# A NEW SPECIES OF *LYCOPUS* (LAMIACEAE) FROM THE WEST GULF COASTAL PLAIN AND A FUTILE EFFORT IN BARCODING

## **AARON FLODEN**

Missouri Botanical Garden 4334 Shaw Blvd St. Louis, MO 63110 afloden@mobot.org

# ABSTRACT

**Lycopus glandulosus** Floden, **sp. nov.**, is described and illustrated from the West Gulf Coastal Plain of eastern Texas, northwest Louisiana, and southwest Arkansas. This species differs from *L. rubellus* in its densely short stipitate glandular indumentum, eucalyptus-scented foliage, and narrower subulate calyx lobes. In addition, an initial effort at barcoding of all native species of *Lycopus* in eastern North America is presented, using the nuclear ribosomal ITS and several chloroplast intergenic spacers. A key to the species is included.

*Lycopus* L. (Lamiaceae: Lycopinae) is a genus comprising ca. 16 herbaceous species with a distribution that is largely circumboreal but also disjunct to Australia (Henderson 1962; Moon & Hong 2006). *Lycopus* is most diverse in North America and East Asia (Henderson 1962). North America harbors 12 of the 16 recognized taxa (Henderson 1962). The International Plant Names Index (IPNI 2022) lists 141 specific and infraspecific names within the genus, which is a reflection of the morphological plasticity most notable in the sinuate-leaved taxa, where leaf shapes differ between the proximal and distal portions of the plant.

In North America most species are broadly distributed over non-physiographically defined areas and ubiquitously in wet habitats. *Lycopus uniflorus* Michx. is widely distributed across the northern portion of North America from the Pacific coast eastward into New England and down the Appalachian Mountains (Henderson 1962). *Lycopus virginicus* L. is widely distributed across eastern North America and is the most frequently encountered species. In contrast some species have restricted ranges and some exhibit highly disjunct occurrences between two regions. *Lycopus amplectens* Raf. and *L. angustifolius* Ell. occur mostly along the Coastal Plain, but *L. amplectens* has a tertiary disjunction — along the coast of New England, the Southeastern Coastal Plain, and also in upper Indiana (Henderson 1962). *Lycopus angustifolius* is only found within the Southeastern Coastal Plain. Narrow endemism is known only in *L. laurentianus* Roll.-Germ. and *L. cokeri* Ahles ex Sorrie, the former native to a small area of Quebec (Henderson 1962), the latter to the Fall-line Sandhills of North and South Carolina, where it grows sympatrically but not syntopically with *L. virginicus* (Sorrie 1997). The new species described here, *L. glandulosus*, occurs within the West Gulf Coastal Plain (WGCP) in northeast Texas, southwest Arkansas, and northwest Louisiana.

Delimitation of *Lycopus* has been facilitated by the use of multiple morphological characters. Henderson (1962) considered *L. uniflorus* and *L. virginicus* closely related based on their short, obtuse calyx lobes which are shorter than or equal to the length of the mature nutlets. The remaining NA species have acuminate to subulate calyx lobes that usually exceed the length of the nutlets (Henderson 1962; Sorrie 1997). Hermann (1936) placed an emphasis on the shape and the apical crest of nutlets to supplement other morphological characters. Nutlet characters have recently been examined in detail (Moon & Hong 2006) and have confirmed Hermann's (1936) observations. Within putatively related species groups delimitation is further enabled by multiple characters, including the stem angle-type, presence of runners (underground) vs. stolons (above ground), tubers present vs. not, sessile vs. petiolate leaves, merely sessile leaves vs. clasping, leaf base shape, inflorescence bract type, and the aforementioned nutlet characters (Henderson 1962; Sorrie 1997; Moon & Hong 2006). Nonetheless, determination of some specimens remains difficult if specimens are immature or when the required characters are not available on a sheet.

Several collections of a morphologically distinct *Lycopus* from southwest Arkansas and northern Louisiana housed at TENN could not be satisfactorily placed within a recognized species and specimens comprising just the upper part of the plant were similar in appearance to glandular forms of *L. amplectens* from the southeastern Coastal Plain, but whole plants were more similar to *L. rubellus* Moench. These plants differ in their copious capitate glandular trichomes throughout the plant, which is also seen frequently in Gulf Coastal Plain samples of *L. angustifolius*. Vestiture is predominantly stipitate- (capitate-) to pilose-glandular (see Moon et al. 2010) and densely distributed on the leaves, stem, stolons, and calyces, which imparts a greyish, canescent appearance to the distal portions of the plant without magnification, but the glands are distinctly yellowish under magnification. This discovery prompted a preliminary molecular investigation of *Lycopus* in the southeastern USA.

Though numerous morphological and anatomical studies have aided in species delimitation (Hermann 1936; Henderson 1962; Sorrie 1997; Moon & Hong 2006), no molecular analyses have sought to test species boundaries or infrageneric classification. Here efforts in barcoding and phylogenetic analyses were undertaken to 1) to estimate the basic relationships of *Lycopus* species within North America, 2) to test the molecular differentiation of this new taxon, and 3) determine whether a barcoding approach using nuclear ribosomal ITS and several plastid markers, including the proposed barcoding *trnH-psbA*, would aid in identification of the species of *Lycopus*.

#### Methods

Specimens of North American *Lycopus* were observed at APSU, BRIT/LL, MO, and TENN (acronyms following Thiers 2022) comprising approximately 1800 collections. Fieldwork was performed in TN from 2009–2016, AR in 2009 and 2011, and east Texas in 2009. Leaf tissue was taken predominantly from herbarium specimens at TENN and some from living collections grown by the author in a common garden. Thirteen taxa and twenty two (17 produced here) samples of *Lycopus* were included. Several Lamiaceae outgroups were included from the sister subtribes of the monotypic Lycopinae — Nepetinae and Prunellinae (*Cedronella canariensis, Horminium pyrenaicum, Lepechina lancifolia*, and *Melissa officinalis*) (Drew & Sytsma 2012).

DNA was extracted using DNeasy Plant Minikits (Qiagen, Valencia CA) following manufacturers protocols. Primers for PCR of ITS used ITS-4 and ITS-Leu with occasional internal 5.8s and 79s primers when PCR failed, and trnH and psbA for the chloroplast *trnH-psbA* intergenic spacer. Additional markers tested were *petA-psbJ*, *rps15-ycf1*, *rpl32-trnL*, and *trnL-trnF*. PCR protocol and sequencing followed Schilling et al. (2007). All downstream analyses of sequence data were performed in Geneious 7.1.9 (Biomatters Ltd, Auckland, New Zealand). Alignment used the MAFFT (Katoh et al. 2002) plugin. Indels were coded using Fastgap 1.2 (Borchsenius 2009). Maximum likelihood (ML) analyses were performed with RaxML using 1000 bootstrap replications (Stamatakis 2014).

#### Results

Examination of specimens showed consistent glandular indumentum on specimens of the putative new species, the collections all confined to a narrow distribution in the WGCP. Other morphology that was noted included the grey-green color of the leaves and stems on herbarium specimens and the absence of purple pigmentation in living collections, the narrower and longer calyx lobes, and slightly larger corollas.

Sequences for nuclear ribosomal ITS (17) and several chloroplast intergenic spacers were produced and submitted to Genbank (Table 1). Sequences of *rpl32-trnL* and *trnL-trnF* for *L. cokeri* and *rps15-ycf1* for *L. virginicus* failed to amplify. Successful sequencing was limited to recent accessions or fresh living material due to low quantity, quality, or secondary chemistry inhibition of DNA in extractions from specimens >10–15 years old. Extraction and subsequent sequencing was limited to one or a few accessions of each species. In some cases sequences of low quality in the chloroplast intergenic spacers were not used in final analyses.

Phylogenetic analyses resolved *Lycopus* into two distinct clades using the cpDNA (Fig. 1). The first clade contained *L. americanus* Muhl., *L. europaeus* L., and *L. lucidus* Turcz. (100 bs). This clade was sister to a larger group consisting of two moderately to low supported clades. One of the sister clades contained *L. angustifolius* and *L. asper* Greene (73 bs) and the other contained the remaining species from eastern North America (63). Within the latter clade two groups were recovered; one contained *L. rubellus* (6348), *L. cokeri* + *L. uniflorus*, and *L. amplectens* and two samples of *L. glandulosus*, while the other clade contained *L. rubellus* samples, the *L. uniflorus* from Genbank (presumably misidentified), and *L. virginicus*.

Several species of *Lycopus*, but not all samples of a species, contained a 17 base pair inversion in *trnH-psbA*, which was inverted for the analyses. The intergenic spacers *rpl32-trnL*, *petA-psbJ*, *rps15-ycf1*, and *trnL-trnF* were largely invariant. In all chloroplast spacer regions *L*. *americanus* was the only species with consistent differences for the North American species. For *rps15-ycf1 L*. *amplectens* and *L*. *angustifolius* both shared a three base pair inversion, and *L*. *amplectens* contained a 6 base pair repeat. A single sample of *L*. *rubellus* contained a 2 base pair repeat.

Lycopus was resolved into two distinct clades by the nrITS DNA data (Fig. 2, 100 bs). The first clade contained L. americanus sister to the Eurasian species. The second clade contained the remaining North American species and these formed two distinct groups (100 bs). The first smaller clade contained L. asper sister (69 bs) to L. cokeri + L. uniflorus (99 bs). The first branch of the moderately to weakly supported sister clade contained L. virginicus sister to a largely unresolved group containing L. amplectens, L. rubellus, L. angustifolius, and L. glandulosus. In this ultimate clade all samples of L. glandulosus formed a cluster but are unresolved with regards to L. angustifolius and L. rubellus.

#### Key to the species of Lycopus in the southeastern USA

Update to Weakley's Flora of the Southeastern United States (2022) from couplet 4, lead 2.

- 4. Nutlet tubercles typically well-developed with 4–5 knuckle-like knobs.
- 6. Leaves evidently petiolate, the petioles narrowly winged.

7. Calyx lobes triangular, attenuate to acute apex; leaves glabrous, glabrate, canescent or	
pubescent, sometimes densely so throughout Lycopus rubellu	IS
7. Calyx lobes subulate, apex acute; leaves pubescent with short eglandular, and also densely	
distributed short stipitate-glandular trichomes, especially distal vegetative parts	
Lycopus glandulosu	S

6. Leaves sessile or subsessile.

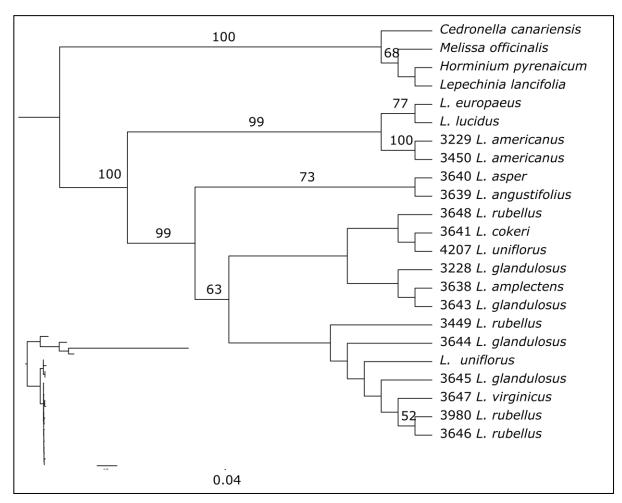


Figure 1. Best-scoring cladogram of *Lycopus* as inferred from Maximum Likelihood analyses of cpDNA data (*tnH-psbA*, *petA-psbJ*, *rpl32-trnL*, and *ycf1-rps15*). The phylogram is inset in the lower left to show the shallow branch structure within *Lycopus* compared to the outgroup taxa. Bootstrap support values >50 are shown at the branches.

# Discussion

The results of herbarium and molecular study provide support for the distinction of a characteristic novel species of *Lycopus*, *L. glandulosus*, closely related to *L. rubellus* though well-differentiated from it in morphological characters. The new species occurs in a narrow region of the West Gulf Coastal Plain of Texas, Arkansas, and Louisiana

Lycopus glandulosus is distinct in its morphology and several characters are sufficient to distinguish it from its congeners. Morphological separation of *L. glandulosus* from *L. rubellus* is facilitated by the presence of abundant stipitate glandular trichomes throughout the plant and live material is noticeably viscidulous and fragrant (reminiscent of mild-scented Eucalyptus-menthol). Glandular trichomes are occasionally observed in the southeastern Coastal Plain in some *L. rubellus*, *L. amplectens*, and *L. angustifolius* populations, but no glandular plants of *L. rubellus* have been observed in the West Gulf Coastal Plain. Lycopus angustifolius and *L. amplectens* in the CP often have densely distributed stipitate glands on the distal portions of the plants, but neither occurs in proximity to *L. glandulosus*. The closest population of *L. rubellus* observed during this study that had glandular trichomes was in southeastern Mississippi — these plants are ambiguous in morphology

and approach forms of *L. amplectens*, which is also known from that area. The calyx lobes and corolla are generally marginally larger in *L. glandulosus* than in *L. rubellus*, but the former also differ in their longer subulate lobes vs. attenuate lobes. In the field and in cultivation the leaves of *L. glandulosus* are distinctly grey-green in color, lacking any of the red pigmentation common in *L. rubellus* (and also *L. virginicus*). In common garden cultivation *L. glandulosus* retains its distinctive appearance and glandular pubescence.

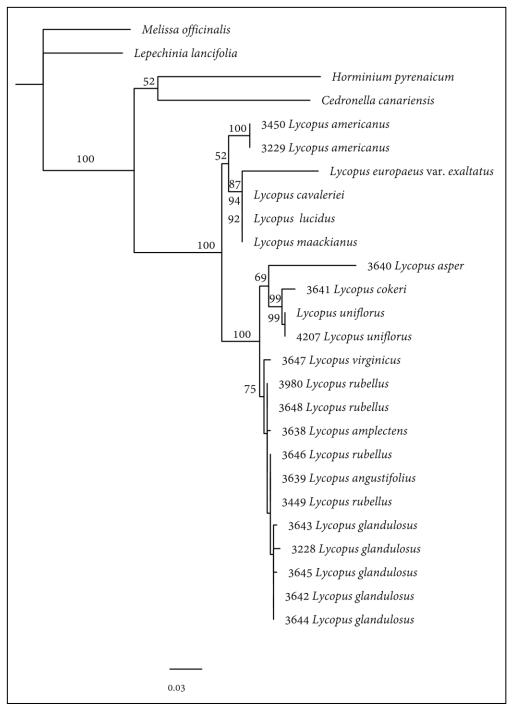


Figure 2. Best-scoring phylogram of *Lycopus* as inferred from Maximum Likelihood analyses of the nuclear ribosomal ITS. Bootstrap support values  $\geq$ 50 are shown at the branches.

# Discussion

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Results of the phylogenetic analyses reveal moderate to low support for three distinct clades in North American *Lycopus* and low support or resolution of the species. In the cpDNA and nrITS phylogenetic analyses *L. americanus* was recovered as sister to the Eurasian species. This clade is distinctive in its sinuate leaf margins and nutlets with round and smooth apices. The next clade consists of two sister clades, one with *L. asper* sister to *L. cokeri* + *L. uniflorus*. *Lycopus cokeri* has only recently been recognized at specific rank separate from *L. uniflorus* and the molecular results confirm this (see Sorrie 1997 and others cited there). The final clade received only moderate to low support ( $\leq 75$ ), in which *L. virginicus* is sister to the large *L. rubellus*-group which consists of unresolved relationships of *L. amplectens*, *L. angustifolius*, *L. rubellus*, and *L. glandulosus*. Though these species are unresolved due to the absence of chloroplast spacer variation observed here and limited ITS differences, it is significant that *L. glandulosus* differs from all samples of *L. rubellus* and the glandular *L. angustifolius* by a single base pair change for ITS.

Molecular variation within *Lycopus* was found to be relatively limited in most samples across all chloroplast loci. *Lycopus americanus* from eastern and western North America were 100% identical to one another though highly divergent from the other North American species. The *Lycopus rubellus* group (incl. *L. amplectens, L. angustifolius,* and *L. glandulosus*) shared a character change not observed in other species for *rpl32-trnL*, though this species group was otherwise invariant among the species across all cpDNA loci. Two samples of *L. rubellus* exhibited unexpected sequence differences compared to the other samples. One sample (3980, *L. rubellus*) was collected in the field in flower without mature nutlets as *L. uniflorus* and only upon sequencing was it apparent that it was not that species. The other is morphologically identical to the other samples but was not placed with them in the phylogenetic analyses.

ITS sequence data provide enough characters as a barcoding marker to accurately identify to species all *Lycopus* sampled except for *L. angustifolius* and *L. rubellus*, even though infrageneric relationships were unresolved. Two of the *L. rubellus* (3648 and 3980) do not share the same bp changes as the others, nor do they share some of their sequence characters with any other species. Despite this, for *L. rubellus* 3648 the chloroplast data places it with *L. uniflorus* + *L. cokeri*, and the

ITS data places it in the large *L. rubellus* clade of several unresolved species. *Lycopus rubellus* 3980 is placed in the terminal clade of unresolved species despite the sequence differences. The samples of *L. glandulosus* have a single bp change shared with the most proximal *L. rubellus* that are not shared with the other *L. rubellus* samples or *L. angustifolius*.

Limited molecular variation of the chloroplast loci sampled in the North American *Lycopus* was observed. This low molecular variation or homogeneity in sequence data, especially within the *L. rubellus*-group may be due to recent speciation, hybridization, or also locus selection used here which does not exhibit significant, or any changes in the several chloroplast markers sampled. The disjunct forms of *L. amplectens*, i.e, the Mid-Atlantic-New England and the southern Coastal Plain forms, and the purported hybrid *L.* × *sherardii* were not investigated (Henderson 1962). There are notable differences in pubescence between the centers of distribution of the former, and the latter is reported from regions where both parents are not always present. Further molecular work should seek informative regions of the chloroplast or whole chloroplasts to resolve species relationships and investigate possible hybridization in the evolution of *Lycopus* in the southeastern USA.

LYCOPUS GLANDULOSUS Floden, sp. nov. TYPE: USA. Arkansas. Nevada Co.: NW of Bluff City, off Co. Road 290 S of Co. Road 16 along Caney Creek, west floodplain in open area growing with *Carex lupulina*, *C. intumescens*, *C. abscondita*, *Rhynchospora* sp., *Cyperus* sp., *Carya* sp., *Ilex opaca*, *Styrax americana*, *Dichanthelium* sp., abundant in floodplain and the dominant species in areas, highly fragrant when touched, 23 Sep 2011, *A. Floden 1797* with S. Hart (holotype: MO; isotypes: AR, BRIT, K, NCU, KY, NY, TENN, TX). Figures 3–4.

Different from *Lycopus rubellus* Moench by its calyx lobes subulate (vs. attenuate) and vestiture comprised nearly of stipitate-glandular trichomes with some non-glandular pubescence (vs. eglandular trichomes of various lengths or glabrate).

**Perennial herbs**, stoloniferous. **Stolons** with fusiform tubers at stolon terminus, stolons densely stipitate-glandular, bracteate leaves obovate, apex obtuse. Stems 0.5-1 m tall, erect, 4angled, angles rounded, unbranched to branched from leaf nodes of upper stem, densely stipitate glandular. Leaves opposite, ovate, to oblong, broadest above the middle, lamina  $5-8 \times 1-2$  cm, 5-8(10) serrate, each tooth upward curved with a rounded obtuse apex, margins strigose, densely capitate-glandular adaxially and more densely abaxially, both surfaces densely punctate-glandular, 25-50 per mm<sup>2</sup>, midvein densely stipitate glandular and strigose, leaf base abruptly attenuate to a winged petiole, 1-3 cm, petiole densely pilose-glandular, nodal band of same vestiture. Inflorescence of sessile axillary cymules, densely many-flowered, bracts linear to lanceolate, margin ciliate, faces strigose and glandular. Calyx 5-lobed, lobes 0.7-1 mm, lanceolate-subulate, apex acute, densely glandular and strigose, apices of calyx lobes just reaching sinuses of corolla lobes. Corolla white, throat bearded, extending 1.5–2.0 mm out of calyx, tube 2.0–2.5 mm, corolla strigose and resin dotted on lobes, lobes 5, 0.5-0.7 mm, apices obtuse to round, lower petal straight upper petal recurved. Stigma; exerted ca. 1–1.5 mm. Stamens (fertile) 2, just exerted from tube, pinkish. Nutlets triangular obovoid,  $1.0-1.25 \times 1-1.2$  mm, glandular-punctate, apex with acute to obtuse tubercles, usually irregular, the outermost tubercles larger than the others.

The epithet *glandulosus* alludes to the abundant stipitate glands present on nearly all parts of the plant, which impart a viscidulous feel to living plants.

*Lycopus glandulosus* and *L. rubellus* are sympatric in the West Gulf Coastal Plain, but at no locality have they been observed to be syntopic. *Lycopus rubellus* was observed in close proximity to populations of *L. glandulosus* in southwest Arkansas, but these proximal populations are clearly distinct in morphology and do not have the short stipitate-glandular trichomes throughout the plant.

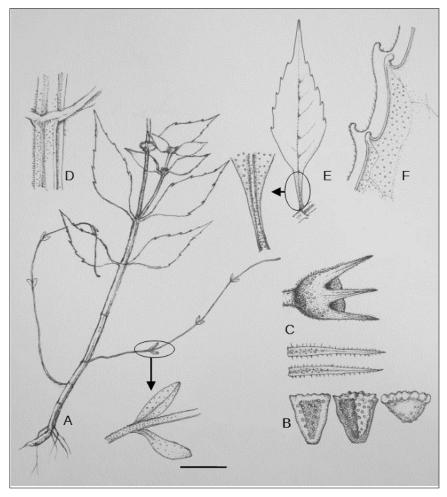


Figure 3. *Lycopus glandulosus (Floden 1797)* showing pertinent features of the new species. Scale bar: A=5 cm, B=1 mm, C=1 mm, D=2 mm, E=3 cm, and F=3 mm.



Figure 4. *Lycopus glandulosus* from the type collection (*Floden 1797*) showing from left to right: (a) glandular stem pubescence and bract pubescence of an inflorescence; (b) calyx lobes, corollas, and glandular stem pubescence; and (c) close-up of the abaxial densely glandular leaf surface. Scale bars: a, 1 mm; b, 0.5 mm; c, 0.25 mm.

Other specimens examined. ARKANSAS. Calhoun Co.: West Gulf Coastal Plain. Pleistocene Fluvial Terraces Ecoregion. 3.1 mi. E of Artestian [sic]. Moro Big Pine Natural Area-Wildlife Managment Area. 0.3 mi. NNW of "Five Points", between CR 31 & CR 34, vicinity of N 33.40194, W 92.41595. Artesian 7.5' quandrangle, upper edge of seep in open loblolly pine woodland (thinned & burned in recent years), near contact between Smithdale Fine Sandy Loam (uphill) & Amy Silt Loam (flats), locally common, forming large colony, stoloniferous, leaves thick with golden punctate glands, with Pinus taeda, Xyris spp., Woodwardia areolata, W. virginica, Osmundastrum cinnamomeum, Osmunda regalis, Magnolia virginiana, Rhynchospora spp., Dichanthelium scabriusculum, Eleocharis spp., Viburnum nudum, Sphagnum spp., fertile material from the same area collected 15 Sep 2014 as Witsell 14-0384, 19 Jul 2012, Witsell s.n. (ANHC, TN); ibidem, 15 Sep 2014, Witsell 14-0384 (ANHC, MO). Lafayette Co.: West Gulf Coastal Plain, Tertiary Uplands Ecoregion, 1.2 mi ENE of Buckner, E side of gravel road, 0.4 mi N of Hwy 82, at jct with spur road to well, between Clear Creek & Bayou Dorcheat, vicinity of N 33.36451, W 93.41601, Buckner 7.5' quandrangle, seep marsh at roadside, locally common, rhizomatous, forming a colony, 21 Jul 2015, Witsell 15-0430 (ANHC, MO). Nevada Co.: Woods along N bank of Caney Creek W of County Rd 290 and south of County Rd 16 NW of Bluff City, Sec. 36, T11S, R21W, 13 Oct 2000, Thomas 168163 (MO, NYBG, TENN); P.O. Bluff City, creek banks, elev. 280 ft, 11 Oct 1939, Demaree 20715 (F). Ouachita Co.: Along N bank of Smackover Creek at Ark. 7 bridge at the Union Co. line S of Camden, sec. 30, T15S, R16W, 8 Oct 2000, Thomas & Doffit 167796 (MO); construction area along north prong of Two Bayou W of Ark. 4 bridge W of US 79, Camden, sec. 28, 11 Sep 1993, Thomas 137242 (TENN). Union Co.: Woods beside paved road and Caney Creek 2.7 mi N of New London; Sec. 22, T17S, R12W, 1 Sep 1989, Thomas 112583 (TENN); salty runoff area beside small stream just E of Lawson and S end of Ark. 129; sec 31, T17S, R13W, area graded in attempt to clean up runoff from oil wells, 22 Oct 1987, Thomas 101113 (GSW). LOUISIANA. Bienville Par.: Disturbed rights-of-way of La. 9 and powerline near Six Mile Creek N of La. 4 and Lucky; Sec. 21, 15N, R6W, 28 October 2000, Thomas 168710 (BRIT). Claiborne Par.: Moist woods between Claiborne Dam on Bayou D'Arbonne and La. 518; Sec. 8, T20N, R5E, 30 Sep 1967, Thomas et al. 4923 (TENN). Jackson Par.: Chatham Lake, sandy peaty shores, 22 Oct 1962, Kral 16101 (BRIT, FSU). Lincoln Par.: Ca. 10 mi E of Ruston, moist sandy clay of ditchbank, 15 Oct 1962, Kral 16019 (BRIT, FSU). Natchitoches Par.: KNF, Winn Dist., Compt. 3, stands 8 & 14, along a small Forest Service road N of USFS 508 at curve W of Eight Mile Creek; N of La. 126 E of Readhimer; Secs. 1 & 13, T13N, R6W, 6 Oct 1995, Thomas 147478 (TENN). Webster Par.: DMC4 68770/36 02630, 3.2 km N of Walker Cemetery, ca. 1.5 km SE of Knottingham Cemetery, 25 Aug 2002, Arnett & Hastings 449 (TENN). Winn Par.: Cleared flatwoods S of La. 156 at Spring Branch east of La. 123 and W of Calvin; Sec. 27, T12N, R5W, 14 Oct 1994, Thomas 143077 (MO, TENN); Cypress Creek Branch Baygall, Winn District, Kitsatchie NF, compartment 5, T13NR5WS8-9, N of FS 506, E of 554, 10 Sep 1999, MacRoberts 4126 (BRIT). TEXAS. Austin Co.: 15 Oct 1939, Tharp (MO). Bowie Co.: Aikens Creek, S of Hwy 67, pine woodland area, stoloniferous, corolla large, white, 23 Sep 1948, Whitehouse 20459 (BRIT). Cass Co.: Along meadow stream, about 2.5 mi E of Hughes Springs, Hwy 11, Sep 1970, Anderson 299 (BRIT); 6.5 mi E of Hughes Springs on Hwy 11 near Black Cypress Creek, 1 Nov 1946, Whitehouse 17692 (BRIT). Hardin Co.: Infrequent in moist soil of sandy woods, 2.5 mi W of Silsebee, 3 Oct 1945, Cory 49878 (BRIT, MI). Harrison Co.: Caddo Lake State Park on bank of bluff between ramp and private holding, 30 Jun 1975, Fleetwood 22869 (BRIT); Davidson Foundation Forest, along ditch, 20 Sep 1974, Fleetwood 11229 (BRIT). Morris Co.: Daingerfield State Park, pine and oak woods, flrs white, 24 Sep 1961, Correll & Correll 24646 (LL). San Jacinto Co.: Ca. 9 mi W of Cleveland on Hwy 105 along Peach Creek, open wooded area, white, locally frequent, 4 Oct 1980, Nixon & Ward 10618 (BRIT). Smith Co.: Swan, 17 Sep 1902, Reverchon s.n. (MO). Tyler Co.: 3.5 mi NW of Woodville, frequent in borrow ditch of highway, stems branched, up to 4.5 dm. high, with long slender leafy stolons, 28 Sep 1948, Cory 54861 (BRIT, LL). Upshur Co.: W side of Big Sandy, sandy ground at edge of water, lake shore, corolla white, selected small plant (larger 6 dm high), 14 Sep 1953, Shinners 15973 (BRIT). Wood Co.: Lake Ellies, in log, aquatic, fls white, 3 Sep 1942, Lundell & Lundell 11759 (BRIT, LL).

# ACKNOWLEDGEMENTS

I thank Theo Witsell and Dwayne Estes for assistance in fieldwork, the Dennis Breedlove fund at the University of Tennessee for monetary support that enabled field studies, the late R. Dale Thomas for his extensive collections from Arkansas and Louisiana, which initiated this study, APSC and BRIT for specimen loans, Nick Mattson in the Schilling Lab at UT for his assistance in sequencing, and Joe Brown and Veronica May at the University of Tennessee Genomics Core.

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**Appendix**. Species, voucher information (Collector and number, State, DNA sequence number), and Genbank numbers for samples of *Lycopus* for the corresponding molecular markers: ITS, petA-psbJ, rps15-ycf1, trnH-psbA, rpl32-trnL, and trnL-trnF). "-" indicates sequence not obtained or not available in Genbank.

Lycopus americanus, Packer 5547, WY 3450, MH558143, MH631984, MH632017, MH632033, MH632001, MH632050; Schilling s.n., TN, 3229, MH558142, MH631983, MH632016, MH632032, MH631983, MH632049; L. amplectens, Slaughter 17171, FL, 3448, MH558148, MH631986, MH632019, MH632035, MH632003, MH632052; L. angustifolius, L& R 2198, FL, 3638, MH558156, MH631998, MH632031, MH632047, MH632014, MH632063; L. asper, McNeilus 98-607, MN 3640, MH558144, MH631985, MH632018, MH632034, MH631985, MH632051; L. cavaleriei, KM886730, -, -, -, -, -; L. cokeri, Nelson 11503, SC, 3641, MH558145, MH631987, MH632020, MH632036, -, -; L. europaeus var. exaltatus, AY506652, -, -, -, -, -; L. glandulosus, Floden, cultivated from type gathering, AR, 3228, MH558149, MH631989, MH632022, MH632038, MH632005, MH632054; Thomas 143077, AR, 3642, MH558150, MH631990, MH632023, MH632039, MH632006, MH632055; A & H 449, AR, 3643, MH558151, MH631991, MH632024, MH632040, MH632007, MH632056; Floden 1803, AR, 3644, MH558152, MH631992, MH632025, MH632041, MH632008, MH632057; Thomas 164287, AR, 3645, MH558153, MH631994, MH632027, MH632043, MH632010, MH632059; L. lucidus, AB763377, -, -, -, -, -; L. lucidus var. maackianus, JQ669110, -, -, -, -, -; L. rubellus Estes & Beck 8367, TN, 3449, MH558154, MH631993, MH632026, MH632042, MH632009, MH632058; Thomas 157861, AR, 3646, MH558156, MH631995, MH632028, MH632044, MH632011, MH632060; McNeilus 97-1038, 3648, MH558157, MH631996, MH632029, MH632045, MH632012, MH632061; Floden, TN, 3980, MH558158, MH631997, MH632030, MH632046, MH632013, MH632062; L. uniflorus, DQ667302, -, -, -, -, -; Halse 5182, OR, 4207, MH558146, MH631988, MH632021, MH632037, MH632004, MH632053; L. virginicus, Thomas 173412, AR, 3647, MH558147, MH631999, -, MH632048, MH632015, MH632064.