A SYSTEMATIC STUDY OF THE FLOWERING PLANT GENUS *MICRANTHES* (SAXIFRAGACEAE) IN THE SOUTHERN APPALACHIANS

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ABSTRACT

A SYSTEMATIC STUDY OF THE FLOWERING PLANT GENUS *MICRANTHES* (SAXIFRAGACEAE) IN THE SOUTHERN APPALACHIANS

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Recent molecular phylogenetic analyses have clearly shown the large, arctic and north-temperate genus *Saxifraga* (Saxifragaceae) sensu lato is polyphyletic with two distinct clades: *Saxifraga* sensu stricto and *Micranthes*. Six species belonging to *Micranthes* exist in the Southern Appalachians, including two questionably distinct species and one rock outcrop endemic. Taxonomists have traditionally distinguished the very similar *M. careyana* and *M. caroliniana* primarily based on geographic locality and four morphological characters: sepal orientation (erect or reflexed), filament shape (uniform or club-shaped), petal coloration (none or 2 yellow spots), and fruit length (2.5-5 or 4-5 mm). The goal of this research was to examine these characters to clarify the taxonomy of these species and look for molecular differences in the nuclear and chloroplast DNA regions, and examine the phylogeny of all six Southern Appalachian species in the context of the entire genus. Several populations of *M. careyana* and *M. caroliniana* from the Blue Ridge Physiographic Province have been examined and material has been collected for molecular analyses. Populations in the counties of Ashe, Alleghany, and

Watauga (North Carolina) and Johnson (Tennessee) displayed reflexed sepals and club-shaped stamen filaments, consistent with *M. caroliniana*. Populations examined in flower in all other counties displayed erect sepals and uniform stamen filaments, consistent with *M. careyana*. The other two characters were not useful in distinguishing these taxa. These differences in floral characters are correlated with mutations in ITS and *trn*L-F sequences. In phylogenetic analyses, populations determined to represent *M. caroliniana* appear in a distinct clade from those determined to represent *M. careyana*, supporting the separation of the two as species. In addition, the high-elevation rock outcrop endemic *M. petiolaris* appears in a distinct clade from the other five Southern Appalachian species, indicating this taxon evolved along a separate lineage and should be placed in the genus *Hydatica* in future systematic treatments.

INTRODUCTION

Science of systematics

The science of systematics consists of two broad areas of investigation, traditional taxonomy (the description, identification, nomenclature, and classification of organisms) and phylogeny (the evolutionary history of life) (Simpson 2006). The goal of systematics is to study biological diversity, describe all species, and discover all branches of the evolutionary tree of life (Judd et al. 1999). In phylogenetic systematics, the subjects of interest for study are defined groups of organisms referred to as taxa and which possess the property of monophyly, defined as all of the descendents of a most recent common ancestor (Simpson 2006). Traditional taxonomy is a descriptive science based on the variation and form of morphological characters and focuses on the similarities and dissimilarities among organisms (Jones and Luchsinger 1986). Classification of life based on overall similarity between and among taxa is referred to as phenetic classification (Simpson 2006).

Phylogeny is the component of systematics that refers to the evolutionary history of a group of organisms and is commonly represented by a phylogenetic tree, a diagram that shows the branching pattern of evolutionary descent, with each line in the tree representing a particular lineage and each node representing a hypothetical ancestor from which the lineage diverged (Simpson 2006). Evolution occurs in lineages over time and represents a change in populations from ancestral character states to new, derived character states (Simpson 2006). Phylogenetic classification is based on the recognition of monophyletic groups, consisting of all of the descendents of a common ancestor (Judd

et al. 1999). In contrast, many traditional systems recognize paraphyletic groups, consisting of a common ancestor but not all descendants, or a polyphyletic group, consisting of two or more separate groups each with a different common ancestor (Simpson 2006). Many systematists believe the two latter types of groups should not be recognized because they do not accurately represent the evolutionary history of life, although they may be useful in identification and to recall biological information. In addition to aiding classification, phylogenetic trees are essential tools and powerfully predictive in examining biogeography, ecological history, speciation, and adaptive character evolution of organisms (Simpson 2006).

Species concepts

Another key aspect to systematics the application of species concepts, whereby the boundaries between individuals of the same versus different species are determined. In many animals the ability to interbreed is usually used as the primary criterion for distinguishing among species, along with other morphological, ecological, and geographic considerations (Judd et al. 1999). This concept of species is commonly referred to as the biological species concept (BSC) and defines species as "groups of actually or potentially interbreeding populations, which are reproductively isolated from other such groups" (Mayr 1963). However, this concept of species may not apply to organisms with different breeding habits, such as plants or asexual organisms. Plant species frequently hybridize, and gene flow between species varies greatly, so applying the biological species concept would result in broadly inclusive species (Judd et al.

1999). Many other species concepts have been proposed, and with the emergence of phylogeny as a principal component of systematics, phylogenetic species concepts have been advanced with at least three different criteria proposed for distinguishing species (Table 1). The autapomorphy species concept (Donoghue 1985; Mishler and Brandon 1987) uses the criterion of monophyly, which specifies that a species contains all of the descendants of one ancestral population and can be recognized by autapomorphies, or unique derived character states. The diagnosability species concept (Nixon and Wheeler 1990) defines a phylogenetic species as the smallest cluster of populations or lineages displaying a unique set of fixed character states. The genealogical species concept (Baum and Shaw 1995) uses basal exclusivity as the criterion for a species, which means that members of a group are more closely related to each other than to any outside that group. All of these concepts have both strengths and flaws, therefore there is no consensus about which species concepts should be used, particularly in plants (Judd et al. 1999).

Phylogenetic studies in Saxifraga

Many systematic questions have arisen with respect to the genus *Saxifraga* L. sensu lato, by far the largest genus in Saxifragaceae with over 440 species, and which traditionally included 15 sections: *Ciliatae* Haworth, *Cotylea* Tausch, *Cymbalaria* Grisebach, *Gymnopera* D. Don, *Heterisia* (Rafinesque ex Small) A. M. Johnson, *Irregulares* Haworth, *Ligulatae* Haworth, *Merkianae* (Engler & Irmscher) Gornall, *Mesogyne* Sternberg, *Micranthes* (Haworth) D. Don, *Odontophyllae* Gornall, *Porphyrion*

Tausch, *Saxifraga* L. (Webb and Gornall 1989), *Trachyphyllum* (Gaudin) W. D. J. Koch, and *Xanthizoon* Grisebach. Most members of *Saxifraga* s.l. are found in the arctic or north temperate regions of the world, with areas of high biodiversity in the European mountains, eastern Asia, and the mountains of western North America (Webb and Gornall 1989). Extreme diversity in morphology, ecology, habit, and cytology has led to difficulties in defining the limits of such a large genus (Mort and Soltis 1999). Some workers divided *Saxifraga* into 16 separate genera (Haworth 1812; Small and Rydberg 1905; Löve 1983), while others preferred a more broad interpretation (Engler 1872; Engler and Irmscher 1916; Gornall 1987; Webb and Gornall 1989). Lord (1961) offers a more detailed history of the various taxonomic treatments of this large, complex genus.

It is now recognized that *Saxifraga* as previously understood is clearly a polyphyletic group and must be divided into two distinct genera: *Saxifraga* L. sensu stricto, consisting of the majority of the species within 14 of the 15 traditional sections; and *Micranthes* Haworth (Brouillet & Gornall 2007) consisting of only 65-100 species, most of which were formerly placed in Section *Micranthes* (Soltis et al. 1993; Johnson and Soltis 1994, 1995; Soltis et al. 1996; Mort and Soltis 1999; Soltis et al. 2001). These findings are based on phylogenetic analyses of chloroplast DNA (cpDNA) restriction sites (Soltis et al. 1993), sequences of the cpDNA gene regions *rbc*L and *mat*K (Johnson and Soltis 1994, 1995; Soltis et al. 1996) (Fig. 1), and nuclear ribosomal sequences from the internal transcribed spacer (ITS) regions (Johnson and Soltis 1998). Two species of *Saxifraga* section *Merkianae*, the monotypic *Cascadia nuttallii* (Small) A. M. Johnson (= *S. nuttallii*) and *S. tolmiei* Torrey & Gray appear to be the closest relatives of the *Micranthes* clade (Soltis et al. 1996). In addition, it has been demonstrated that

Micranthes is more closely related to the genera *Heuchera* L., *Mitella* L., *Tiarella* L., and others than to the majority of *Saxifraga* (Soltis et al. 1996). *Micranthes* was formally circumscribed as a genus by Brouillet and Gornall (2007) and this study focuses on the Southern Appalachian species belonging to this genus.

Sub-generic classification of Micranthes

Four sections are recognized within the *Micranthes* clade: *Stellares* (Engler & Irmscher) Gornall, *Micranthes* (Haworth) Gornall, *Rotundifoliatae* A. M. Johnson, and *Cuneifoliatae* A. M. Johnson (Webb and Gornall 1989), based primarily on the nature of the seed coat, in addition to inflorescence characteristics, leaf shape, trichome type, and ovary position (Kaplin 1981). These sectional designations have been adapted from classification schemes of Webb and Gornall (1989) by elevating each level up one rank (i.e. subsection to section). More recently, the chloroplast gene *mat*K was sequenced in 26 species of *Micranthes* to test the phylogenetic relationships among species within the genus and the monophyly of each of the four sections (Mort and Soltis 1999). Results from this study indicate that sections *Micranthes* and *Stellares* are each monophyletic, while sections *Rotundifoliatae* and *Cuneifoliatae* may not be distinct from each other. A strongly supported sister group relationship between the *Micranthes* and *Rotundifoliatae* clades is also evident. Only two of the six Southern Appalachian species, *M. pensylvanica* (L.) Haworth and *M. virginiensis* (Michx.) Small, were included in this phylogenetic study, so it is unclear what the relationships of the other four species are in the context of the entire genus.

Southern Appalachian species of Micranthes

There are six species of the genus *Micranthes* Haworth found in the Southern Appalachians: *M. careyana* (Gray) Small (Carey saxifrage), *M. caroliniana* (Gray) Small (Carolina saxifrage), *M. micranthidifolia* (Haworth) Small (branch-lettuce), *M. pensylvanica* (swamp saxifrage), *M. petiolaris* (syn. *Saxifraga michauxii*) (Raf.) Brouillet & Gornall (mountain saxifrage), and *M. virginiensis* (early saxifrage). Four of the six are Southern Appalachian endemics; *M. careyana*, *M. caroliniana*, *M. micranthidifolia*, and *M. petiolaris* (Weakley 2008). A seventh species, *M. tennesseensis* Small (golden-eye saxifrage), was described by Small (1896) who thought it was closely related to *M. virginiensis* based on morphological characters but distinct enough to be recognized at the species level. This taxon was later synonymized with *M. careyana* by Lord (1961), who showed that the two taxa had many characters in common including their overlapping measurements of petal and fruit length. All six Southern Appalachian species are currently classified in section *Micranthes*, except *M. petiolaris*, which is in section *Stellares* with six other North American species. In addition, section *Micranthes* includes three subsections: *Aulaxis* (Haworth) Gornall containing *M. micranthidifolia*; *Dermasea* (Haworth) Gornall containing *M. careyana*, *M. caroliniana*, and *M. virginiensis*; and *Micranthes* (Haworth) Gornall containing *M. pensylvanica* (Gornall 1987; Webb and Gornall 1989) (Table 2). All six species are perennial herbaceous, scapose plants that possess an extremely shortened main stem with a basal rosette of leaves, the leaf margins having some degree of serration, and a terminal long-pedunculate inflorescence of several to many cymose units with many small flowers. Each flower has five green sepals, five white petals with or without yellow spots, ten stamens, a single

pistil comprised of two carpels fused at the base, and a superior to semi-inferior ovary position.

According to Weakley (2008), *Micranthes careyana* grows in moist soil at the base of rock outcrops and overhanging cliffs from southwestern Virginia south to eastern Tennessee, western North Carolina, and northwestern South Carolina. It is a species of concern (GA Special Concern, NC Watch List, SC Rare, VA Rare) that flowers from May-June. *Micranthes caroliniana* also grows in moist soil at the base of rock outcrops and overhanging cliffs from West Virginia south to northeastern Tennessee and western North Carolina. It is a rare species (US Species of Concern, NC Rare, VA Rare) that flowers from May-June. *Micranthes micranthidifolia* grows commonly in wet and rocky soils of seepages and the beds of high-elevation brooks and brookbanks from eastern Pennsylvania and West Virginia south to eastern Tennessee, western North Carolina, northwestern South Carolina, and northeastern Georgia. It flowers from May-June. *Micranthes pensylvanica* is uncommon (NC Rare) and is found growing in mountain bogs and mucky seeps from Maine west to Minnesota, south to eastern Virginia, central and western North Carolina, and Missouri. It flowers from April-June. *Micranthes petiolaris* commonly grows on exposed high-elevation rock outcrops and in rocky seeps from northwestern Virginia (rare in VA Piedmont), West Virginia, and Kentucky south to eastern Tennessee, western North Carolina, southwestern South Carolina, and northeastern Georgia. It flowers from June-August. *Micranthes virginiensis*, with the widest distribution, commonly grows in rock outcrops, streambanks, and riverbanks from New Brunswick west to Manitoba, south to central Georgia, Louisiana, and Arkansas (rare in Coastal Plain). It flowers from March-May.

The Micranthes careyana – caroliniana species complex

Traditional studies of the genus *Saxifraga* sensu lato have employed four major criteria to differentiate among species: floral structure, fruit structure, vegetative characters, and habit. In order to morphologically distinguish among the six Southern Appalachian *Micranthes* species, some of which are very similar in habit and appearance, Lord (1961) noted differences among stems, leaves and petioles, presence or absence of a hypanthium (floral tube below the free petals), sepal orientation, corolla type, and filament length and shape. She also noted that *M. careyana* and *M. micranthidifolia* have rhizomatous stems, unlike the other four Southern Appalachian species. Other more specific differences were noted between the very similar *M. careyana* and *M. caroliniana* in sepal orientation (erect to spreading or fully reflexed), filament shape (filiform or clavate), petal coloration (none or 2 yellow spots), and fruit length (2.5–5 mm or 4–5 mm) (Radford 1964, 1968; Massey et al. 1983; Wofford 1989; Weakley 2008).

Micranthes careyana and *M. caroliniana* were first described by Asa Gray in 1841 and 1846 respectively, who placed them in the genus *Saxifraga*. He had collected plants in the fall of 1843 and cultivated them, not realizing he had collected two different species until the following spring when they flowered (Lord 1961). Gray determined that the two species can only be distinguished in flower by sepal orientation and filament shape. *Micranthes careyana* has erect to spreading sepals and a filiform filaments, while *M. caroliniana* has completely reflexed sepals and clavate filaments at full anthesis. Lord (1961) determined that Gray's assessment of these characters was correct and also retained the two taxa as distinct species. However, many of these characters may not be consistent, are confusing to field botanists, and all are often difficult to interpret on dried

herbarium specimens. Therefore, it appears that many populations have been misidentified and many herbarium specimens have been annotated at some time as both species, calling the status of the two as distinct taxa into question (e.g. Weakley 2008, p. 527).

The case of Micranthes petiolaris

It has been hypothesized that many Southern Appalachian rock outcrop plant species may be more closely related to northern and alpine species than to other geographically proximate species, possibly being disjunct arctic species that survived in high-elevation communities after warming since the last ice age, approximately 20,000 years ago (Wiser 1994; Wiser et al. 1996). These plant communities likely contain Pleistocene relicts of an alpine flora that was much more widespread than the smaller isolated patches that currently exist today (Wiser 1998). It is possible that these communities were spread to lower latitudes from the north as the ice sheet moved south, and remained in high-elevation communities of the Southern Appalachians after the glaciers receded (Ramseur 1960). Therefore, it has been proposed that *M. petiolaris*, the only high-elevation rock outcrop species of *Micranthes* in the Southern Appalachians, may be more closely related to other arctic species, than to the other Southern Appalachian species. This hypothesis is reflected in its placement in section *Stellares* with *M. stellaris*, *M. ferruginea*, *M. bryophora*, and *M. folioliosa* and supported by its unique characters, including zygomorphic (bilaterally symmetric) flowers, a primarily bracteate inflorescence, coarsely dentate leaf margins, and multiseriate glandular hairs.

These characters are not found in the other Southern Appalachian species and are uniquely shared by the five North American species placed in section *Stellares*. In addition, Small (1933) placed *M. petiolaris* in the genus *Hydatica* Necker with eleven other species world wide, segregating this species from *Micranthes*. More recently, phylogenetic analyses of the chloroplast gene *matK* have clearly shown that the other four North American species in section *Stellares* are well represented in a distinct clade within the genus *Micranthes* (Mort and Soltis 1999). However, *M. petiolaris* was not included in this analysis.

Objectives of Study

There are several objectives of this research: (1) to examine the circumscriptions of the two similar species, *M. careyana* and *M. caroliniana*, to test the validity of the four reported morphological differences and to look for phenological and geographic differences in their distributions; (2) to look for molecular differences between the two similar species; (3) to examine the evolutionary relationships of the six Southern Appalachian species of *Micranthes* in the context of a larger phylogenetic analysis of the genus (Mort and Soltis 1999) using molecular markers and cladistic methods, especially M. petiolaris in context of the refugia hypothesis; and (4) to write a new identification key to the representative species of the region. The first hypothesis proposes that *M. careyana* and *M. caroliniana* are distinct taxa in accordance with the diagnosability species concept based on differences in morphological and molecular characters, and a possible phenological and geographic isolation. The second hypothesis proposes that

there is more than one distinct evolutionary lineage, or monophyletic group, among the species of *Micranthes* in the Southern Appalachians, with *M*. *petiolaris* belonging to a separate clade from the other five species.

MATERIALS AND METHODS

Plant Material

Leaf material from at least one population of all six species was collected in the field and preserved either as frozen (-70°C) or silica gel-dried tissue, along with whole plant specimens deposited as herbarium vouchers (Table 1). Additional leaf material of *M. careyana* and *M. caroliniana* was collected from at least one population from the following counties of the Blue Ridge Physiographic Province: Alleghany, Ashe, Avery, Graham, Haywood, Macon, Madison, McDowell, Mitchell, Polk, Swain, Transylvania, Watauga (North Carolina), Greenville and Pickens (South Carolina), Blount, Knox, Johnson, and Sevier (Tennessee) for a total of 21 accessions of the two species combined. Plants in each population were examined for the state of the three floral characters (sepal orientation, petal coloration, and filament shape). In addition, numerous dried specimens of the two species from the herbaria of North Carolina State University [NCSC], University of North Carolina-Chapel Hill [NCU], and Western Carolina University [WCUH] were used for morphological analysis of floral characters and 21 specimens were used for measurements of fruit length (Table 2). Approximately five capsules per specimen were measured, with the longest recorded, and plotted against the collection date of the specimen. A two-sample t-test (assuming equal variance) was conducted to determine whether or not there is a significant difference in the means of these measurements. Herbarium specimens were also used to document population localities and flowering times, and were annotated by the end of this research.

Table 1. Total new accessions examined for this study.

Species	Population	County/State	Coll. Date	Collection #
M. careyana	Nantahala Gorge	Swain/NC	09 Apr 08	015
M. careyana	Winding Stairs Rd.	Macon/NC	09 Apr 08	016
M. careyana	Slickrock Trail	Graham/NC	01 Aug 08	030-031
M. careyana	Elkhollow Branch	Avery/NC	07 Aug 08	032-033
M. careyana	Cullasaja Falls	Macon/NC	21 Aug 08	034
M. careyana	Crow Creek	Macon/NC	04 Sept 08	035
M. careyana	Clawhammer Mtn.	Transylvania/NC	16 Oct 08	037
M. careyana	Slickrock Trail	Graham/NC	09 Apr 09	046-048
M. careyana	Ivy River Bluffs	Madison/NC	17 Apr 09	053
M. careyana	Pigeon River Gorge	Haywood/NC	17 Apr 09	054
M. careyana	Ijam's Nature Center	Knox/TN	07 May 09	057-059
M. careyana	Little River Gorge	Blount/TN	11 May 09	060-063
M. careyana	Cliff Ridge	Macon/NC	13 May 09	064-066
M. careyana	Gouges Creek Falls	Mitchell/NC	21 May 09	067-068
M. careyana	Elkhollow Branch	Avery/NC	21 May 09	069
M. careyana	Profile Trail	Avery/NC	23 May 09	072-073
M. careyana	Linville Gorge	McDowell/NC	23 May 09	074
M. careyana	Unaka Springs	Unicoi/TN	04 June 09	079-081
M. caroliniana	Mt. Jefferson	Ashe/NC	30 May 08	024
M. caroliniana	New River St. Park	Alleghany/NC	30 May 08	025
M. caroliniana	Shady Valley	Johnson/TN	06 Mar 09	044
M. caroliniana	Howard Creek Falls	Watauga/NC	22 May 09	070-071
M. caroliniana	Shady Valley	Johnson/TN	28 May 09	075-077
M. micranthidifolia	Shook Cove Rd.	Jackson/NC	16 June 08	028
M. micranthidifolia	Dark Cove Farm	Jackson/NC	16 Apr 09	052
M. pensylvanica	Tater Hill Bog	Watauga/NC	30 May 08	022
M. petiolaris	Whiteside Mtn.	Macon/NC	01 Apr 08	011
M. petiolaris	Silver Run Falls	Jackson/NC	08 Apr 08	013-014
M. petiolaris	Blue Valley	Macon/NC	12 Apr 08	017
M. petiolaris	Cedar Rock Mtn.	Pickens/SC	19 Apr 08	019-020
M. petiolaris	Mt. Jefferson	Ashe/NC	30 May 08	023
M. petiolaris	Whiteside Mtn.	Macon/NC	01 June 08	026
M. petiolaris	Blue Valley	Macon/NC	08 June 08	027
M. petiolaris	Dry Falls	Macon/NC	29 June 08	029
M. virginiensis	Wadakoe Mtn.	Pickens/SC	19 Apr 08	018
M. virginiensis	Village Creek	Jefferson/AL	02 Apr 09	045
M. virginiensis	Melrose	Polk/NC	27 Apr 09	055
M. virginiensis	Gap Creek Rd.	Greenville/SC	27 Apr 09	056

Table 2. Measurements of fruit length from herbarium specimens.

Species	County/State	Voucher	Day/Month	Length (mm)
M. careyana	Pickens/SC	Lanning 018	19 Apr	4
M. careyana	Graham/NC	Pittillo 10148	22 Apr	4
M. careyana	Macon/NC	Mathews et al.	29 Apr	4
M. careyana	Macon/NC	Ramsey 40	01 May	3
M. careyana	Grundy/TN	Clark 1805	15 May	3
M. careyana	Haywood/NC	Pittillo 6560	15 May	3
M. careyana	Haywood/NC	Pittillo 102447	15 May	3.5
M. careyana	Graham/NC	Radford 11893	29 May	4
M. careyana	Madison/NC	Sather 1264	04 June	4
M. careyana	Henderson/NC	Radford 7116	05 June	3
M. careyana	Swain/NC	Radford 7232	07 June	4
M. careyana	McDowell/NC	Govus 112	08 June	3
M. careyana	Macon/NC	Pittillo 7529	12 June	3.5
M. careyana	Mitchell/NC	Brown 566515	22 June	4.5
M. caroliniana	Smyth/VA	Britton et al.	01 June	4.5
M. caroliniana	Ashe/NC	Radford 45390	03 June	3.5
M. caroliniana	Ashe/NC	N/A	07 June	4.5
M. caroliniana	Washington/VA	Harrill 18580	11 June	4.5
M. caroliniana	Smyth/VA	Small	13 June	4
M. caroliniana	Ashe/NC	Radford 43954	23 June	3.5
M. caroliniana	Ashe/NC	Poindexter 05-855	26 June	4.5

DNA isolation, amplification, and sequencing

Leaf tissue from fresh, frozen, silica gel-dried materials or herbarium vouchers were ground to powder in liquid nitrogen using either a mortar and pestle or the BioMasher mini-pestle and electric drill, and total DNA was isolated via the DNeasy Plant Mini Kit (Qiagen). Total DNA concentrations achieved were quantified using a NanoDrop ND-1000 Full-spectrum UV/Vis spectrophotometer and run on a 1% w/v agarose gel including 3μL of ethidium bromide for visualization on a U.V. light box. The nrDNA ITS region was amplified using external primer pairs N-nc18S10 and C26A following the protocol from Johnson and Soltis (1998), and chloroplast (*trn*L-*trn*F and *mat*K) DNA markers were amplified using external primer pairs *trn*L-F*f* and *trn*L-F*c* following the protocol of Soltis et al. (2001) and *trnK-3914F* and *trnK-2R* following the protocol from Soltis et al. (1996). Fifty-microliter PCR reactions were prepared using 10 μl Taq Master adjuvant, 5 μl 10x buffer, 2 μl dNTP (10 mM), 2.5 μl $MgCl_2$ (25 mM), 2 μl forward primer (10 μM), 2 μl reverse primer (10 μM), 0.25 μl Taq polymerase (5 U/μl), 1 μl of diluted (1/10, 1/50, or 1/100) DNA template with concentration ≤ 30 ng/μl, and ddH_2O to fill.

Amplifications for the ITS region were performed as follows: one denaturing cycle of 3 minute at 95°C; 30 cycles including a 1 minute denaturing at 95°C, a 1 minute annealing at 45°C, and a 1 minute and 20 second extension at 72°C; followed by a final extension of 5 minutes at 72°C. Amplifications for the *trn*L-*trn*F intergenic spacer region were performed as follows: one denaturing cycle of 2 minutes at 95°C; 30 cycles including a 50 second denaturing at 95°C, a 50 second annealing at 50°C, and a 1 minute and 50 second extension at 72°C; followed by a final extension of 7 minutes at 72°C

Amplifications for the chloroplast gene *mat*K were performed as follows: one denaturing cycle of 3 minutes at 94°C; 30 cycles including a 1.5 minute denaturing at 94°C, a 2 minute annealing at 48°C, and a 3 minute extension at 72°C; followed by a final extension of 15 minutes at 72°C. The same primer pairs were used for sequencing of the ITS and *trn*L-F regions, however the primers *trn*K-390F and *mat*K-2000R were used for sequencing of the *mat*K gene.

PCR products were run on a 1% w/v agarose gel with a DNA ladder for sizing and visualization, then cleaned and prepared for sequencing using the QIAquick PCR Purification Kit (Qiagen). Concentrations of PCR products were quantified using NanoDrop ND-1000 in order to determine the amount of DNA template to be used in sequencing reactions. Ten microliter sequencing reactions were prepared with 1 µl DNA template with a concentration ≤ 30 ng/µl, 4 µl Big Dye premix (Applied Biosystems), 3.2 µl of a 1 µM solution of each primer, and 1.8 µl ddH_2O. Cycle-sequencing reactions were performed with an initial denaturing cycle of 60 seconds at 96°C; followed by 24 cycles including a denaturing cycle of 10 seconds at 96°C; an annealing cycle of 5 seconds at 50°C; an extension cycle of 240 seconds at 60°C; followed by an indefinite hold at 4°C. Reactions were purified by an Ethanol – Sodium Acetate precipitation or Jephadex columns (Illustra GEH Sciences) and dried down in a vacuum centrifuge. Dried sequencing reactions were resuspended in 10 µl of Hi-Di formamide and microcentrifuged. Reaction tubes were incubated at 95° C for five minutes to denature DNA. Tubes were then snap-chilled in ice for at least two minutes to prevent re-annealing of the DNA strands. The entire 10 µl volume of denature samples was loaded into a 96-well reaction plate and electrophoresed on a 4-capillary 3130 Genetic Analyzer

(Applied Biosystems, Foster City CA). Resulting chromatograms were visualized using the Sequencing Analysis software (Applied Biosystems) and downloaded for further analysis.

Sequence alignments and phylogenetic analyses

The forward and reverse primer sequences for each sample were uploaded into and viewed in the sequence editing program Sequencher (GeneCodes Corp.) in order to compare and confirm the sequences of the opposite complementary strands of each sample and combine into a consensus sequence. A large ITS data set was created using outgroup sequences obtained from GenBank (Table 3) and all generated sequences of Southern Appalachian species (Table 4). Simple ITS and *trn*L-F data sets were created using the outgroup sequences (Table 3) combined with representative sequences for the Southern Appalachian species (Table 4), and subsequently combined for a forth data set of these sequences. Lastly, a *mat*K data set was created by combining three sequences of Southern Appalachian species to the data set of outgroup *Micranthes* sequences from Mort and Soltis (1999) (Table 5). Sequences were loaded into ClustalX (Thompson et al. 1997) for a complete alignment and generation of a Nexus file of aligned sequences and gaps. Phylogenetic analyses were performed by loading all sequences into PAUP* (Swofford 2003) to run a heuristic parsimony search on the data using 100 replicates of random taxon addition. Pairwise differences among the taxa were viewed in a triangle matrix to analyze the percent nucleotide differences. A bootstrap analysis (Felsenstein 1985) was conducted as a measure of confidence for the level of support for the various

clades in the tree, by randomly resampling characters from the original data matrix using 100 replicates of taxon addition.

Table 3. Outgroup *Micranthes* sequences used in both ITS and *trn*L-F data sets obtained from either Johnson and Soltis (1998), Soltis et al. (2001), or directly from GenBank.

Taxon	Voucher	ITS(1/2)	*trn*L-F
M. integrifolia	Soltis & Soltis 2253	Johnson & Soltis 1998	AF374801
M. punctata	Soltis & Soltis 2217	Johnson & Soltis 1998	AF374800
M. stellaris	Horandl 2703	AF374827/28	AF374802
M. tolmiei	WS 32167	Johnson & Soltis 1998	AF374799

Table 4. Sequences representing various populations of Southern Appalachian *Micranthes* species used in the ITS data set along with four outgroup *Micranthes* sequences obtained from GenBank (see Table 2). Populations with an asterisk (*) were used as representative sequences in the simple ITS and *trn*L-F data sets.

Taxon	Population	County/State	Voucher
M. careyana	Nantahala Gorge 1	Swain/NC	Mathews (2007)
	*Nantahala Gorge 2	Swain/NC	Lanning 015
	Slickrock Trail	Graham/NC	Lanning 030
	Elkhollow Branch	Avery/NC	Lanning 032
	Cullasaja Falls	Macon/NC	Lanning 034
	Crow Creek	Macon/NC	Lanning 035
	Clawhammer Mtn.	Transylvania/NC	Lanning 037
	Ivy River Bluffs	Madison/NC	Lanning 053
	Pigeon River Gorge	Haywood/NC	Lanning 054
	Ijam's sp. 1	Knox/TN	Lanning 057
	Ijam's sp. 2	Knox/TN	Lanning (2009)
	Little River Gorge	Blount/TN	Lanning 060
	Groto Falls	Sevier/TN	Stoehrel (2009)
	Cliff Ridge	Macon/ NC	Lanning 064
	Gouges Creek Falls	Mitchell/NC	Lanning 067
	Profile Trail	Avery/NC	Lanning 072
	Linville Gorge	McDowell/NC	Lanning 074
M. caroliniana	*Mt. Jefferson	Ashe/NC	Lanning 024
	New River St. Park	Alleghany/NC	Lanning 025
	Shady Valley	Johnson/TN	Lanning 044
	Howard Creek Falls	Watauga/NC	Lanning 070
M. micranthidifolia	*Fisher Creek (1)	Jackson/NC	Lanning (2008)
	Elkhollow Branch (2)	Avery/NC	Lanning (2008)
M. pensylvanica	*Tater Bog	Watauga/NC	Lanning 022
M. petiolaris	Cedar Rock Mtn. (1)	Pickens/SC	Lanning 017
	Silver Run Falls (2)	Jackson/NC	Lanning 013
	Mt. Jefferson (3)	Ashe/NC	Lanning 023
	*Whiteside Mtn. (4)	Macon/NC	Lanning 026
M. virginiensis	*Village Creek	Jefferson/AL	Lanning 045
Unknown spp.	Wadakoe Mtn.	Pickens/SC	Lanning 019
	Melrose	Polk/NC	Lanning 055
	Gap Creek Rd.	Greenville/SC	Lanning 056

Table 5. Species of *Micranthes* used in phylogenetic analyses of *mat*K sequences. Sequences generated by this study are indicated with an asterisk (*). All other sequences obtained are from Soltis et al. (1996) or Mort and Soltis (1999) and were obtained from GenBank.

Taxon	Voucher	GenBank No.
Cascadia tolmiei (Small) A.M. Johnson	WS 21562	AF115483
M. aprica (Greene) Small	HSU 85431	AF114179
M. bryophora (A.Gray) Brouillet & Gornall	Ertter 7775	AF114180
M. californica (Greene) Small	HSU 85868	AF114172
M. calycina (Sternb.) Gornall & H. Ohba	Murray & Kelso 11309	AF115492
**M. careyana* (A. Gray) Small	Mathews (2007)	----------
M. ferruginea (Graham) Brouillet & Gornall	Soltis & Soltis 2233	L34141
M. folioliosa (R. Brown) Gornall	Murray et al. 7058	AF115494
M. fusca Maximowicz	Soltis 2524	AF115490
M. hieracifolia (Waldst. & Kit.) Haworth	Brochman 92-4-8	AF115485
M. howellii (Greene) Small	Cronquist 6863	AF114178
M. integrifolia (W.J. Hooker) Small	Johnson & Soltis (1994)	L20131
M. lyallii (Engler) Small	Soltis & Soltis 223	AF115487
**M. micranthidifolia* (Haworth) Small	Lanning (2008)	----------
M. nidifica (Greene) Small	Daubenmire 46107	AF114182
M. nivalis (L.) Small	Murray et al. 11350	AF115491
M. occidentalis (S. Watson) Small	Soltis & Soltis 2504	AF114177
M. odontoloma (Piper) A.A. Heller	WS 11639	AF114184
M. oregana (Howell) Small	Norment 318	AF114181
M. pensylvanica (L.) Haworth	Layser 1737	AF114175
**M. petiolaris* (Raf.) Brouillet & Gornall	Lanning 026	----------
M. punctata D. Don (Small) (=*M. nelsonia*)	Soltis & Soltis 2217	L34144
M. reflexa (W.J. Hooker) Small	Murray & Kelso 11308	AF115486
M. rhomboidea (Greene) Small	Weber & Randolph 17371	AF114176
M. rufidula Small	Arp s.n.	AF114183
M. spicata (D. Don) Small	Murray et al. 11111	AF114173
M. stellaris (L.) Galasso, Banfi & Soldano	Horandl 2703	AF374802
M. tenuis (Wahlenberg) Small	Brochman s.n.	AF115489
M. texana Buckley (Small)	Freeman 3055	AF114174
M. tolmiei (T. & G.) Brouillet & Gornall	WS 32167	AF115484
M. virginiensis (Michaux) Small	OBG 75-449-5	AF115488

RESULTS

Morphology

Field observations of *Micranthes careyana* and *M. caroliniana* populations support differences in only two of the four reported floral characters: sepal orientation and filament shape. Populations in Avery, Graham, Haywood, Macon, Madison, McDowell, Mitchell, Polk, Swain, Transylvania counties of North Carolina, and Blount, Knox, and Sevier counties of Tennessee exhibit an erect, later spreading sepal orientation and filiform (uniformly-shaped) anther filaments at anthesis (Table 6; Figure 1). Populations in Alleghany, Ashe, and Watauga counties of North Carolina and Johnson County, Tennessee exhibit a fully reflexed sepal orientation and clavate (club-shaped) anther filaments at anthesis (Table 6; Figure 2). Populations of these two species are extremely difficult to identify in the field based on vegetative characters alone (Figures 4 and 5).

Table 6. State of floral character states of populations of both species examined in flower.

Population Name	County	State	Sepals	Filaments	Petals
Nantahala Gorge	Swain	NC	erect	filiform	2 yellow spots
Winding Stairs Rd.	Macon	NC	erect	filiform	2 yellow spots
Slickrock Trail	Graham	NC	erect	filiform	2 yellow spots
Ivy River Bluffs	Madison	NC	erect	filiform	2 yellow spots
Pigeon River Gorge	Haywood	NC	erect	filiform	2 yellow spots
Ijam's Nature Center	Knox	TN	erect	filiform	2 yellow spots
Little River Gorge	Blount	TN	erect	filiform	2 yellow spots
Cliff Ridge	Macon	NC	erect	filiform	2 yellow spots
Gouges Creek Falls	Mitchell	NC	erect	filiform	2 yellow spots
Elkhollow Branch	Avery	NC	erect	filiform	2 yellow spots
Linville Gorge	McDowell	NC	erect	filiform	2 yellow spots
Profile Trail	Avery	NC	erect	filiform	2 yellow spots
Mount Jefferson	Ashe	NC	reflexed	clavate	2 yellow spots
New River St. Park	Alleghany	NC	reflexed	clavate	2 yellow spots
Shady Valley	Johnson	TN	reflexed	clavate	2 yellow spots
Backbone Rock	Johnson	TN	reflexed	clavate	2 yellow spots
Howard Creek Falls	Watauga	NC	reflexed	clavate	2 yellow spots
Wadakoe Mountain	Pickens	SC	erect	filiform	no spots
Melrose	Polk	NC	erect	subulate	no spots
Gap Creek Rd.	Greenville	SC	erect	filiform	no spots

Figure 1. *Micranthes careyana* flower displaying erect sepals and filiform filaments (white arrows).

Figure 2. *Micranthes caroliniana* flower displaying fully reflexed sepals and clavate filaments (black arrows).

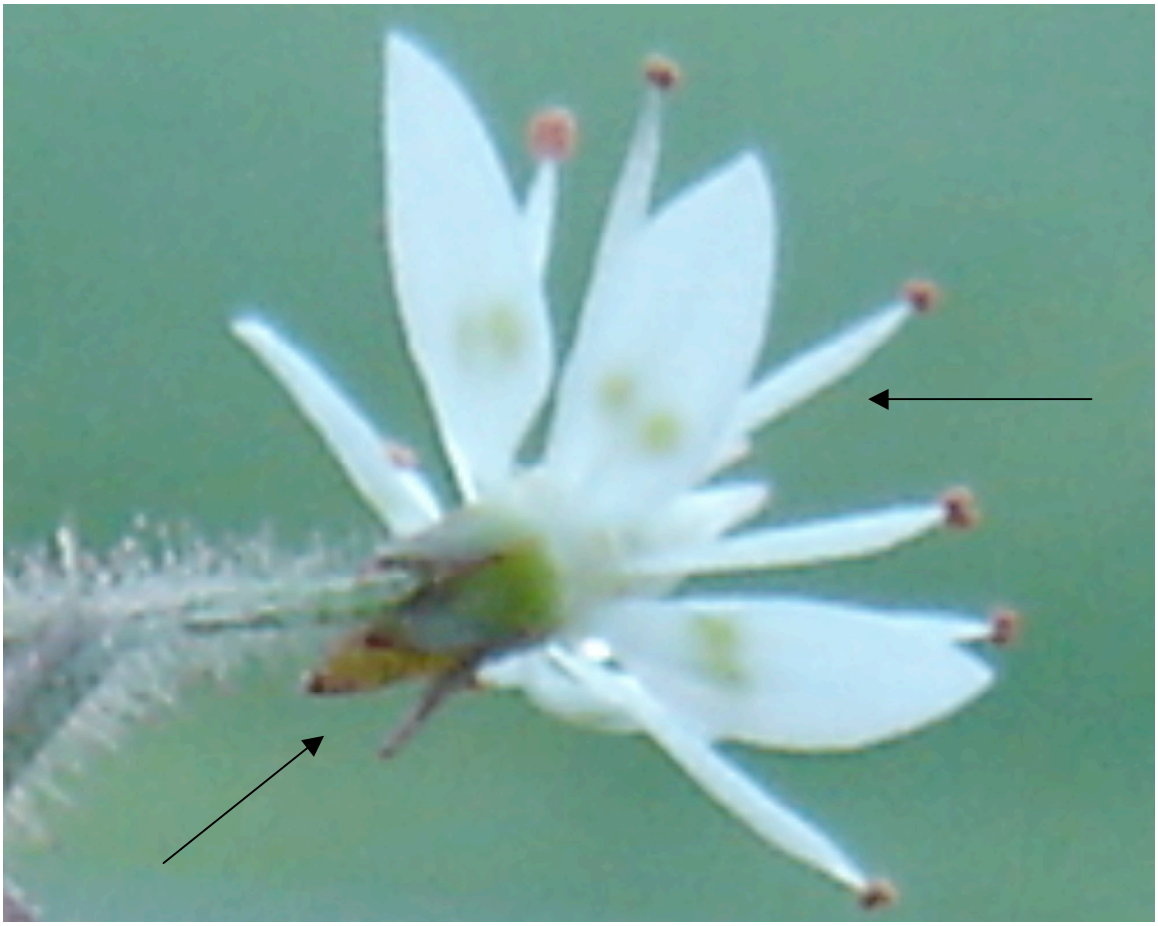


Figure 3. *Micranthes careyana* in flower, Nantahala Gorge, Swain County, NC.

Figure 4. *Micranthes caroliniana* in flower, New River State Park, Alleghany County, North Carolina.

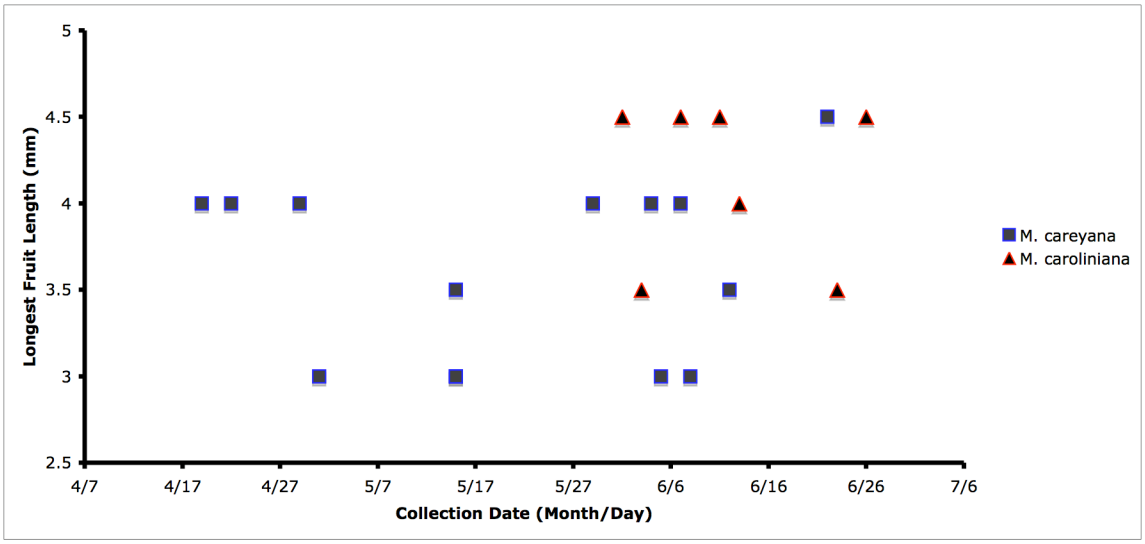
Three populations from Polk (NC), Greenville and Pickens (SC) counties exhibit characters not consistent with descriptions of either *M. careyana* or *M. caroliniana* (Table 6). The population in Polk County ("Melrose") appears to share more characters in common with *M. virginiensis* than with either of the two other taxa, particularly in the length of the petals (4-4.5 mm vs. 3-3.5 mm), the absence of coloration on the petals, the length of the filaments (1-1.5 mm vs. 3-3.5 mm), and the shape of the filaments (subulate vs. filiform or clavate). However, there is one defining character of *M. virginiensis* that is not shared by this population; the fusion of petals at the base forming a partial hypanthium. The petals of individuals of this population are not fused at the base and are actually recurved back, possibly because the flowers appear to hang over upside down (Figure 5). It is unclear at this point whether this population represents a local variety or subspecies of *M. virginiensis* or an entirely new species altogether.

Figure 5. *Micranthes* sp. flower preserved in ethanol in natural orientation, Melrose, Polk County, NC.

The populations in Greenville and Pickens County were observed and collected in the late flowering – early fruiting stage and therefore some of the floral characters were not observable (e.g. some missing petals). Both populations appear to have erect sepals, filiform filaments, no coloration on the petals, long filaments (ca. 3-3.5 mm), and a large fruit size (4-4.5 mm). These populations are especially problematic, because it is unclear what species they belong to based on their floral characters alone. The lack of coloration on the petals suggests *M. virginiensis*, but the erect sepal orientation, filiform filament shape, and long filament and fruit lengths suggest *M. careyana*. It must be noted however that these two populations have previously been identified as *M. virginiensis* (P. McMillan, pers. com.)

All of the populations determined to represent *M. careyana* or *M. caroliniana* exhibit flowers with 2 yellow spots on each petal (Table 6). Measurements of fruit size did differ slightly ($t = 0.54$; $P = 0.04$), but is not an informative field character (Figure 6). In fact, the distinguishing fruit length measurements used in field identification keys are overlapping (3.5-5 vs. 4.5-5 mm) and measurements from the same individual or individuals of the same population are often indicative of both species. Therefore petal coloration and fruit length are not informative characters for distinguishing the two taxa. Measurements of fruit size from herbarium sheets confirm field observations by showing a great deal of overlap when plotted against the collection date (Figure 6).

Figure 6. Measurements of fruit length from herbarium specimens plotted against the collection date.



An interesting population can be found at Ijam's Nature Center along the Tennessee River in Knoxville, Tennessee. In addition to individuals belonging to *M. careyana*, there are individuals which key out to *M. virginiensis* growing together in a mixed population. There is no apparent introgression based on morphology, although both species were found to be flowering at the same time. This discovery is not particularly surprising though, since Knoxville is located in a unique position geographically that lies on the edge of the distribution of both species.

Several populations of *M. careyana* that were previously misidentified by various botanists as *M. caroliniana* in natural heritage data should be noted: Cliff Ridge in Macon County, NC; Winding Stairs Road, Macon, NC; Slickrock Trail, Graham County, NC; Ivy River Bluffs, Madison County, NC; Pigeon River Gorge, Haywood County, NC; Gouges Creek Falls, Mitchell County, NC; Elkhollow Branch, Avery County, NC; Profile Trail (Grandfather Mountain), Avery County, NC; and Chimney Rock Park, Rutherford County, NC. In addition, one population of *M. petiolaris* was misidentified as *M. caroliniana* from Judaculla Ridge, Jackson County, NC. These determinations were made based on observations in the field and examination of herbarium specimens.

The populations in Greenville and Pickens County, South Carolina and Polk County, North Carolina appear to belong to or be closely related to *M. virginiensis*. This taxon has a large distribution in eastern North America, as far north as Canada and as far south as Louisiana, but is extremely rare in the mountains of the Blue Ridge. These three populations are found in a transitional area between the Mountain and Piedmont Physiographic Provinces, which could explain their unique floral characters.

Phenology

There appears to be a slight difference in flowering times among populations of *M*. *careyana* and *M*. *caroliniana*, with some overlap in those populations found on the edge of the distributions of the two taxa in northwestern North Carolina. Generally, *M*. *careyana* flowers from late March through May and *M*. *caroliniana* flowers from late April to early June. The three populations of questionable identity do fall into the range of flowering time reported for *M*. *virginiensis* (early saxifrage) from early March through April, as the common name implies. Both populations in Greenville and Pickens County were in the fruiting stage in late April and the population in Polk County was in late anthesis, or late flowering stage, at the end of April (Table 7).

Table 7. Date observed and reproductive state of populations examined in the field.

M. careyana	County	State	Date Observed	Reproductive State
Nantahala Gorge	Swain	NC	09 Apr 2008	flower
Winding Stairs Rd.	Macon	NC	09 Apr 2008	flower
Slickrock Trail	Graham	NC	09 Apr 2009	flower
Pigeon River Gorge	Haywood	NC	17 Apr 2009	flower
Ivy River Bluffs	Madison	NC	24 Apr 2009	flower
Ijam's Nature Center	Knox	TN	07 May 2009	flower
Little River Gorge	Sevier	TN	11 May 2009	flower
Cliff Ridge	Macon	NC	13 May 2009	flower
Gouges Creek Falls	Mitchell	NC	21 May 2009	flower
Elkhollow Branch	Avery	NC	21 May 2009	flower
Linville Gorge	McDowell	NC	23 May 2009	flower
Profile Trail	Avery	NC	23 May 2009	flower

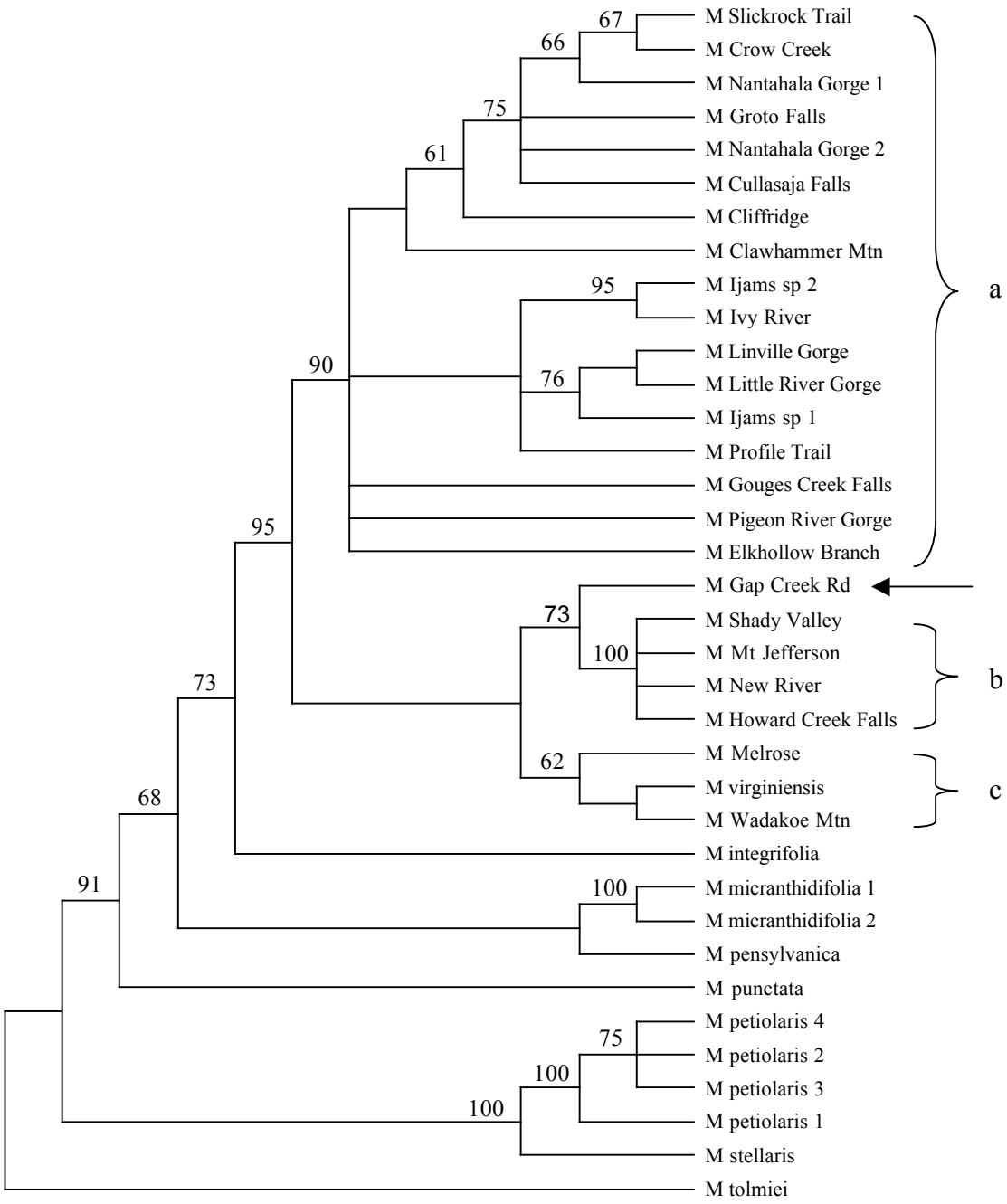
M. caroliniana	County	State	Date Observed	Reproductive State
Mount Jefferson	Ashe	NC	30 May 2008	flower
New River Bluffs	Alleghany	NC	30 May 2008	flower
Shady Valley	Johnson	TN	28 May 2009	flower
Backbone Rock	Johnson	TN	28 May 2009	flower
Howard Creek Falls	Watauga	NC	22 May 2009	flower

Others	County	State	Date Observed	Reproductive State
Wadakoe Mountain	Pickens	SC	19 Apr 2008	fruit
Melrose	Polk	NC	27 Apr 2009	flower
Gap Creek Rd.	Greenville	SC	27 Apr 2009	fruit

Phylogeny

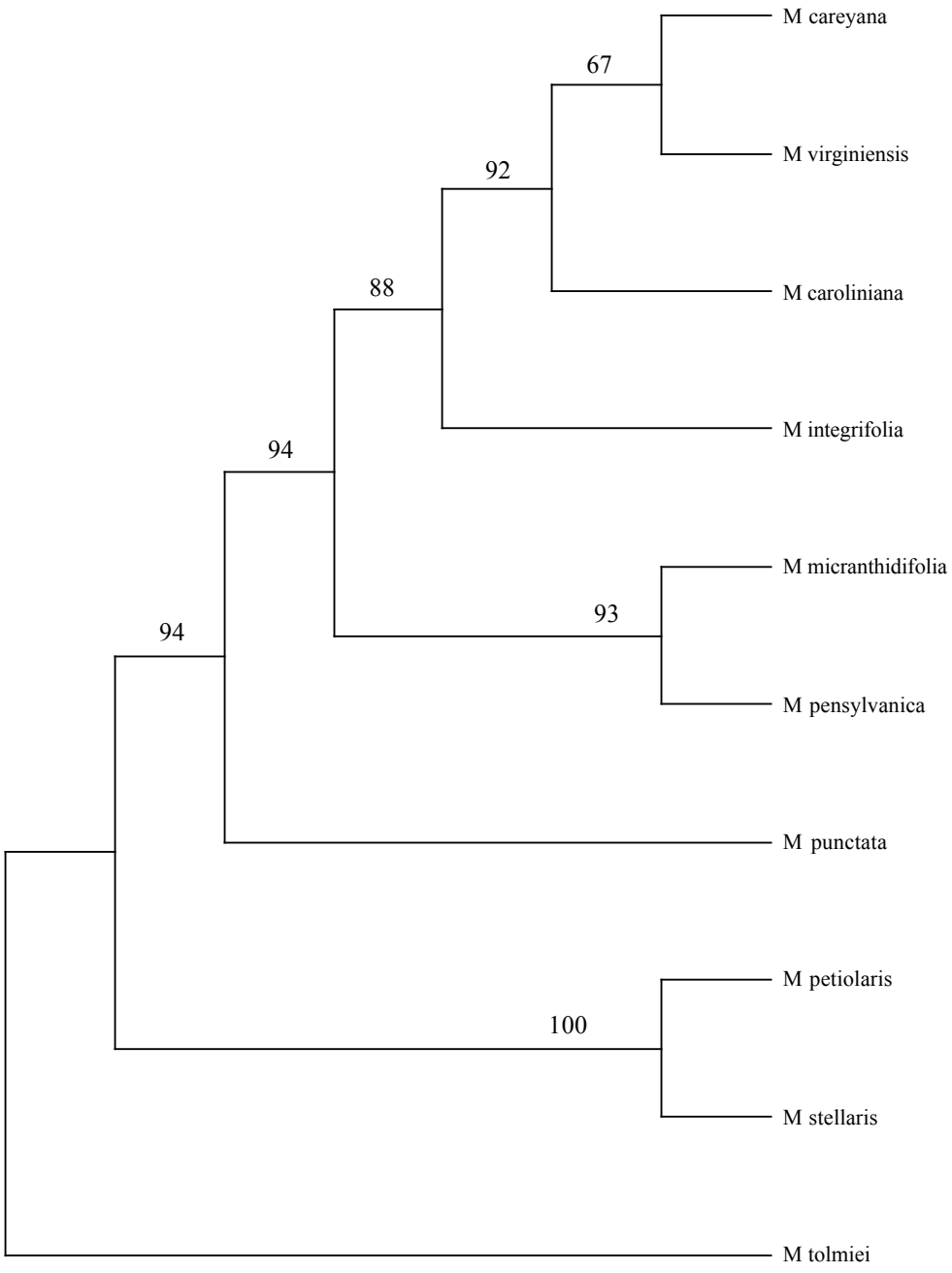
The aligned ITS data matrix contained 711 total characters, 214 of which were parsimony informative. Pairwise divergence values among ingroup taxa ranged from 0.00612 (0.61%) to 0.30851 (30.85%). Parsimony analysis of the large ITS data set yielded 144 shortest trees of length 739 (CI = 0.5946; RI = 0.7692). In the strict consensus tree (Figure 7), all populations identified as *M. careyana* in the field are represented in a well-supported clade (90%), with the exception of "Ijam's sp. 2" which was identified as *M. virginiensis* based on floral characteristics. All populations identified as *M. caroliniana* in the field are represented in a well-supported clade (100% BS), to the exclusion of the "Gap Creek Rd." population (73% BS), which was difficult to identify in the field. Another clade is represented by a known population of *M. virginiensis* (Village Creek, Jefferson County, Alabama) + the "Wadakoe Mtn." population (69%) with the "Melrose" population, both of which were also difficult to identify in the field. The close relationship of the three clades representing *M. careyana*, *M. caroliniana*, and *M. virginiensis* is also well-supported (95%). A sister-group relationship was found between *M. caroliniana* and *M. virginiensis*, although this is not supported by bootstrap analyses.

Figure 7. Strict consensus tree obtained through parsimony analyses of all ITS sequences (length = 739 steps; CI = 0.5946; RI = 0.7692). Bootstrap values >50% indicated above branches. Species designations indicated by brackets for sequences named by population: (a) *M. careyana*, (b) *M. caroliniana*, and (c) *M. virginiensis*. Arrow indicates problematic population (c.f. *virginiensis*).



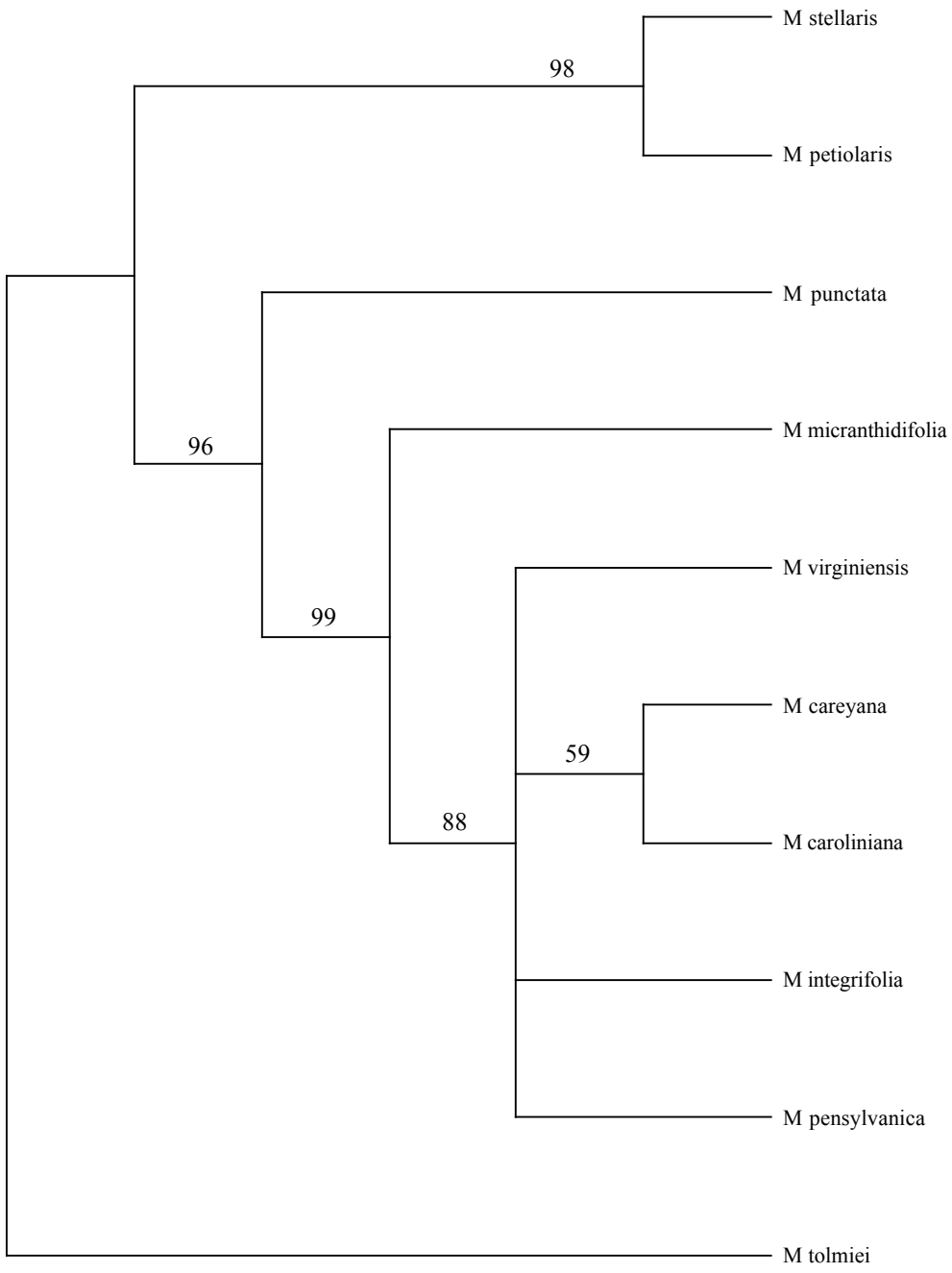
The aligned ITS data matrix using only representative species contained 810 total characters, 135 of which were parsimony informative. Pairwise divergence values among ingroup taxa ranged from 0.03357 (3.36%) to 0.35084 (35.08%). Parsimony analyses of this data set yielded 1 shortest tree of length 425 (CI = 0.8235; RI = 0.6696). The single shortest tree (Figure 8) revealed two major well-supported clades: *M. stellaris* + *M. petiolaris* of Section *Stellares* (100%) and all other ingroup taxa (94%) including members of section *Micranthes*, and *M. punctata* of section *Rotundifoliatae*. Within section *Micranthes*, there are an additional two well-supported clades: *M. virginiensis* + *M. careyana* + *M. caroliniana* (subsection *Dermasea*) (92%), subsection *Dermasea* + *M. integrifolia* (subsection *Micranthes*) (88%); and *M. micranthidifolia* (subsection *Aulaxis*) + *M. pensylvanica* (subsection *Micranthes*) (93%). In this phylogeny, *M. careyana* and *M. virginiensis* are sister taxa with bootstrap support of 67%.

Figure 8. Single shortest tree obtained through parsimony analyses of ITS sequences (length = 425 steps; CI = 0.8235; RI = 0.6696). Bootstrap values >50% indicated above branches.



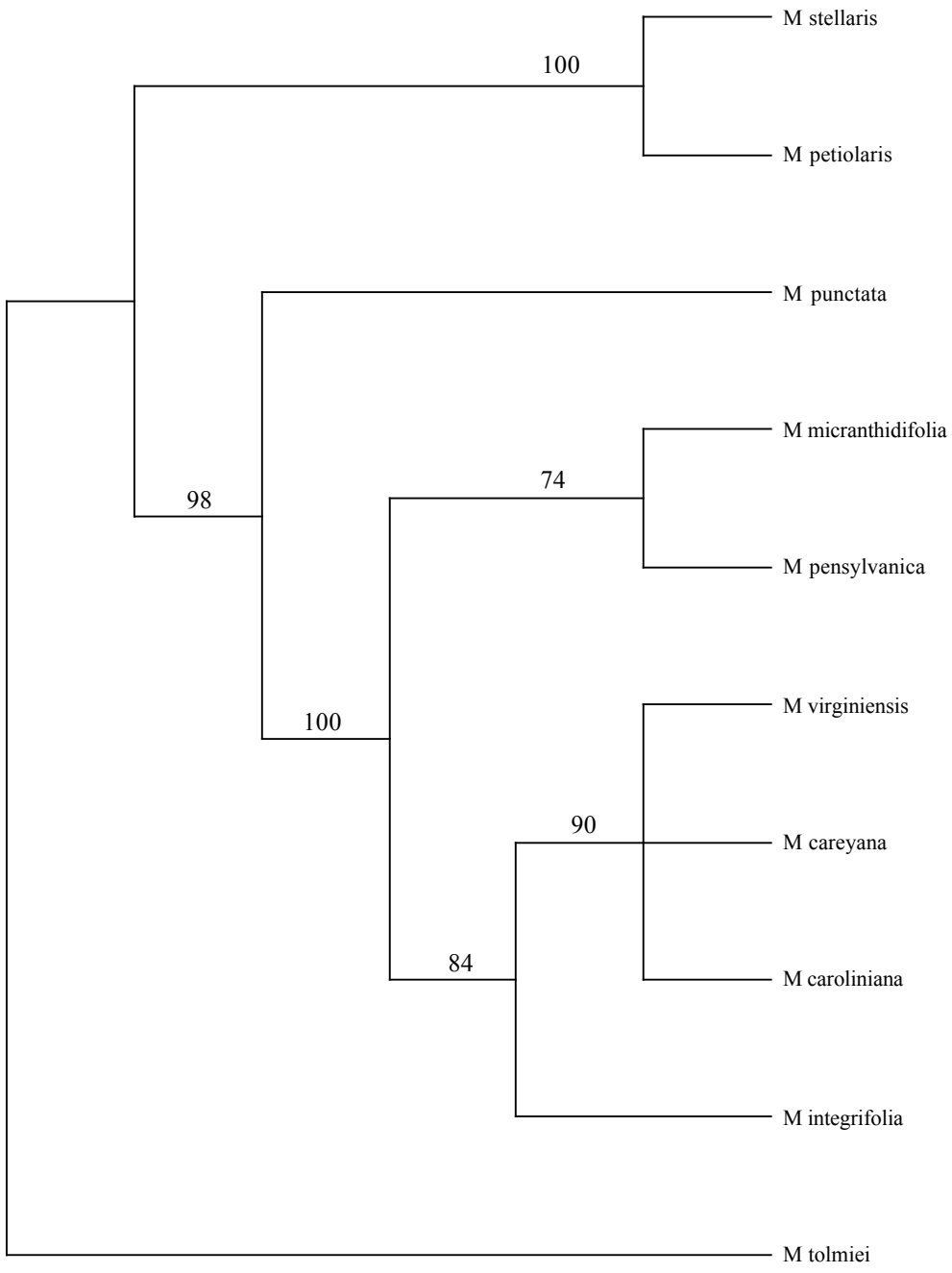
The aligned *trn*L-*trn*F data matrix using only representative species contained 911 total characters, 46 of which were parsimony informative. Pairwise divergence values among ingroup taxa ranged from 0.01051 (1.05%) to 0.22210 (22.21%). Parsimony analyses of this region yielded 5 shortest trees of length 258 (CI = 0.9535; RI = 0.8378). In the strict consensus tree (Figure 9), *M. stellaris* + *M. petiolaris* (section *Stellares*) form a well-supported clade (98% BS). Section *Micranthes* is also supported as monophyletic (99% BS) with *M. punctata* (section *Rotundifoliatae*) as its sister group (96% BS). Within section *Micranthes*, *M. micranthidifolia* is sister to a clade containing all other taxa (88% BS). *M. careyana* and *M. caroliniana* form a sister group with low bootstrap support (59%).

Figure 9. Strict consensus tree obtained through parsimony analyses of *trn*L-F sequences (length = 258 steps; CI = 0.9535; RI = 0.8378). Bootstrap values >50% indicated above branches.



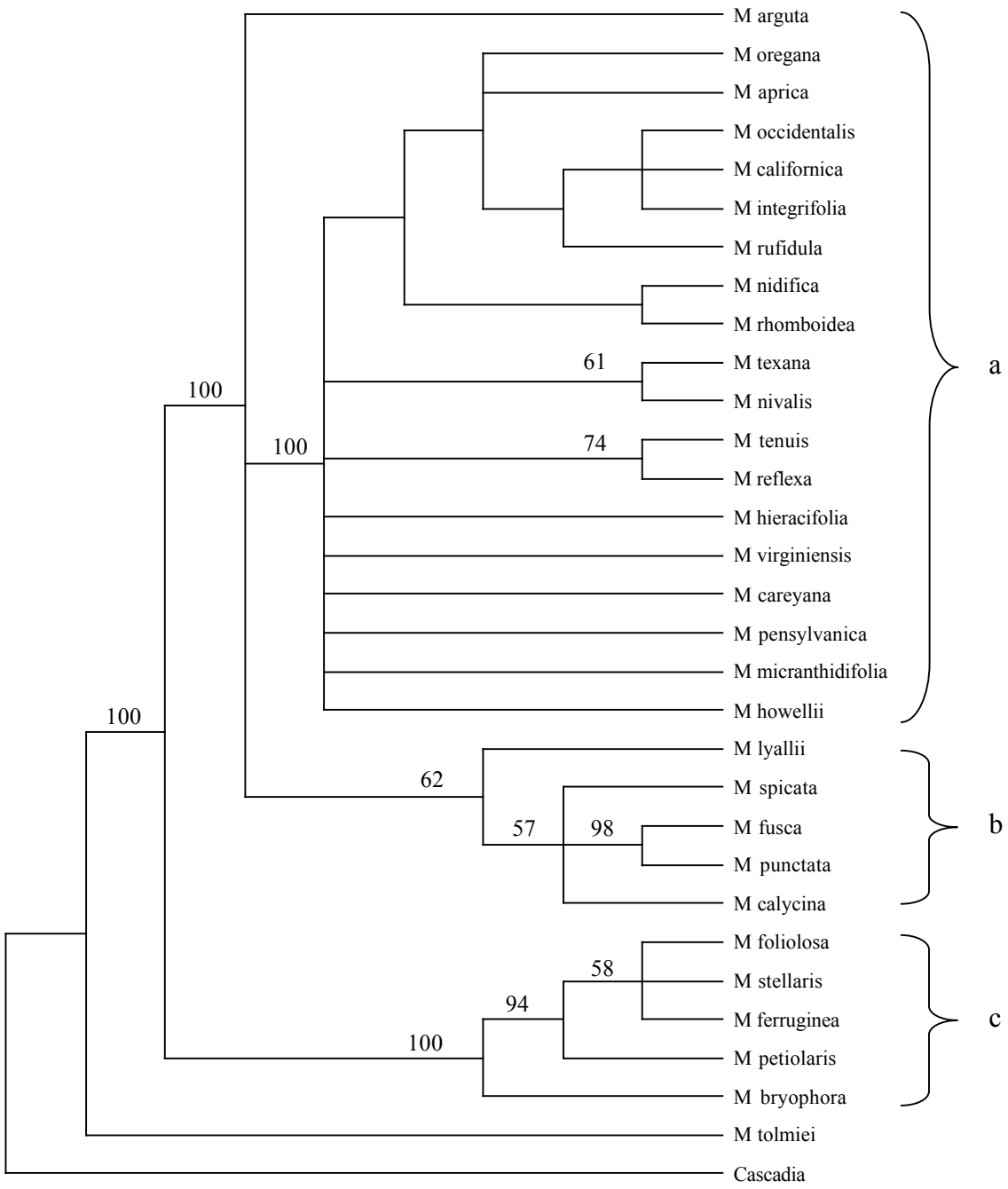
The aligned data set of combined *trn*L-F and ITS sequences included 1721 total characters, 181 of which were parsimony informative. Pairwise divergence values among ingroup taxa ranged from 0.02629 (2.63%) to 0.26453 (26.45%). Parsimony analyses of the combined regions yielded three shortest trees of length 689 (CI = 0.8650; RI = 0.6910). In the strict consensus tree (Figure 10), *M. stellaris* + *M. petiolaris* (section *Stellares*) form a well-supported clade (100% BS). Section *Micranthes* is also supported as monophyletic (100% BS) with *M. punctata* (section *Rotundifoliatae*) as its sister group (98% BS). Within section *Micranthes*, *M. micranthidifolia* and *M. pensylvanica* are sister taxa (74% BS) and *M. virginiensis* + *M. careyana* + *M. caroliniana* for a clade (90% BS) to the exclusion of *M. integrifolia*.

Figure 10. Strict consensus tree obtained through parsimony analyses of combined *trn*L-F and ITS sequences (length = 689 steps; CI = 0.8650; RI = 0.6910). Bootstrap values >50% indicated above branches.



The aligned data set of *mat*K sequences included 1523 total characters, 181 of which were parsimony informative. Pairwise divergence values among ingroup taxa ranged from 0.00113 (0.11%) to 0.18845 (18.85%). Parsimony analyses of this region yielded 16,043 shortest trees of length 479 (CI = 0.8330; RI = 0.8802). The strict consensus tree (Figure 11) revealed three major well-supported clades representing sections *Micranthes* (100% BS) to the exclusion of *M. arguta* (section *Rotundifoliatae*) (51% BS); *Rotundifoliatae* (62% BS), with the inclusion of *M. calycina* (section *Cuneifoliatae*); and *Stellares* (100% BS). In addition a strong sister relationship is supported between the *Micranthes* and *Rotundifoliatae* clades (100% BS). All of the Southern Appalachian species appear within the *Micranthes* clade, except *M. petiolaris*, which appears in the *Stellares* clade.

Figure 11. Strict consensus tree obtained through parsimony analyses of matK sequences (length = 479 steps; CI = 0.8330; RI = 0.8802). Bootstrap values >50% indicated above branches. Sectional designations indicated with brackets: (a) *Micranthes*, (b) *Rotundifoliatae*, and (c) *Stellares*.



DISCUSSION

Taxonomic boundaries and species relationships

The presence of both morphological and molecular synapomorphies in the Alleghany, Ashe, Watauga (North Carolina) and Johnson (Tennessee) county populations, determined to represent *Micranthes caroliniana*, support the status of this taxon as distinct from *M. careyana*, in accordance with the diagnosablity species concept (Nixon and Wheeler 1990). Completely reflexed sepals at anthesis and clavate (club-shaped) filaments can be used as field characters to identify *M. caroliniana*. Petal coloration and fruit size are not informative characters for distinguishing these two species in the field. Misidentification of these two species seems to stem from misinterpretation of the characters used in dichotomous keys, rather than the characters themselves being unreliable. *Micranthes careyana* can exhibit a spreading sepal orientation during late anthesis that is often interpreted as "reflexed" and subsequently leads to its misidentification as *M. caroliniana*. In addition, these two species are geographically and phenologically isolated, with *M. caroliniana* being restricted to just seven counties in northwestern North Carolina, northeastern Tennessee, and southwestern Virginia and flowering at a slightly later date. Based on the molecular phylogenetic analyses of ITS and *trn*L-F sequences, as well as morphological similarity, both *M. careyana* and *M. caroliniana* appear to be closely related to *M. virginiensis*, which supports the taxonomic classification of these three species in section *Micranthes*, subsection *Dermasea*. *Micranthes caroliniana* is seemingly the most well defined

species in this complex, possibly due to its limited geographical distribution and unique floral characters.

Molecular analyses of ITS and *trn*L-F sequences also support the separation of *Micranthes careyana* and *M. caroliniana* as distinct species. In fact, in the large ITS tree (Fig. 9) *M. caroliniana* and *M. virginiensis* are sister species to the exclusion of *M. careyana*. In the simple ITS tree (Fig. 10), *M. careyana* and *M. virginiensis* are sister species to the exclusion of *M. caroliniana*. However, there is little bootstrap support for either of these sister group relationships, although the support for the *M. careyana* + *M. caroliniana* + *M. virginiensis* clade is strong in each analysis. Furthermore in the *trn*L-F tree (Fig. 11) *M. careyana* and *M. caroliniana* appear as sister species with only 59% bootstrap support and the relationship of the three species cannot be determined in the combined ITS - *trn*L-F tree (Fig. 12), but the support for all three taxa being closely related is well supported by a 90% bootstrap value. From these molecular analyses, it is unclear which of these species are sister lineages and which is the more ancestral. However, it is clear that *M. careyana* and *M. caroliniana* are in fact distinct lineages and both are closely related to *M. virginiensis*. There is strong support for the monophyly of all three taxa based on morphology, biogeography, and phylogeny.

Micranthes virginiensis is an extremely complex species and it is difficult to determine what is included in this taxon. It has a very widespread distribution in the eastern United States and is known to have some named varieties in eastern North America. The two populations in Greenville County ("Gap Creek Rd.") and Pickens County, South Carolina ("Wadakoe Mtn.") were difficult to identify in the field due to a late observation of the flowering state, and appear to have characters in common with

both *M. careyana* and *M. virginiensis*. Both populations have previously been identified as (P. McMillan, pers. com.) and appear to be closely related to *M. virginiensis*, although the "Gap Creek Rd." population appears to be closely related to the *M. caroliniana* clade (73% BS) in the large ITS data set (Fig. 9). However, it is extremely unlikely this population should actually be included in *M. caroliniana* based on morphology and biogeography. The population in Polk County, North Carolina ("Melrose") also appears to belong to or be closely related to *M. virginiensis* based on morphology and ITS data, but possesses unique floral characters. Flowers from individuals of this population exhibit petals that are not fused at the base, a defining character of *M. virginiensis*, and which are actually reflexed back away from the carpels.

Micranthes micranthidifolia of the monotypic subsection *Aulaxis* and *M. pensylvanica* of subsection *Micranthes* appear to be closely related based on parsimony analyses of ITS sequences. However, the relationship of these taxa is unresolved in the *trn*L-F and *mat*K phylogenies. These species both have long, oblanceolate shaped leaves and are sometimes confused for one another. However, there are many characters that easily distinguish these two species. *Micranthes micranthidifolia* has leaves with serrated margins, white petals each with 2 yellow spots, and strongly clavate stamen filaments, while *M. pensylvanica* has leaves with entire to crenate margins, greenish petals with no spots, and filiform stamen filaments. These species also differ in habitat and distribution. *Micranthes micranthidifolia* is commonly found growing along streams, brooks, and branches in the Southern Appalachians. However, *M. pensylvanica* is only found growing in swamps and bogs and is common in the northeastern U.S., but extremely rare

in the Southern Appalachians. This species is only known to occur in two counties of the Southern Appalachians: Watauga, North Carolina and Johnson, Tennessee.

Micranthes petiolaris is the only Southern Appalachian species placed in section *Stellares* along with four other North American species, *M. stellaris*, *M. ferruginea*, *M. bryophora*, and *M. foliolosa*, as well as seven species found in Europe and western Asia. This placement is based on morphological characters which are unique among the Southern Appalachian species, but shared by all species of section *Stellares*, and is now supported by molecular data. Parsimony analyses of ITS and *trn*L-F sequences reveal that *M. petiolaris* forms a clade with *M. stellaris*, far removed from the other Southern Appalachian species. Similarly, parsimony analyses of *mat*K sequences reveal that *M. petiolaris* belongs to a clade with the other four North American species of section *Stellares*, supporting the hypothesis that this species belongs to a separate lineage from the other Southern Appalachian species of *Micranthes*.

It is clear based on molecular analyses of *mat*K sequences that the sectional groupings of *Micranthes* and *Stellares* established by Webb and Gornall (1989) are each monophyletic. Section *Rotundifoliatae* with the inclusion of *M. calycina* and the exclusion of *M. arguta* is also monophyletic and section *Cuneifoliatae* may not be distinct, but more sequences from this section are needed to adequately address this question. Webb and Gornall (1989) note the latter section is poorly studied and many species therein morphologically resemble those placed in either section *Micranthes* or section *Rotundifoliatae*. In addition, in the *mat*K phylogeny, the *Micranthes* and *Rotundifoliatae* clades are sister with 100% bootstrap support to the exclusion of the *Stellares* clade (100% BS). There is little support for the various subsections within

section *Micranthes*; *Aulaxis*, *Dermasea*, and *Micranthes*. Species belonging to each of these subsections appear in various places throughout the clade representing section *Micranthes*, and this clade is poorly resolved.

Biogeography

The pattern of disjunct distributions of many flowering plant genera between eastern Asia and eastern North America has been well documented (Graham 1972; Raven 1972; Raven and Axelrod 1974; Boufford and Spongberg 1983; Davidse 1983). This pattern is often referred to as the "Asa Gray disjunction" (Krutzch 1989) as a result of the floristic comparisons made by Gray between eastern North America, western North America, Japan, and Europe (Gray 1840; 1846; 1856, 1857; 1859; 1878). A similar disjunct distribution has also been established in fungi (Hongo and Yokoyama 1978; Wu and Mueller 1997), arachnids (Suzuki et al. 1977), millipedes (Enghoff 1993), insects (Nordlander et al. 1996), and freshwater fishes (Patterson 1981). More recent studies of disjunct distributions of flowering plant genera in eastern North America, western North America, and eastern Asia (Wen et al. 1996; Wen et al. 1998; Xiang et al. 1998) reveal a closer relationship between the flora of eastern North America and western North America, than between eastern North America and eastern Asia (Wen 1999). Causes of this pattern are hypothesized to be vicariance (Gould and Donoghue 1998; Li et al. 1998), long-distance dispersal (Iltis 1983), migration through the Bering (Hopkins 1967) and North Atlantic land bridges (McKenna 1983a, 1983b; Tiffney 1985b), extinction and speciation (Wen 1999). This latter pattern of disjunction is also seen in the flowering

plant genus *Micranthes*, which is represented by species in the mountains of Europe, eastern Asia, the mountains of western North America, and the mountains and piedmont of eastern North America. While not all relationships can be fully addressed here due to lack of sequence data, the similarities between western North American and eastern North American species of *Micranthes*, as well as some eastern Asian and circumpolar species, can be discussed and compared.

There are in fact multiple eastern North American and western North American disjunct distributions within groups of related species of *Micranthes*, revealing that this pattern has occurred several times in various lineages within this genus and that phylogenetic relationships do not always correlate to geographic distributions. Phylogenetic analyses of *mat*K sequences reveal that the *Stellares* clade includes both eastern and western North American, as well as two circumpolar species. These species include: *M. stellaris* with a circumpolar distribution in sub-arctic eastern Canada and western Europe; *M. foliolosa* with a circumpolar distribution in arctic Canada, the Rocky Mountains, and western Europe; *M. ferruginea* with a western North American distribution from Alaska to northwestern California and also from the Rocky Mountains to northern Idaho and western Montana; *M. bryophora* endemic to the Klamath, Cascades, and Sierra Nevada ranges of northern California; and *M. petiolaris* endemic to the Southern Appalachians.

Analyses of *mat*K sequences further reveal that the *Micranthes* clade also includes eastern and western North American, as well as two circumpolar species. These species include: *M. hieracifolia* with a circumpolar distribution, *M. nivalis* and *M. tenuis* with arctic distributions in Canada and Europe; *M. careyana* and *M. micranthidifolia*

endemic to the Southern Appalachians; *M. pensylvanica* and *M. virginiensis* with widespread distributions in the northeastern United States; *M. texana* from southwestern Missouri to northern Texas; and *M. aprica*, *M. arguta*, *M. californica*, *M. howellii*, *M. integrifolia*, *M. nidifica*, *M. occidentalis*, *M. odontoloma*, *M. oregana*, *M. reflexa*, *M. rhomboidea*, and *M. rufidula* of various distributions in the mountains of western North America.

Finally, phylogenetic analyses of matK sequences reveal that the *Rotundifoliatae* clade includes species from both eastern Asia and western North America, but no extant species from eastern North America, a slightly different biogeographic pattern from the other two clades. These species include: *M. fusca* of Japan in eastern Asia; *M. calycina*, *M. punctata*, and *M. spicata* of the mountains of Alaska, the Yukon, and eastern Siberia; and *M. lyallii* from Alaska to Washington. No extant species within this clade exist in the Southern Appalachians.

Four species belonging to section *Stellares* in western North America and one in the Southern Appalachians of eastern North America are more closely related to each other than species in section *Micranthes* of the same geographical area. Similarly, all of the eastern and western North American species of section *Micranthes* are more closely related to each other than to species of sections *Stellares* or *Rotundifoliatae* in the same geographic area. Although there are no species of section *Rotundifoliatae* in eastern North America, the western species of this group are more closely related to each other than to species in either of the other two clades in western North America. Therefore, the Southern Appalachian species are not all each other's closest relatives and several groups

are more closely related to species found in western North America, eastern Asia, and Europe.

The pattern of disjunction among multiple genera of flowering plants among the temperate areas of the northern hemisphere can be explained by the widespread circumboreal distribution of temperate forests during the mid-Tertiary and the climatic cooling of the late Tertiary-Quaternary, which caused a decline of temperate flora in western North America and western Europe (Graham 1993; Manchester 1999). The presence of multiple disjunct lineages within the genus *Micranthes* is consistent with the hypothesis that these patterns of disjunction most likely occurred several times through different pathways (Tiffney 1985a), which has been supported by recent phylogenetic studies (reviewed in Wen 1999). The five major periods proposed by Tiffney (1985a) that floristic exchanges most likely occurred between eastern Asia and eastern North America include the Pre-Tertiary, the Early Eocene, the Late Eocene-Oligocene, the Miocene, and the Late Tertiary-Quaternary periods. It is probable that the family Saxifragaceae evolved by the mid-Miocene (approximately 12-16 million years ago) based on fossil evidence of fruits and seeds (Tiffney 1985a). The temperate flora that was continuous across the land-masses of the northern hemisphere became fragmented in the late-Miocene (approximately 5-7 million years ago) due to climatic changes, vulcanism, and mountain-building (Tiffney 1985a,b). Several other genera in the tribe Saxifrageae including *Boykinia*, *Mitella*, *Tiarella* display similar disjunctions in eastern and western North America and likely evolved in the late-Miocene (Web and Gornall 1989).

It is possible that the various sections of *Micranthes* evolved prior to the widespread distribution of the temperate forests in the northern hemisphere in the mid-Tertiary, because representatives of each of the sections show disjunct distributions across various continents and all sections have at least one arctic species. Species of section *Stellares* are found in eastern and western North America and Europe, with the majority of species in arctic Canada and Europe, implying a North Atlantic center of origin. Species of section *Micranthes* can be found in both eastern and western North America, eastern Asia, and Europe. Species of section *Rotundifoliatae* can only be found in western North America and eastern Asia. The majority of species in these clades are found on both sides of the Northern Pacific Rim in eastern Asia and western North America, implying a Beringian center of origin. Therefore, it is likely that the genus *Micranthes* evolved sometime in the mid-Miocene, shortly after the evolution of the family Saxifragaceae, and later diversified into at least three distinct lineages.

The most ancestral lineage, *Stellares*, probably originated in the North Atlantic and spread eastward into Europe and westward into North America. The sister lineages, *Micranthes* and *Rotundifoliatae*, probably originated in Beringia and later diversified into the respective lineages. Species of the *Rotundifoliatae* seemingly remained in the higher latitudes of eastern Asia and western North America, while species of the *Micranthes* lineage spread southward to lower latitudes and eastward to eastern North America. The later fragmentation of the temperate forests in the late-Miocene could explain the disjunct distribution of section *Micranthes* in eastern and western North America. As the climate began to significantly cool in the Pliocene, resulting in the glaciations of the Pleistocene, many arctic plant species were pushed southward and remained in high-elevation refugia

when the climate began to warm again. This could explain the disjunct distribution of the mostly arctic section *Stellares* in eastern and western North America, with *M. petiolaris* in the higher elevations of the Southern Appalachians, *M. bryophora* in the higher elevations of the Klamath, Cascades, and Sierra Nevada ranges, and *M. ferruginea* of the higher elevations of the Cascades and Rockies.

Taxonomic and conservation status of Southern Appalachian species

Micranthes careyana (Gray) Small (Carey saxifrage) is currently designated as a Watch List species in North Carolina, Rare in South Carolina, Rare in Virginia, and should remain at the species status in order to ensure its long-term protection and viability. *Micranthes caroliniana* (Gray) Small (Carolina saxifrage) is currently designated as a U.S. Species of Concern, Significantly Rare in North Carolina, Endangered in Tennessee, a Watch List species in Virginia, and should remain the species status in order to conserve populations of this species. Results of this study indicate this species may be even more rare than previously known, due to the large number of misidentifications discovered. All possible measures should be taken to conserve and avoid impact to populations of this globally vulnerable species.

Micranthes virginiensis (Michaux) Small (early saxifrage) is known to be a complex species with a widespread distribution in eastern North America, with a few morphological varieties. Results of this study indicate a new variety of this species should be recognized, presently only known to Polk County, North Carolina, based on unique floral characters and molecular differences.

Micranthes micranthidifolia (Haworth) Small (branch-lettuce) is common throughout its range in the Central and Southern Appalachians. There are no known threats to the viability of this species, but it has been historically harvested and eaten as greens by Native Americans and European settlers. *Micranthes pensylvanica* (L.) Small (swamp saxifrage) is common in the northeastern United States, but extremely rare in the Southern Appalachians, restricted to mountain bogs or swamps. It is currently designated as Significantly Rare in North Carolina and Endangered in Tennessee and should remain at this status in order to conserve populations of this species in the Southern Appalachians.

Micranthes petiolaris (Raf.) Brouillet and Gornall (mountain saxifrage) clearly belongs to a distinct evolutionary lineage than the other five Southern Appalachian species, based on both morphological and molecular analyses. Therefore, the placement of this species in the genus *Hydatica* by Small and Rydberg (1905), along with the eleven other species in section *Stellares*, seems warranted. Future taxonomic treatments in North America and Europe should recognize the following twelve species formerly placed in *Hydatica*: *H. brunoniana* (Bong.) Small, *H. bryophora* Small, *H. cuneifolia* Raf., *H. ferruginea* (Graham) Small, *H. foliolosa* (Raf.) Small, *H. geum* Raf., *H. hirsuta* Raf., *H. newcombei* Small, *H. petiolaris* (Raf.) Small, *H. stellaris* Gray, *H. umbrosa* Raf., *H. vreelandii* Small. Placement of these species in *Hydatica* would also require recognizing the sister species to this group, *M. tolmiei*, in the genus *Leptasea* Haworth as *L. tolmiei* Small.

CONCLUSIONS

Distribution of M. careyana and M. caroliniana

Data from morphological analyses of floral characters in the field and from herbarium specimens, molecular analyses, and previously published distributions (Lord 1961) indicate that the taxon *M. careyana* can be found in most of the counties in the Blue Ridge Physiographic Province including: Avery, Buncombe, Burke, Graham, Haywood, Henderson, Jackson, Macon, Madison McDowell, Mitchell, Polk, Rutherford, Transylvania, Watauga, and Yancey in North Carolina; Anderson, Bledsoe, Blount, Carter, Cocke, Franklin, Grainger, Greene, Grundy, Hamilton, Hancock, Knox, Marion, Monroe, Polk, Rhea, Roane, Sevier, Sullivan, Unicoi, Union, Van Buren, and Washington in Tennessee (Figure 12). A population of *M. careyana* has been reported in Scott County, Virginia (J. Townsend, pers. com.) based on the presence of an erect sepal orientation and a filiform filament shape, but this has not been confirmed by this study. The taxon *M. caroliniana* is restricted to just seven counties of the Blue Ridge Physiographic Province and does not overlap with *M. careyana*: Ashe, Alleghany and Watauga in North Carolina; Johnson in Tennessee; and Russell, Smyth and Washington in Virginia (Figure 13). Corrections or additions have not been made for the other four Southern Appalachian species from the distributions determined by Lord (1961).

Figure 12. Geographical distribution of *Micranthes careyana*.

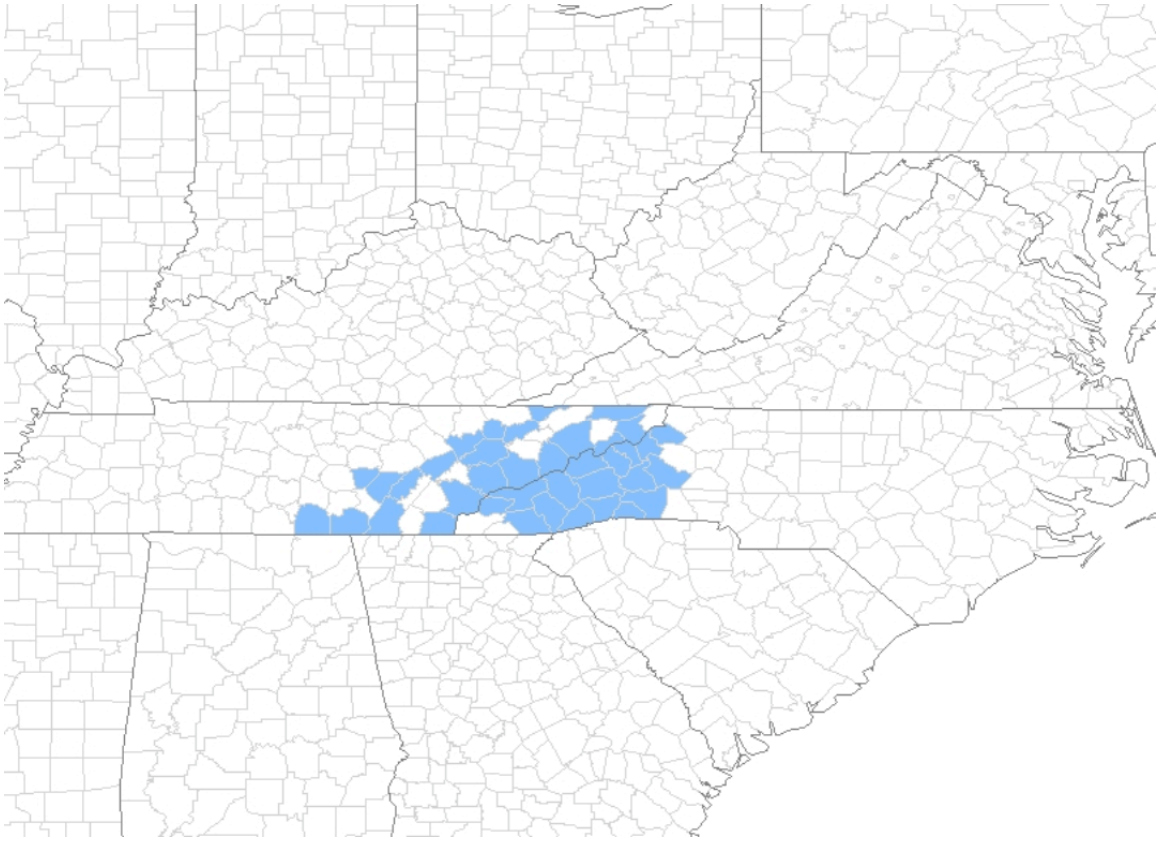
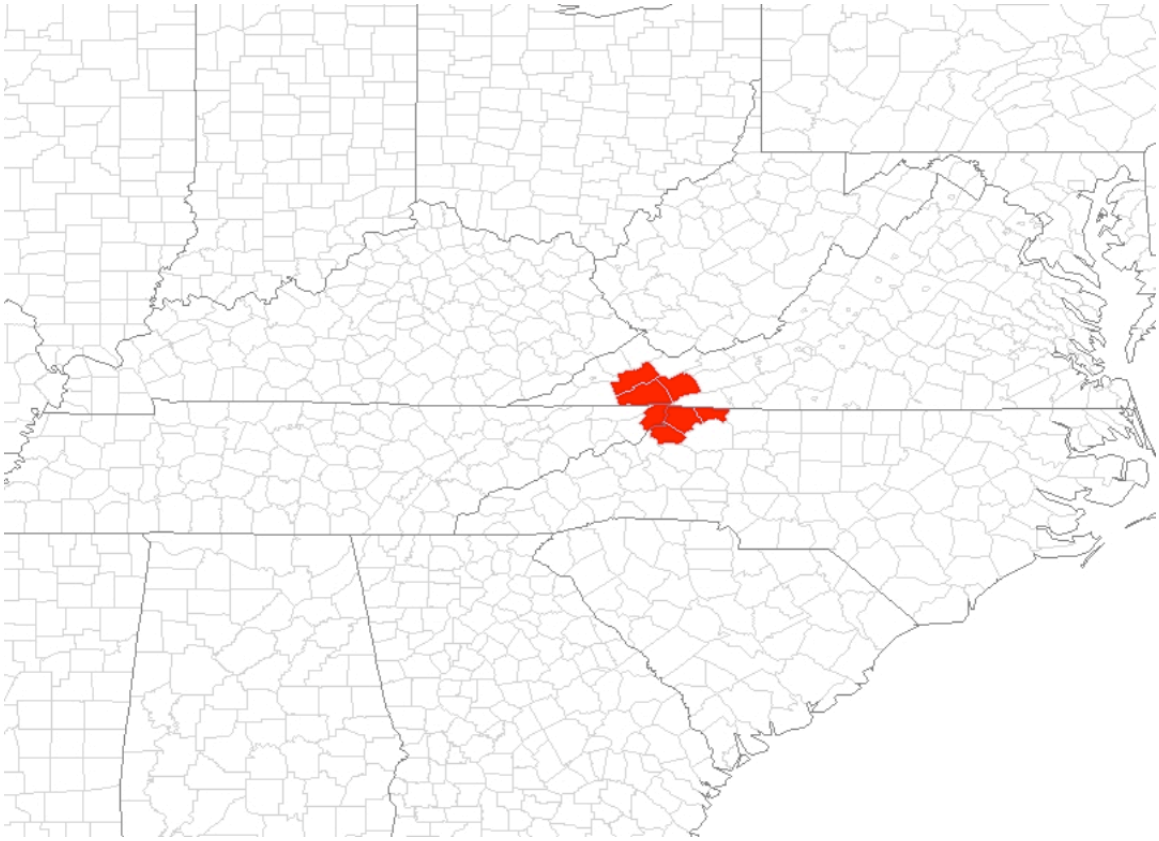


Figure 13. Geographical distribution of *Micranthes caroliniana*.



Field identification key

The following key to the species of *Saxifraga* sensu lato in the Southern Appalachians (Figure 14) has been constructed by utilizing field observations, examination of herbarium specimens, and previous treatments of the genus by Gray (1841; 1846), Small (1905; 1933), Fernald (1950), Lord (1961), Radford, Ahles and Bell (1968), Wofford (1989), and Weakley (2008):

Figure 14. Key to the species of *Saxifraga* sensu lato in the Southern Appalachians.

1 Corrolla zygomorphic; leaf margins coarsely dentate..... ***Hydatica***
1 Corolla actinomorphic; leaf margins entire to serrate .....***Micranthes***

Hydatica Necker 1790 MOUNTAIN-SAXIFRAGE

H. petiolaris (Rafinesque) Small MICHAUX'S SAXIFRAGE. Exposed, wet rock outcrops, cliffs, and overhangs: GA, NC, SC, TN, VA. Common in mountains.
A Central and Southern Appalachian endemic. [syn. *Saxifraga michauxii*]

Micranthes Haworth 1812 SMALLFLOWER

1 Leaf blades oblanceolate, 4-10x as long as wide; petioles narrowing slightly into a psuedopetiole 2
  2 Leaf margins entire to crenate; petals greenish, lacking spots; filaments distinctly filiform.....***M. pensylvanica***
  2 Leaf margins coarsely serrate; petals white, each with 2 yellow spots; filaments strongly clavate.....***M. micranthidifolia***
1 Leaf blades ovate or obovate, about as long as wide; petioles narrowing abrubtly into a distinctly winged petiole 3
  3 Petals fused into a slight hypanthium, not spotted, 4-5.5 mm long; filaments subulate, 1-1.5 mm long; ovary partly inferior .....***M. virginiensis***
  3 Petals free, clawed and each with 2 yellow spots, 3-3.5 mm long; filaments 3.5 mm long; ovary superior 4
    4 Sepals erect to spreading at anthesis; filaments distinctly filiform (use 10x)..... ***M. careyana***
    4 Sepals fully reflexed at anthesis; filaments slightly clavate (use 10x).....***M. caroliniana***

M. careyana (Gray) Small CAREY SAXIFRAGE. Dripping cliffs, overhangs, and seeps: GA, NC, SC, TN. Rare (GA Special Concern; NC Watch List; SC Rare). A Southern Appalachian endemic. [syn. *Saxifraga careyana*]

M. caroliniana (Gray) Small CAROLINA SAXIFRAGE. Dripping cliffs, overhangs, and seeps: NC, TN, VA. Very Rare (NC Rare; TN Endangered, VA Watch List). A Southern Appalachian endemic. [syn. *Saxifraga caroliniana*]

M. micranthidifolia (Haworth) Small BRANCH-LETTUCE. Seeps, springs, and branches: GA, NC, SC, TN, VA. Common throughout. A Central and Southern Appalachian endemic. [syn. *Saxifraga micranthidifolia*]

M. pensylvanica (Linnaeus) Haworth SWAMP SAXIFRAGE. Swamps and bogs: NC, TN, VA. Very Rare (NC Rare; TN Endangered). [syn. *Saxifraga pensylvanica*]

M. virginiensis (Michaux) Small EARLY SAXIFRAGE. Wet cliffs and limestone bluffs: GA, NC, SC, TN, VA. Uncommon in mountains. [syn. *Saxifraga virginiensis*]

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APPENDICES

APPENDIX A: HERBARIUM SPECIMENS EXAMINED

Micranthes careyana (Gray) Small (Carey saxifrage)

NORTH CAROLINA

Avery County: *W. W. Ashe*, 29 April 1893 [NCU 71796]

Buncombe County: *A.E. Radford, C.E. Wood, & B.M. Taylor 6947*, 03 May 1953 [NCSC 45959]; *A. E. Radford et al. 6947*, 03 May 1953 [NCU 56690]

Graham County: *A. E. Radford 11893*, 29 May 1956 [NCU 129601]; *Dan Pittillo s.n.,* 29 March 1981 [NCU 392286]; *R. Johnson 19*,20 April 1985 [WCUH 19773]; *K. Lathrop 26*, 03 April 1993 [WCUH 22484]; *D. Pittillo 10148*, 22 April 1989 [WCUH 21155]

Haywood County: *D. Pittillo & B. Dellinger 10247*, 15 May 1989 [WCUH 21237]; *D. Pittillo & S. McCall 16440*, 15 May 1974 [WCUH 16440]

Henderson County: *D. Pittillo 7013*, May 1956 [NCU 129583]; *A. E. Radford & J. G. Haesloop 7116*, 05 June 1953 [NCU 56598]

Jackson County: *D. Pittillo & S. Hagar 10719*, 01 Dec 1990 [WCUH 21470]

Macon County: *J. F. Mathews et. al*, 29 April 1967 [NCU 421626]; *D. Pittillo 7529*, 12 June 1977 [WCUH 17677]

Madison County: *D. Sather 994*, 18 April 1980 [NCU 508454]; *D. Sather 1264*, 04 June 1981 [NCU 516521]; *H. E. Ahles w/ J. A. Duke 38922*, 26 April 1958 [NCU 129604]

McDowell County: *T. Govus & D. Pittillo 112*, 08 June 197 [NCU 567920]

Mitchell County: *R. Brown 566515*, 22 June 2000 [NCU 566515]; 04 May [NCU 71743]; *J. W. Chickerberg Jr.*, 05 July 1880 [NCU 30126]

Polk County: *W. W. Ashe*, 23 April 1916 [NCU 71761]; *D. Pittillo 7652*, 27 July 1978 [WCUH 17846]

Rutherford County: *H. McIver 5*, 27 April 1981 [NCU 519873]; H. McIver 7, 28 April 1981 [NCU 519875]; *H. A. Ahles w/ C. R. Bell 11256*, 21 April 1956 [NCU 129607]; *D. Pittillo 6749*, 25 April 1975 [WCUH 15909]

Swain County: *A. E. Radford & J. G. Haesloop 7232*, 07 June 1953 [NCU 55199]; *T. Kiser III 159*, 18 April 1971 [WCUH 12377]; *D. Pittillo 2898*, 21 April 1968 [WCUH

8472]; *D. Pittillo 2898*, 21 April 1968 [WCUH 8590]; *D. Pittillo* 3582, 18 April 1971 [WCUH 9923]; *D. Pittillo 11789*, 14 April 1995 [WCUH 23623]

Watauga County: *K. I. Miller & I. W. Carpenter 1390*, 03 May 1968 [NCU 324563*]; H. A. Ahles w/ R. P. Ashworth 39514*, 04 May 1958 [NCU 129679]; *K. Miller & I. Carpenter 1390*, 03 May 1968 [WCUH 7268]

Yancey County: *D. McLeod 1187*, 28 April 1971 [NCU 480397]

TENNESSEE

Cocke County: *R. Dale Thomas et. al 22918*, 21 April 1971 [NCU 442040]

Franklin County: *D. W. Krickbaum*, 23 April 1959 [NCU 246308]

Grundy County: *R. C. Clark 1805*, 15 May 1965 [NCU 299414]

Knox County: April 1894 [NCU 571995]

Marion County: *R. C. Clark w/ L. Mays 1703a*, 14 May 1965 [NCU 299413]

Sevier County: *A.J. Sharp & H.H. Iltis 1725*, 17 May 1943 [NCSC 37173]; *L. Stewart*, 15 June 1936 [NCU 71801]; *A. Ruth*, May 1897 [NCU 71778]; *F. Bartley*, 28 April 1956 [NCU 137140]

Micranthes caroliniana (Gray) Small (Carolina saxifrage)

NORTH CAROLINA

Alleghany County: *A. E. Radford 32669*, 02 May 1958 [NCU 129566]

Ashe County: *G. L. Nesom 1015*, 04 May 1968 [NCSC 102089]; 07 June [NCU 71744]; *A. E. Radford 43954*, 23 June 1961 [NCU 215211]; *S. Spongberg 67-111*, 25 April 1967 [NCU 294919]; *D. B. Poindexter 05-325*, 25 May 2005 [NCU 582928]; *D. B. Poindexter 05-855*, 26 June 2005 [NCU 582927]; *A. E. Radford 45390*, 03 June 1967 [WCUH 6169]; *J. W. Hardin 13257*, 23 May 1968 [NCSC 72099]

VIRGINIA

Smyth County: *J.K. Small*, 13 June 1892 [NCSC 24478]; *N. L. & E. G. Britton & A. M. Vail*, June 1892 [NCU 574644]; *A. M. Harrill 18580*, 11 June 1968 [NCU 318031]

APPENDIX B: DNA SEQUENCES

i. Sequences used in ITS data set:

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[                                10        20        30]

M_Clawhammer_Mtn        GGGGTCGCGAA--TAGCGTCATTTCAACGT   [28]
M_Cliffridge            ...........A-.................   [29]
M_Crow_Creek            .........C.--.................   [28]
M_Cullasaja_Falls       ...........--.................   [28]
M_Elkhollow_Branch      ...........AA.................   [30]
M_Gap_Creek_Rd          ...........--.................   [28]
M_Gouges_Creek_Falls    ...........A-.................   [29]
M_Groto_Falls           .........T.--.............-...   [27]
M_Howard_Creek_Falls    ...........--.............-...   [27]
M_Ijams_sp_1            .........T.A-.................   [29]
M_Ijams_sp_2            ...........A-.................   [29]
M_Ivy_River             .........T.--.............-...   [27]
M_Linville_Gorge        -.C......T.--..........CA----C   [23]
M_Little_River_Gorge    C...GT...TTA-A....-....CA---..   [25]
M_Melrose               .........-.A-.................   [28]
M_Mt_Jefferson          ---------..--.............-...   [18]
M_Nantahala_Gorge_1     ...........A-.................   [29]
M_Nantahala_Gorge_2     ...........A-.............-...   [28]
M_New_River             .A.......T.--.............-...   [27]
M_Pigeon_River_Gorge    ...........--.................   [28]
M_Profile_Trail         ...........A-.................   [29]
M_Shady_Valley          ...........--.................   [28]
M_Slickrock_Trail       .....TCGCC.A-.............-...   [28]
M_Wadakoe_Mtn           ----------.A-.................   [19]
M_virginiensis          ...........A-.................   [29]
M_micranthidifolia_1    ...........A-........A........   [29]
M_micranthidifolia_2    ...........--........A........   [28]
M_pensylvanica          ----------..A-........A........   [20]
M_petiolaris_1          ----------.AA..A...ATGA.......   [20]
M_petiolaris_2          ----------.AA..A...ATGA.......   [20]
M_petiolaris_3          ----------.AA..A...ATGA.......   [20]
M_petiolaris_4          ----------AA..A...ATGA.......   [19]
M_integrifolia          ----------.AA.......T.........   [20]
M_punctata              ----------.AA........A...N....   [20]
M_stellaris             ----------.AA......ATGA.......   [20]
M_tolmiei               ----------.AA..........C......   [20]

[                                40        50        60]

M_Clawhammer_Mtn        TTGGGGATTAATTTGAAGC-ATATCTCAAG   [57]
M_Cliffridge            ...........C.GAG---A.A..A.....   [56]
M_Crow_Creek            ...........C.C.-...-....A.....   [56]
M_Cullasaja_Falls       ...........C.C.-...-....A.....   [56]
M_Elkhollow_Branch      ...........C.C.GG..A..........   [60]
M_Gap_Creek_Rd          ...........C.C..G---.......G..   [55]
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M_Gouges_Creek_Falls    ..........C.A..GA--.........   [57]
M_Groto_Falls           .A.........C.-.-...-........   [54]
M_Howard_Creek_Falls    G.........C.C..GC--.........A   [55]
M_Ijams_sp_1            .........-.AG-.GGA--GGG.AAA...   [55]
M_Ijams_sp_2            ..........GAAA.GA--..GGAGA...   [57]
M_Ivy_River             ........--.AGC..GC--...A.A....   [53]
M_Linville_Gorge        G......--------.-.--.G.CA..CG.   [43]
M_Little_River_Gorge    .G...A..C-.AG-.GC.--C.C.....G.   [51]
M_Melrose               ..........CAG....--...G--A...   [54]
M_Mt_Jefferson          G.........C.C..GC--.........   [46]
M_Nantahala_Gorge_1     ............TT...-....A.....   [58]
M_Nantahala_Gorge_2     ..........C.C.-...-....A.....   [56]
M_New_River             G.........C.C..GC--......T...   [55]
M_Pigeon_River_Gorge    ..........C.C...---.A.G......   [55]
M_Profile_Trail         .........-.AC...GA--.GGG.A..G.   [56]
M_Shady_Valley          G.........C.C..GC--.........   [56]
M_Slickrock_Trail       ..........C.C.-...-....A.T...   [56]
M_Wadakoe_Mtn           ..........C.A..GC--.........   [47]
M_virginiensis          ............C..GC--.........   [57]
M_micranthidifolia_1    G.........C.C..GC--.........   [57]
M_micranthidifolia_2    G..........--..GC--.........   [54]
M_pensylvanica          G.........C....GC--......T...   [48]
M_petiolaris_1          AA.........C.A..GT--..T.......   [48]
M_petiolaris_2          AA.........C....GT--..T.......   [48]
M_petiolaris_3          AA.........AA-..G.--..T.......   [47]
M_petiolaris_4          AA.........C....GT--..T.......   [47]
M_integrifolia          G.........C.C..G--C.........   [48]
M_punctata              G.........C....G--C....T.....   [48]
M_stellaris             AA.........C....G--T..T.......   [48]
M_tolmiei               G.AA..G..T.A....GAAC.......G.T   [50]

[                                70        80        90]

M_Clawhammer_Mtn        TGATAAACGTT-CGACTCGATTTGGTAAAA   [86]
M_Cliffridge            ..........-............C....   [85]
M_Crow_Creek            ..........-............C....   [85]
M_Cullasaja_Falls       ..........-............C....   [85]
M_Elkhollow_Branch      ........A..-............C....   [89]
M_Gap_Creek_Rd          ..........-............C....   [84]
M_Gouges_Creek_Falls    ..........-............C....   [86]
M_Groto_Falls           ..........-............C....   [83]
M_Howard_Creek_Falls    ..........-............C....   [84]
M_Ijams_sp_1            ........A..-..............-.   [83]
M_Ijams_sp_2            ..........-............CCC..   [86]
M_Ivy_River             ..........-..............   [82]
M_Linville_Gorge        A.......A..---CA.........C....   [70]
M_Little_River_Gorge    A......GTCGAT..T..TG...CAC.TC.   [81]
M_Melrose               ..........-............C....   [83]
M_Mt_Jefferson          ..........-............C....   [75]
M_Nantahala_Gorge_1     ..........-............C....   [87]
M_Nantahala_Gorge_2     ..........-............C....   [85]
M_New_River             ..........-............C....   [84]
M_Pigeon_River_Gorge    G.........-............C....   [84]
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M_Profile_Trail          ........A..-............C....   [85]
M_Shady_Valley           ..........-............C....   [85]
M_Slickrock_Trail        ..........-............C....   [85]
M_Wadakoe_Mtn            ..........-............C....   [76]
M_virginiensis           ..........-............C....   [86]
M_micranthidifolia_1     ...C.......-..................   [86]
M_micranthidifolia_2     ...C.......-............C....   [83]
M_pensylvanica           ...C.......-............C....   [77]
M_petiolaris_1           C..C....T.A-............C....   [77]
M_petiolaris_2           C..C....T.A-............C....   [77]
M_petiolaris_3           C..C....T.A-............C....   [76]
M_petiolaris_4           C..C....T.A-............C....   [76]
M_integrifolia           ..........-............C....   [77]
M_punctata               ...C.......-............C....   [77]
M_stellaris              C..C....T.A-............CG...   [77]
M_tolmiei                ...C....T---..........--------   [69]

[                                  100       110       120]

M_Clawhammer_Mtn         ACAAAAAAG-GCTTATAAACCACCACTTAT   [115]
M_Cliffridge             C......GA-.....................   [114]
M_Crow_Creek             C........-.....................   [114]
M_Cullasaja_Falls        C........-.....................   [114]
M_Elkhollow_Branch       C........-.....................   [118]
M_Gap_Creek_Rd           C........-......G..............   [113]
M_Gouges_Creek_Falls     C........-.....................   [115]
M_Groto_Falls            C........-.....................   [112]
M_Howard_Creek_Falls     C........-......G..............   [113]
M_Ijams_sp_1             .A.......-.....................   [112]
M_Ijams_sp_2             C........-.....................   [115]
M_Ivy_River              C........-.....................   [111]
M_Linville_Gorge         .A...G.G.T.G...................   [100]
M_Little_River_Gorge     GA.G.G...-......-..............   [109]
M_Melrose                C........-......G..............   [112]
M_Mt_Jefferson           C........-......G..............   [104]
M_Nantahala_Gorge_1      CA.......-.....................   [116]
M_Nantahala_Gorge_2      C........-.T...................   [114]
M_New_River              C........-......G..............   [113]
M_Pigeon_River_Gorge     C........-.....................   [113]
M_Profile_Trail          C........-...A.................   [114]
M_Shady_Valley           C........-......G..............   [114]
M_Slickrock_Trail        C........-.....................   [114]
M_Wadakoe_Mtn            C........-......G..............   [105]
M_virginiensis           C........-......G..............   [115]
M_micranthidifolia_1     ..C.....AG.....................   [116]
M_micranthidifolia_2     ..C.....AG.....................   [113]
M_pensylvanica           ..-.....AG.....................   [106]
M_petiolaris_1           ..C...G.AG.A....GT.............   [107]
M_petiolaris_2           ..C...G.AG.A....GT.............   [107]
M_petiolaris_3           ..C...G.AG.A....GT.............   [106]
M_petiolaris_4           ..C...G.AG.A....GT.............   [106]
M_integrifolia           C.....CT.-......G..............   [106]
M_punctata               C........-.....................   [106]
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M_stellaris             C....G...-......GT.............   [106]
M_tolmiei               --------..-.T...G.T.............   [91]

[                                130       140       150]

M_Clawhammer_Mtn        CGTGACGTTTCATCACTCG-TACTCCTTTT   [144]
M_Cliffridge            ....................-.........   [143]
M_Crow_Creek            ....................-.........   [143]
M_Cullasaja_Falls       ....................-.........   [143]
M_Elkhollow_Branch      ....................-.........   [147]
M_Gap_Creek_Rd          ....................-.....T....   [142]
M_Gouges_Creek_Falls    ....................-.........   [144]
M_Groto_Falls           ....................-.........   [141]
M_Howard_Creek_Falls    ....................-.....T....   [142]
M_Ijams_sp_1            ....................-.........   [141]
M_Ijams_sp_2            ....................-.........   [144]
M_Ivy_River             ....................-.........   [140]
M_Linville_Gorge        ....................-.........   [129]
M_Little_River_Gorge    ....................-.........   [138]
M_Melrose               ....................-.....T....   [141]
M_Mt_Jefferson          ....................-.....T....   [133]
M_Nantahala_Gorge_1     ....................-.........   [145]
M_Nantahala_Gorge_2     ....................-.........   [143]
M_New_River             ....................-.....T....   [142]
M_Pigeon_River_Gorge    ....................-.........   [142]
M_Profile_Trail         ....................-.........   [143]
M_Shady_Valley          ....................-.....T....   [143]
M_Slickrock_Trail       ....................-.........   [143]
M_Wadakoe_Mtn           ......A.............-.....T....   [134]
M_virginiensis          ....................-.....T....   [144]
M_micranthidifolia_1    ....................-.....T....   [145]
M_micranthidifolia_2    ....................-.....T....   [142]
M_pensylvanica          ....................-.....T....   [135]
M_petiolaris_1          ..................A.-.....A....   [136]
M_petiolaris_2          ..................A.-.....A....   [136]
M_petiolaris_3          ..................A.-.....A....   [135]
M_petiolaris_4          ..................A.-.....A....   [135]
M_integrifolia          ....................-.....T....   [135]
M_punctata              ....................-.....T....   [135]
M_stellaris             ..................A.-.....A....   [135]
M_tolmiei               ....................G.....TA...   [121]

[                                160       170       180]

M_Clawhammer_Mtn        TAGGTCAACCACA--CGT-AAGTGTATGGG   [171]
M_Cliffridge            .............--...-..........   [170]
M_Crow_Creek            .............--...-..........   [170]
M_Cullasaja_Falls       .............--...-..........   [170]
M_Elkhollow_Branch      .............--.A.-..........   [174]
M_Gap_Creek_Rd          .............--...-.T.G.......   [169]
M_Gouges_Creek_Falls    .............--.A.-..........   [171]
M_Groto_Falls           .............--...-..........   [168]
M_Howard_Creek_Falls    .............--...-...G.......   [169]
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M_Ijams_sp_1            ..........C..--.C.-...........   [168]
M_Ijams_sp_2            ............C--.A.-...........   [171]
M_Ivy_River             .............--.A.-...........   [167]
M_Linville_Gorge        .............--...-...........   [156]
M_Little_River_Gorge    .........TCTTATTC.-....AG.....   [167]
M_Melrose               .............--...-...G.......   [168]
M_Mt_Jefferson          .............--...-...G.......   [160]
M_Nantahala_Gorge_1     .............--...-...........   [172]
M_Nantahala_Gorge_2     .............--...-...........   [170]
M_New_River             .............--...-...G.......   [169]
M_Pigeon_River_Gorge    .............--.A.-....A......   [169]
M_Profile_Trail         .............--.A.-...........   [170]
M_Shady_Valley          .............--...-...G.......   [170]
M_Slickrock_Trail       .............--...-...........   [170]
M_Wadakoe_Mtn           C............--...-...G.......   [161]
M_virginiensis          .............--.C.-...G.......   [171]
M_micranthidifolia_1    .............--..C-...G.......   [172]
M_micranthidifolia_2    .............--..C-...G.......   [169]
M_pensylvanica          .............--.AC-...G.......   [162]
M_petiolaris_1          ....C.........--.AC-GT.G.C.....   [163]
M_petiolaris_2          ....C.........--.AC-GT.G.C.....   [163]
M_petiolaris_3          ....C.........--.AC-GT.G.C.....   [162]
M_petiolaris_4          ....C.........--.AC-GT.G.C.....   [162]
M_integrifolia          .............CGTA---..G.......   [162]
M_punctata              .............CG.AAGG..G.......   [165]
M_stellaris             ....C........CA.---GT.G.C.....   [162]
M_tolmiei               .G.N.G.......CAAA--TGA..C.C...   [149]

[                                 190       200       210]

M_Clawhammer_Mtn        AGGCCAATCTTTTCTCACCACAAATCAT--   [199]
M_Cliffridge            ............................--   [198]
M_Crow_Creek            ............................--   [198]
M_Cullasaja_Falls       ............................--   [198]
M_Elkhollow_Branch      ............................--   [202]
M_Gap_Creek_Rd          ........................T.--   [197]
M_Gouges_Creek_Falls    ............................--   [199]
M_Groto_Falls           ............................--   [196]
M_Howard_Creek_Falls    ........A.A.............T.--   [197]
M_Ijams_sp_1            ............................--   [196]
M_Ijams_sp_2            ............................--   [199]
M_Ivy_River             ............................--   [195]
M_Linville_Gorge        ............................--   [184]
M_Little_River_Gorge    ............................--   [195]
M_Melrose               ....................T...T.--   [196]
M_Mt_Jefferson          ........A.A.............T.--   [188]
M_Nantahala_Gorge_1     ............................--   [200]
M_Nantahala_Gorge_2     ............................--   [198]
M_New_River             ........A.A......T........T.--   [197]
M_Pigeon_River_Gorge    ............................--   [197]
M_Profile_Trail         ............................--   [198]
M_Shady_Valley          ........A.A.............T.--   [198]
M_Slickrock_Trail       ............................--   [198]
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M_Wadakoe_Mtn           ........................T.--   [189]
M_virginiensis          ........................TA--   [199]
M_micranthidifolia_1    ...........C...........TC.TA   [202]
M_micranthidifolia_2    ...........C...........TC.TA   [199]
M_pensylvanica          ................T.......T.--   [190]
M_petiolaris_1          .........A.CC...C..G....CGCA--   [191]
M_petiolaris_2          .........A.CC...C..G....CGCA--   [191]
M_petiolaris_3          .........A.CC...C..G....CGCA--   [190]
M_petiolaris_4          .........A.CC...C..G....CGCA--   [190]
M_integrifolia          ..............-..AC..ATC.TA   [191]
M_punctata              ...........C....T.......GCCTA   [195]
M_stellaris             .........A.CC...G..G....CGCAAA   [192]
M_tolmiei               ..A.....T........A..A...ATGATA   [179]

[                                220       230       240]

M_Clawhammer_Mtn        AAACCTAAATAGGAATGGTTT-GTGGGAGA   [228]
M_Cliffridge            ......................T........   [228]
M_Crow_Creek            ......................T........   [228]
M_Cullasaja_Falls       ......................T........   [228]
M_Elkhollow_Branch      .......................-........   [231]
M_Gap_Creek_Rd          ..T...G.G............-........   [226]
M_Gouges_Creek_Falls    .......................-........   [228]
M_Groto_Falls           ......................T........   [226]
M_Howard_Creek_Falls    ..-...G..............-........   [225]
M_Ijams_sp_1            .......................-........   [225]
M_Ijams_sp_2            .......................-........   [228]
M_Ivy_River             .......................-........   [224]
M_Linville_Gorge        ................A.G..-........   [213]
M_Little_River_Gorge    ...........A.........T........   [225]
M_Melrose               ......G..............-........   [225]
M_Mt_Jefferson          ..-...G..............-........   [216]
M_Nantahala_Gorge_1     ......................T........   [230]
M_Nantahala_Gorge_2     ......................T........   [228]
M_New_River             ..-...G..............-........   [225]
M_Pigeon_River_Gorge    .......................-........   [226]
M_Profile_Trail         .......................-........   [227]
M_Shady_Valley          ..-...G..............-........   [226]
M_Slickrock_Trail       ......................T........   [228]
M_Wadakoe_Mtn           ..-...G..A...........-....A...   [217]
M_virginiensis          ......G..............-........   [228]
M_micranthidifolia_1    .....A....G....A-.G..T........   [231]
M_micranthidifolia_2    .....A....G....A-.G..T........   [228]
M_pensylvanica          .....A....G....AA.G..T........   [220]
M_petiolaris_1          ..CA..TC..GT...A..A..-........   [220]
M_petiolaris_2          ..CA..TC..GT...A..A..-........   [220]
M_petiolaris_3          ..CA..TC..GT...A..A..-........   [219]
M_petiolaris_4          ..CA..TC..GT...A..A..-........   [219]
M_integrifolia          ..C.TA..TA.....A..G..-........   [220]
M_punctata              ..C.AA..-.G....A.....-........   [223]
M_stellaris             C.CTAC.--.GT...A..A..-........   [219]
M_tolmiei               T.T.A...GAGAT.CAA....-T.......   [208]
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[                                250       260       270]

M_Clawhammer_Mtn        AGTGTGTACGTGACGCCCAGGCAGACGTGC   [258]
M_Cliffridge            ............................     [258]
M_Crow_Creek            ............................     [258]
M_Cullasaja_Falls       ............................     [258]
M_Elkhollow_Branch      ............................     [261]
M_Gap_Creek_Rd          ............................     [256]
M_Gouges_Creek_Falls    ............................     [258]
M_Groto_Falls           ............................     [256]
M_Howard_Creek_Falls    ............................     [255]
M_Ijams_sp_1            ............................     [255]
M_Ijams_sp_2            ............................     [258]
M_Ivy_River             ............................     [254]
M_Linville_Gorge        ............................     [243]
M_Little_River_Gorge    ............................     [255]
M_Melrose               ............................     [255]
M_Mt_Jefferson          ............................     [246]
M_Nantahala_Gorge_1     ............................     [260]
M_Nantahala_Gorge_2     ............................     [258]
M_New_River             ............................     [255]
M_Pigeon_River_Gorge    ............................     [256]
M_Profile_Trail         ............................     [257]
M_Shady_Valley          ............................     [256]
M_Slickrock_Trail       ............................     [258]
M_Wadakoe_Mtn           ............................     [247]
M_virginiensis          ............................     [258]
M_micranthidifolia_1    ............................     [261]
M_micranthidifolia_2    ............................     [258]
M_pensylvanica          ............................     [250]
M_petiolaris_1          ............................     [250]
M_petiolaris_2          ............................     [250]
M_petiolaris_3          ............................     [249]
M_petiolaris_4          ............................     [249]
M_integrifolia          .......NNNNNNNNNNNNNNNNNNNNN     [250]
M_punctata              ....----T..NNNNNNNNNNNNNNNNNN     [249]
M_stellaris             ....----T..NNNNNNNNNNNNNNNNNN     [245]
M_tolmiei               ....G..NNNNNNNNNNNNNNNNNNNNN     [238]

[                                280       290       300]

M_Clawhammer_Mtn        CCTCAA-CCAGAGGCTTC-GGGCGCAACTT   [286]
M_Cliffridge            ......C..........-..........     [287]
M_Crow_Creek            ......-..........-..........     [286]
M_Cullasaja_Falls       ......-..........-..........     [286]
M_Elkhollow_Branch      ......C.-........-..........     [289]
M_Gap_Creek_Rd          ......-..........-..........     [284]
M_Gouges_Creek_Falls    ......-..........-..........     [286]
M_Groto_Falls           ......-..........-..........     [284]
M_Howard_Creek_Falls    ......-..........-..........     [283]
M_Ijams_sp_1            ......-..........-..........     [283]
M_Ijams_sp_2            ......-..........-..........     [286]
M_Ivy_River             ......-..........-..........     [282]
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M_Linville_Gorge        ......-.........-.........   [271]
M_Little_River_Gorge    ......A..........C.........   [285]
M_Melrose               ......-.........-.........   [283]
M_Mt_Jefferson          ......-.........-.........   [274]
M_Nantahala_Gorge_1     ......-.........-.........   [288]
M_Nantahala_Gorge_2     ......-.........-.........   [286]
M_New_River             ......-.........-.........   [283]
M_Pigeon_River_Gorge    ......-.........-.........   [284]
M_Profile_Trail         ......-.........-.........   [285]
M_Shady_Valley          ......-.........-.........   [284]
M_Slickrock_Trail       ......-.........-.........   [286]
M_Wadakoe_Mtn           ......-.........-.........   [275]
M_virginiensis          ......-.........-.........   [286]
M_micranthidifolia_1    ......-...A.....-.........   [289]
M_micranthidifolia_2    ......-...A.....-.........   [286]
M_pensylvanica          ......-.........-.........   [278]
M_petiolaris_1          ......C.A.........-A.......   [279]
M_petiolaris_2          ......C.A.........-A.......   [279]
M_petiolaris_3          ......C.A.........-A.......   [278]
M_petiolaris_4          ......C.A.........-A.......   [278]
M_integrifolia          NNNNNN--NNNNNNNNNN-NNNNNNNNNNN   [277]
M_punctata              NNNNNN--NNNNNNNNNN-NNNNNNNNNNN   [276]
M_stellaris             NNNNNN--NNNNNNNNNN-NNNNNNNNNNN   [272]
M_tolmiei               NNNNNN--NNNNNNNNNN-NNNNNNNNNNN   [265]

[                              310       320       330]

M_Clawhammer_Mtn        GCGTTCAAAAACTCGATGGTTCACGGGATT   [316]
M_Cliffridge            ....-........................   [316]
M_Crow_Creek            ............................   [316]
M_Cullasaja_Falls       ............................   [316]
M_Elkhollow_Branch      ............................   [319]
M_Gap_Creek_Rd          ............................   [314]
M_Gouges_Creek_Falls    ..........-.................   [315]
M_Groto_Falls           ............................   [314]
M_Howard_Creek_Falls    ............................   [313]
M_Ijams_sp_1            ............................   [313]
M_Ijams_sp_2            ............................   [316]
M_Ivy_River             ............................   [312]
M_Linville_Gorge        ............................   [301]
M_Little_River_Gorge    ............................   [315]
M_Melrose               ............................   [313]
M_Mt_Jefferson          ............................   [304]
M_Nantahala_Gorge_1     ............................   [318]
M_Nantahala_Gorge_2     ............................   [316]
M_New_River             ............................   [313]
M_Pigeon_River_Gorge    ............................   [314]
M_Profile_Trail         ............................   [315]
M_Shady_Valley          ............................   [314]
M_Slickrock_Trail       ............................   [316]
M_Wadakoe_Mtn           ............................   [305]
M_virginiensis          ............................   [316]
M_micranthidifolia_1    ............................   [319]
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M_micranthidifolia_2    ............................   [316]
M_pensylvanica          ............................   [308]
M_petiolaris_1          ............................   [309]
M_petiolaris_2          ............................   [309]
M_petiolaris_3          ............................   [308]
M_petiolaris_4          ............................   [308]
M_integrifolia          NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [307]
M_punctata              NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [306]
M_stellaris             NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [302]
M_tolmiei               NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [295]

[                              340       350       360]

M_Clawhammer_Mtn        CTGCAATTCACACCAAGTATCGCATTTCGC   [346]
M_Cliffridge            ............................   [346]
M_Crow_Creek            ............................   [346]
M_Cullasaja_Falls       ............................   [346]
M_Elkhollow_Branch      ............................   [349]
M_Gap_Creek_Rd          ............................   [344]
M_Gouges_Creek_Falls    ............................   [345]
M_Groto_Falls           ............................   [344]
M_Howard_Creek_Falls    ............................   [343]
M_Ijams_sp_1            ............................   [343]
M_Ijams_sp_2            ............................   [346]
M_Ivy_River             ............................   [342]
M_Linville_Gorge        ............................   [331]
M_Little_River_Gorge    ............................   [345]
M_Melrose               ............................   [343]
M_Mt_Jefferson          ............................   [334]
M_Nantahala_Gorge_1     ............................   [348]
M_Nantahala_Gorge_2     ............................   [346]
M_New_River             ............................   [343]
M_Pigeon_River_Gorge    ............................   [344]
M_Profile_Trail         ............................   [345]
M_Shady_Valley          ............................   [344]
M_Slickrock_Trail       ............................   [346]
M_Wadakoe_Mtn           ............................   [335]
M_virginiensis          T...........................   [346]
M_micranthidifolia_1    ............................   [349]
M_micranthidifolia_2    ............................   [346]
M_pensylvanica          ............................   [338]
M_petiolaris_1          ............................   [339]
M_petiolaris_2          ............................   [339]
M_petiolaris_3          ............................   [338]
M_petiolaris_4          ............................   [338]
M_integrifolia          NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [337]
M_punctata              NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [336]
M_stellaris             NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [332]
M_tolmiei               NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [325]

[                              370       380       390]

M_Clawhammer_Mtn        TACGTTCTTCATCGATGCGAGAGCCGAGAT   [376]
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M_Cliffridge              ............................   [376]
M_Crow_Creek              ............................   [376]
M_Cullasaja_Falls         ............................   [376]
M_Elkhollow_Branch        ............................   [379]
M_Gap_Creek_Rd            ............................   [374]
M_Gouges_Creek_Falls      ............................   [375]
M_Groto_Falls             ............................   [374]
M_Howard_Creek_Falls      ............................   [373]
M_Ijams_sp_1              ............................   [373]
M_Ijams_sp_2              ............................   [376]
M_Ivy_River               ............................   [372]
M_Linville_Gorge          ............................   [361]
M_Little_River_Gorge      ............................   [375]
M_Melrose                 ............................   [373]
M_Mt_Jefferson            ............................   [364]
M_Nantahala_Gorge_1       ............................   [378]
M_Nantahala_Gorge_2       ............................   [376]
M_New_River               ............................   [373]
M_Pigeon_River_Gorge      ............................   [374]
M_Profile_Trail           ............................   [375]
M_Shady_Valley            ............................   [374]
M_Slickrock_Trail         ............................   [376]
M_Wadakoe_Mtn             ............................   [365]
M_virginiensis            ............................   [376]
M_micranthidifolia_1      ............................   [379]
M_micranthidifolia_2      ............................   [376]
M_pensylvanica            ............................   [368]
M_petiolaris_1            ............................   [369]
M_petiolaris_2            ............................   [369]
M_petiolaris_3            ............................   [368]
M_petiolaris_4            ............................   [368]
M_integrifolia            NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [367]
M_punctata                NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [366]
M_stellaris               NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [362]
M_tolmiei                 NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [355]

[                                  400       410       420]

M_Clawhammer_Mtn          ATCCGTTGCCGAG-AGTCATTAAAGACATA   [405]
M_Cliffridge              ............-.............   [405]
M_Crow_Creek              ............-.............   [405]
M_Cullasaja_Falls         ............-.............   [405]
M_Elkhollow_Branch        ............-.............   [408]
M_Gap_Creek_Rd            ............-.............   [403]
M_Gouges_Creek_Falls      ............-.............   [404]
M_Groto_Falls             ......G--....-.............   [401]
M_Howard_Creek_Falls      ............-.............   [402]
M_Ijams_sp_1              ............-.............   [402]
M_Ijams_sp_2              ............-.............   [405]
M_Ivy_River               ............G.............   [402]
M_Linville_Gorge          ............-.............   [390]
M_Little_River_Gorge      ............-.............   [404]
M_Melrose                 ............-.............   [402]
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M_Mt_Jefferson          ............-.............   [393]
M_Nantahala_Gorge_1     ............-.............   [407]
M_Nantahala_Gorge_2     ............-.............   [405]
M_New_River             ............-.............   [402]
M_Pigeon_River_Gorge    ............-.............   [403]
M_Profile_Trail         ............-.............   [404]
M_Shady_Valley          ............-.............   [403]
M_Slickrock_Trail       ............-.............   [405]
M_Wadakoe_Mtn           ............-.............   [394]
M_virginiensis          ............-.............   [405]
M_micranthidifolia_1    ............-.............   [408]
M_micranthidifolia_2    ............-.............   [405]
M_pensylvanica          ............-.............   [397]
M_petiolaris_1          ............-............C   [398]
M_petiolaris_2          ............-............C   [398]
M_petiolaris_3          ............-............C   [397]
M_petiolaris_4          ............-............C   [397]
M_integrifolia          NNNNNNNNNNNNN-NNNNNNNNN.......   [396]
M_punctata              NNNNNNNNNNNNN-NNNNNNNNNNNNN...   [395]
M_stellaris             NNNNNNNNNNNNN-NNNNNNNNNNNNN..C   [391]
M_tolmiei               NNNNNNNNNNNNN-NNNNNNNNN......C   [384]

[                                430       440       450]

M_Clawhammer_Mtn        AAGAAGATAACATTTTTTTCATTTTATGCA   [435]
M_Cliffridge            .............................   [435]
M_Crow_Creek            .............................   [435]
M_Cullasaja_Falls       .............................   [435]
M_Elkhollow_Branch      ..A..........................   [438]
M_Gap_Creek_Rd          ................-T..........   [432]
M_Gouges_Creek_Falls    .............................   [434]
M_Groto_Falls           ....-........................   [430]
M_Howard_Creek_Falls    ............A.....-T..........   [431]
M_Ijams_sp_1            .............................   [432]
M_Ijams_sp_2            .............................   [435]
M_Ivy_River             .............................   [432]
M_Linville_Gorge        .............................   [420]
M_Little_River_Gorge    .............................   [434]
M_Melrose               ............G....-..........   [431]
M_Mt_Jefferson          ............A.....-T..........   [422]
M_Nantahala_Gorge_1     .............................   [437]
M_Nantahala_Gorge_2     .............................   [435]
M_New_River             ............A.....-T..........   [431]
M_Pigeon_River_Gorge    .............................   [433]
M_Profile_Trail         .............................   [434]
M_Shady_Valley          ............A.....-T..........   [432]
M_Slickrock_Trail       .............................   [435]
M_Wadakoe_Mtn           ................-...........   [423]
M_virginiensis          ................-.....-.....   [433]
M_micranthidifolia_1    ..............C-..A........   [437]
M_micranthidifolia_2    ..............C-..A........   [434]
M_pensylvanica          .............CC-..A.C......   [426]
M_petiolaris_1          T............C.C.C..C...G....   [428]
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M_petiolaris_2          T............C.C.C..C...G....   [428]
M_petiolaris_3          T............C.C.C..C...G....   [427]
M_petiolaris_4          T............C.C.C..C...G....   [427]
M_integrifolia          ......-.........--C.G.........   [423]
M_punctata              ..T.....T.......-.C..A..A.....   [424]
M_stellaris             T............C.C.C..C...G....   [421]
M_tolmiei               ....G...G..C..CC..CAC..GN.....   [414]

[                                460       470       480]

M_Clawhammer_Mtn        AAATAACATAAAATGAAA--AATGCTCT-T   [462]
M_Cliffridge            ..................--........-.   [462]
M_Crow_Creek            ..................--........-.   [462]
M_Cullasaja_Falls       ..................--........-.   [462]
M_Elkhollow_Branch      ..................--........-.   [465]
M_Gap_Creek_Rd          .--..............G-........-.   [458]
M_Gouges_Creek_Falls    ..................--........-.   [461]
M_Groto_Falls           ..................--........-.   [457]
M_Howard_Creek_Falls    ..-...........A...--........-.   [457]
M_Ijams_sp_1            ..................--........-.   [459]
M_Ijams_sp_2            ..................--........-.   [462]
M_Ivy_River             ..................--........-.   [459]
M_Linville_Gorge        ..................--........-.   [447]
M_Little_River_Gorge    ..................--........-.   [461]
M_Melrose               ..-...............--........-.   [457]
M_Mt_Jefferson          ..-...........A...--........-.   [448]
M_Nantahala_Gorge_1     ..................--........-.   [464]
M_Nantahala_Gorge_2     ..................--........-.   [462]
M_New_River             ..-...........A...--........-.   [457]
M_Pigeon_River_Gorge    ..................--........-.   [460]
M_Profile_Trail         ..................--........-.   [461]
M_Shady_Valley          ..-...........A...--........-.   [458]
M_Slickrock_Trail       ..................--........-.   [462]
M_Wadakoe_Mtn           ..-...........C.....--........-.   [449]
M_virginiensis          ..-...............--........-.   [459]
M_micranthidifolia_1    T.T--.............A-.....A..-.   [463]
M_micranthidifolia_2    T.T--.........R...A-.....A..-.   [460]
M_pensylvanica          T.T..............A-........-.   [454]
M_petiolaris_1          ---.GG...G..G..G.GAA.G...C..C.   [455]
M_petiolaris_2          ---.GG...G..G..G.GAA.G...C..C.   [455]
M_petiolaris_3          ---.GG...G..G..G.GAA.G...C..C.   [454]
M_petiolaris_4          ---.GG...G..G..G.GAA.G...C..C.   [454]
M_integrifolia          T.T............N..--.....A.-C.   [450]
M_punctata              T.CAC.T-G...TG..G.--........T.   [451]
M_stellaris             TG-GC.T-G..GTG..G.--........C.   [447]
M_tolmiei               C..A....GG..TA..GG--N.-.T..AA.   [441]

[                                490       500       510]

M_Clawhammer_Mtn        TTTTAAAAAAAATTCCTTGGCGCAATTCAC   [492]
M_Cliffridge            .............................   [492]
M_Crow_Creek            .............................   [492]
M_Cullasaja_Falls       .............................   [492]
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M_Elkhollow_Branch      ............................   [495]
M_Gap_Creek_Rd          ....T....--...........GN.....   [486]
M_Gouges_Creek_Falls    ............................   [491]
M_Groto_Falls           ............................   [487]
M_Howard_Creek_Falls    ....T....--.................   [485]
M_Ijams_sp_1            ............................   [489]
M_Ijams_sp_2            ............................   [492]
M_Ivy_River             ............................   [489]
M_Linville_Gorge        ............................   [477]
M_Little_River_Gorge    ............................   [491]
M_Melrose               ....T....--.................   [485]
M_Mt_Jefferson          ....T....--.................   [476]
M_Nantahala_Gorge_1     ............................   [494]
M_Nantahala_Gorge_2     ............................   [492]
M_New_River             .........--.................   [485]
M_Pigeon_River_Gorge    ............................   [490]
M_Profile_Trail         ............................   [491]
M_Shady_Valley          ....TT...--.................   [486]
M_Slickrock_Trail       ............................   [492]
M_Wadakoe_Mtn           ....T....--.................   [477]
M_virginiensis          ....T....--.................   [487]
M_micranthidifolia_1    A...T....T-.................   [492]
M_micranthidifolia_2    A...T....T-.................   [489]
M_pensylvanica          ....T....--.................   [482]
M_petiolaris_1          C...TTT.....................G.   [485]
M_petiolaris_2          C...TTT.....................G.   [485]
M_petiolaris_3          C...TTT.....................G.   [484]
M_petiolaris_4          C...TTT.....................G.   [484]
M_integrifolia          .........--.................   [478]
M_punctata              ....T....--..........T......   [479]
M_stellaris             ....T....--.................G.   [475]
M_tolmiei               C...T....--T................   [469]

[                                520       530       540]

M_Clawhammer_Mtn        GCCGGGGTTT--GTTACAATGT---AAGGA   [517]
M_Cliffridge            ..........--.........---.....   [517]
M_Crow_Creek            ..........--.........---.....   [517]
M_Cullasaja_Falls       ..........--.........---.....   [517]
M_Elkhollow_Branch      ..........--.........---.....   [520]
M_Gap_Creek_Rd          ........-C.--......G..CAAG.....   [513]
M_Gouges_Creek_Falls    ....A.....--.........---.....   [516]
M_Groto_Falls           ..........--.........---.....   [512]
M_Howard_Creek_Falls    .......G..--.........AGG---..   [510]
M_Ijams_sp_1            ..........--...-.....---.....   [513]
M_Ijams_sp_2            .....T...G-AAGA.....---.....   [518]
M_Ivy_River             .........GGAAGA.A....---.....   [516]
M_Linville_Gorge        ..........--.........---.....   [502]
M_Little_River_Gorge    .........G--.........C---.....   [516]
M_Melrose               .......G..--......GAAATGT.....   [513]
M_Mt_Jefferson          .......G..--.........AGG---..   [501]
M_Nantahala_Gorge_1     ..........--.........---.....   [519]
M_Nantahala_Gorge_2     ..........--.........---.....   [517]
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M_New_River             .......G..--.........AGG---..   [510]
M_Pigeon_River_Gorge    ..........--.........---.....   [515]
M_Profile_Trail         ..........--.........---.....   [516]
M_Shady_Valley          .......G..--.........AGG---..   [511]
M_Slickrock_Trail       ..........--.........---.....   [517]
M_Wadakoe_Mtn           .......G..--.........AAG.....   [505]
M_virginiensis          .......G..--.........AAG.....   [515]
M_micranthidifolia_1    .......G..--...G.....---.....   [517]
M_micranthidifolia_2    .......G..--...G.....---.....   [514]
M_pensylvanica          .......G..--.........---.....   [507]
M_petiolaris_1          .......G..--......T..C---.G...   [510]
M_petiolaris_2          .......G..--......T..C---.G...   [510]
M_petiolaris_3          .......G..--......T..C---.G...   [509]
M_petiolaris_4          .......G..--......T..C---.G...   [509]
M_integrifolia          .......G..--.........---.....   [503]
M_punctata              .......G..--...G.....---.....   [504]
M_stellaris             .......G..--......T..---.G...   [500]
M_tolmiei               ........GN--...GTG.A..---C.A.G   [494]

[                                 550       560       570]

M_Clawhammer_Mtn        ACATTCAATTACACACA--CATGCATGCAT   [545]
M_Cliffridge            ............T....--..........   [545]
M_Crow_Creek            ............T....--..........   [545]
M_Cullasaja_Falls       ............T....--..........   [545]
M_Elkhollow_Branch      ..................--..........   [548]
M_Gap_Creek_Rd          G.....T.....T....--....TG..TG.   [541]
M_Gouges_Creek_Falls    ..................--..........   [544]
M_Groto_Falls           ............T....--..........   [540]
M_Howard_Creek_Falls    ............T....--..........   [538]
M_Ijams_sp_1            .........AT......--...T.......   [541]
M_Ijams_sp_2            ..................--..........   [546]
M_Ivy_River             ..................--..........   [544]
M_Linville_Gorge        ..................--..........   [530]
M_Little_River_Gorge    ............T....--..........   [544]
M_Melrose               ............T..A.--..........   [541]
M_Mt_Jefferson          ............T....--..........   [529]
M_Nantahala_Gorge_1     ............T....--..........   [547]
M_Nantahala_Gorge_2     ............T....--..........   [545]
M_New_River             ............T....--..........   [538]
M_Pigeon_River_Gorge    ..................--..........   [543]
M_Profile_Trail         ..................--..........   [544]
M_Shady_Valley          ............T....--..........   [539]
M_Slickrock_Trail       ............T....--..........   [545]
M_Wadakoe_Mtn           ............T..A.--..........   [533]
M_virginiensis          ............T..A.--..........   [543]
M_micranthidifolia_1    ...C........TT...--.CC..GC..G.   [545]
M_micranthidifolia_2    ...C.S......TT...--.CC..GC..G.   [542]
M_pensylvanica          ...C........TT...--.CC..G.....   [535]
M_petiolaris_1          ..GC.....-..T.CG.--.G.........   [537]
M_petiolaris_2          ..GC.....-..T.CG.--.G.........   [537]
M_petiolaris_3          ..GC.....-..T.CG.--.G.........   [536]
M_petiolaris_4          ..GC.....-..T.CG.--.G.........   [536]
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M_integrifolia          ...C........T....--...........   [531]
M_punctata              ...........TTTG..CCT.CA.....G.   [534]
M_stellaris             ...C.....ACTC.GA---.G.........   [527]
M_tolmiei               ...CC.G..ACAT...--C..CT.......   [522]

[                                 580       590       600]

M_Clawhammer_Mtn        AAAAGCAA-CACA-AGGTTAAAGAAATCAA   [573]
M_Cliffridge            .........-.G..-................   [573]
M_Crow_Creek            .........-.G..-............-TC.   [572]
M_Cullasaja_Falls       .........-.G..-............-TC.   [572]
M_Elkhollow_Branch      .........-.G..-................   [576]
M_Gap_Creek_Rd          .T....GC-.G..CGA..A.G...T....T   [570]
M_Gouges_Creek_Falls    .........-.G..-................   [572]
M_Groto_Falls           .........-.G..-.......C....-TC.   [567]
M_Howard_Creek_Falls    .........-.G..-.A..............   [566]
M_Ijams_sp_1            .........-.G.T-.TT..TTTATTT.TTT   [569]
M_Ijams_sp_2            .........-.G..-.......T......TT   [574]
M_Ivy_River             .........-.G..-..............TT   [572]
M_Linville_Gorge        .........-.G..-................   [558]
M_Little_River_Gorge    .........-.G..-.......T.C...TTT   [572]
M_Melrose               .........-.G..-..A.....TT....TT   [569]
M_Mt_Jefferson          .........-.G..-.-..............   [556]
M_Nantahala_Gorge_1     .........-.G..-............ATC.   [575]
M_Nantahala_Gorge_2     .........-.G..-............-TC.   [572]
M_New_River             .........-.G..-.A..............   [566]
M_Pigeon_River_Gorge    .........-.G..-................   [571]
M_Profile_Trail         .........-.G..-................   [572]
M_Shady_Valley          .........-.G..-.A..............   [567]
M_Slickrock_Trail       .........-.G..-............-TC.   [572]
M_Wadakoe_Mtn           .........-.G-.-..A..........A..   [560]
M_virginiensis          .........-.G..-..A.............   [571]
M_micranthidifolia_1    ........A.G..-................   [574]
M_micranthidifolia_2    ........A.G..-................   [571]
M_pensylvanica          ........A.G..-...G............   [564]
M_petiolaris_1          GG.......--G.CC.C.AC....G...G..   [565]
M_petiolaris_2          GG.......--G.CC.C.AC....G...G..   [565]
M_petiolaris_3          GG.......--G.CC.C.AC....G...G..   [564]
M_petiolaris_4          GG.......--G.CC.C.AC....G...G..   [564]
M_integrifolia          N.......ATG..-....-........T..   [559]
M_punctata              ........ATG..-G.......A.....G.   [563]
M_stellaris             GG......G....-C.AC-....G...G..   [555]
M_tolmiei               .-......GGGTG-..---.G.AGT...GC   [547]

[                                 610       620       630]

M_Clawhammer_Mtn        A----TGCCTCCCCACAAAG--GCGAGAA-   [596]
M_Cliffridge            .----..............--........-   [596]
M_Crow_Creek            .A--TGC.-..........--........-   [596]
M_Cullasaja_Falls       .A--TGC.-..........--........-   [596]
M_Elkhollow_Branch      .----..............--........-   [599]
M_Gap_Creek_Rd          .----..--........C..--........-   [591]
M_Gouges_Creek_Falls    .----..............--........-   [595]
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M_Groto_Falls           .A--AG.............--.......-   [592]
M_Howard_Creek_Falls    .----..............--.......-   [589]
M_Ijams_sp_1            .AAGA.........C.....--.......-   [596]
M_Ijams_sp_2            T----.C............--.......-   [597]
M_Ivy_River             T----.C............--.......-   [595]
M_Linville_Gorge        .----..............---......-   [580]
M_Little_River_Gorge    -----.T............--.......-   [594]
M_Melrose               T----CC............--.......-   [592]
M_Mt_Jefferson          .----..............--.......-   [579]
M_Nantahala_Gorge_1     .A--TGC............--.......-   [600]
M_Nantahala_Gorge_2     .A--TGT............--.......-   [597]
M_New_River             .----..............--.......-   [589]
M_Pigeon_River_Gorge    .----..............--.......-   [594]
M_Profile_Trail         .---TGC............--.......-   [596]
M_Shady_Valley          .----..............--.......-   [590]
M_Slickrock_Trail       .A--TGC.-..........--.......-   [596]
M_Wadakoe_Mtn           .----..T...........--....A..-   [583]
M_virginiensis          .----C.............--......G-   [594]
M_micranthidifolia_1    .----..A...........AT.T.....-   [599]
M_micranthidifolia_2    .----..G...........AT.T.....-   [596]
M_pensylvanica          .----..T...........--.T.....-   [587]
M_petiolaris_1          .-----CAAAT..A.....--.TA.A..-   [587]
M_petiolaris_2          G-----CGAAT..A.....--.TA.A..-   [587]
M_petiolaris_3          G-----CGAAT..A.....--.TA.A..-   [586]
M_petiolaris_4          G-----CGAAT..A.....--.TA.A..-   [586]
M_integrifolia          .----..........N..--.A..AG.-   [582]
M_punctata              .----..A.A...A.....--.T.....-   [586]
M_stellaris             G----C.A-AT..A.....--.TA.A..A   [578]
M_tolmiei               .----CA.....TG...TC.--.G.....-   [570]

[                              640       650       660]

M_Clawhammer_Mtn        GAGACATGAAATG---CCCC--TCCTCGAT   [621]
M_Cliffridge            ............---....--.....A..   [621]
M_Crow_Creek            ............---....T--....A..   [621]
M_Cullasaja_Falls       ...........-----TG..CC.....A..   [621]
M_Elkhollow_Branch      ............---....CTC.......   [626]
M_Gap_Creek_Rd          .........G...---.G..TCC-TCACT.   [617]
M_Gouges_Creek_Falls    ..........T..---....--........   [620]
M_Groto_Falls           ...............-----TG..CC.....A..   [617]
M_Howard_Creek_Falls    ............---T...TCC-TCA.T.   [615]
M_Ijams_sp_1            .TTT.-.C-C.CC---....--C..CTTC.   [619]
M_Ijams_sp_2            ..........C.---.............T-.   [621]
M_Ivy_River             .....T.....CC---...T--.....T..   [620]
M_Linville_Gorge        ..-....CCCCCC---TTTT--...CTCC.   [604]
M_Little_River_Gorge    .......T-T.AC---.....--.T.CTC..   [618]
M_Melrose               .......CCC.AC---.....--CT.CTCT.   [617]
M_Mt_Jefferson          ............---T...TCC.TCAAT.   [606]
M_Nantahala_Gorge_1     ............---....CTC....A..   [627]
M_Nantahala_Gorge_2     ..........A----....CC.....A..   [623]
M_New_River             ............---T...TCC-TCA.T.   [615]
M_Pigeon_River_Gorge    ..........T---.....--........   [619]
M_Profile_Trail         .........C.AC---G...--CT.C.C..   [621]
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M_Shady_Valley          .............---T...TCC-TCA.T.   [616]
M_Slickrock_Trail       .............GCG.TT.CTC....A..   [626]
M_Wadakoe_Mtn           ............----....--CT.CTC.A   [607]
M_virginiensis          ............---....--CT.CTC.A   [619]
M_micranthidifolia_1    ..A.........---....-TC.TCAAT.   [625]
M_micranthidifolia_2    ..A.........---....-TC.TCAAT.   [622]
M_pensylvanica          ..A..........---....CTC..C.A..   [614]
M_petiolaris_1          .GA...AAG....---....TCCT..TTT.   [614]
M_petiolaris_2          .GA...AAG....---.T..TCCT..TTT.   [614]
M_petiolaris_3          .GA...AAG....---.T..TCCT..TTT.   [613]
M_petiolaris_4          .GA...AAG....---.T..TCCT..TTT.   [613]
M_integrifolia          --..........---....TCCT.AATT.   [607]
M_punctata              ..A..........---....TCCT.AAT..   [613]
M_stellaris             .GA...AAG....---.T..TTCT..TTT.   [605]
M_tolmiei               .GA..........---....CCC..C.TGG   [597]

[                                670       680       690]

M_Clawhammer_Mtn        TTTTTTAACATG-TTCTC----AATGTTT-   [645]
M_Cliffridge            ...........-.....----.......-   [645]
M_Crow_Creek            ...........-.....----.......T   [646]
M_Cullasaja_Falls       ...........-.....----.......T   [646]
M_Elkhollow_Branch      ...........-.....----.......T   [651]
M_Gap_Creek_Rd          ....--....A.-A.G..-----..G.A..T   [640]
M_Gouges_Creek_Falls    ...........-.....----.......-   [644]
M_Groto_Falls           ...........--.....----...-G..T   [640]
M_Howard_Creek_Falls    .....-......-.....-----..G....T   [639]
M_Ijams_sp_1            ..--------------------------   [622]
M_Ijams_sp_2            ...........-.....----.......-   [645]
M_Ivy_River             ...........-.....----.......-   [644]
M_Linville_Gorge        CGA...T.----------------------   [612]
M_Little_River_Gorge    ...........-.....----.......T   [643]
M_Melrose               .....A......-.....-----..G....T   [642]
M_Mt_Jefferson          .....-.......-G.TC.TCCA..GT...C   [634]
M_Nantahala_Gorge_1     ...........-.....----...-...T   [651]
M_Nantahala_Gorge_2     ...........-.....----...-G..T   [647]
M_New_River             .....-......-.....---A.GGT...T   [640]
M_Pigeon_River_Gorge    .......C.T..-.....----.......-   [643]
M_Profile_Trail         ...........-.....----.......T   [646]
M_Shady_Valley          ...........-.....-----..G.G.AC   [641]
M_Slickrock_Trail       ...........-.....----...-...T   [650]
M_Wadakoe_Mtn           ...........-.....-----..G....-   [631]
M_virginiensis          ...........-.....-----..G....T   [644]
M_micranthidifolia_1    ...........-.....-----..G....-   [649]
M_micranthidifolia_2    ...........-.....-----..G....T   [647]
M_pensylvanica          ...........G.....----.-------   [633]
M_petiolaris_1          .GAGG.C..GC.-...A.----.CG....-   [638]
M_petiolaris_2          .GAGG.C..GC.-...A.----.CG....T   [639]
M_petiolaris_3          .GAGG.C..GC.-...A.----.CG....T   [638]
M_petiolaris_4          .GAGG.C..GC.-...A.----.CG....-   [637]
M_integrifolia          ..--.....-.....----.TG....T   [630]
M_punctata              ..--..T.....-.....-----..G....T   [636]
M_stellaris             .GAGG.C..G..-...A.----.CG....T   [630]
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M_tolmiei                .N..GN......-...G.-----.TG....T   [622]

[                                  700       710]

M_Clawhammer_Mtn         CTGCAT--GCAGGG-TT-CAG   [662]
M_Cliffridge             .......--......-.--...   [661]
M_Crow_Creek             ......T-.......--.T...   [664]
M_Cullasaja_Falls        ........--.......--.T...   [663]
M_Elkhollow_Branch       ......T-.......-..T...   [670]
M_Gap_Creek_Rd           ....T.T-.......-.G-..T   [658]
M_Gouges_Creek_Falls     ........--.......-..-...   [661]
M_Groto_Falls            ........--.......-..-...   [657]
M_Howard_Creek_Falls     ......T-.......-..T...   [658]
M_Ijams_sp_1             --------------------   [622]
M_Ijams_sp_2             .......-T.......-..C...   [664]
M_Ivy_River              ........--....A.-..A...   [662]
M_Linville_Gorge         --------------------   [612]
M_Little_River_Gorge     .......-T.......-..T...   [662]
M_Melrose                ........--.....A-.--...   [658]
M_Mt_Jefferson           ...G.A---------------   [640]
M_Nantahala_Gorge_1      ......T-C........--.T...   [669]
M_Nantahala_Gorge_2      ......T-.......-..T...   [666]
M_New_River              ......T-.......-..T...   [659]
M_Pigeon_River_Gorge     ........--.......-..-...   [660]
M_Profile_Trail          TCTTGCAT.......-A.A...   [666]
M_Shady_Valley           --------------------   [641]
M_Slickrock_Trail        ......T-......G..T...   [670]
M_Wadakoe_Mtn            ........--.......-..-...   [648]
M_virginiensis           ......T-.......-..T...   [663]
M_micranthidifolia_1     ........--.....T---T...   [665]
M_micranthidifolia_2     ......T-.....T---T...   [664]
M_pensylvanica           --------------------   [633]
M_petiolaris_1           ......T-.....T---T...   [655]
M_petiolaris_2           ........--.....T---T...   [655]
M_petiolaris_3           ........--.....T---T...   [654]
M_petiolaris_4           ........--.....T---T...   [653]
M_integrifolia           ......T-.....T--.T...   [648]
M_punctata               ......T-.....T--.T...   [654]
M_stellaris              ......T-.....T--.T...   [648]
M_tolmiei                ....TAA-.....T--.T.GA   [640]
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ii. Sequences used in *trn*L-*trn*F data set:

```
[                              10        20        30]

M_careyana             ---TNCGAANNNGGGTAGACGCTACGGACT   [27]
M_caroliniana          ---.T....ATNC...T.............   [27]
M_virginiensis         --TCGNA..TTC....T.............   [28]
M_micranthidifolia     --T.CGA..ATC..................   [28]
M_pensylvanica         -TTCGNA.NANG..................   [29]
M_petiolaris           NNNNNNNTNNNNNNNNNA............   [30]
M_integrifolia         -----------------------------   [0]
M_punctata             -----------------------------   [0]
M_stellaris            ----------------------------G..   [3]
M_tolmiei              ----------------------------G..   [3]

[                              40        50        60]

M_careyana             TAATT-GAATTGAGCCTYK-GTATGGAACC   [55]
M_caroliniana          .....--G.A.......TG-........A.   [54]
M_virginiensis         ...A.-...........TG-..M....RA.   [56]
M_micranthidifolia     .....-...........TG-........A.   [56]
M_pensylvanica         ...A.T...........TGC........A.   [59]
M_petiolaris           .....-...........TG-........A.   [58]
M_integrifolia         ----------......TG-........A.   [18]
M_punctata             ----------------.TG-........A.   [13]
M_stellaris            .....-...........TG-........A.   [31]
M_tolmiei              .....-...........TC-........A.   [31]

[                              70        80        90]

M_careyana             -TACTAAGTGATA-CTTTCAAATTCAGAGA   [83]
M_caroliniana          C............A................   [84]
M_virginiensis         C............-................   [85]
M_micranthidifolia     C............A................   [86]
M_pensylvanica         -............A................   [88]
M_petiolaris           -............AT...............   [87]
M_integrifolia         C............A................   [48]
M_punctata             C............A................   [43]
M_stellaris            C............AT...............   [61]
M_tolmiei              C............A................   [61]

[                              100       110       120]

M_careyana             AACCCTGGAATTAAAAATGGGCAATCCTGA   [113]
M_caroliniana          ..............................   [114]
M_virginiensis         ..............................   [115]
M_micranthidifolia     ..............................   [116]
M_pensylvanica         ..............................   [118]
M_petiolaris           ............T.................   [117]
M_integrifolia         ..............................   [78]
M_punctata             ..............................   [73]
M_stellaris            ............T.................   [91]
M_tolmiei              ............T.................   [91]
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[                          130       140       150]

M_careyana            GCCAAATCCTGTTTT--ATGAAAAAAAAAA   [141]
M_caroliniana         ..............--............-   [141]
M_virginiensis        ..............--............-   [142]
M_micranthidifolia    ..............T-............    [145]
M_pensylvanica        ..............--............    [146]
M_petiolaris          ..............----CT.........   [143]
M_integrifolia        ..............--............    [106]
M_punctata            ..............ACGAA.........    [103]
M_stellaris           ..............----CT........G   [117]
M_tolmiei             ..............--.CA....G....C   [119]

[                          160       170       180]

M_careyana            GGATAGGTGCAGAGACTCAATGGAAGCTAT   [171]
M_caroliniana         ............................    [171]
M_virginiensis        ............................    [172]
M_micranthidifolia    ............................    [175]
M_pensylvanica        ............................    [176]
M_petiolaris          ...G.....................A.G.   [173]
M_integrifolia        ............................    [136]
M_punctata            ............................    [133]
M_stellaris           ...G.....................A.G.   [147]
M_tolmiei             .C.AG...T....A.GAT.GAAA..AAA.G   [149]

[                          190       200       210]

M_careyana            TCTAACAAAGGGAGTTGACTGCGACTGCGG   [201]
M_caroliniana         ............................    [201]
M_virginiensis        ............................    [202]
M_micranthidifolia    ............................    [205]
M_pensylvanica        ............................    [206]
M_petiolaris          ......................------.   [197]
M_integrifolia        ............................    [166]
M_punctata            ............................    [163]
M_stellaris           ..............G.....------.    [171]
M_tolmiei             GAKGG-GTG...M.ACTC..TG.WGGCT.T   [178]

[                          220       230       240]

M_careyana            TGCGTTGGTAAAGGAATCCTTACATCGAAA   [231]
M_caroliniana         ............................    [231]
M_virginiensis        ............................    [232]
M_micranthidifolia    ............................    [235]
M_pensylvanica        ............................    [236]
M_petiolaris          ...A........A................   [227]
M_integrifolia        ............................    [196]
M_punctata            ........................T....   [193]
M_stellaris           Y..A........A................   [201]
M_tolmiei             .TTWNYAAYGG....-.GAC.G.KG..GGT   [207]
```

```
[                          250       260       270]

M_careyana            AAGGCGGAAGGATAAACCTAACCCCTATGT   [261]
M_caroliniana         --............................   [259]
M_virginiensis        --............................   [260]
M_micranthidifolia    --............................   [263]
M_pensylvanica        --............................   [264]
M_petiolaris          --...T......G..........T......   [255]
M_integrifolia        --.......................T.....   [224]
M_punctata            --------G.C............TA.....   [215]
M_stellaris           --...T......G..........T......   [229]
M_tolmiei             ----TNKT.AAGG.GGAM.....T......   [233]

[                          280       290       300]

M_careyana            TGAA---------GAAAGAATCAAATATT-   [281]
M_caroliniana         ...----------.................-   [279]
M_virginiensis        ....GAAGTTGAA.................-   [289]
M_micranthidifolia    ...----------.................-   [283]
M_pensylvanica        ....T--GTTGAA.................-   [291]
M_petiolaris          ...----------.................A   [276]
M_integrifolia        ...----------.................-   [244]
M_punctata            CCT.TATGTTGAA.................-   [244]
M_stellaris           ...----------.................A   [250]
M_tolmiei             ...----------.............W..-   [253]

[                          310       320       330]

M_careyana            -----CATTGAAAAACCCACTCACTCCCCA   [306]
M_caroliniana         -----............................   [304]
M_virginiensis        -----............................   [314]
M_micranthidifolia    -----............................   [308]
M_pensylvanica        -----............................   [316]
M_petiolaris          ATATT......T................G..   [306]
M_integrifolia        -----............................   [269]
M_punctata            -----......T................G..   [269]
M_stellaris           ATATT......T................G..   [280]
M_tolmiei             ------....N.T.......T.......GG.   [278]

[                          340       350       360]

M_careyana            GTCTGATAAATCCTGTGAAAAGCTGATTAA   [336]
M_caroliniana         ................................   [334]
M_virginiensis        ................................   [344]
M_micranthidifolia    ................................   [338]
M_pensylvanica        ................................   [346]
M_petiolaris          .....C.......CT................   [336]
M_integrifolia        ................................   [299]
M_punctata            ..............T.................   [299]
M_stellaris           .....C.......CT................   [310]
M_tolmiei             ..Y..C.......CT.............T.   [308]

[                          370       380       390]
```

```
M_careyana            TCAGATGAGAATAAAGATAGAGTCCCATTC   [366]
M_caroliniana         .............................   [364]
M_virginiensis        ............................T   [374]
M_micranthidifolia    .............................   [368]
M_pensylvanica        .............................   [376]
M_petiolaris          ....CC.......................   [366]
M_integrifolia        ......T......................   [329]
M_punctata            .............................   [329]
M_stellaris           ....CC.......................   [340]
M_tolmiei             .NN...TNNNNNNNNNNNNNN....G...   [338]

[                              400       410       420]

M_careyana            TACATGTCAATCCAGACAACAATGAAATTT   [396]
M_caroliniana         .............................   [394]
M_virginiensis        .............................   [404]
M_micranthidifolia    .............................   [398]
M_pensylvanica        .............................   [406]
M_petiolaris          .............................   [396]
M_integrifolia        .............................   [359]
M_punctata            .............................   [359]
M_stellaris           ...................G.........   [370]
M_tolmiei             .M..G.Y...KN......N...........   [368]

[                              430       440       450]

M_careyana            ATAGTAAGAGGAAAATCCGTCGACTTTAGA   [426]
M_caroliniana         .............................   [424]
M_virginiensis        .............................   [434]
M_micranthidifolia    .............................   [428]
M_pensylvanica        .............................   [436]
M_petiolaris          .............................   [426]
M_integrifolia        .............................   [389]
M_punctata            .............................   [389]
M_stellaris           .............................   [400]
M_tolmiei             ..R............Y......M...A...   [398]

[                              460       470       480]

M_careyana            AATCATGAGGGTTCAAGTCCCTCTATCCCC   [456]
M_caroliniana         .............................   [454]
M_virginiensis        .............................   [464]
M_micranthidifolia    .............................   [458]
M_pensylvanica        .............................   [466]
M_petiolaris          .............................   [456]
M_integrifolia        .............................   [419]
M_punctata            .............................   [419]
M_stellaris           .............................   [430]
M_tolmiei             ............Y..........W......   [428]

[                              490       500       510]
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```
M_careyana            ATC-GACTCCTTTATTATTTATCTTATCCT   [485]
M_caroliniana         ...-.........................   [483]
M_virginiensis        ...-.........................   [493]
M_micranthidifolia    ...-A.........C..............   [487]
M_pensylvanica        ...-.........................   [495]
M_petiolaris          ..T-T.........C..............   [485]
M_integrifolia        ...-............--...........   [446]
M_punctata            ...-..........C..............   [448]
M_stellaris           ..T-T.........C..............   [459]
M_tolmiei             .AAA.G.CGG..KGCCCCC.TA.CC..Y.C   [458]

[                              520       530       540]

M_careyana            ATACCT-CTCTTTTCCTTAGTGGTTCAAAA   [514]
M_caroliniana         ......-.......................   [512]
M_virginiensis        ......-.......................   [522]
M_micranthidifolia    ......-.......................   [516]
M_pensylvanica        ......-.......................   [524]
M_petiolaris          ......-.......................   [514]
M_integrifolia        ......-.......................   [475]
M_punctata            ......-........A.....T........   [477]
M_stellaris           ......-.......................   [488]
M_tolmiei             .A.NN.TY.....Y.A.A..G...A....R   [488]

[                              550       560       570]

M_careyana            GTCCTTATGTTTCTCATTCACTCTACGCTT   [544]
M_caroliniana         .........C.....................   [542]
M_virginiensis        ..............................   [552]
M_micranthidifolia    ..............................   [546]
M_pensylvanica        ..............................   [554]
M_petiolaris          ............................---.   [541]
M_integrifolia        ..............................   [505]
M_punctata            ............T............T....   [507]
M_stellaris           ............................---.   [515]
M_tolmiei             ..GG.AN.N...YC...C.C...NCS.---   [515]

[                              580       590       600]

M_careyana            TTTTCCCAAAAGGGAAATTTTTGTTTTT--   [572]
M_caroliniana         ..............................--   [570]
M_virginiensis        ..............................--   [580]
M_micranthidifolia    ..............................--   [574]
M_pensylvanica        ..............................--   [582]
M_petiolaris          C............................CT   [571]
M_integrifolia        ...............T............--   [533]
M_punctata            C..--------------------------   [511]
M_stellaris           C......C.....................CT   [545]
M_tolmiei             Y...T.....G..----..CCAG.-------   [535]

[                              610       620       630]

M_careyana            ---------TATCATATCACAAACAAGTCT   [593]
```

```
M_caroliniana          ---------.....................   [591]
M_virginiensis         ---------.....................   [601]
M_micranthidifolia     ---------.....................   [595]
M_pensylvanica         ---------.....................   [603]
M_petiolaris           CATATCTTT.C...................   [601]
M_integrifolia         ---------.....................   [554]
M_punctata             -----------------------------   [511]
M_stellaris            CATATCTTT.C..----.............   [571]
M_tolmiei              ----------G.A.A..T.AYCC.......   [555]

[                               640       650       660]

M_careyana             TATGAT----ACACAC-GTACACATGAACA   [618]
M_caroliniana          .....----.....---.............   [614]
M_virginiensis         .....----.....---.............   [624]
M_micranthidifolia     .....----.....---.............   [618]
M_pensylvanica         .....----.....---.............   [626]
M_petiolaris           ......TATG.T....-.............   [630]
M_integrifolia         .....----.....---.............   [577]
M_punctata             ---..----.....---.............   [531]
M_stellaris            .....------....-.............   [594]
M_tolmiei              GG.A.AAGTT.AW.C.G.NNNN.....C..   [585]

[                               670       680       690]

M_careyana             TCTTTA----AAGCAAG-AATTCCCATTTG   [643]
M_caroliniana          .....----.......G.............   [640]
M_virginiensis         .....----.......G.............   [650]
M_micranthidifolia     .....----.......G.............   [644]
M_pensylvanica         .....----.......G.............   [652]
M_petiolaris           .....----.......G...C....G...   [656]
M_integrifolia         .....----.......G.............   [603]
M_punctata             .....----.......G...C........   [557]
M_stellaris            .....----.......G...C........   [620]
M_tolmiei              ......GGTT..A.C.AT...C..A..AA.   [615]

[                               700       710       720]

M_careyana             AATGATTCACAGTCCATA-TAATT------   [666]
M_caroliniana          ..................-...------   [663]
M_virginiensis         ..................-.C..------   [673]
M_micranthidifolia     ..................-.C..------   [667]
M_pensylvanica         ...T..............-.C..------   [675]
M_petiolaris           .........T........-.C...ACTCAT   [685]
M_integrifolia         ..................-.C..------   [626]
M_punctata             ..................-.T..------   [580]
M_stellaris            ...T.....T........-.C...ACTCAT   [649]
M_tolmiei              ...T....C.......C.A.C...CCTCAN   [645]

[                               730       740       750]

M_careyana             -TTGAAACTTACAAAGTTTTTTTTT-AGAT   [694]
M_caroliniana          -.........................--....   [690]
```

```
M_virginiensis        -.................G.....-....   [701]
M_micranthidifolia    -..............C........-....   [695]
M_pensylvanica        -.......................--....   [702]
M_petiolaris          A.......................--....   [713]
M_integrifolia        -.......................T....   [655]
M_punctata            -.......................-....   [608]
M_stellaris           A.......................-....   [678]
M_tolmiei             N.....C...C.............-TT..   [674]

[                             760       770       780]

M_careyana            CCAAGAAATACCAAGGCCTGGATAAGACTT   [724]
M_caroliniana         ..C............................   [720]
M_virginiensis        ...............................   [731]
M_micranthidifolia    ...............................   [725]
M_pensylvanica        ...............................   [732]
M_petiolaris          .........T.....................   [743]
M_integrifolia        ...............................   [685]
M_punctata            ....A....T.....T.T.............   [638]
M_stellaris           .........T.....................   [708]
M_tolmiei             .........T.....................   [704]

[                             790       800       810]

M_careyana            TTTAATTCTTTGTAATTGCCATAGACCCAG   [754]
M_caroliniana         ..................A...........   [750]
M_virginiensis        ..................A...........   [761]
M_micranthidifolia    .......A...-......A...........   [754]
M_pensylvanica        ..................A...........   [762]
M_petiolaris          ...C...A....G.....A..........T   [773]
M_integrifolia        ..................A...........   [715]
M_punctata            .......A..........A..G........   [668]
M_stellaris           ...C...A....G.....A..........T   [738]
M_tolmiei             .G.....A..........A.........TA   [734]

[                             820       830       840]

M_careyana            GTCATCTAGTAAAATGAGAATGATGCGTCG   [784]
M_caroliniana         ...............................   [780]
M_virginiensis        ...............................   [791]
M_micranthidifolia    ...............................   [784]
M_pensylvanica        ...............................   [792]
M_petiolaris          ...............................   [803]
M_integrifolia        ......-------------------------   [721]
M_punctata            ....G..........................   [698]
M_stellaris           ...............G..............   [768]
M_tolmiei             ......C........................   [764]

[                             850       860       870]

M_careyana            G----------GATAG-TCAGCTGGAA--G   [801]
M_caroliniana         .----------.....C........T.GA.   [800]
M_virginiensis        .----------.....C........T.GA.   [811]
```

```
M_micranthidifolia    .----------....-.......A..AAN   [803]
M_pensylvanica        .----------....-.........GA.   [810]
M_petiolaris          .----------....-.......AR.--.   [820]
M_integrifolia        ------------------------------   [721]
M_punctata            .-----------------------------   [699]
M_stellaris           .----------..-----------------   [771]
M_tolmiei             .AAATGGTCGG.....C....T...T.GA.   [794]

[                              880       890       900]

M_careyana            CACAAGACTGAAAATCCTCGTGTCNCNNNN   [831]
M_caroliniana         ..G.G...................A.AANA   [830]
M_virginiensis        ..G.G...................A.ANAN   [841]
M_micranthidifolia    ..A.G........NN.TC.NN.GNCAANAN   [833]
M_pensylvanica        ..G.G.................NNC.-CNA   [839]
M_petiolaris          ..A....ACTG........N.N.NCNNNAN   [850]
M_integrifolia        ------------------------------   [721]
M_punctata            ------------------------------   [699]
M_stellaris           ------------------------------   [771]
M_tolmiei             ..G.G...----------------------   [802]

[                              910]

M_careyana            TTNNAAAN---   [839]
M_caroliniana         N.TNC..ANT-   [840]
M_virginiensis        ..NC...NTA-   [851]
M_micranthidifolia    N.TNN..TNTN   [844]
M_pensylvanica        ..NN..NATA-   [849]
M_petiolaris          NNNA..NAA--   [859]
M_integrifolia        -----------   [721]
M_punctata            -----------   [699]
M_stellaris           -----------   [771]
M_tolmiei             -----------   [802]
```

iii. Sequences used in *mat*K data set:

```
[                              10        20        30]

M_tolmiei              ATGGGGGAATTTCAAGGATATTTAGAATTC   [30]
Cascadia               ------------------------------   [0]
M_arguta               ....A.A...A................C..   [30]
M_aprica               ---------------------------C..   [3]
M_bryophora            ....A.....A................C..   [30]
M_californica          ....A.....A................C..   [30]
M_calycina             ....A.....A.A..............C..   [30]
M_careyana             ------------------------------   [0]
M_ferruginea           ....A.....A................C..   [30]
M_foliolosa            ....A.....A................C..   [30]
M_fusca                ....A.....C................C..   [30]
M_hieracifolia         ....A.....A................C..   [30]
M_howellii             ....N.....A....N...........C..   [30]
M_integrifolia         ....A.....A................C..   [30]
M_lyallii              ....NNNN..A................C..   [30]
M_micranthidifolia     ------------------------------   [0]
M_nidifica             ....A.....A................C..   [30]
M_nivalis              ....A.....A................C..   [30]
M_occidentalis         ------------------------...C..   [6]
M_oregana              ....A.....A................C..   [30]
M_pensylvanica         ------------------------------   [0]
M_petiolaris           ------------------------------   [0]
M_punctata             ....A.....C................C..   [30]
M_reflexa              ....A.....A................C..   [30]
M_rhomboidea           ....A.....A................C..   [30]
M_rufidula             ------------------------------   [0]
M_spicata              ....A.....A....N...........C..   [30]
M_stellaris            ....A.....A................C..   [30]
M_tenuis               ....A.....A................C..   [30]
M_texana               ------------------------------   [0]
M_virginiensis         ....A..N..A....N...........C..   [30]

[                               40        50        60]

M_tolmiei              AATCAATTTCGACAACATGATTTCCTATAT   [60]
Cascadia               ------------------.......NNNNN   [12]
M_arguta               ...A.......T...A....C.........   [60]
M_aprica               ...A.......T...A....C.........   [33]
M_bryophora            GN...NN....G...N.......N..C...   [60]
M_californica          ...A.......T...A....C.........   [60]
M_calycina             ...A.......G...A....C.........   [60]
M_careyana             ------------------------------   [0]
M_ferruginea           ..........G...A..A....A..C...   [60]
M_foliolosa            ..........G...A..A....A..C...   [60]
M_fusca                ...A.....T.G...A....C.........   [60]
M_hieracifolia         ...A.......T...A....C.........   [60]
M_howellii             G..G.......G...Y....C...G.....   [60]
M_integrifolia         ...A.......T...A....C.........   [60]
M_lyallii              ...A.......G...A....C.........   [60]
```

```
M_micranthidifolia    ----------------------------   [0]
M_nidifica            ...A.......T...A....C.........  [60]
M_nivalis             C..A.......T...A....C.......C.  [60]
M_occidentalis        ...A.......T...A....C.........  [36]
M_oregana             ...A.......T...A....C.........  [60]
M_pensylvanica        ---------..T...A....C.........  [21]
M_petiolaris          ----------------------------   [0]
M_punctata            ...A.....TTG...A....C.........  [60]
M_reflexa             C..A.......T...A....C.........  [60]
M_rhomboidea          ...A.......T...A....C.........  [60]
M_rufidula            ----------------------------   [0]
M_spicata             R..A.......G...N....S.........  [60]
M_stellaris           ...........G...A..A....A..C...  [60]
M_tenuis              C..A.......T...A....C.........  [60]
M_texana              ----------------------------   [0]
M_virginiensis        ...A.......T...A....C.........  [60]

[                              70        80        90]

M_tolmiei             CCACTTATCTTTCAGGAGTATATTTATGCA  [90]
Cascadia              ..............N..........NN   [42]
M_arguta              ......C.......................  [90]
M_aprica              ..............................  [63]
M_bryophora           N....NC.N.....................  [90]
M_californica         ..............................  [90]
M_calycina            ......C.......................  [90]
M_careyana            ----------------------------   [0]
M_ferruginea          ......C.A...A.................  [90]
M_foliolosa           ......C.A...A.................  [90]
M_fusca               ......C.......................  [90]
M_hieracifolia        ..............................  [90]
M_howellii            ............R.................  [90]
M_integrifolia        ..............................  [90]
M_lyallii             ......C.......................  [90]
M_micranthidifolia    ----------------------------   [0]
M_nidifica            ..............................  [90]
M_nivalis             ..............................  [90]
M_occidentalis        ..............................  [66]
M_oregana             ..............................  [90]
M_pensylvanica        ..............................  [51]
M_petiolaris          ----------------------------   [0]
M_punctata            ..N...C.......................  [90]
M_reflexa             ...N..........................  [90]
M_rhomboidea          ..............................  [90]
M_rufidula            --------------------.........  [10]
M_spicata             GN....C.......................  [90]
M_stellaris           ......C.A...A.................  [90]
M_tenuis              ..............................  [90]
M_texana              ----------------------------   [0]
M_virginiensis        .......N........N.............  [90]

[                              100       110       120]
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```
M_tolmiei              CTTGCTCATGATCATGTTTTAAATAGATGC   [120]
Cascadia               ...........NN....N...........   [72]
M_arguta               ..............A..............   [120]
M_aprica               ..............GA.......A.A....   [93]
M_bryophora            ..............A.G............   [120]
M_californica          ..............GA.......A.A....   [120]
M_calycina             ..............A..............   [120]
M_careyana             ------------------------------   [0]
M_ferruginea           ..............A..............   [120]
M_foliolosa            ..............A..............   [120]
M_fusca                ..............A..............   [120]
M_hieracifolia         ..............GA.......A.A....   [120]
M_howellii             ..............A....G....A..C.   [120]
M_integrifolia         ..............GA.......A.A....   [120]
M_lyallii              ..............A..............   [120]
M_micranthidifolia     ------------------------------   [0]
M_nidifica             ..............GA.......A.A....   [120]
M_nivalis              ..............GA.......A.A....   [120]
M_occidentalis         ..............GA.......A.A....   [96]
M_oregana              ..............GA.......A.A....   [120]
M_pensylvanica         ..............GA.......A.A....   [81]
M_petiolaris           ------------------------------   [0]
M_punctata             G.............A..............   [120]
M_reflexa              ..............GA.......A.A....   [120]
M_rhomboidea           ..............GA.......A.A....   [120]
M_rufidula             ..............GA.......A.A....   [40]
M_spicata              ..............A.G.....W......   [120]
M_stellaris            ..............A..............   [120]
M_tenuis               ..............GA.......A.A....   [120]
M_texana               ------------------------------   [0]
M_virginiensis         ..............AA.......A.A....   [120]

[                               130       140       150]

M_tolmiei              ATTTTTTTGGATTATTTTGGTTATGATAAT   [150]
Cascadia               ............................   [102]
M_arguta               .......CA...A.CC..A.......C...   [150]
M_aprica               .....G.CA...A..C..A.......C...   [123]
M_bryophora            .....K.C....A............C...   [150]
M_californica          .....G.CA...A..C..A.......C...   [150]
M_calycina             .....G.CA...A..C..A.......C...   [150]
M_careyana             ------------------------------   [0]
M_ferruginea           .......C....A.....A.......C...   [150]
M_foliolosa            .......C....A.....A.......C...   [150]
M_fusca                .....G.CA...A..C..A.......C...   [150]
M_hieracifolia         .....G.CA...A..C..A.......C...   [150]
M_howellii             .....G.M....A..C..........C...   [150]
M_integrifolia         .....G.CA...A..C..A.......C...   [150]
M_lyallii              .....G.CA...A..C..A.....N.C...   [150]
M_micranthidifolia     ------------------------------   [0]
M_nidifica             .....G.CA...A..C..A.......C...   [150]
M_nivalis              .....G.CA...A..C..A.......C...   [150]
M_occidentalis         .....G.CA...A..C..A.......C...   [126]
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M_oregana             .....G.CA...A..C..A.......C...   [150]
M_pensylvanica        .....G.CA...A..C..A.......C...   [111]
M_petiolaris          ----------------------------     [0]
M_punctata            .....G.CA...A..C..A.......C...   [150]
M_reflexa             .....G.CA...A..C..A.......C...   [150]
M_rhomboidea          .....G.CA...A..C..A.......C...   [150]
M_rufidula            .....G.CA...A..C..A.......C...   [70]
M_spicata             .....G.CA...A..C..AR......C...   [150]
M_stellaris           .......C....A.....A.......C...   [150]
M_tenuis              .....G.CA...A..C..A.......C...   [150]
M_texana              -----G.CA...A..C..A.......C...   [25]
M_virginiensis        .....G.CA...A..C..A.......C...   [150]

[                              160       170       180]

M_tolmiei             AAATCCANNTCACTAATTGTGAAACGTTTA    [180]
Cascadia              .......NN......N.......N.....N   [132]
M_arguta              .....T.GC.................C...   [180]
M_aprica              .....T.GC.................C...   [153]
M_bryophora           .....T.GC.........H...G..C...   [180]
M_californica         .....T.GC.................C...   [180]
M_calycina            .....T.GC.................C...   [180]
M_careyana            ----------------------------     [0]
M_ferruginea          .....T.GC.........C......C...   [180]
M_foliolosa           .....T.GC.........C......C...   [180]
M_fusca               .....T.GC.................C...   [180]
M_hieracifolia        .....T.GC.................C...   [180]
M_howellii            .....N.GY......G.K.........G..   [180]
M_integrifolia        .....T.GC.................C...   [180]
M_lyallii             .....T.GC.................C...   [180]
M_micranthidifolia    ----------------------------     [0]
M_nidifica            .....T.GC.................C...   [180]
M_nivalis             .....T.GC.................C...   [180]
M_occidentalis        .....T.GC.................C...   [156]
M_oregana             .....T.GC.................C...   [180]
M_pensylvanica        .....T.GC.................C...   [141]
M_petiolaris          ----------------------------     [0]
M_punctata            .....T.GC.................C...   [180]
M_reflexa             .....T.GC.................C...   [180]
M_rhomboidea          .....T.GC.................C...   [180]
M_rufidula            .....T.GC.................C...   [100]
M_spicata             .....T.GC.............WR..C...   [180]
M_stellaris           .....T.GC.........C......CN..   [180]
M_tenuis              .....T.GC.................C...   [180]
M_texana              .....T.GC.................C...   [55]
M_virginiensis        .....T.GC.................N...   [180]

[                              190       200       210]

M_tolmiei             ATTACTCGAATGTGTCAACCAAATCATTTG    [210]
Cascadia              ..............................   [162]
M_arguta              ............C......G........C   [210]
M_aprica              .......A.....C.....TG........A   [183]
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M_bryophora            ..................G..C.....A   [210]
M_californica          .......A.....C.....TG........A   [210]
M_calycina             ............C......G........A   [210]
M_careyana             ----------------------------   [0]
M_ferruginea           ............A.........C.....A   [210]
M_foliolosa            ............A.........C.....A   [210]
M_fusca                ............C......G........A   [210]
M_hieracifolia         .......A.....C.....TG........A   [210]
M_howellii             .......A.....C.....TG........A   [210]
M_integrifolia         .....N.A.....C.....TG........A   [210]
M_lyallii              ............C......G........C   [210]
M_micranthidifolia     ----------------------------   [0]
M_nidifica             .......A.....C.....TG........A   [210]
M_nivalis              .......A.....C.....TG........A   [210]
M_occidentalis         .......A.....C.....TG........A   [186]
M_oregana              .......A.....C.....TG........A   [210]
M_pensylvanica         .......A.....C.....TG........A   [171]
M_petiolaris           ------------------NNNNN..NANN   [11]
M_punctata             ............C......G........A   [210]
M_reflexa              .......A.....C.....TG........A   [210]
M_rhomboidea           .......A.....C.....TG........A   [210]
M_rufidula             .......A.....C....NNNNNNNNNNNN   [130]
M_spicata              .....G.......C..............A   [210]
M_stellaris            ............C.........C.....A   [210]
M_tenuis               .......A.....C.....TG........A   [210]
M_texana               .......A.C...C.....TG........A   [85]
M_virginiensis         .......A.....C.....TG........A   [210]

[                              220       230       240]

M_tolmiei              ATTATTTATGCTAATGATTCCACCCCAAAT   [240]
Cascadia               .............................   [192]
M_arguta               .......C..A...........A.......   [240]
M_aprica               .......C..A...........A..A....   [213]
M_bryophora            ...C...CG.A...........A.NA....   [240]
M_californica          .......C..A...........A..A....   [240]
M_calycina             .......C..A...........A..A....   [240]
M_careyana             ----------------------------   [0]
M_ferruginea           ...C...CG.A...........A.AA....   [240]
M_foliolosa            ...C...CG.A...........A.AA....   [240]
M_fusca                .......C..A...........A..A....   [240]
M_hieracifolia         .......C..A...........A..A....   [240]
M_howellii             .......C..A...........A..A....   [240]
M_integrifolia         .......C..A...........A..A....   [240]
M_lyallii              .......C..A...........A..A....   [240]
M_micranthidifolia     ----------------------------   [0]
M_nidifica             .....C.C..A...........A..A....   [240]
M_nivalis              .......C..A...........A..A....   [240]
M_occidentalis         .......C..A...........A..A....   [216]
M_oregana              .......C..A...........A..A....   [240]
M_pensylvanica         .......C..A...........A..A....   [201]
M_petiolaris           NNNCCN.NN.T.NGNNNANTNNNAGG....   [41]
M_punctata             .......C..A...........A..A....   [240]
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M_reflexa             .......C..A..........A..A....   [240]
M_rhomboidea          .......C..A..........A..A....   [240]
M_rufidula            NNNNNNNNNNNNNNNNNNNNNNNNNNNNN   [160]
M_spicata             .......C..A.R........A..A.N..   [240]
M_stellaris           ...C...CG.A..........A.AA....   [240]
M_tenuis              .......C..A..........A..A....   [240]
M_texana              .......C..A..........A..A....   [115]
M_virginiensis        .......C..A..........A..A....   [240]

[                              250       260       270]

M_tolmiei             AAATTTTTGGGGCATAACAAGAATTTAGAT   [270]
Cascadia              ............................    [222]
M_arguta              ..........C.................    [270]
M_aprica              .C.........A....C............   [243]
M_bryophora           CC...G....N....N....N.T...C...   [270]
M_californica         .C.........A....C............   [270]
M_calycina            C.........................T...   [270]
M_careyana            ----------------------------    [0]
M_ferruginea          CC...G....T..........TA..C...   [270]
M_foliolosa           CC...G....T..........TA..C...   [270]
M_fusca               C......G............A.....T...   [270]
M_hieracifolia        .C.............C............   [270]
M_howellii            .C.............C............   [270]
M_integrifolia        .C.........A....C............   [270]
M_lyallii             .........................T...   [270]
M_micranthidifolia    ----------------------------    [0]
M_nidifica            .C.........A....C............   [270]
M_nivalis             .C.........A....C............   [270]
M_occidentalis        .C.........A....C............   [246]
M_oregana             .T.........A....C............   [270]
M_pensylvanica        .C.........A....C............   [231]
M_petiolaris          TCGA...AAAAATTC..TTGA...C---.G   [68]
M_punctata            C.........N.........A.....T...   [270]
M_reflexa             .C.............C............   [270]
M_rhomboidea          .C.............C............   [270]
M_rufidula            NNNNNNNNNNNNNNNNNNNNNNNNNNNNN   [190]
M_spicata             CC.....................R...T...   [270]
M_stellaris           CC...G....T.T.........TA..C...   [270]
M_tenuis              .C.............C............   [270]
M_texana              .C.........A....C............   [145]
M_virginiensis        .C......N...G...C..........N.   [270]

[                              280       290       300]

M_tolmiei             T------ATCAAATCATATCAGAGGGATTT   [294]
Cascadia              .------.....................    [246]
M_arguta              .ATCAAA.......G.............    [300]
M_aprica              .ATCAAA..A....G.............    [273]
M_bryophora           .CTCAAA.......G...AKS.......NN   [300]
M_californica         .ATCAAA..A....G.............    [300]
M_calycina            .ATCAAA.......G.............    [300]
M_careyana            ----------------------------    [0]
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M_ferruginea          .CTCAAA.......G..............   [300]
M_foliolosa           .CTCAAA.......G..............   [300]
M_fusca               .ATCAAA.......G..............   [300]
M_hieracifolia        .ATCAAA..A....G..............   [300]
M_howellii            .CTCAAA..A....G..............   [300]
M_integrifolia        .ATCAAA..A....G..............   [300]
M_lyallii             .ATCAAA.......G..............   [300]
M_micranthidifolia    ----------------------------   [0]
M_nidifica            .ATCAAA..A....G..............   [300]
M_nivalis             .ATCAAA..A....G..............   [300]
M_occidentalis        .ATCAAA..AC...G..............   [276]
M_oregana             .ATCAAA..A....G..............   [300]
M_pensylvanica        .ATCAAA..A....G..............   [261]
M_petiolaris          AGGGGAAT.TGC.GTTC..TG.T..A.A..   [98]
M_punctata            .ATCAAA.......G..............   [300]
M_reflexa             .ATCAAA.......G..............   [300]
M_rhomboidea          .ATCAAA..A....G..............   [300]
M_rufidula            NNNNNNNNNNNNNNNNNNNNNN.......   [220]
M_spicata             .ATCAWA.......G..............   [300]
M_stellaris           .ATCAAA......CG..............   [300]
M_tenuis              .ATCAAA.......G..............   [300]
M_texana              .ATAAAA..A....G..............   [175]
M_virginiensis        .ATCAAA..A....T..............   [300]

[                              310       320       330]

M_tolmiei             ACAGTCGTTGTGGAAATTCCATTTTATCTA   [324]
Cascadia              ............................   [276]
M_arguta              G.................A....CC....   [330]
M_aprica              G.................A.....C....   [303]
M_bryophora           NNN...N.N..........N....CC...   [330]
M_californica         G.................A.....C....   [330]
M_calycina            G.....A...........A.....CC...   [330]
M_careyana            ----------------------------   [0]
M_ferruginea          G.....A...........A.....CC...   [330]
M_foliolosa           G.....A...........A.....CC...   [330]
M_fusca               G.................A.....CC...   [330]
M_hieracifolia        G.................A.....C....   [330]
M_howellii            G.................A.....C....   [330]
M_integrifolia        G.................A.....C....   [330]
M_lyallii             G.......................CC...   [330]
M_micranthidifolia    ------------NNCGA.TAG.A..T..CT   [18]
M_nidifica            G.................A.....C....   [330]
M_nivalis             G.................A.....C....   [330]
M_occidentalis        G.................A.....C....   [306]
M_oregana             G.................A.....C....   [330]
M_pensylvanica        G.................A.....C....   [291]
M_petiolaris          C..T.--..CCTAC....TAG.A..T..CT   [126]
M_punctata            G.................A.....CC...   [330]
M_reflexa             G.................AN....C....   [330]
M_rhomboidea          G.................A.....C....   [330]
M_rufidula            G.................A.....C....   [250]
M_spicata             G.................A.....CC...   [330]
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M_stellaris           GN....A.....A.....CC...   [330]
M_tenuis              G.....A.....C....   [330]
M_texana              G.....A.....C....   [205]
M_virginiensis        G.....A.....C....   [330]

[                          340       350       360]

M_tolmiei             CGATTAGTATCTTCCTTAGAAAGGAAAGAA   [354]
Cascadia              ............................   [306]
M_arguta              ..........T.............G....   [360]
M_aprica              ..........T......NNNNNN..G....   [333]
M_bryophora           ST.....K..T........Y....R....R   [360]
M_californica         ..........T............CG....   [360]
M_calycina            ..........T...................   [360]
M_careyana            --------------------NG...N.NN.   [10]
M_ferruginea          .A........T........T..........   [360]
M_foliolosa           .A........T........T..........   [360]
M_fusca               ..........T...................   [360]
M_hieracifolia        ..........T.............G....   [360]
M_howellii            ..........T.............G....   [360]
M_integrifolia        ..........T.............G....   [360]
M_lyallii             ..........T...................   [360]
M_micranthidifolia    TAGAA..G.GGAGAAAATAGT.A.G.GAG.   [48]
M_nidifica            ..........T.............G....   [360]
M_nivalis             ..........T.............G....   [360]
M_occidentalis        ..........T.............G....   [336]
M_oregana             ..........T.............G....   [360]
M_pensylvanica        ..........T.............G....   [321]
M_petiolaris          TAG.A..G.C...AG.A---.G.A..GA..   [153]
M_punctata            ..........T...................   [360]
M_reflexa             ..........T.............G....   [360]
M_rhomboidea          ..........T.............G....   [360]
M_rufidula            ..........T.............G....   [280]
M_spicata             ..........T...................   [360]
M_stellaris           .A........T........T..........   [360]
M_tenuis              ..........T.............G....   [360]
M_texana              ..........T.............G....   [235]
M_virginiensis        ..........T.............G....   [360]

[                          370       380       390]

M_tolmiei             ATAGTAAAATCTCAGAATTTACGATCAATT   [384]
Cascadia              ............................   [336]
M_arguta              ............................   [390]
M_aprica              ............................   [363]
M_bryophora           .....CT...S...NW.............   [390]
M_californica         ............................   [390]
M_calycina            ............T...............   [390]
M_careyana            ............................   [40]
M_ferruginea          .....C........T.............   [390]
M_foliolosa           .....C........T.............   [390]
M_fusca               ............T...............   [390]
M_hieracifolia        ............................   [390]
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M_howellii              ............................   [390]
M_integrifolia          ............................   [390]
M_lyallii               ............T...............   [390]
M_micranthidifolia      ............................   [78]
M_nidifica              ............................   [390]
M_nivalis               ............................   [390]
M_occidentalis          ............................   [366]
M_oregana               ............................   [390]
M_pensylvanica          ............................   [351]
M_petiolaris            .....C........T..............   [183]
M_punctata              ............T...............   [390]
M_reflexa               ............................   [390]
M_rhomboidea            ............................   [390]
M_rufidula              ............................   [310]
M_spicata               ............T...............   [390]
M_stellaris             .....C........T..............   [390]
M_tenuis                ............................   [390]
M_texana                ............................   [265]
M_virginiensis          ............................   [390]

[                                400       410       420]

M_tolmiei               CATTCAATATTTCCTTTTTTAGAGGATCAT   [414]
Cascadia                ....N.....................G..   [366]
M_arguta                ......................A..CA..   [420]
M_aprica                ......................A..CA.N   [393]
M_bryophora             ............C.....C..A..CA..   [420]
M_californica           ......................A..CA..   [420]
M_calycina              ......................A..CA..   [420]
M_careyana              ..........-.......A-..A..CA..   [68]
M_ferruginea            ............C.....C..A..CA..   [420]
M_foliolosa             ..................C..A..CA..   [420]
M_fusca                 ......................A..CA..   [420]
M_hieracifolia          ......................A..CA..   [420]
M_howellii              .........NNNNNNNNNNNNNNNNNNN   [420]
M_integrifolia          ......................A..CA..   [420]
M_lyallii               ......................A..CA..   [420]
M_micranthidifolia      ..................A-..A..CA..   [107]
M_nidifica              ......................A..NNNN   [420]
M_nivalis               ......................A..CA..   [420]
M_occidentalis          ......................A..CA..   [396]
M_oregana               ......................A..CA..   [420]
M_pensylvanica          ......................A..CA..   [381]
M_petiolaris            ..................C..A..CA..   [213]
M_punctata              ......................A..CA..   [420]
M_reflexa               ......................A.NNNNN   [420]
M_rhomboidea            ......................A..CA..   [420]
M_rufidula              ......................A..CA..   [340]
M_spicata               .....C................A..CA..   [420]
M_stellaris             ..................C..A..CA..   [420]
M_tenuis                ......................A..CA..   [420]
M_texana                ..............NNNNNNNNNNNNN   [295]
M_virginiensis          ......................A..CA..   [420]
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[                              430       440       450]

M_tolmiei              TTTTTAC--ATTTAAA-TTATGTGTCAGAT   [441]
Cascadia               ......N--N......-.............   [393]
M_arguta               NNNNNNN--NNNNNNN-NNNNNNNNNNNNN   [447]
M_aprica               NNNNNNN--NNN....-.........G...   [420]
M_bryophora            .......--.......-.........G...   [447]
M_californica          .......--.......-.........G...   [447]
M_calycina             .......--.......-.........G...   [447]
M_careyana             ....ACA---......-.........G...   [94]
M_ferruginea           .......--.......-.........G...   [447]
M_foliolosa            .......--.......-.........G...   [447]
M_fusca                .......--.......-.........G...   [447]
M_hieracifolia         .......--.......-.........G...   [447]
M_howellii             NNNNNNN--NNN....-.........G...   [447]
M_integrifolia         .......--.......-.........G...   [447]
M_lyallii              .......--.......-.........T...   [447]
M_micranthidifolia     ....ACA---.....--.........G...   [132]
M_nidifica             NNNNNNN--NNN....-.........G...   [447]
M_nivalis              .....NN--NNNNNNN-NNNNNNNNNNNNN   [447]
M_occidentalis         .......--.......-...NNN...G...   [423]
M_oregana              .......--.......-.........G...   [447]
M_pensylvanica         NNNNNNN--NNNNNNN-NNNNNNNNNNNNN   [408]
M_petiolaris           .....TACA.......T.........G...   [243]
M_punctata             .......--.......-.........G...   [447]
M_reflexa              NNNNN..--.......-.........G...   [447]
M_rhomboidea           .......--.......-.........G...   [447]
M_rufidula             .......--.......-.........G...   [367]
M_spicata              .......--.......-.........G...   [447]
M_stellaris            .......--.......-.........G...   [447]
M_tenuis               .......--.......-.........G...   [447]
M_texana               NNNNNNN--NNNNNNN-NNNNNNNNNNNNN   [322]
M_virginiensis         .......--.......-.........G...   [447]

[                              460       470       480]

M_tolmiei              ATACTAATACCCCACCCAATCCATCTGGAA   [471]
Cascadia               ............................     [423]
M_arguta               NNNNNNNNNNNNNNNNNNNNNNNNNNNNN   [477]
M_aprica               ................C............   [450]
M_bryophora            ................C............   [477]
M_californica          ................C............   [477]
M_calycina             ................CC...........   [477]
M_careyana             ................C............   [124]
M_ferruginea           ................C............   [477]
M_foliolosa            ................C............   [477]
M_fusca                ..............T.CC...........   [477]
M_hieracifolia         ................C............   [477]
M_howellii             ................C............   [477]
M_integrifolia         ................C............   [477]
M_lyallii              ................CC...........   [477]
M_micranthidifolia     ................C............   [162]
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M_nidifica              .................C............   [477]
M_nivalis               NNNNNNNNNNNNNNNNNNNNNNNNN....   [477]
M_occidentalis          .................C............   [453]
M_oregana               .................C............   [477]
M_pensylvanica          NNNNNNNNNNNNNNNNNNNNNNNNNNNNN   [438]
M_petiolaris            .................C............   [273]
M_punctata              .........NN....T.CC...........   [477]
M_reflexa               .................C........NNNN   [477]
M_rhomboidea            .................C............   [477]
M_rufidula              .................C............   [397]
M_spicata               .................CC...........   [477]
M_stellaris             .................C............   [477]
M_tenuis                .................C............   [477]
M_texana                NNNNNNNNNNNNNNNNNNNNNNNNNNN...   [352]
M_virginiensis          .................C............   [477]

[                                490       500       510]

M_tolmiei               ATATTGGTTCAAACTCTTCGCTACTGGGTG   [501]
Cascadia                .............................   [453]
M_arguta                NNNNNNNNNNNNNNNNNNNNNNNNNNNNN   [507]
M_aprica                .............................   [480]
M_bryophora             ........................C.....   [507]
M_californica           .............................   [507]
M_calycina              ..C.........................   [507]
M_careyana              .............................   [154]
M_ferruginea            ..C.............G..T...C....A   [507]
M_foliolosa             ..C.............G..T...C....A   [507]
M_fusca                 ..C.........................   [507]
M_hieracifolia          .............................   [507]
M_howellii              .............................   [507]
M_integrifolia          .............................   [507]
M_lyallii               ..C.........................   [507]
M_micranthidifolia      .............................   [192]
M_nidifica              .............................   [507]
M_nivalis               .............................   [507]
M_occidentalis          .............................   [483]
M_oregana               .............................   [507]
M_pensylvanica          NNNNNNNNNNNNN...............   [468]
M_petiolaris            ..C.............G..T...C..A.A   [303]
M_punctata              ..C.........................   [507]
M_reflexa               N............................   [507]
M_rhomboidea            .............................   [507]
M_rufidula              .............................   [427]
M_spicata               ..C.........................   [507]
M_stellaris             ..C.............G..T..TC....A   [507]
M_tenuis                .............................   [507]
M_texana                .............................   [382]
M_virginiensis          .................N...........   [507]

[                                520       530       540]

M_tolmiei               AAAGATGCCTCTTCTTTACATTTATTACGA   [531]
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```
Cascadia              ..............................   [483]
M_arguta              NNNNNNNNNNNNNNNNNNNNNNNNNNNNNN   [537]
M_aprica              ................G.............   [510]
M_bryophora           ........T.....A..G.............   [537]
M_californica         ................G.............   [537]
M_calycina            ................G.............   [537]
M_careyana            ................G.C...........   [184]
M_ferruginea          ........T........G.............   [537]
M_foliolosa           ........T........G.............   [537]
M_fusca               ................G.............   [537]
M_hieracifolia        ................G.............   [537]
M_howellii            ................G.............   [537]
M_integrifolia        ................G.............   [537]
M_lyallii             ..C.............G.............   [537]
M_micranthidifolia    ................G.............   [222]
M_nidifica            ................G.............   [537]
M_nivalis             ................G.............   [537]
M_occidentalis        ................G.............   [513]
M_oregana             ................G.............   [537]
M_pensylvanica        ................G.............   [498]
M_petiolaris          ........T........G.............   [333]
M_punctata            ................G.............   [537]
M_reflexa             ................G.............   [537]
M_rhomboidea          ................G.............   [537]
M_rufidula            ................G.............   [457]
M_spicata             ................G.............   [537]
M_stellaris           ........T........G.............   [537]
M_tenuis              ................G.............   [537]
M_texana              ................G.............   [412]
M_virginiensis        ................G.............   [537]

[                                550       560       570]

M_tolmiei             TTCTTTCTCTATGAGTATCAGAATTGGAAT   [561]
Cascadia              ..............................   [513]
M_arguta              NNNNNNNNNNNNNNNNNNNNNNNNNNNNNC.   [567]
M_aprica              ........................C.A.C.   [540]
M_bryophora           ........................N.....   [567]
M_californica         ........................C.A.C.   [567]
M_calycina            ........................C..N..   [567]
M_careyana            ........................C.A.C.   [214]
M_ferruginea          ........................A.....   [567]
M_foliolosa           ........................A.....   [567]
M_fusca               ........................C.....   [567]
M_hieracifolia        ........................C.A.C.   [567]
M_howellii            ........................C.A.C.   [567]
M_integrifolia        ........................C.A.C.   [567]
M_lyallii             ..............................   [567]
M_micranthidifolia    ........................C.A.C.   [252]
M_nidifica            ........................C.A.C.   [567]
M_nivalis             ........................C.A.C.   [567]
M_occidentalis        ........................C.A.C.   [543]
M_oregana             ........................C.A.C.   [567]
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M_pensylvanica        .....................C.A.C.   [528]
M_petiolaris          .....................A.....   [363]
M_punctata            .....................C.....   [567]
M_reflexa             .....................NC.A.C.   [567]
M_rhomboidea          .....................C.A.C.   [567]
M_rufidula            .....................C.A.C.   [487]
M_spicata             .....................C.....   [567]
M_stellaris           .....................A.....   [567]
M_tenuis              .....................C.A.C.   [567]
M_texana              .....................C.A.C.   [442]
M_virginiensis        ..................T..C.A.C.   [567]

[                              580       590       600]

M_tolmiei             TGTCTTAGTATTCCAACTCAAAAAAAATCA   [591]
Cascadia              ................C.........   [543]
M_arguta              A.....T..C.......C.......GA.   [597]
M_aprica              A.....T..C.......C.........   [570]
M_bryophora           A.....TG.C.......C...G...G.C   [597]
M_californica         A.....T..C.T.....C.......G..   [597]
M_calycina            A.....T..C.......C.........   [597]
M_careyana            A.....T..C.......C.........   [244]
M_ferruginea          A.....CTG.C..T.....C.......GA.   [597]
M_foliolosa           A.....CTG.C..T.....C.......GA.   [597]
M_fusca               A.....T..C.......C.......G..   [597]
M_hieracifolia        A.....T..C.......C.........   [597]
M_howellii            A.....T..C.......C.........   [597]
M_integrifolia        A.....T..C.T.....C.......G..   [597]
M_lyallii             A.....T..C.......C.........   [597]
M_micranthidifolia    A.....T..C.......C.........   [282]
M_nidifica            A.....T..C.T..G...C......TG..   [597]
M_nivalis             A.....T..C.T.....C.........   [597]
M_occidentalis        A.....T..C.T.....C.......G..   [573]
M_oregana             A.....T..C.......C.........   [597]
M_pensylvanica        A.....T..C.......C.........   [558]
M_petiolaris          A.....CTG.C..T.....C.......GA.   [393]
M_punctata            A.....T..C.......C.........   [597]
M_reflexa             A.....T..C.......C.........   [597]
M_rhomboidea          A.....T..C....G...C.........   [597]
M_rufidula            A.....T..C.......C.......G..   [517]
M_spicata             A.....T..C.......C.........C   [597]
M_stellaris           A.....CTG.C..T.....C.......GA.   [597]
M_tenuis              A.....T..C.......C.........   [597]
M_texana              A.....T..C.T.....C.........   [472]
M_virginiensis        A.....T..C.................   [597]

[                              610       620       630]

M_tolmiei             ATTTCCATTGTTTCAAAAAGGAATCAAAGA   [621]
Cascadia              ..............................   [573]
M_arguta              ..............................   [627]
M_aprica              ................G.......C...   [600]
M_bryophora           ..............................   [627]
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M_californica          .................G.......C...   [627]
M_calycina             ............................   [627]
M_careyana             .................G.......C...   [274]
M_ferruginea           ............................   [627]
M_foliolosa            .......G....................   [627]
M_fusca                ............................   [627]
M_hieracifolia         .........A.......G.......C...   [627]
M_howellii             .................G.......C...   [627]
M_integrifolia         .................G.......C...   [627]
M_lyallii              .................A..........   [627]
M_micranthidifolia     .................G.......C...   [312]
M_nidifica             .................G.......C...   [627]
M_nivalis              .................G.......C...   [627]
M_occidentalis         .................G.......C...   [603]
M_oregana              .................G.......C...   [627]
M_pensylvanica         .................G.......C...   [588]
M_petiolaris           ............................   [423]
M_punctata             ............................   [627]
M_reflexa              .................G.......C...   [627]
M_rhomboidea           .................G.......C...   [627]
M_rufidula             .................G.......C...   [547]
M_spicata              ............................   [627]
M_stellaris            ............................   [627]
M_tenuis               .................G.......C...   [627]
M_texana               .................G.......C...   [502]
M_virginiensis         .................G.......C...   [627]

[                              640       650       660]

M_tolmiei              TTATTCTTGTTTCTATATAATTCTTTTGTA   [651]
Cascadia               ............................   [603]
M_arguta               .........A...NNNNNNNNNNNNNNNN   [657]
M_aprica               .........A..............A.T..   [630]
M_bryophora            ..................Y.M..A....   [657]
M_californica          .........A..............A.T..   [657]
M_calycina             .........A..............A....   [657]
M_careyana             .........A..............A.T..   [304]
M_ferruginea           ............C.......A..A....   [657]
M_foliolosa            ............C.......A..A....   [657]
M_fusca                .........A..............A....   [657]
M_hieracifolia         .........A..............A.T..   [657]
M_howellii             ..C......A..............A.T..   [657]
M_integrifolia         .........A..............A.T..   [657]
M_lyallii              .........AA.............A.NNN   [657]
M_micranthidifolia     .........A..............A....   [342]
M_nidifica             .........A..............A.T..   [657]
M_nivalis              .........A..............A.T..   [657]
M_occidentalis         .........A..............A.T..   [633]
M_oregana              .........A..............A.T..   [657]
M_pensylvanica         .........A..............A.T..   [618]
M_petiolaris           ............C..........A....   [453]
M_punctata             .........A..............A....   [657]
M_reflexa              .........A..............A.N..   [657]
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M_rhomboidea          .........A..............A.T..   [657]
M_rufidula            .........A..............A.T..   [577]
M_spicata             .........A.....G.....N...A....   [657]
M_stellaris           ..............C.......A..A....   [657]
M_tenuis              .........A..............A....   [657]
M_texana              .........A..............A.T..   [532]
M_virginiensis        .........A..........N....A.T..   [657]

[                                670       680       690]

M_tolmiei             TGTGAATACGAATCCATCTTTATTTTTCTT   [681]
Cascadia              ............................    [633]
M_arguta              NNNNNNNNNNNNNNNNNNNNNNNNNNNN   [687]
M_aprica              ..............T.....C......A..   [660]
M_bryophora           ..............T.....C....-----   [682]
M_californica         ..............T.....C......A..   [687]
M_calycina            ..............T.....C......N..   [687]
M_careyana            ..............T.....C......A..   [334]
M_ferruginea          ..............T.....C.........   [687]
M_foliolosa           ..............T.NNNNNNNN......   [687]
M_fusca               ............N..T....NNNN....A..   [687]
M_hieracifolia        ..............T.....A......A..   [687]
M_howellii            ..............T.....C......A..   [687]
M_integrifolia        ..............T.....C......A..   [687]
M_lyallii             NNNNNNNNNNNN..T.....NN.....A..   [687]
M_micranthidifolia    ..............T.....C......A..   [372]
M_nidifica            ..............T.....C......A..   [687]
M_nivalis             ..............T.....C......A..   [687]
M_occidentalis        ..............T.....C......A..   [663]
M_oregana             ..............T.....C......A..   [687]
M_pensylvanica        ....NNNNNNNNNNNNNNNNNNNNNN..   [648]
M_petiolaris          ........T.....T.....C.........   [483]
M_punctata            ..............T.....C......A..   [687]
M_reflexa             ............NNNNNNNNNNNN..A..   [687]
M_rhomboidea          ..............T.....C......A..   [687]
M_rufidula            ..............T.....C......A..   [607]
M_spicata             .....N.....----------------------   [668]
M_stellaris           ..............T.....C.........   [687]
M_tenuis              ..............T.....C......NNN   [687]
M_texana              ..............T.....C......A..   [562]
M_virginiensis        ..............T.....C.NNN..A..   [687]

[                                700       710       720]

M_tolmiei             TGTAACCAATCTTCTCATTTACGATCAACA   [711]
Cascadia              ............................    [663]
M_arguta              NNNNNNNNNNN.................T.   [717]
M_aprica              ............................T.   [690]
M_bryophora           ----------------------------   [682]
M_californica         ............................T.   [717]
M_calycina            ..NNN...................N...T.   [717]
M_careyana            ............................T.   [364]
M_ferruginea          ....G........T......G.......T.   [717]
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M_foliolosa           .............T......G.......T.   [717]
M_fusca               ............................T.   [717]
M_hieracifolia        ............................T.   [717]
M_howellii            ............................T.   [717]
M_integrifolia        ............................T.   [717]
M_lyallii             ............................T.   [717]
M_micranthidifolia    ............................T.   [402]
M_nidifica            .............NNNNNNNN.......T.   [717]
M_nivalis             ..........C..................T.   [717]
M_occidentalis        ............................T.   [693]
M_oregana             ............................T.   [717]
M_pensylvanica        ............................T.   [678]
M_petiolaris          .............T......G.......T.   [513]
M_punctata            ............................T.   [717]
M_reflexa             ............................T.   [717]
M_rhomboidea          ............................T.   [717]
M_rufidula            ............................T.   [637]
M_spicata             ------------------------------   [668]
M_stellaris           .............T......G.......T.   [717]
M_tenuis              NNNNNNNNNNNNNN..............T.   [717]
M_texana              ............................T.   [592]
M_virginiensis        .....N......................T.   [717]

[                                730       740       750]

M_tolmiei             TCTTCTGGAACTCTTTTTGAGCGAATATAT   [741]
Cascadia              .............................   [693]
M_arguta              .............................   [747]
M_aprica              ......................A......   [720]
M_bryophora           ------------------------------   [682]
M_californica         ......................A......   [747]
M_calycina            ....N.......................   [747]
M_careyana            ......................A......   [394]
M_ferruginea          ....T......................C...   [747]
M_foliolosa           ....T......................CC..   [747]
M_fusca               .............................   [747]
M_hieracifolia        ......................A......   [747]
M_howellii            ......................A......   [747]
M_integrifolia        ......................A......   [747]
M_lyallii             .............................   [747]
M_micranthidifolia    ......................A......   [432]
M_nidifica            ......................A......   [747]
M_nivalis             ......................A......   [747]
M_occidentalis        ......................A......   [723]
M_oregana             ......................A......   [747]
M_pensylvanica        ......................A......   [708]
M_petiolaris          ....T......................C...   [543]
M_punctata            .............................   [747]
M_reflexa             ......................A......   [747]
M_rhomboidea          ......................A......   [747]
M_rufidula            ......................A......   [667]
M_spicata             ------------------------------   [668]
M_stellaris           ....T......................C...   [747]
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M_tenuis                .....................A.........   [747]
M_texana                .....................A.........   [622]
M_virginiensis          .....................A.........   [747]

[                                760       770       780]

M_tolmiei               TTCTGTAGAAAAATAAAGCATCTTATCAAA   [771]
Cascadia                ............................     [723]
M_arguta                ....A.G..........A......G.AG..   [777]
M_aprica                ..T.ACG..........AA.....G.AG..   [750]
M_bryophora             ----------------------------     [682]
M_californica           ..T.ACG..........AA.....G.AG..   [777]
M_calycina              ....A.G..........AA..A.NN.AG..   [777]
M_careyana              ....ACG..........A......G.AG..   [424]
M_ferruginea            ....A.G..........A......G.AG..   [777]
M_foliolosa             ....A.G..........A......G.AG..   [777]
M_fusca                 ....A.G..........A......G.AG..   [777]
M_hieracifolia          ....ACG..........A......G.AG..   [777]
M_howellii              ....ACG..........A......G.AG..   [777]
M_integrifolia          ..T.ACG..........AA.....G.AG..   [777]
M_lyallii               ....A.G..........A......G.AG..   [777]
M_micranthidifolia      ....ACG..........A......G.AG..   [462]
M_nidifica              ..TCACG..........A......G.AG..   [777]
M_nivalis               ....ACG..........A......G.AG..   [777]
M_occidentalis          ..T.ACG..........AA.....G.AG..   [753]
M_oregana               ..T.ACG..........AA.....G.AG..   [777]
M_pensylvanica          ....ACG..........A......G.AG..   [738]
M_petiolaris            ....A.G..........A......G.AG..   [573]
M_punctata              ....A.G..........A......G.AG..   [777]
M_reflexa               ....ACG..........A......G.AG..   [777]
M_rhomboidea            ..TCACG..........A......G..G..   [777]
M_rufidula              ..T.ACG..........AA.....G.AG..   [697]
M_spicata               ----------------------------     [668]
M_stellaris             ....A.G..........A......G.AG..   [777]
M_tenuis                ....ACG..........A......G.AG..   [777]
M_texana                ....ACG..........A......G.AG..   [652]
M_virginiensis          ....ACG..........A......G.AG..   [777]

[                                790       800       810]

M_tolmiei               GTATTTTCTAATGATTTTCCTATCGTCCTA   [801]
Cascadia                ............................     [753]
M_arguta                ..C....A..C...........CG......   [807]
M_aprica                ..C....A..C...........CT......   [780]
M_bryophora             ----------------------------     [682]
M_californica           ..C....A..C...........CT......   [807]
M_calycina              ..C....A..C...........C.......   [807]
M_careyana              ..C....A..C...........CT......   [454]
M_ferruginea            ........C.C.T.........C.......   [807]
M_foliolosa             ........C.C...........CT..N...   [807]
M_fusca                 ..C....A..C...........C.......   [807]
M_hieracifolia          ..C....A..C...........CT......   [807]
M_howellii              ..CG...A..C...........CT......   [807]
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M_integrifolia        ..C....A..C...........CT......   [807]
M_lyallii             ..C....A..C.......N...C..C....   [807]
M_micranthidifolia    ..C....A..C...........CT......   [492]
M_nidifica            ..C....A..C...........CT......   [807]
M_nivalis             ..C....A..C...........CT......   [807]
M_occidentalis        ..C....A..C...........CT......   [783]
M_oregana             ..C....A..C...........CT......   [807]
M_pensylvanica        ..C....A..C...........CT......   [768]
M_petiolaris          ........C.C...........C.......   [603]
M_punctata            ..C....A..C...........C.......   [807]
M_reflexa             ..C....A..C...........CT......   [807]
M_rhomboidea          ..C....A..C...........CT......   [807]
M_rufidula            ..C....A..C...........CT......   [727]
M_spicata             ------------------------------   [668]
M_stellaris           ........C.C...........C.......   [807]
M_tenuis              ..C....A..C...........CT......   [807]
M_texana              ..C....A..C...........CT......   [682]
M_virginiensis        ..C....A..C...........CT......   [807]

[                              820       830       840]

M_tolmiei             TGGTTATTCAAAGNNCCATTCATACATTAT   [831]
Cascadia              ...N.........AT...............   [783]
M_arguta              .....G.......AC.....T..G......   [837]
M_aprica              .....G.......CC..G..T..GT.....   [810]
M_bryophora           ------------------------------   [682]
M_californica         .....G.......CC..G..T..GT.....   [837]
M_calycina            .....G.......AC..G..TN.GTN....   [837]
M_careyana            .....G.......CC..G..T..GT.....   [484]
M_ferruginea          .....G.......AC..G..T.........   [837]
M_foliolosa           .....G.......AC..G..T.........   [837]
M_fusca               .....G.......AC..G..T..G......   [837]
M_hieracifolia        .....G.......CC..G..T..GT.....   [837]
M_howellii            .....G.......CC..G..T..GT.....   [837]
M_integrifolia        .....G.......CC..G..T..GT.....   [837]
M_lyallii             .....G.......AC..G..T..G......   [837]
M_micranthidifolia    .....G.......CC..G..T..GT.....   [522]
M_nidifica            .....G.......CC..G..T..GT.....   [837]
M_nivalis             .....G.......CC..G..T..GT.....   [837]
M_occidentalis        .....G.......CC..G..T..GT.....   [813]
M_oregana             .....G.......CC..G..T..GT.....   [837]
M_pensylvanica        .....G.......CC..G..T..GT.....   [798]
M_petiolaris          .....G.......AC..G..T.........   [633]
M_punctata            .....G.......TC..G..T..G......   [837]
M_reflexa             .....G.......CC..G..T..GT.....   [837]
M_rhomboidea          .....G.......CC..G..T..GT.....   [837]
M_rufidula            .....G.......CC..G..T..GT.....   [757]
M_spicata             ------------------------------   [668]
M_stellaris           .....G.......AC..G..T.........   [837]
M_tenuis              .....G.......CC..G..T..GT.....   [837]
M_texana              .....G.......CC..G..T..GT.....   [712]
M_virginiensis        .....G.......CC..G..T..GT.....   [837]
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[                              850       860       870]

M_tolmiei              GTTAGGTATCAAGAAAAATCAATTCTGGCA   [861]
Cascadia               ............................    [813]
M_arguta               .....A.......G..............    [867]
M_aprica               .....A.......G.............N   [840]
M_bryophora            ----------------------------    [682]
M_californica          .....A.......G..............    [867]
M_calycina             .....A.......G..............    [867]
M_careyana             .....A.......G..............    [514]
M_ferruginea           ............G..............    [867]
M_foliolosa            ............G...G............    [867]
M_fusca                .....A.......G..............    [867]
M_hieracifolia         .....A.......G..............    [867]
M_howellii             .....A.......G..............    [867]
M_integrifolia         .....A.......G..............    [867]
M_lyallii              .....A......AG..............    [867]
M_micranthidifolia     .....A.......G..............    [552]
M_nidifica             .....A.......G..............    [867]
M_nivalis              .....A.......G..............    [867]
M_occidentalis         .....A.......G..............    [843]
M_oregana              .....A.......G..............    [867]
M_pensylvanica         .....A.......G..............    [828]
M_petiolaris           ..........G..G...G..........    [663]
M_punctata             ...C.A.......G..............    [867]
M_reflexa              .....A.......G..............    [867]
M_rhomboidea           .....A.......G..............    [867]
M_rufidula             .....A.......G..............    [787]
M_spicata              ----------------------------    [668]
M_stellaris            ............G..............    [867]
M_tenuis               .....A.......G..............    [867]
M_texana               .....A.......G..............    [742]
M_virginiensis         .....A.......G..............    [867]

[                               880       890       900]

M_tolmiei              TCAAAAGGTACGCCTCTTCTTCTCAATAAA   [891]
Cascadia               ..................NN........    [843]
M_arguta               .....G...G.........G..G......    [897]
M_aprica               NNNNNN...G.........G..G......    [870]
M_bryophora            ----------------------------    [682]
M_californica          .....T...G.........G..G......    [897]
M_calycina             .........G.........G..G......    [897]
M_careyana             .....T...G.........G..G......    [544]
M_ferruginea           ........GG.........G..G......    [897]
M_foliolosa            ........GG.........G..G......    [897]
M_fusca                .........G.........G..G......    [897]
M_hieracifolia         .....T...G.........G..G......    [897]
M_howellii             .....T...G.........G..G......    [897]
M_integrifolia         .....T...G.........G..G......    [897]
M_lyallii              .........G.........G..G......    [897]
M_micranthidifolia     .....T...G.........G..G......    [582]
M_nidifica             .....T...G.T.......G..G......    [897]
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M_nivalis             ...NNNNNNNNNNNNNNNNNNNNNNNNN   [897]
M_occidentalis        .....T...G.........G..G......   [873]
M_oregana             ...NNNNNNG.........G..G......   [897]
M_pensylvanica        ..T..T...G.........G..G......   [858]
M_petiolaris          ........GG.........G..G......   [693]
M_punctata            .........G.........G..G......   [897]
M_reflexa             .....T...G.........G..G......   [897]
M_rhomboidea          .....T...G.........G..G......   [897]
M_rufidula            .....T...G.........G..G......   [817]
M_spicata             ----------------------------   [668]
M_stellaris           ........GG..T.......G..G......   [897]
M_tenuis              .....T...G.........G..G......   [897]
M_texana              .....T...G.........G..G......   [772]
M_virginiensis        .....T...G.........G..G......   [897]

[                              910       920       930]

M_tolmiei             TGGAAATATTACCTTCTAAATTGCTGGCAA   [921]
Cascadia              ............................   [873]
M_arguta              ..............G......T.......   [927]
M_aprica              ..............G......T.......   [900]
M_bryophora           ----------------------------   [682]
M_californica         ..............G......T.......   [927]
M_calycina            ..............G......T.......   [927]
M_careyana            ..............G......T.......   [574]
M_ferruginea          ..............G......T.......   [927]
M_foliolosa           ..............G......T.......   [927]
M_fusca               ..............G......T.......   [927]
M_hieracifolia        ..............G......T.......   [927]
M_howellii            ..............G......T.......   [927]
M_integrifolia        ..............G......T.......   [927]
M_lyallii             ..............G......T.......   [927]
M_micranthidifolia    ..............G......T.......   [612]
M_nidifica            ..............G......T.......   [927]
M_nivalis             NNNNNNNNNNNNNNNNNNNNNNN.......   [927]
M_occidentalis        ..............G......T.......   [903]
M_oregana             ..............G......T.......   [927]
M_pensylvanica        ...NNNNNNNNN...G......T.......   [888]
M_petiolaris          ..............G......T.......   [723]
M_punctata            ..............G......T.......   [927]
M_reflexa             ..............G......T.......   [927]
M_rhomboidea          ..............G......T.......   [927]
M_rufidula            ..............G......T.......   [847]
M_spicata             ----------------------------   [668]
M_stellaris           ........C......G......T.......   [927]
M_tenuis              ..............G......T.......   [927]
M_texana              ..............G......T...T---   [799]
M_virginiensis        ..............G......T.......   [927]

[                              940       950       960]

M_tolmiei             TGTAATTTTTATGTATGGTCTCAGCCAGAA   [951]
Cascadia              ...........N..................   [903]
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M_arguta              ..........CC.........AT...G.   [957]
M_aprica              ..........CC.........A....G.   [930]
M_bryophora           ------------------------------   [682]
M_californica         ..........CC.........A....G.   [957]
M_calycina            ..........CC.........AN...G.   [957]
M_careyana            ..........CC.........A....G.   [604]
M_ferruginea          .........G.CC.........A....G.   [957]
M_foliolosa           .........G.CC.........A....G.   [957]
M_fusca               ..........CC.........AT...G.   [957]
M_hieracifolia        ..........CC.........A....G.   [957]
M_howellii            ..........CC.........A....G.   [957]
M_integrifolia        ..........CC.........A....G.   [957]
M_lyallii             ..........CC.........AT...G.   [957]
M_micranthidifolia    ..........CC.........A....G.   [642]
M_nidifica            ..........CC.........A....G.   [957]
M_nivalis             ..........CC.........A......   [957]
M_occidentalis        ..........CC.........A....G.   [933]
M_oregana             ..........CC.........A....G.   [957]
M_pensylvanica        ..........CC.........A....G.   [918]
M_petiolaris          .........G.CC.........A....G.   [753]
M_punctata            ..........CC.........AT...G.   [957]
M_reflexa             ..........CC.........A......   [957]
M_rhomboidea          ..........CC.........A....G.   [957]
M_rufidula            ..........CC.........A....G.   [877]
M_spicata             ------------------------------   [668]
M_stellaris           .........G.CC.........A....G.   [957]
M_tenuis              ..........CC.........A......   [957]
M_texana              ------------------------------   [799]
M_virginiensis        ..........CC.........A....G.   [957]

[                              970       980       990]

M_tolmiei             AGGATCCATATAAACCAATTATCCAAAAGA   [981]
Cascadia              ........................NN...   [933]
M_arguta              ............................AT   [987]
M_aprica              ............................AT   [960]
M_bryophora           ------------------------------   [682]
M_californica         ............................AT   [987]
M_calycina            ............................AT   [987]
M_careyana            ............................AT   [634]
M_ferruginea          .T...G................A....AT   [987]
M_foliolosa           .T...G................A....AT   [987]
M_fusca               ............................AT   [987]
M_hieracifolia        .....T......................AT   [987]
M_howellii            ............................AT   [987]
M_integrifolia        ............................AT   [987]
M_lyallii             ............................AT   [987]
M_micranthidifolia    .........C..................AT   [672]
M_nidifica            ............................AT   [987]
M_nivalis             ............................AT   [987]
M_occidentalis        ............................AT   [963]
M_oregana             ............................AT   [987]
M_pensylvanica        ............................AT   [948]
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M_petiolaris          .T...G.................A....AT   [783]
M_punctata            ............................AT   [987]
M_reflexa             ............................AT   [987]
M_rhomboidea          ............................AT   [987]
M_rufidula            ............................AT   [907]
M_spicata             ----------------------------   [668]
M_stellaris           .T...G.................A....AT   [987]
M_tenuis              ............................AT   [987]
M_texana              ----------------------------   [799]
M_virginiensis        ............................AT   [987]

[                              1000      1010      1020]

M_tolmiei             CCCCTCGACTTTCTGGGCTATCTTTCTAGT   [1011]
Cascadia              ............................    [963]
M_arguta              T.....A..................G...   [1017]
M_aprica              T.T...A..................G...   [990]
M_bryophora           ----------------------------   [682]
M_californica         T.T...A..................G...   [1017]
M_calycina            T.....A..................G...   [1017]
M_careyana            T.T...A..................G...   [664]
M_ferruginea          T......................G..G..G   [1017]
M_foliolosa           T......................G..G..G   [1017]
M_fusca               T.....A..................G...   [1017]
M_hieracifolia        T.T...A..................G...   [1017]
M_howellii            T.T...A..................G...   [1017]
M_integrifolia        T.T...A..................G...   [1017]
M_lyallii             T.....A..................G...   [1017]
M_micranthidifolia    T.T...A..................G...   [702]
M_nidifica            T.T...A..................G...   [1017]
M_nivalis             T.T...A..................G...   [1017]
M_occidentalis        T.T...A..................G...   [993]
M_oregana             T.T...A..................G...   [1017]
M_pensylvanica        T.T...A..................G...   [978]
M_petiolaris          T......................G..G..G   [813]
M_punctata            T.....A..................G...   [1017]
M_reflexa             T.T...A..................G...   [1017]
M_rhomboidea          T.T...A..................G...   [1017]
M_rufidula            T.T...A..................G...   [937]
M_spicata             ----------------------------   [668]
M_stellaris           T......................G..G..G   [1017]
M_tenuis              T.T...A..................G...   [1017]
M_texana              ----------------------------   [799]
M_virginiensis        T.T...A..................G...   [1017]

[                              1030      1040      1050]

M_tolmiei             ATGCGACTAAATCCTTCAGCAGTACGGAGT   [1041]
Cascadia              ............................    [993]
M_arguta              G.......C............G.........   [1047]
M_aprica              G.....T.............G.........   [1020]
M_bryophora           ----------------------------   [682]
M_californica         G.....T.............G.........   [1047]
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M_calycina            G.................G.........   [1047]
M_careyana            G.....T.............G.........   [694]
M_ferruginea          ........C....T................   [1047]
M_foliolosa           ........C....T................   [1047]
M_fusca               G.........C.........G.........   [1047]
M_hieracifolia        G.....T.............G.........   [1047]
M_howellii            G.....T.............G.........   [1047]
M_integrifolia        G.....T.............G.........   [1047]
M_lyallii             G........N..........G.........   [1047]
M_micranthidifolia    G.................G.........   [732]
M_nidifica            G.....T.............G.........   [1047]
M_nivalis             G.....T.............G.........   [1047]
M_occidentalis        G.....T.............G.........   [1023]
M_oregana             G.....T.............G.........   [1047]
M_pensylvanica        G.....T.............G.........   [1008]
M_petiolaris          ........C....T................   [843]
M_punctata            G.................G.........   [1047]
M_reflexa             G.....T.............G.........   [1047]
M_rhomboidea          G.....T.............G.........   [1047]
M_rufidula            G.....T.............G.........   [967]
M_spicata             ------------------------------   [668]
M_stellaris           ........C....T................   [1047]
M_tenuis              G.....T.............G.........   [1047]
M_texana              ------------------------------   [799]
M_virginiensis        G....NT.............G.........   [1047]

[                              1060]

M_tolmiei             CAACTGCTAGAAA   [1054]
Cascadia              .............   [1006]
M_arguta              ...A.........   [1060]
M_aprica              ...A..T......   [1033]
M_bryophora           -------------   [682]
M_californica         ...A..T......   [1060]
M_calycina            ...A.........   [1060]
M_careyana            ...A..T......   [707]
M_ferruginea          ...A.........   [1060]
M_foliolosa           ...A.........   [1060]
M_fusca               ...A.........   [1060]
M_hieracifolia        ...A..T......   [1060]
M_howellii            ...A..T......   [1060]
M_integrifolia        ...A..T......   [1060]
M_lyallii             ...A.........   [1060]
M_micranthidifolia    ...A..T......   [745]
M_nidifica            ...A..T......   [1060]
M_nivalis             ...A..T......   [1060]
M_occidentalis        ...A..T......   [1036]
M_oregana             ...A..T......   [1060]
M_pensylvanica        ...A..T......   [1021]
M_petiolaris          ...A.........   [856]
M_punctata            ...A.........   [1060]
M_reflexa             ...A..T......   [1060]
M_rhomboidea          ...A..T......   [1060]
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M_rufidula            ...A..T......   [980]
M_spicata             ------------    [668]
M_stellaris           ...A.........   [1060]
M_tenuis              ...A..T......   [1060]
M_texana              ------------    [799]
M_virginiensis        ...A..T......   [1060]
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