAAAT.GTAACAAAGAGAGCC.TTG
Polemonium foliosissima     GTGCA...CG.CACTGCC.....TCGGCAA.TGCTTKC.CTTCCCGACACATAAACGAA.CCCCGGCGCGG.TATGCG.CAAGGAATT.GTAACAAAGACAG...TTG
Linanthus aureus            GAGCA....TC.CATGGTC......TCGGCTA.TGGTTGTYCTTCCCGGCACCTTAACGAA.CCCCGGCACG..AATGTG.CAAGGAATT.GTAACAAAGAGAGC..TTG
Linanthus parryae           GTACA....TT.CATAGCT......TTGGCGA.TGCTTGTGCTTCCCGGCACCCTAACGAA.CCCCGGCGCGG.AATGTGCCAAGGAATT.GTAACATAGAGAGC..TTG
Gilia maculata              GTGCA....TT.CAT.GCC.....TCGGCCA.TGCTTG.GCTTCCCGGCACCTAACGA..CCCCGGCGCGG.TATGGC.CAAGGAATT.GTAACATAGAGAGC..TTG
Gilia campanulata           GCGCA....TT.CATGGCC.....TTGGCAA.TGTTTGCGCTTCCCGGCACCTAAACGAA.CCCCGGCGCGG.AATGCGCCAAGGAATT.GTAACAAAGAGACAGC..TTG
Gilia inyoensis             GTGCA....TT.CATTGCC.....TCGGCAA.TGCTTGCGCTTCCCGGCACCTTAACGAA.CCCCGGCGCGG.TATGCGCCAAGGAATT.GTAACAAAGAGAGC..GTG
Gilia filiformis            GTGCA....TT.CATGGCC.....TCGGTCA.TGGTTGCGCTTCCCGGCACCTAAACGAA.CCCCGGCGCGG.TATGCGCCAAGGAATT.GTAACAAAGACAGC..TTG
Phlox gracilis              GTGCA....TC.CATTGCC.....CCGGCAA.TGCATGCGCTTCCCGG...CTTAACTAA.CCCCGGCACGG.TATGTGCCAAGGAATT.GTAACAAAGAGAGC..TTG
Phlox standsburyi           GTGCA....TT.CATGGCC.....TCGGCAA.TGCGTGCGCTTCCCGGCA.CTTAACTAA.CCCCGGCACGG.TATGTGCCAAGGAATT.GTAACAAAGAGAGC..TTG
Leptodactylon watsonii      GTGCA....TT.CATGGCC.....TCGGCAA.TGCTTGTGCTTCCCGGCACCTAAACGAA.CCCCGGCGCGG.TATGCG.CAAGGAATT.GTAACATAGACAGC..TTG
Leptodactylon pungens       GTGCA....TC.CATGGCC.....TCGGCGA.TGCTTGCGCTTCCCGGCACCTAAACGAA.CCCCGGCGCGG.TATGCGCCAAGGAATT.GTAACATAGACAGC..TTG
Linanthus bicolor           GAGCA....TC.CATT.CC......TTGTGTA.TGGTTGTTCTTCCCGGCAACTTAACGAA.CCCCGGCACGG.AATGTGCCAAGGAATT.GTAACAAAGAGAGC..TTG
Linanthus nuttallii         GAGCA....TC.CATTGCC......TAGTGTA.TGGGTGTTCTTCCCGGCAACTTAACGAA.CCCCGGCACGG.AATGTG.CAAGGAATT.GTAACAAAGAGAGC..TTG
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Appendix 1. Continued.

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                      230       240       250       260       270       280       290       300       310       320       330
                       .         .         .         .  ITS1   .  <----][.---->   .  ITS2  .         .         .         .
Fouqueria columnaris   CCCCCGGCGCTCC.GTTCGCGGATGCACG..GGGACGGTTGCATCTTTTC.ACGAAACATCGCGTCGCTCCCCACCCC.TCGCCCCGTTGC.GGGCGCGGGTGTTCGAGG
Fouqueria splendens    CCCCCGACGCTCC.GTTTGCGGATGCACG..GGGACGGTTGCATCTTTTC.ACGAAACATCGCGTCGCTCCCCACCCC.TCGCCCCTTGT.GGGCGCGGGTGTCCGAGG
Diapensia lapponica    TTCCCTCTTGCCCCGTTTACTCGGGTGCTAGGGGGAAGTTGG.TGTCTCCTGAAAACATCGCGTCGCCCCCAACCCCCTCTCCCTTTTGTGGTTAGTGGAGTGTTGGGG
Acanthogilia gloriosa  TC.CGGAGGCCCC.GTTCGCGGGTGA.CGAAGGGCGG.TGCCATCTGA.C.ACAAAACATCGCGTCGCCCCCAGCC...TCTGCCCTCAC.GGGGAGCGGGCAATG.GGA
Cantua quercifolia     TCCGTCCCTCGCCCGTTCACGGGTGT.CGAAGGGGCG.CGGCATCD.A.C.GCAAATGATTGCGTCGCCCCAAACC...TGTCCCCTCGG.GGAAATCRG..ATTG.GS.
Bonplandia geminiflora  CC.CGGAGGCCGG.CTTCG..GGT.C.CGAAGGGCAA.TGCCATCTTA.C.TCAATACATCGCGTCGCCCCCAACCCC.TTACCCCTTCG.GGCGTGACGGGCTTG.GGA
Cobaea biaurita        TC.CTCCTTTCCCATTTTATGG..GT.CAAAGGGCAA.TGTCRTCTTA.C.ACAATACATCTCGTCGCTCCCAACC...TATTTCCACAA.GAAAKYRTTGTTGTT.CGA
Cobaea scandens        TC.CTCCTTTCCCATTTTATGGGTGT.CAGAGGGCAA.TGTCGTCTTA.CAACAATACATCTCGTCGCTCCCAACC...TATTTCYACAA.GAAATTGTTGTTGTT.CGA
Gilia caespitosa       TT.TGGAAGCCCC.GTTCGCGGGTGC.CGAAGTGCAA.TGCCATCTKA.C.AAAAAACATCGCGTCGCCCCCAACC...TATTCCCTTTG.GGATTTGAGGTATTG.GGA
Gilia subnuda          TT.TGGAAGCCCC.GTTCGCGGGTGC.CGAAGG.CAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTTCCCTTTG.GGATTTGATGTATTG.GGA
Gilia pentstemonoides  TT.TGGAAGTCCC.GTTCGCGGGTGC.CGAAGGACAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TATTCCCTTTG.GGATTTAAGGTATTG.GGA
Gilia mcvickerae       TT.TGGAAGCCCC.GTTCGCGGGTGC.CGAA.GACAA.TGCCATCTTA.C..AAAAAACATCGCGTCGCCCCCAACC...TATTCCCTTTG.GGATTTAAGGTATTG.GGA
Gilia nyensis          TT.CGAAGGCCCC.GTTCGCGGGTGC.CGAAGAACAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TATTCCCTTTG.GGATTTAAGGTATAG.GGA
Gilia hutchinsifolia   TT.CGGAGGCCCC.GTTCGCG.GTGC.TGAAGGGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TATTCCCTTTG.GGAATTAMGGTCCTG.GGA
Gilia latifolia        TC.TCAAAGCCCC.GTTCGC.GCTGC.CGAAGG.CAA.TG.CATCTTA.C.ACGAAACATCGCGTCGCCCCCAACC...TATTCCCTCTG.GGAGTTACGGTTTTT.GGA
Gilia foetida          TC.CGGATGCCTC.GTTCGCGGKKGC.CGAAGGGCAA.TGCCATCTTA.C.TCAAAACATCGCGTCGCCCCCAACC...CTTCCCTATG.GGAATCGTGGTGCTG.GGA
Gilia rigidula         ....AGAAGCCTC.GTTCGCGAGTGC...AAGGGTAT.TGCCATCTTA.C.WMAAAACATCGCGTCGCCCCCAATC..TCTTCCCATTG.GGAA.GATGGTTTTG.GGA
Gilia scabra           TC.CGGAAGCCCC.GTTCGCGGGCGT.GGAAGGGCAA.CGCCATCTTG.C.AACAAACATCGCGTCGCCCCCAACCC...CTTCCCTCTG.GGGATTGCGGCGTTG.GGA
Loeselia glandulosa    CC.TGGAAGCCCC.GTTCGCGGGTGT.GGAAGGGCAGATGCGATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TATTTCCTTCG.GGAACTATGGTCTTG.GGA
Ipomopsis longiflora   TTGCGGAA.CCCC.GTTCAGGGGTGC.AGGAAGGGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCCACCC...TATTTCCTTTG.GAAATTGTGGTGTTT.GGA
Ipomopsis gunnisonii   CT.CGAAAGCCCC.GTTCGCGGRWGCAAGAAGGGCAA.TGCAATCTTA.C.AAAAAACATCGCGTCGCCCCCCACCC...TATTTCCTTAG.GGAACTACGGTGTTG.GGA
Ipomopsis multiflora   TT.CRGAAGCCCC.GTTCGCGGGTGC.GGAAGTGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TATTTCCTTTG.GAAATTGTGGTGTTT.GGA
Ipomopsis tenuifolia   TC.CGGAAGCCCC.GTTCGCGGGTGC.GGAAG.GCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTTCCCTTCG.GGAAATGTGGTCTTG.GGA
Ipomopsis sonorae      TT.CGGAAGCCCC.GTTCGCGGGTGC.AGAAG.GACATTGCCATCTTA.C.AAAAA.CTAAACGTCGCGTCGCCCCAACTCTTCCCTCCG.GGAAGCACGGTCTTG.GGA
Eriastrum diffusum     TC.CGGAAG.CCC.GTTCGC.GGTGCCGGAAGGGCAG.TG.CATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTT.CCTCT..GGAATTACGGTCTTG.GGA
Loeseliastrum matthewsii TT.CGGAAGCCCC.GTTCGCGGGTGC.CGAAGKKCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTT.CCTCT..GGAATTACGGTCTT..GGA
Loeseliastrum schottii TT.CGGAAG.CCC.GTTCGCGGGTGC.CGAAGG.CAA.TG.CATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTTCCCTCTG.GGGATTACGGTCTTG.GGA
Langloisia setosissima TC.CGGAAGCCCC.GTTCGCGGGTGC.GGAAGGGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTTCCCTCTG.GGAATTACGGTCTTG.GGA
Langloisia set. punctata TC.CGGAAGCCCC.GTTCGCGGGTGC.GGAAGGGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCTT.CCTCT..GGAATTACGGTCTTG.GGA
Gilia stellata         CT.TGGAAGCCCC.GTCCGCGGGTGC.TGAAGGGCAT.TGCCATCTTA.C.AAAAAATATCGCGTCGCCCCCAACC...TCTTCCCTACG.GGAATCATGG.TTCG.GGG
Gilia crassifolia      TT.TCGATGCCCC.GTCCGCGGGTGC.TGAAGGGCAAG.GCCATCTTA.C.AAAAAATATCGCGTCGCCCCCAACC...TCTTCCCTAAG.GGAATCATGG.TTCG..GG
Gilia scopulorum       CT.TGGAAGCCCC.GTCCGCGGGTGC.TGAAGGGCAAG.GCCATCTTA.C.AAAAAATATCGCGTCGCCCCCAACC...TCTTCCCTACG.GGAATCATG.GTG.GGG
Gilia ochroleuca       CT.TAGATGCCCC.GTTC.CGCGTGC.TGAACGGCAAG.GCCATCTTA.C.AAAAAATATCGCGTCGCCCCCAACC...TCTTCCCTACG.GGAATCATGG.TTCG.GGG
Gilia flavocincta      CT.TGGATGCCCC.GTTCGCGGGTGC.TGAAAGGCAAG.GCCATCTTA.C.AAAAAATATCGCGTCGCCCCCAACC...TCTTCCTTAAG.GGAATCATGGTTTCG.GGA
Gilia tenerrima        CC.TGGATGCCCG.GTTCGCGGGTGW.WGAAAGGCTAGTGACATC.TA.CAAAAAAAACATCGCGTCGCCTCCAACC...TCTTCCCTATG.GGAATCATGG.TTTG.GGA
Gilia capitata         CT.TGGATGCCCG.GTYCGCGGGTGC.TGAAGGGCAAG.GCCATCTTA.CAAAAAAAATATCGCGTCGCCCCCAACC...TCTTCCCCACG.GGAATCATGG.TTG.GGK
Gilia tricolor         CT.TGGATGCCCC.GTTCGCGGGTGC.TGAAGGTCAAC.GCCATCTTA.C.AAAAAATATCGCGTCGCCCCCAACC...TCTTCCCTACG.GGAATCATGG.TTCG.GGG
Allophyllum gilioides  TT.CGGAAGCCCC.GTTCGCGGGTGC.TGAAAGGCAA.TGCCATCTTA.C.AATATACATCGCGTCGCCCCCAACC...TCTTCCCTATG.GGAATCATGK.TCTG.GGA
Allophyllum divaricatum TT.CGGAAGCCCC.GTTCGCGGGTGC.TGAAAGGCAA.TGCCATCTTA.C.AATATATTACGCGTCGCCCCCAACC...TCTTCCCTATG.GGAATCATGG.TTTG.GGA
Gilia splendens        CT.TGGATGCCCC.GTCCGCGGGTGC.TGAAGGGCAA.TGCCATCTTA.C.AAAAA.CATCGCGTCGCCCCCAACC...TCTTCCCTATG.GGAATCATGG.TGTG.GGA
Collomia linearis      CT.TGGAAGCCCC.GTCCGCGGGTGC.TGAAGTGCAA.TGCCATCTTG.C.AAAAAACATCGCGTCGCCCCCAACC...TCATCCCTATG.GGAATCTTGG.TTTG.GGA
Collomia grandiflora   CT.TGGATGCCCC.GTTTGCGGGTGC.TGAAGAGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACC...TCATCCCTATG.GGAGCATGG.TTTG.GGA
Navarretia breweri     CT.TGGACGCCCCGGTTCGCGGGTGC.TGAAGTGCAA.TGCCATCTTA.C.AAAAAACATCGCGTCGCCCCCAACCC..TA.TCCCTACG.GGAATCATGG.TTTG.GGA
Polemonium foliosissima C..CAGAAGCCC..GTTCGCGGGTGC..GAAGTGAAC..GCG.TCTTA.C.ACAAAACATCGCGTCGCCCCCAACC...TCATTCCCTTTG.GGAATCACGGTCTTG.GGA
Linanthus aureus       TC.TGGAAGCCCC.GTTCGCGGGTGC.CAAAGGGCAT.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAACCC...ATTCCTTTTG.GAAAACGGGG.TTG.GGA
Linanthus parryae      TC.CGGAAGCCCC.GTTCGCGGGTGC.GGAAGGGCAT.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAACC...TTTCCCTCTG.GGAATCATGGGCTTG.GGA
Gilia maculata         TC.CAGAAGCCCC.GTTTGCGGGTGC.CGAAGGGCAT.TG.CATCTTA.C.ACAAAACATCGCGTCGCCCCCAACCC...TTTTCCTTTG.GGAATCACGGGGATTG.GGA
Gilia campanulata      TC.CGGAAGCCCC.GTTCGCGGGTGC.CGAAGGGCAT.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAACCC...TTTCCCTTTG.GGAATCACGGGTTTG.GGA
Gilia inyoensis        TC.TCGAAGCCCC.GTTCGCGGGTGC.TGAAGGGCAT.TGCCATCTTA.C.AAAAACTTCGCGTCGCCCCCAAACC.CTTTCCCTTTG.GGAATCACGGGTTTG.GGA
Gilia filiformis       TC.CGGAAGCCCC.GTCCGCGGGTGC.CGAAGGGCAT.TGCCATCTTA.C.ACAAACTTCGCGTCGCCCCCAACCC...TTTCCCTTTG.GGAA.CACGGGCTTG.GGA
Phlox gracilis         TC.TGGAAGCCCC.GTTCGCGGGTGC.CGAAGGGCAC.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAACCC...ATTCCCTTCG.GGATTCAATGGTTTG.GGA
Phlox standsburyi      TC.CGGAAGCCCC.GTTCGCGGGTGC.CGAAGGGCAT.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAACCC...ATTTCCTTCG.GGATTCAACGGTTTG.GGA
Leptodactylon watsonii TC.CGGAAGCCTC.GTTCGCGGRWGC.GGAAGGGCAT.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAACCC...TTTTCCTTTT.GGAATCACGGGCTTG.GGA
Leptodactylon pungens  TC.CGGAAGCCTC.GTTCGCGGGTGC.GGAAGGGCAT.TGCCATCTTA.C.ACAAAACTACGCGTCGCCCCCAACCC...TTTCCCTTTG.GGAATCACGGGCTTG.GGA
Linanthus bicolor      TC.TTGAAGCCCC.GTTCGGGGGTGC.AAAAGGACAA.TGCCATCTTA.C.ACAAACTACGCGTCGCCCCCAACCA...ATT.CCTTTT.GGAATCACGGTCTTG.GGA
Linanthus nuttallii    CC.TGGAAGTCCC.GTTCGCGGGTGC.CAAAGGGCAT.TGCCATCTTA.C.ACAAAACATCGCGTCGCCCCCAATCC...ATT.CCTCTT.GGAATCATGGTTTTG.GGA
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Appendix 1. Continued.

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                              340       350       360       370       380       390       400       410       420       430       440
                               .         .         .         .         .         .         .         .         .         .         .

Fouqueria columnaris          GCGGAAG.ATGG..CCCCCCGTGCGCGTCAGTGCTGCGGTCGGCCTAAAAAGAGAGTCCC.CGGCGACTGACG.TCACGA.CAAGTGGTGGTTGAC.AAA.CCGTT.GCG
Fouqueria splendens           GCGGAAG.ATGG..CCCCCCGTGCACATCAGTG.CGCGGTCGGCCTAAAAAGCGAGTCCT.CGACGACTGACG.TCACGA.CAAGTGGTGGTTGAC.AGA.CCGTT.GCA
Diapensia lapponica           GCGGAAG.TTGG..CCCCCCGTGAGCACAGTCTCTGCGGYCGGCCAAAATA..CAGTCCATCGACGAAAWTATGTCACGA.CATCTGGTGGTTGAC.AAA.CTTAC.GAG
Acanthogilia gloriosa         GCGGAAA.TTGG..TCTCCCGTGCACAC.GCTG.CGGGTTGGCCTAAAAA.AGAGTCGA.CGTGGACGAGCG.TCACAG.CATGTGGTGGTTGTC.AAA.CCGTT.GCG
Cantua quercifolia            GCRGAGA.TTGG..CCTCCCGTGCACATCGTTG.AGCGGTTGGCCTAAATA...AGTGGA.CGTTGATGGGCG.CATGTGGTGGTTGTKCCCAACCGTT.GCG
Bonplandia geminiflora        GCGGAAA.TTGG..TCTCCCGTGCGCCTAGCCG.TGCGGTTGGCCTAAAAA.CGAGTGCA.CGTGGACGGACG.TCACAA.CATGTGGTGGTTGAACAAA.CCGTT.GCG
Cobaea biaurita               GCGGAAAGTTGGCCTCTCCTGTGCACATTGTTG.TGTGGTTGGCCTAAAAT.TGAGTSCA.CGTGGATGAGAGTTCGCAA.CAAGTGGTGGTTGTC.AAG.CCGTT.GCG
Cobaea scandens               GCGGAAG.TTGG..TCTCCTGTGCACATTGTTG.TGTGGTTGGCCTAAAAT.TGAGTGCA.CGTGGATGAGAGTTCGCAA.CAAGTGGTGGTTGTC.AAG.CCGTT.GCA
Gilia caespitosa              GCGGAAA.TTGG..TCTCCCGTGCTTTT.TCTG.TGCGGTTGGCCTAAAAA.CGAGTCGA.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.SAG
Gilia subnuda                 GCGGAAA.TTGG..TCTCCCGTGCATTT.TCTG.TGCGGTTGGCCTAAAAA.CAAGTCGA.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCR
Gilia pentstemonoides         GCRGAAA.TTGG..TCTCCCGTGCTCTT.WTTG.TGCGGTTGGCCTAAAAA.AGAGTAGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia mcvickerae              GCGGAAA.TTGG..TCTCCCGTGCTCTT.TCTG.TTCGGTTGGCCTAAAAA.AGAGTMGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia nyensis                 GCGGAAA.TTGG..TCTCCCGTGCTTTTCTCTG.TGCGGTTGGCCTAAAAA.AGAGTCGA.CGTGGAMGAACG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia hutchinsifolia          GCGGAAA.TTGG..TCTCCCGTGCATTT.TCTG.TGCGGTTGGCCTAAAAA.AGAGTCGA.CGTGGACGAACG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia latifolia               GCGGAAA.TTGG..TCTCCCGTGCACTTTGTTG.TGCGGTTGGCCTAAATA.TGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia foetida                 GCGGAAA.TTGG..TCTCCCGTGCACTCTGTTG.TGCGGTTGGCCTAAATA.AGAG.CGG.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTG.C.AAA.CCGTT.GCG
Gilia rigidula                GCGGAGA.TTGG..TCTCCCGTGCACTATGTTG.TGCGGTTGGCCTAAAAA.AGAGTCGG.CGTGGATGAGCG.TCACAA.CWAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia scabra                  GCGGAAA.TTGG..TCTCCCGTGCACTCTGTTG.AGCGGTTGGCCTAAATA.CGAGTCGA.CGTGAACGAGCG.TCACAA.CATGTGGTGGTTGAC.TGA.CCGTT.GCG
Loeselia glandulosa           GCGGAAA.TTGG..TCTCCCGTGCACTCAGATG.CGCGGTTGGCCTAAATA.AGAGTCGG.CGTTGACGAACG.TCACAA.CAAGTGGTGGTTGAC.AAA.TCGTT.GCG
Ipomopsis longiflora          GCGGAAA.TTGG..TCTCCCGTGCACTCTG.TG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Ipomopsis gunnisonii          GCGGAAA.TTGG..TCTCCCGTGCACTCTGTTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGATGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCA
Ipomopsis multiflora          GCGGAAA.TTGG..TCTCCCGTGCACTTTGC..CTKCGGTTGGCCTAAATA.AGAGTCGA.CGTGGACGACG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Ipomopsis tenuifolia          GCGGAAA.TTGG..TCTCCCGTGCACTCTGC.G.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Ipomopsis sonorae             GCGGAAA.TTGG..TCTCCCGTGCACTCGTTTG.TGCGGCTGGCCTAAACA.AGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Eriastrum diffusum            GCGGAAA.TTGG..TCTCCCGTGCACTCTGATG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGGKGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTTGGCG
Loeseliastrum matthewsii      GCGGAAA.TTGG..TCTCCCGTGCACTCTGTTGCTGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Loeseliastrum schottii        GCGGAAA.TTGG..TCTCCCGTGCACTCTGTTG.CGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Langloisia setosissima        GCGGAAA.TTGG..TCTCCCGTGCACTCTGCTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Langloisia set. punctata      GCGGAAA.TTGG..TCTCCCGTGCACTCTGCTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGGCGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia stellata                GCGGAAA.TTGG..TCTCCCGTGCACACCGATG.CGCGGTTGGCCTAAATA.TGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia crassifolia             GCGGAAA.TTGG..TCTCCCGTGCACACCGATG.CGCGGTTGGCCTAAATA.TGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia scopulorum              GCGGAAA.TTGG..CCTCCCGTGCACACYGATG.CGCGGTTGGCCTAAATA.AGAGTYGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia ochroleuca              GCGGAAA.TTGG..TCTCCCGTGCACACCGATG.CGCGGTTGGCCTAAATA.AGAGTTGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia flavocincta             GCGGAAA.TTGG..TCTCCCGTGCACATCGATG.CGCGGTTGGCCTAAATA.AGAGTTGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT..CG
Gilia tenerrima               GCGGAAA.TTGG..TCTCCCGTGTGCACCGTTG.YGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia capitata                ...GAGA.TTGG..TCTCCCGTGCACTCCGATG.CGCGGTTGGCCTAAATA.AGAGTTGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia tricolor                ACGGAAA.TTGG..TCTCCCGTGCACCTCGATG.CGCGGTTGGCCTAAATA.AGAGTTGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Allophyllum gilioides         GCGGAAA.TTGG..TCTCCCGTGCACTTCGTTG.TGCGGTTGGCCTAAATA.AGAGTTGA.CGTGGACGAGCG.TCATAA.CATGTGGTGGTTGTC.AAA.CCGTT.GCG
Allophyllum divaricatum       GCGGAAA.TTGG..TCTCCCGTGCACTACGTTG.TGCGGTTGGCCTAAATA.AGAGTTGA.CGTGAACGAGCG.TCATAA.CATGTGGTGGTTGTC.AAA.CCGTT.GCG
Gilia splendens               GCGGAAG.TTGG..TCTCCCGTGCACAATGCTG.CGCGGTTGGCCTAAACA.AGAGTTGA.CGTGGACGAGCG.TTACAA.C?AGTGGTGGTTGTC.AAA.CCGTT.GCG
Collomia linearis             GCGGAAA.TTGG..TCTCCCGTGCACATCGTTG.CGTGGTTGGCCTAAATA.AGAGTGGA.CGTGAAACGAGC.GTCACAACAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Collomia grandiflora          GCGGAAA.TTGG..TCTCCCGTGCACATCGTTG.CGTGGTTGGCCTAAATA.AGAGTGGA.CGTGAACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Navarretia breweri            GCGGAAA.TTGG..TCTCCCGTGCACATTGTTG.TGTGGTTGGCCTAAATA.AGAGTGGA.CGTGGACGAGCG.TCACAA.CAAGTGGTGGTTGTC.AAA.CCGTT.GCG
Polemonium foliosissima       GCGGAAA.TTGG..TCTCCCGTGCACGCTGTTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGKACGAGCG.TCACAA.CAAGTGGTGGTTGTT.AAA.CCGTT.GCG
Linanthus aureus              .CGGAGA.TTGG..TCTCCCGTGCACAT.GTTG.TGTGGTTGGCCTAAATA.AGAGTTGG.CGTGTACGAGTG.CSACAA.CRATTGGTGGTTGTTAAWRGCCTTTGCCC
Linanthus parryae             GCGGAGA.TTGG..TCTCCCGTGCACTT.GTTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGTGCGTGCG.TCACAA.CAATTGGTGGTTGTTATAAACTTTT.GCC
Gilia maculata                GCGGAGA.TTGG..TCTCCCGTGCACTT.GCTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGACGTGCG.TCACAA.CAATTGGTGGTTGTT.AAAACCAGT.GCC
Gilia campanulata             GCGGAGA.TTGG..TCTCCCGTGCACAACGCTG.CGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGACGTGCG.TCACAA.CAATTGGTGGTTGTT.AAAACCGAT.GCC
Gilia inyoensis               GCGGAGA.TTGG..TCTCCCGTGCACTTCGCTG.CGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGAGGAGCG.TCACAA.CAATTGGTGGTTGTT.AAGACCTTTTGCC
Gilia filiformis              GCGGAGA.TTGG..TCTCCCGTGCACTT.GTTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGGACAAGCG.TCACAA.CAATTGGTGGTTGTT.AAAACCGTT.GCC
Phlox gracilis                GCGGAAA.TTGG..TCTCCCGTGCACTTCGGTG.TGCGGTTGGCCCAAATT.AGAGT.GA.CGCGGACTAGCG.ACACAA.CAATTGGTGGTTGTT.AAAACCTCA.GCC
Phlox standsburyi             GCGGAAA.TTGG..TCTCCCGTGCACTTCGGTG.TGCGGTTGGCCTAAATA.AGAGT.GA.CGTGGACTAGCG.ACACAA.CAATTGGTGGTTGTC.AAAACCTTA.TCC
Leptodactylon watsonii        GCGGAGA.TTGG..TCTCCCGTGCACTT..GTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGTACGTGCG.TCACAA.CAATTGGTGGTTGTTAAAAACCTTT.GCC
Leptodactylon pungens         GCGGAGA.TTGG..TCTCCCGTGCACTT.GTTG.TGCGGTTGGCCTAAATA.AGAGTCGA.CGTGTACGTGCG.TCACAA.CAATTGGTGGTTGTTAAAAACCTTT.CCC
Linanthus bicolor             GCGGAGA.TTGG..TCTCCCATACACAT.GTTG.TGTGGTTGGCCTAAATA.AGAGTCGG.CATGAGTGAGTG.TCACAA.CAATTGGTGGTTGTT.AAAACCTTT.GCC
Linanthus nuttallii           GCGGAGA.TTGG..TCTCCCGTACACAT.GCTG.TGTGGTTGGCCTAAATA.AGAGTCGG.CGTGAATGAGTG.CCACAG.CAATTGGTGGTTG.TAAAAACCTTT.GCC
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Appendix 1. Continued.

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                                450       460       470       480       490       500       510       520
                                 .         .         .         .         .         .         .         .

Fouqueria columnaris          TTTCCTGTCG.TGCGTCGCA.TCGTCGCAACGGGCTGG...CTCAC...GTGACCC.TGAAG.TG..CGGTAACAG.TG..CCTCGACG
Fouqueria splendens           TTTCCTGTCG.TGCGTCGCA.TCGTCGCTACGGGCCGG...CTCAT...GTGACCC.TGAAG.TG..CGGTAACAGCTGTGCCTCGACC
Diapensia lapponica           CCT..TGTCG.TGA.TC....TTCTCGCCGCATGT?GCA..CTTTA....TGACCC.TGATG.TG..CCATTCTTACGGTGCTTCGACC
Acanthogilia gloriosa         CTGT.TGTTG.TGCGT.GCT.TCGTCTCCC.TCGT.AA..CTCCA...ACGACCC.TGTTA.CA..TCGCGCAAGCGATGCCTCGACC
Cantua quercifolia            TTGCC.GTTG.TGCGT..CG.TCGTCTCCGG.....AA..GCKCCA...TAGACCC.TCTTG.CG..CCGCTTCAGCGGTGCCTCGATC
Bonplandia geminiflora        TT.CCTGTTGTTG.GT..CG.TTCTCCCCGG..TT.AG.CGCTCCG...AAGACCC.AGACG.CG..TTGCGCAAGCGACGCCTCGACC
Cobaea biaurita               TTAT.TGTTG.TCTGA..CC.TCGTCCTC....TACAA...CWCCA...TTGACCT.CGTTG.TA..TTATTAAAGTGATACCTCGTCT
Cobaea scandens               TTAT.TGTTG.TCTGA..CC.TCGTCCTCG...AAAA...CTCCA...TTGACCT.TGTTG.CA..TTATAAAAGTGATGCCTCGACT
Gilia caespitosa              TTGC.TGTTG.TGCGT..TT.TCATCCTCT.TCGTCGA...CCCCA...TTGACCC.TGTCG.CA..TTGCGCAAGCGATGCCTCGACC
Gilia subnuda                 TTGC.TGATG.TGCGT..TT.TCATCCTCT.TCGTCGA...CCCCA...TTGACCC.TGTCG.CA..TTGCGCAAGCGATGCCTCGACC
Gilia pentstemonoides         TTGC.TGTTG.TGCGT..TT.TCATCCTCA.TCGTCGA...CTACA...TTGACCC.TRTTG.CA..TTGCGTAAGCGATGCCTCGACC
Gilia mcvickerae              TTGC.TGTTG.TGCGT..TT.TCATCCTCT.TCGTCGA...CTCCA...TTGACCC.TRTTG.CA..TTGCGCAAGCGATGCCTCGACC
Gilia nyensis                 TTGC.TGTTG.TACGT..TT.TCATCCTCT.TCGTCGA...CTCCA...TTGACCC.TGTTG.CA..TTGCGCAAGCGATGCCTCGACC
Gilia hutchinsifolia          TTGC.TGTTG.TACGT..TT.TCATCCTTT.TCGTCGA...CTTCA...TTGACCC.TGTTG.CA..TTGCGAAAGCGATGCCTCGACC
Gilia latifolia               TTGC.TGTTG.TGCGT..TA.CCGTCCTCT.TCGCCAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGCAAGTGATGCCTCGACC
Gilia foetida                 TTGT.TGTTG.TGCGTG.TA.AGTCGTGCTY.CGTCCAGCCTTACATAGTTGACCC.TGTTG.CA..TTGCGTGAGCAATGCCTCGACC
Gilia rigidula                TTGT.TGTTG.TGCGC..TA.A.TCCT.CT.TCGTCAA...CTCCA...TTGACCC.TGTTG.CA..TTGTGTAAGCAATGCATCGACC
Gilia scabra                  TGGT.TGTTG.TGCGT..CA.TCGTACTCA.CCGTTGG...TTCCG...TGGACCC.TTTTGACA..CTGCGCAAGCAATGCCTCGACC
Loeselia glandulosa           TTGT.TGTTGTAGCGT..CAGCCGTCTTGT.ACGCSAA...CTCAA...TTGACCC.TTTTG.CATGTTGTGAAAGCAATGCCACGACC
Ipomopsis longiflora          TTGC.TGTTG.TGCGT..CA.CCGTCCTC.ACGTCAA...CTCCA...ATGACCC.TGTTG.AA..TTGCGAAAGTAATGTCACGACC
Ipomopsis gunnisonii          ATTC.TGTTG.WGCGT..AA.KCATCCTCT.ATGTMGA...CTCCA...ATGACCC.TGTTG.CA..TTGTGCAAGCAATGCCACGACC
Ipomopsis multiflora          TTGC.TGTTG.TGCGT..CA..TGTCCTCT.CCGTCAA...CTCCA...ATGACCC.TGTTG.GA..TTGCGCAAGCAATGCCTCGACC
Ipomopsis tenuifolia          TTGC.TGTTG.TGCGT..CA.TCGTCCTCT.CCGTCAA...CTCAA...TTGACCC.TGTTG.CA..TTGCGCAAGTGCCGCGACGACC
Ipomopsis sonorae             TTGT.TGTTG.TGCGT..CG.TCGTCCGCT.CAGTCGA...CTCCA...ACGACCC.TGTTG.CG..TTGTGCAAGCAATGCCTCGACC
Eriastrum diffusum            TTGCGTGTTG.TGCGT.GCGGTCGTCCTCT.CCGTMGA...CTCCA...TTGACCC.TGTTG.CA..CTGTGCAAGCAGTGCCWCGACC
Loeseliastrum matthewsii      TTGC.TGTTG.AGCGT..CA.GCGTCCTCT.CCGTCGA...CTCCA...ATGACCC.TGTTG.CA..TTGCGCAAGCAATGCCACGACC
Loeseliastrum schottii        TTGM.TGTTG.TGCGT..CA.TCGTCCTCT.CCGTCGA...CTCCA...TTGACCC.TGTTG.TA..TTGCGCAAGCAATGCCTCGACC
Langloisia setosissima        TTGC.TGTTG.TGCGT..CA.TCGTCCTCY.CCGTCGA...CTCCA...TTGACCC.TGTTG.CA..CTGCGCAAGCAATGCCTCGACC
Langloisia set. punctata      TTGC.TGTTG.TGCGT..CA.TCGTCCTCC.CCGTCGA...CTCCA...TTGACCC.TGTTG.CA..CTGCGCAAGCAATGCCTCGACC
Gilia stellata                TTGT.TGTTG.TGCGT..TA.TCGTCCTCT.CCGTCAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Gilia crassifolia             TTGT.TGTTG.TGCGT..TA.TCGTCCTCT.CCGTCAG...CTCCA...TTCACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Gilia scopulorum              TTGT.TGTTG.TGCGT..TA.TCGTCCTCT.CCGTCAG...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Gilia ochroleuca              TTGT.TGTTG.TGCGT..TA.TCGTCCTMT.CCGTCRA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Gilia flavocincta             TTGT.TGTTG.TGCGT..TA.TCGTCCTCT.CTGTCAG...CTCCA...TTGACCC.TGTTG.CA..TTC.GAA.GTGATGCCTCGACC
Gilia tenerrima               TTGT.TGTTG.TGCGT..TT.TCGTCCTCT.WCGTCWA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Gilia capitata                TTGT.TGTTG.TGCGT..TA.TCGTCCTCT.CGTACAG...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTACACC
Gilia tricolor                TTGT.TGTTG.TGCGT..TA.TCGTCCTCT.CGTACAT...CTCCA...TTGACCC.TGTTG.CA..TTG.GAAAGT.ATGCCTACACA
Allophyllum gilioides         TTAT.TGTTGTAGCGT..TAGTTGTTCTCT.TCGTTAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCWCGACC
Allophyllum divaricatum       TTAT.TGTTGT.GCGT..TA.TTGTTCTCT.TCGTTAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Gilia splendens               TTGC.TGTTG.TGCGT..CT.TCGCCCTCT.TCGTCAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Collomia linearis             TTGT.TGTTG.TGCGT..TA.TTGTTCTCT.CCGTCAA...CTCCA...TTGACCC.TGATG.CA..TTGCGTAAGTAATGCCTCGACC
Collomia grandiflora          TTGT.TGTTG.TGCGT..TG.TTGTTCTCT.CCGTCAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTAATGCCTCGACC
Navarretia breweri            TTGT.TGTTG.WGCGT..TA.TTGTTCTCT.TCGTCAA...CTCCA...TTGACCC.TGTTG.CA..TTGCGACCGTGATGCCAGTACA
Polemonium foliosissima       TTGC.TGTTG.WGCGT..TA.SCGTCCTCT.TCGTMGG...CTCCA...TTGACCC.TGTTG.CA..TTATAAAAGTGATGCCTCGACT
Linanthus aureus              TTACATGTTG.TG.....TGCTTGTTTCCG....MTGT...CTCCA...AKGACCCTTGTTG.CA..TTGCGAAAGTGATGCCACGACC
Linanthus parryae             TTGC.TGTTG.TGCGT..TG.TCGTCCTCG.TC..CGA...CTCCA...TTGACCC.TGTTG.CA..TTGCGTAAGTGATGCCTCGACC
Gilia maculata                TTAC.TGTTG.TGCGT..TA.ACGTCCTTG.TCGTCGA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGCAATGCCTCGACC
Gilia campanulata             TTAC.TGTTG.TGCGT..TG.TCGTCCTCT.TCGTCGT...CTCCA...TTGATCC.TGTTG.CA..TTGCGAAAGCGATGCCTCGACC
Gilia inyoensis               TTAC.TGTTG.TGCGT..CG.TTGTCCTCT.TCGTCGG...CTCCA...TTGACCC.TGTTG.CA..TTGCGTAAGCGATGCCTCGACC
Gilia filiformis              TTAC.TGTTG.TGCGT..TG.TCGTCTTCT.TCGTCGA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGCGATGCCTCGACC
Phlox gracilis                TTAC.TGTTG.TTCGT..CTTTGCTCCTCG....TCTA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTGATGCCTCGACC
Phlox standsburyi             TTAC.TGTWG.TGCGT..CTTTGCTCCTCG....TCTA...CTCCA...TTGACCC.TGTTG.CA..TCGCGAAAGTGATGCCACGACC
Leptodactylon watsonii        TTGC.TGTTG.TGCGT..TG.TCGTCCTCG...TCCGA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTAATGCCTCGACC
Leptodactylon pungens         TTGC.TGTTG.TGCGT..TG.TCGTCCTCG...TCCGA...CTCCA...TTGACCC.TGTTG.CA..TTGCGAAAGTAATGCCTCGACC
Linanthus bicolor             T..C.TGTTG.TGCG...CT.TGCTTAT.....GCTGT...TTCCA...ATGACCC.TGTTG.CG..TTGCGAAAGTGATGCCTCGACC
Linanthus nuttallii           TTACATGTTG.TGCG...CT.TGCTCAT.....GTTAT...CTCCA...ATGACCC.TGTTG.CG..TTGCGAAAGTGATGCCWCGACC
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Appendix 1. Continued.