To the Graduate Council:

I am submitting herewith a thesis written by Frank Fusiak entitled "A Systematic Study of the Prenanthes roanensis Complex of the Southern Appalachians." I have examined the final copy of this thesis for form and content and recommend that it be accepted in partial fulfillment of the requirements for the degree of Master of Science, with a major in Botany.

Edward E. Schilling, Major Professor

We have read this thesis
and recommend its acceptance:

Accepted for the Council:

Vice Chancellor
Graduate Studies and Research

A SYSTEMATIC STUDY OF THE PRENANTHES ROANENSIS COMPLEX OF THE SOUTHERN APPALACHIANS

A Thesis

Presented for the

Master of Science

Degree

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Linnaean Herbarium (LINN)

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ABSTRACT

A systematic study was conducted on the Prenanthes roanensis complex of the southern Appalachians. Investigations involving scanning electron microscopy, flavonoid chemistry, cytology and numerical taxonomy coupled with traditional taxonomic techniques were undertaken to evaluate species validity, limits and relationships, to determine if natural interspecific hybridization was occurring amongst members of this complex and to assess the endangered status of P. roanensis.

With scanning electron microscopy, all taxa were determined to have pollen that was tricolporate and echinolophate and ligules that were transversely striate and distally rugose. With flavonoid analysis, a total of seven luteolin glycosides were isolated and characterized. Both scanning electron microscopy and flavonoid chemistry studies indicated that intraspecific differences were as great as interspecific differences. Meiotic analysis showed all individuals to be $n = 8$ with no irregularities. No species-specific characters could thus be discerned using these techniques.

Multivariate analyses were performed on 100 samples for 18 morphological characters. Cluster and principal coordinates analyses indicated that the samples can be divided into three groups that correspond to the eventually recognized species. These results from traditional taxonomic techniques and numerical analysis, especially those results derived from cluster analysis, support the recognition

of the following three closely related but morphologically distinct species: P. altissima, P. roanensis and P. serpentaria.

The occurrence of natural interspecific hybridization could not be discerned from the aforementioned analyses. Its occurrence, however, cannot be conclusively ruled out at this time.

Lastly, P. roanensis should not be considered federally endangered or threatened. It is presently known from 20 counties in North Carolina, Tennessee and Virginia and is abundant at most of these sites that are federally protected public lands.

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CHAPTER I

INTRODUCTION

The genus Prenanthes L. (Compositae-Lactuceae) is composed of 26 species distributed throughout the northern temperate regions of Asia, Europe and North America (Milstead, 1964). The species are usually simple stemmed, fall flowering perennials or biennials of deciduous woodlands and alpine regions. The genus is divided into two subgenera, Prenanthes and Nabalus. Subgenus Prenanthes is distributed throughout portions of Europe and Asia, while species of subgenus Nabalus are found in North America and northeastern Asia (Milstead, 1964). In a recent revision of the North American taxa, Milstead (1964) recognized 13 species, with 11 occurring east of the Mississippi River.

Many taxonomic and nomenclatural problems exist within the genus primarily due to the extreme variation in some morphological characters (i.e., leaves), the suspected but never documented natural hybridization with subsequent introgression of morphological characters between species and the paucity of distinguishing characters (Fernald, 1950; Milstead, 1961, 1964; Cronquist, 1980; Johnson, 1980).

An added dimension to these problems is that Ayensu and DeFilipps (1978) listed two species, P. boottii (DC) Gray and P. roanensis (Chickering) Chickering, as federally endangered or threatened. These two taxa grow in similar open habitats at high

elevations (above 4000 feet) of the Appalachian Mountains. Prenanthes boottii occurs from Maine to northern New York (Gleason and Cronquist, 1963), and P. roanensis inhabits areas of North Carolina, Tennessee and Virginia (Cronquist, 1980; Johnson, 1980).

Recently, the endangered status of the southern taxon, P. roanensis, was re-evaluated by Wofford and Smith (1981, Special Report, U.S. Fish and Wildlife Service). In their report, they stated:

> The taxonomy of Prenanthes roanensis is complicated by what appears to be numerous intermediates between P. roanensis and P. altissima. In our opinion, these intermediates represent P. cylindrica sensu Small. However, until further study, we accept the conservative treatments of Milstead (1964) and Cronquist (1980) and include P. cylindrica within the definition of P. roanensis.

Their report has been an impetus to initiate field and herbarium studies on these taxa. A survey of herbarium material revealed a particularly close morphological resemblance among specimens of P. altissima and P. roanensis, with many appearing to be of intermediate morphology and, therefore, possibly of hybrid origin. Characters used in descriptive keys (Fernald, 1950; Gleason and Cronquist, 1963; Milstead, 1964; Radford, Ahles and Bell, 1968; Cronquist, 1980; Johnson, 1980) to separate the two taxa also appeared to be confusing and overlapping. Preliminary investigations also revealed that another taxon, P. serpentaria Pursh, differed from P. roanensis only in its slightly larger capitulum and its corymbiform inflorescence. Since interspecific hybrids often exhibit intermediate morphology between parental taxa (Stebbins, 1950;

Grant, 1981), hybrids derived from P. altissima and P. serpentaria could also fit within the circumscription of P. cylindrica sensu Small (1933). Thus the systematic study was expanded to include P. serpentaria, as well as P. altissima and P. roanensis. Investigations involving scanning electron microscopy, flavonoid chemistry, cytology and numerical taxonomy coupled with traditional taxonomic techniques were initiated to evaluate species validity, limits and relationships, to determine if interspecific hybridization was occurring amongst members of this complex and to assess the endangered and threatened status of P. roanensis.

CHAPTER II

SCANNING ELECTRON MICROSCOPY

Recent systematic literature includes many examples of the usefulness of the scanning electron microscope (SEM) to taxonomic studies. The great resolution and depth of focus achieved with the SEM have provided the systematist with an additional tool for the study of microscopic characters that may prove taxonomically and phylogenetically important. Analyses of pollen are most frequently cited along with seeds, spores and the microcharacters of leaves and ligules.

The pollen morphology of the family Compositae, tribe Lactuceae, to which Prenanthes belongs, has been extensively surveyed with the SEM (Tomb, Larson and Skvarla, 1974; Tomb, 1975; Feuer and Tomb, 1977; Blackmore, 1981). These studies have demonstrated the usefulness of employing pollen characters in the classification of the Lactuceae. The tribe contains both echinate and echinolophate pollen. The former has spines spaced evenly over the surface except in the regions near the apertures, and the latter has spines confined to ridges surrounding spineless depressions called lacunae (Wodehouse, 1935).

Microcharacters of the ligules of the Compositae have also been suggested to be taxonomically important (Baggøe, 1977a, 1977b, 1978, 1980). The tribe Lactuceae exhibits the widest range of variation in the ligule epidermis characters within the family

(Baggøe, 1980), and these thus show the greatest promise to be taxonomically useful.

In the present study, pollen and ligule microcharacters were examined with the SEM for all taxa of the P. roanensis complex in hope of finding characters that would prove taxonomically useful at or below the species level.

Materials and Methods

All material examined by SEM was obtained from dried herbarium specimens. Voucher specimens are listed in Table II-1. Pollen and ligules were affixed to standard 3/4-inch aluminum stubs with double-faced cellophane tape. The specimens were then coated with vaporized gold:palladium (60:40) in vacuo using a Denton model DV-512 vacuum evaporator equipped with a random rotating head and were subsequently viewed on an ETEC Autoscan scanning electron microscope at the Electron Microscopy Facility of The University of Tennessee, Knoxville. Photographs were taken using Polaroid Type 655 Positive/Negative film.

Results and Discussion

The pollen is characterized by spines on ridges surrounding lacunae (echinolophate; Figure II-1 A). The grains possess a distinct equatorial ridge and three apertures that are divided into lacunae connected by narrow inter-lacunal gaps (tricolporate; Figure II-1 B). The pollen is also characterized by a spine covered

Table II-1. Populations of the Prenanthes roanensis Complex Examined with the Electron Microscope.

Species	Subject	Voucher[a]
P. altissima	pollen	MA Hampshire Co.: Ahles 79941
		PA Bucks Co.: Morton 2317
		WV Grant Co.: Downs 7257 (UNC)
		VA Patrick Co.: Ahles and Clark 60153 (UNC)
		TN Sevier Co.: Thomas s.n.
		LA Caldwell Parish: Marx 2755A (UNC)
	ligule	NC Montgomery Co.: Radford 19403 (UNC)
		NC Watauga Co.: Smyth 2555 (VPI)
		VA Montgomery Co.: Smyth 5435 (VPI)
		VA Buchanan Co.: Kral 14284 (VDB)
P. roanensis	pollen	NC Swain Co.: Ramseur 1455 (UNC)
		NC Mitchell Co.: Smyth 3494 (VPI)
		VA Grayson Co.: Croy 675 (VPI)
		TN Unicoi Co.: Wofford and Smith 79-298
	ligule	NC Avery Co.: Fusiak, Smith and McFarland 322

Table II-1 (continued)

Species	Subject	Voucher[a]
(*P. roanensis*)	(ligule)	NC Avery Co.: *Fusiak*, *Smith* and *McFarland* 317
		TN Unicoi Co.: *Fusiak s.n.*
		TN Carter Co.: *Fusiak s.n.*
P. serpentaria	pollen	PA Montgomery Co.: *Berkheimer* 6708 (VDB)
		VA Charlotte Co.: *Ahles* and *James* 60891 (UNC)
		VA Loudon Co.: *Ahles* and *James* 61526 (UNC)
		TN Blount Co.: *Thomas* and *Rogers s.n.*
		MS Tishmingo Co.: *Rogers* 46214
		AL Mobile Co.: *Lelong* 3907 (UNC)
	ligule	NC Northampton Co.: *Ahles* 52452 (UNC)
		NC Lincoln Co.: *Bell* 15356 (UNC)
		NC Burke Co.: *Bell s.n.* (UNC)
		VA Fairfax Co.: *Bradley* 11409 (UNC)
		TN Grundy Co.: *Wofford* 77-301

[a]Unless otherwise indicated, all specimens are deposited in The University of Tennessee Herbarium (TENN).

Figure II-1. Scanning Electron Micrographs of Pollen Grains of *Prenanthes*.

A. *P. altissima*, lateral view, 1250 X.

B. *P. roanensis*, apertural view, 1250 X.

C. *P. serpentaria*, polar view, 1200 X.

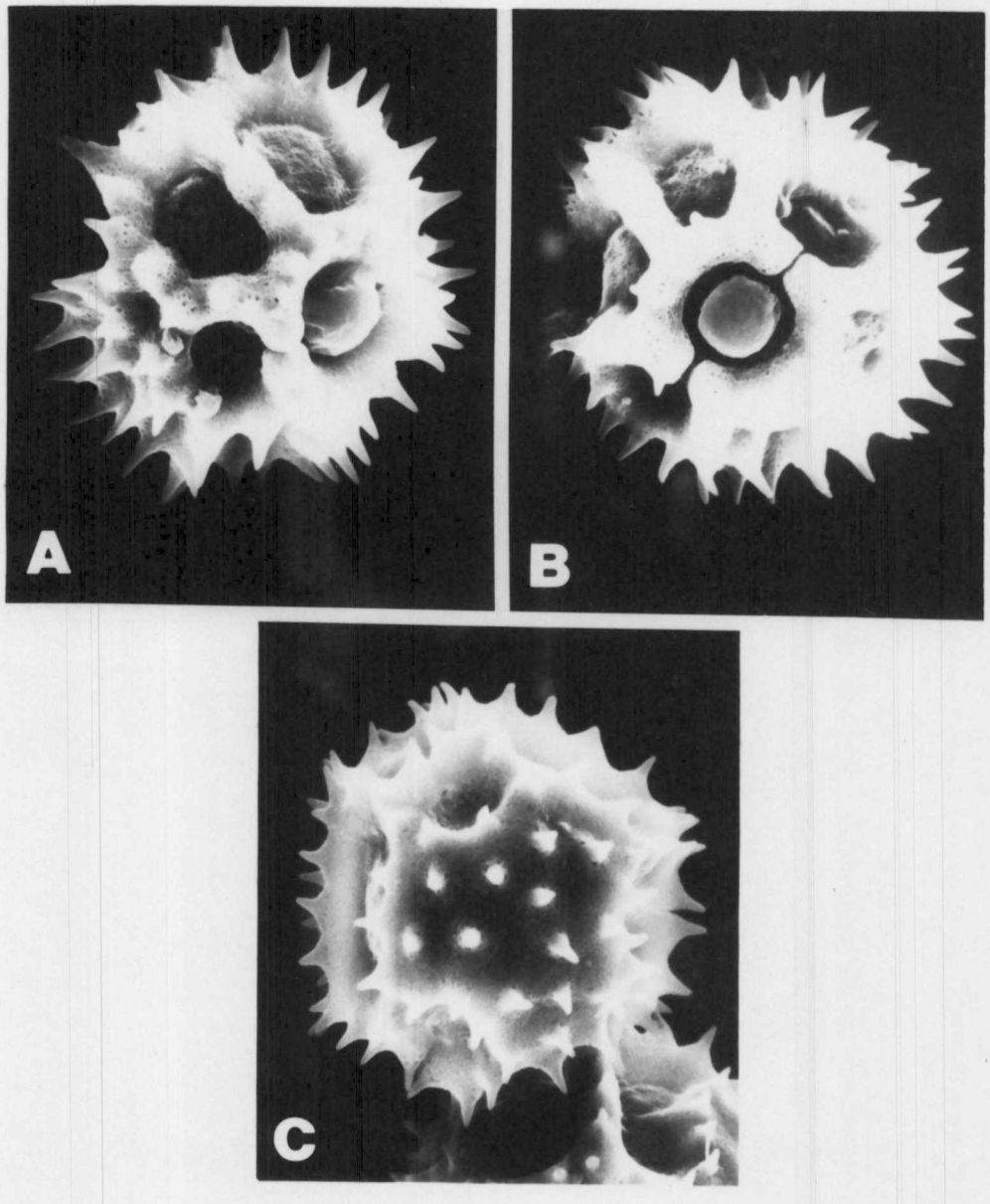


Figure II-1

area at the pole of the grain termed the "polar thickening" (Figure II-1 C).

These observations are in accord with previous SEM studies of Prenanthes pollen which have shown grains to be tricolporate and echinolophate (Tomb, 1975; Skvarla, Turner, Patel and Tomb, 1977). No significant differences were observed in the pollen wall architecture between species examined. No discriminating taxonomic characters could thus be discerned from the pollen grains at the specific level.

All Prenanthes specimens examined had a transversely striate and distally rugose ligule. Abaxial and adaxial surfaces have a similar appearance (Figures II-2 A,B). These results are in accordance with previous SEM studies of ligules in Prenanthes (Baggøe, 1980). No significant differences were observed in the micromorphology of the ligules. No useful species-specific characters could thus be discerned from the micromorphology of the ligules.

Figure II-2. Scanning Electron Micrographs of the Ligules of *Prenanthes*.

A. *P. roanensis*, abaxial surface, 1100 X.

B. *P. altissima*, adaxial surface, 1100 X.

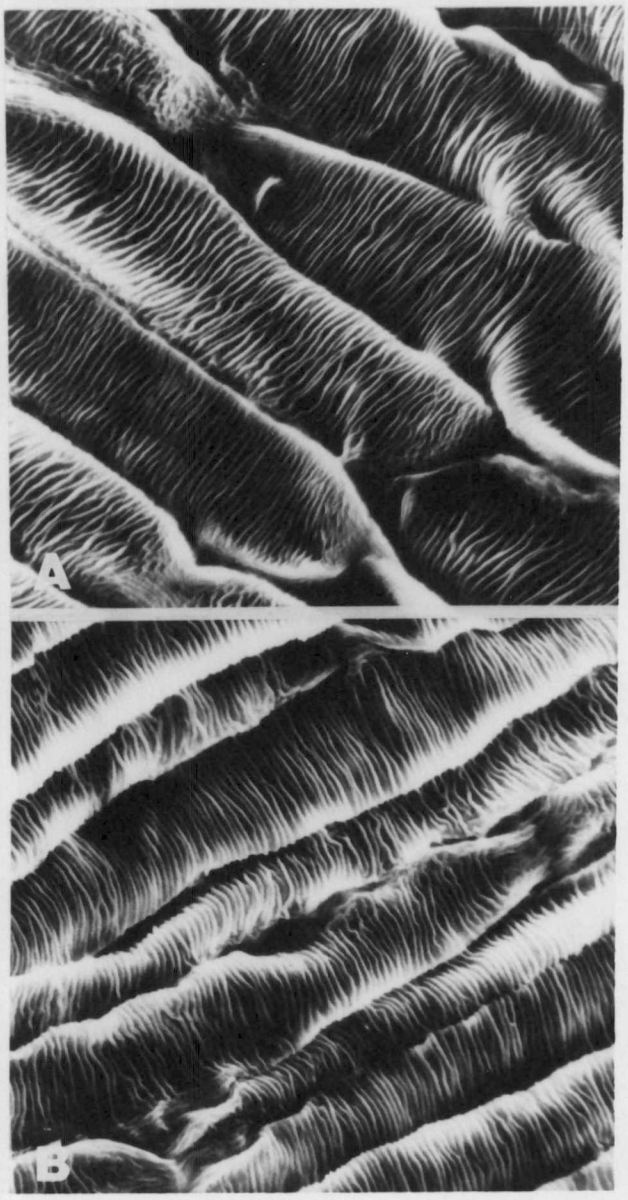


Figure II-2

CHAPTER III

FLAVONOID CHEMISTRY

Since Alston and Turner's (1963) pioneering work on Baptisia showing the usefulness of biochemical constituents to plant systematics, flavonoids have been widely used in taxonomic studies. Because flavonoids are extremely stable, structurally diverse, universally distributed and easily identified by simple methods (Harborne, Mabry and Mabry, 1975; Markham, 1982), they have been employed at all levels of classification. Since species often exhibit species-specific flavonoid profiles, their use has been especially rewarding in detecting interspecific hybridization because full or partial complementation of parental flavonoids is often observed in hybrids (Smith and Levin, 1963).

Flavonoid studies were initiated as part of a systematic investigation of species relationships and to determine if interspecific hybridization was occurring amongst members of the Prenanthes roanensis complex of the southern Appalachians.

Materials and Methods

Material for flavonoid analysis was collected from natural populations. Flavonoids were isolated from 100 g of dried leaf material of P. roanensis (TN: Carter Co., Fusiak 125, TENN). Extraction of plant material, isolation by paper chromatography and identification by ultraviolet spectral analysis for flavonoids

followed the generalized procedures as outlined by Mabry, Markham and Thomas (1970). For other taxa and population samples, 0.3 g of leaf material from at least two individuals per population was combined and extracted in 85% methanol for 24 hours and taken to dryness on a flash evaporator. The extract was then taken up in 100% methanol. The solution was then spotted on Whatman 3 MM chromatography paper and developed in the standard solvents for paper chromatography as outlined by Mabry, Markham and Thomas (1970). Identification of flavonoids from these samples was by comparison of R_f values and color characteristics of 2-dimensional paper chromatograms. Voucher specimens are listed in Table III-1.

Results and Discussion

Seven flavonoids were isolated and characterized from dried leaves of species of the P. roanensis complex (Figure III-1). All compounds were mono- or diglycosides of luteolin, except for one compound that was based on apigenin. Glucose was the most common sugar moiety and was attached at the 7 position in monoglycosides and at the 7 and 4' and the 7 and 3' positions in diglycosides. Luteolin-7-O-glucuronide and luteolin-7-O-diglycoside (based on xylose and glucose) were also detected. The aglycone, luteolin, was also found but probably does not occur as the aglycone in vivo. A chloroform rinse of fresh leaves failed to yield the free form of luteolin, and it is regarded here as an artifact (Wollenweber and Dietz, 1981; Markham, 1982).

Table III-1. Populations of *Prenanthes* Examined for Flavonoids.

Species	Populations Number and Voucher Specimen[a]
P. altissima	1. NJ Wayne Co.: Nyman 566
	2. IN Brown Co.: Fusiak 600
	3. TN Carter Co.: Fusiak 160
	4. TN Carter Co.: Fusiak 174
	5. TN Cocke Co.: Fusiak 201
	6. TN Cocke Co.: Fusiak 213
	7. NC Haywood Co.: Fusiak 227
	8. NC Buncombe Co.: Fusiak 251
	9. NC Avery Co.: Fusiak 315
	10. TN Knox Co.: Fusiak 449
	11. TN Knox Co.: Fusiak 81-36
	12. TN Unicoi Co.: Fusiak 567
	13. TN Polk Co.: Fusiak 374
	14. TN McMinn Co.: Fusiak 590
	15. NC Transylvania Co.: Fusiak 249
P. roanensis	16. TN Carter Co.: Fusiak 125
	17. NC Avery Co.: Fusiak 559
	18. VA Grayson Co.: Patrick s.n.
	19. VA Grayson Co.: Fusiak 184
	20. NC Jackson Co.: Fusiak 230
	21. NC Haywood Co.: Fusiak 239
	22. NC Buncombe Co.: Fusiak 251
	23. NC Buncombe Co.: Fusiak 256
	24. NC Buncombe Co.: Fusiak 280
	25. NC Yancey Co.: Fusiak 547
	26. NC Yancey Co.: Fusiak 298
	27. NC Yancey Co.: Fusiak 309
	28. TN Carter Co.: Fusiak 580
	29. TN Polk Co.: Fusiak 571
	30. NC Avery Co.: Fusiak 318
	31. NC Avery Co.: Fusiak 322
	32. VA Grayson Co.: Fusiak 329
	33. VA Grayson Co.: Fusiak 331
	34. NC McDowell Co.: Fusiak 310
	35. VA Washington Co.: Fusiak 344
	36. VA Smyth Co.: Fusiak 346
	37. TN Unicoi Co.: Fusiak 625
	38. TN Unicoi Co.: Fusiak 628
P. serpentaria	39. AL Lee Co.: Harper 021 (UNC)
	40. AL Mobile Co.: Rogers 1852 (UNC)

Table III-1 (Continued)

Species	Populations Number and Voucher Specimen[a]
(*P. serpentaria*)	41. VA Charlotte Co.: Ahles 60891 (UNC)
	42. VA Loudoun Co.: Ahles 61526 (UNC)
	43. TN McMinn Co.: Fusiak 592
	44. TN Polk Co.: Fusiak 592
	45. TN Morgan Co.: Schmalzer 1448
	46. TN Grundy Co.: Wofford 77-301
	47. MS Tishmingo Co.: Rogers 46214
	48. TN Coffee Co.: Patrick 1465

[a]Unless otherwise indicated, all specimens are on deposit at TENN or will be distributed later.

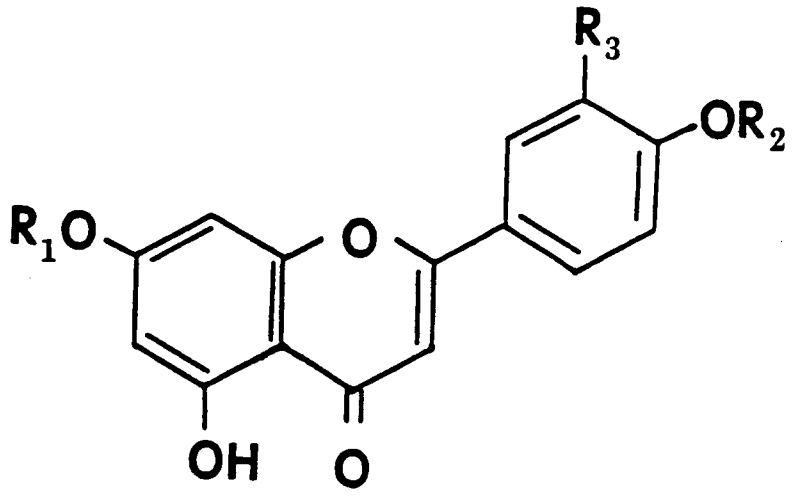


Figure III-1. Flavonoid Compounds Isolated from Leaves of Prenanthes.

1. Luteolin-7-O-diglycoside (R_1: (xylose and glucose); R_2: H; R_3: OH).
2. Luteolin-7-O-glucoside (R_1: glucose; R_2: H; R_3: OH)
3. Luteolin-7-O-glucuronide (R_1: glucuronic acid; R_2: H; R_3: OH)
4. Luteolin-7,4'-O-diglucoside (R_1, R_2: glucose; R_3: OH)
5. Luteolin-7,3'-O-diglucoside (R_1, R_3: glucose, R_2: H)
6. Apigenin-7-O-glucoside (R_1: glucose; R_2, R_3: H)
7. Luteolin (R_1, R_2: H; R_3: OH)

No species-specific flavonoid patterns were observed. As shown in Table III-2, intraspecific variability was as great as interspecific variability. Populations that fit the circumscription of P. cylindrica sensu Small (1933) varied as much as the other taxa examined; thus no discriminating taxonomic information at the specific level could be obtained from flavonoid data. The variability detected here is possibly the result of environmental or physiological factors, but until further study the causes of the variability remain unanswerable.

No previous study of flavonoid chemistry of Prenanthes has been performed. At the generic level, the type of flavonoids present agrees with previous investigations of Lactuceae genera. Of the 100 Lactuceae species in which flavonoids have been identified, 89 (89.0%) contain luteolin or its glucosides (Guppy and Bohm, 1976; Gonzalez, 1977; Harborne, 1977, 1978; Mansour, Saleh and Boulos, 1983). A surprising note is the presence of luteolin-7,4'-O-diglucoside in Prenanthes which has never been reported for any Lactuceae species, even though both the 7 and 4' monoglucosides of luteolin have been reported together. Since it is a fairly common flavone glycoside, it is a good possibility that it has not been reported because of a small data base. Only 2.3% (100 of 2300 species) of all species in the Lactuceae have been examined. The luteolin-7,3'-O-diglucoside is a rare compound that has been reported only once previously in Reseda luteola L. of the Papaverales (Harborne and Williams, 1975). Although these

Table III-2. Distribution of Flavonoids in *Prenanthes* Populations.

Species and Populations[a]	Compounds[b]						
	1	2	3	4	5	6	7
P. altissima							
1	+	+					+
2	+	+	+	+	+	+	+
3	+	+					
4	+	+	+				+
5	+			+			+
6	+	+				+	+
7	+	+					
8	+	+		+			
9	+	+	+	+	+	+	+
10	+	+	+	+	+	+	+
11	+	+					
12	+	+		+		+	
13	+	+	+	+	+	+	+
14	+	+	+	+			
15	+	+					
P. roanensis							
16	+	+	+	+	+	+	+
17	+	+	+	+	+	+	+
18*	+	+					+
19	+	+	+	+			
20*	+	+					+
21*	+	+		+			
22	+	+	+	+	+	+	+
23*	+	+	+	+			
24*	+	+					
25	+	+	+	+	+	+	+
26*	+	+	+				
27	+	+	+				
28	+	+	+	+	+	+	+
29	+	+	+				+
30*	+	+					+
31*	+	+		+			
32*	+	+	+	+	+	+	+
33	+	+					+
34	+	+	+	+		+	
35*	+	+	+	+	+	+	+
36	+	+	+	+	+	+	+
37*	+	+	+	+	+	+	+
38*	+	+	+				+

Table III-2 (Continued)

Species and Populations[a]	Compounds[b] 1	2	3	4	5	6	7
P. serpentaria							
39	+	+	+	+			
40	+	+	+	+			
41	+	+					
42	+	+	+				+
43	+	+	+			+	+
44	+	+	+	+	+	+	+
45	+	+	+				+
46	+	+		+			+
47	+	+		+			
48	+	+	+			+	+

[a]Populations numbers correspond to those listed in Table III-1. Asterisk (*) denotes populations that fit within circumscription of *P. cylindrica sensu* Small (1933).

[b]Compounds numbers correspond to those listed in Figure III-1.

compounds are relatively rare, they offer no insight into the relationship of Prenanthes within the Lactuceae.

CHAPTER IV

CYTOLOGY

Cytology has been an important mainstay of systematic investigations for the last half century. Cytological characters are often regarded as having special importance in taxonomy since chromosomes are more closely connected with the mechanism of heredity than any other feature commonly employed in taxonomy (Stebbins, 1950, 1971; Davis and Heywood, 1963). There is often a relationship between chromosomal change and speciation (Stebbins, 1950, 1971; White, 1978; Grant, 1981). If major structural chromosomal changes (e.g., translocations, inversions) have occurred during speciation, analysis of the meiotic process can reveal the presence of interspecific hybrids. If these changes have occurred, interspecific hybrids often exhibit aberrant meiosis (i.e., formation of multivalents, univalents, bridges, fragments).

Within the genus Prenanthes, two base chromosome numbers with both diploids and tetraploids have been reported, namely $x = 8$ (Babcock, Stebbins and Jenkins, 1937; Löve and Löve, 1966; Powell, Kyhos and Raven, 1974; Tomb, Chambers, Kyhos, Powell and Raven, 1978; Parfitt and Harriman, 1981) and $x = 9$ (Babcock, Stebbins and Jenkins, 1937; Strida and Frazen, 1981; Kirschner, Stepanek and Stepankova, 1982; Löve and Löve, 1982). The former reports of $x = 8$ are confined to species in North America of subgenus Nabalus, while the latter reports of $x = 9$ are for Eurasian species of subgenus

Prenanthes. Within the *P. roanensis* complex, only two species have had chromosome counts reported previously; both *P. altissima* and *P. serpentaria* have been reported as *n* = 8 (Babcock, Stebbins and Jenkins, 1937; Löve and Löve, 1966; Tomb, Chambers, Kyhos, Powell and Raven, 1978).

In the present study, a cytological investigation was undertaken for two reasons. One was to determine the chromosome number of all taxa of the *P. roanensis* complex. Since polyploidy and variation in the chromosomal base number have been reported within *Prenanthes*, it is important to determine the chromosome number of each taxon before proceeding with any cytotaxonomic study. Secondly, the meiotic process of all individuals sampled were examined for irregularities, with particular emphasis placed on examining morphological intermediates that fit the circumscription of *P. cylindrica sensu* Small (1933). These individuals have been suggested to be of intermediate morphology between *P. altissima* and *P. roanensis* and are possibly of hybrid origin between these taxa. Major chromosomal differences between species are common in the Lactuceae subtribe Crepidinae, to which *Prenanthes* belongs (Babcock and Cameron, 1934; Babcock and Emsweller, 1936; Stebbins, Jenkins and Walters, 1953). If *P. altissima* and *P. roanensis* exhibit major structural differences in chromosomes, then irregularities may be apparent during meiosis if these intermediates are truly interspecific hybrids.

Materials and Methods

Young capitula were collected from plants in natural populations, fixed in a mixture of methanol-chloroform-propionic acid (6:3:2, v/v/v; Cooperrider and Morrison, 1967) and stored under refrigeration until examinations. Voucher specimens are listed in Table IV-1. Suitable flowers were dissected out of immature capitula (1-3 mm in length) and squashed in 2% lactic-acetic-orcein stain (Cooperrider and Morrison, 1967). Photographs were taken of some configurations with an Olympus PM-10 A 35-mm photographic system using Kodak Panatomic-X, ASA 32, black and white film.

In an attempt to obtain mitotic chromosome counts, 50-100 seeds of each taxon were placed in petri dishes on moistened filter paper and placed under refrigeration (5°C). After 6 weeks the seeds were removed from refrigeration and placed at room temperature under fluorescent lights (12 hours light, 12 hours dark). However, after 2 months, only one seed of P. roanensis germinated and it survived only 2 weeks. All other seeds failed to germinate and were discarded. Before mitotic chromosome counts can be made from seeds, the germination behavior must be elucidated.

Results and Discussion

Chromosome counts were obtained for all taxa of the P. roanensis complex. The chromosome counts of $n = 8$ for both P. altissima and P. serpentaria are confirmed. The chromosome

Table IV-1. Specimens of Prenanthes Examined Cytologically.

Species	Voucher[a]
P. altissima	NC Buncombe Co.: 544 (81-10)
	TN Carter Co.: 178, 581
	TN Knox Co.: 81-36, 81-37, 81-39, 81-44, 81-45, 449
	TN McMinn Co.: 590, 590a
	TN Polk Co.: 574, 575
	TN Unicoi Co.: 81-55, 565, 566, 567, 568, 569
	VA Washington Co.: 175
P. roanensis	NC Avery Co.: 559, 560
	NC Buncombe Co.: 543 (81-9)
	NC Yancey Co.: 547 (81-13, 81-14, 81-15)
	TN Carter Co.: 99, 113, 133, 184, 580
	TN Polk Co.: 571, 572, 573
	TN Unicoi Co.: 561, 562, 563, 564, 81-48, 81-56, 81-57, 81-58
	VA Grayson Co.: 184, 185
P. serpentaria	TN McMinn Co.: 592, 593
	TN Polk Co.: 576, 577

[a]All collection numbers are of Fusiak and vouchers are on deposit at The University of Tennessee herbarium (TENN).

count of P. roanensis is reported for the first time as n = 8.

No meiotic irregularities were observed in any of the plants examined. If these morphological intermediates are truly interspecific hybrids, major structural changes of chromosomes have not accompanied speciation. However, cryptic structural changes (sensu Stebbins, 1971) undetectable through meiotic analyses may have occurred. Interspecific hybridization, however, appears unlikely. The morphological intermediacy observed is best regarded as representing the extremes of the taxa involved. Numerical taxonomic analyses, especially the results of principal coordinate analysis, support this contention and suggest that the P. roanensis complex contains three morphologically overlapping species (see Chapter V).

CHAPTER V

NUMERICAL TAXONOMY

Taxonomy primarily deals with the laws and principles of classifying objects into groups. One of the major criticisms regarding taxonomists has been that so-called "classical taxonomy" lacks repeatability, testability and objectivity. In order to resolve some of these complaints and to standardize the process by which groups are recognized, the field of numerical taxonomy has arisen. This "new taxonomy" attempts to produce these results by use of statistical techniques in the classification of objects.

The results of numerical taxonomy, however, have not achieved these goals of being objective and repeatable. Classifications have been shown to vary depending on the choice, definition and measurement of characters and on the statistical methods employed (Johnson, 1970; Farris, 1976; Duncan and Baum, 1981). There is also at present no method to evaluate the classifications produced to determine which is "best" or optimal.

Although there are problems with numerical taxonomy, some of its basic methodology has provided useful taxonomic tools. With the aid of computers, it allows the simultaneous evaluation of a large number of characters and individuals. Objectivity is introduced because explicit definitions and decisions are needed in character scoring. Lastly, its results can be tested against initial hypotheses derived from results of other systematic techniques

(Schilling, 1978). Thus, numerical taxonomy when coupled with the results of other systematic techniques can become an important component in the classification process.

A numerical taxonomic investigation was performed to examine the morphological groups of the Prenanthes roanensis complex of the southern Appalachians. An initial null hypothesis was that three species and a series of their hybrids are present within the complex. Along with data from other systematic techniques, the data from these numerical analyses will aid in the classification of the P. roanensis complex.

Materials and Methods

Data for 100 samples or OTUs (= operational taxonomic units: the lowest ranking taxon employed in a study; Sokal and Sneath, 1963) from throughout the range of the P. roanensis complex were obtained for 18 morphological characters (Table V-1) from dried herbarium specimens (Appendix A). Only characters that varied between taxa were included in the numerical analyses. A coefficient of resemblance was calculated between each pair of OTUs. Gower's coefficient (Gower, 1971) was used as the coefficient of resemblance, since it allows the use of mixed qualitative and quantitative data. Although no standardized method of analysis has arisen in numerical taxonomy, utilizing both a method of cluster analysis and one of ordination are often regarded as giving the best results (Sneath and Sokal, 1973; Clifford and Stephenson, 1975).

Table V-1. List of Characters Scored for Numerical Taxonomic Analysis.

Characters

1. Number of inner phyllaries per capitulum (average of four capitula per plant).

2. Length of the inner phyllaries (average of four from four different capitula).

3. Width of the inner phyllaries (average of four from four different capitula).

4. Shape of the inner phyllary tips.
   0 = rounded
   1 = acute

5. Inner phyllary pubescence.
   0 = absent
   1 = marginal clear hispid/ciliate
   2 = marginal dark hispid/ciliate
   3 = midrib clear pilose
   4 = midrib dark pilose
   5 = scattered hispid
   6 = scattered pilose

6. Inner phyllary tip coloration.
   0 = dark or black
   1 = light or green

7. Number of outer phyllaries per capitulum (average of four capitula per plant).

8. Shape of the outer phyllaries.
   0 = triangular
   1 = short lanceolate

9. Outer phyllary pubescence.
   0 = absent
   1 = marginal clear hispid/ciliate
   2 = marginal dark hispid/ciliate
   3 = midrib clear pilose
   4 = midrib dark pilose
   5 = scattered hispid
   6 = scattered pilose

Table V-1 (Continued)

Characters
10. Number of florets per capitulum (average of four capitula per plant).
11. Pappus length.
12. Arrangement of the capitula. 0 = paniculate 1 = corymbiform 2 = thyrsoid/racemiform
13. Plant height.
14. Percentage of total plant height occupied by the inflorescence.
15. Main stem pubescence. 0 = absent 1 = upper portion within the inflorescence 2 = lower portion below the inflorescence 3 = entire stem
16. Leaf shape of leaves below the inflorescence. 0 = entire 1 = lobed 2 = mixed
17. Leaf base shape. 0 = ovate 1 = cordate 2 = hastate/sagittate
18. Peduncle pubescence. 0 = absent 1 = hispid wooly 2 = pilose

These methodologies are primarily useful in the classification process (i.e., arranging OTUs into groups of phenetically similar individuals) with cluster analysis depicting relationships between similar OTUs and ordination portraying the relationships of more distant groups (Clifford and Stephenson, 1975). In the present analyses, the unweighted pair-group using arithmetic means (UPGMA) and principal coordinates analysis (PCA) were used for clustering and ordination, respectively. All computational procedures are given by Sneath and Sokal (1973). Factor analysis was also performed in order to examine the characters primarily responsible for the separation of OTUs along the principal axes. Computations were performed on the DEC-10 and IBM 360/370 computers at The University of Tennessee, Knoxville Computer Center using The University of Tennessee system for analysis of taxonomic data (NUMTAX) programs developed by Dr. Edward E. Schilling and the Statistical Package for the Social Sciences (SPSS) package programs.

Results and Discussion

The phenogram derived from cluster analysis of the 100 OTUs examined is shown in Figure V-1. Clusters corresponding to the species as eventually recognized occur in the phenogram. The OTUs are designated in Figure V-1 by the number corresponding to those in Appendix A and labeled by species as eventually recognized in this study.

The graph of the first two principal axes from PCA for the 100 OTUs is shown in Figure V-2. Results from PCA are approximately

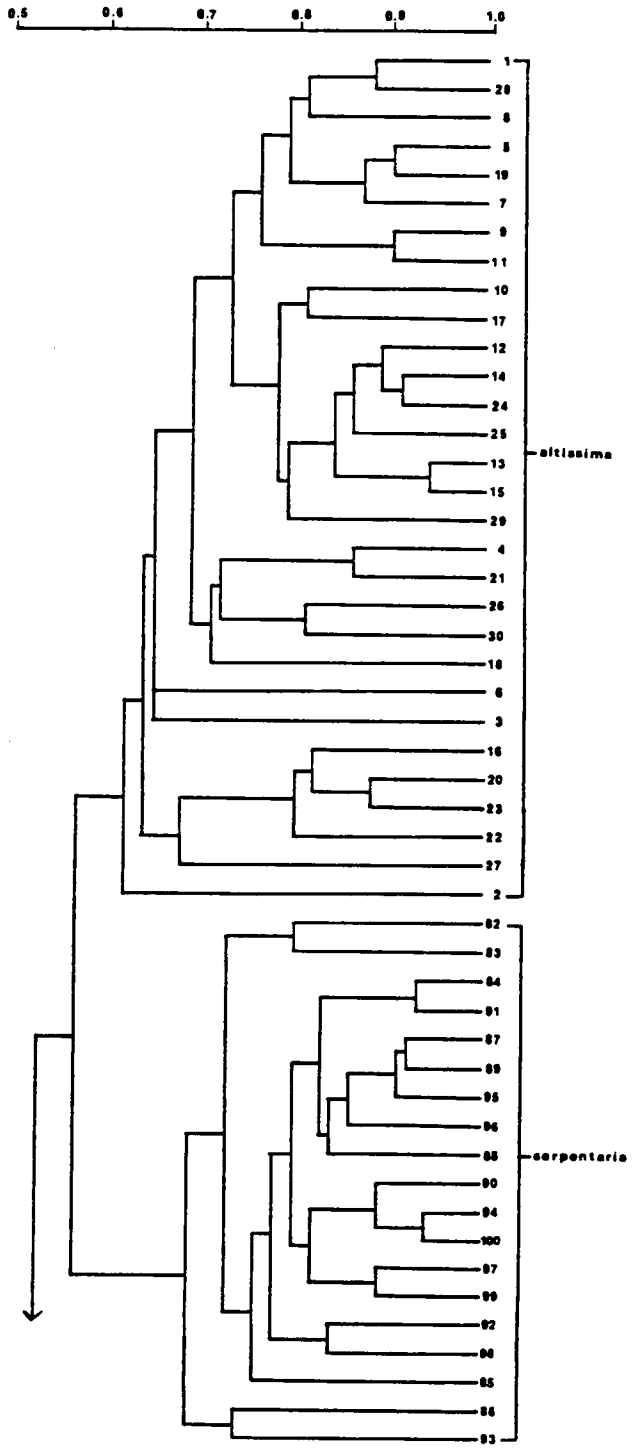


Figure V-1. Phenogram Derived from UPGMA Cluster Analysis of 100 OTUs of the Prenanthes roanensis Complex Using 18 Characters and Gower's Coefficient of Similarity. Stars indicated individuals that fit within the circumscription of P. cylindrica sensu Small (1933).

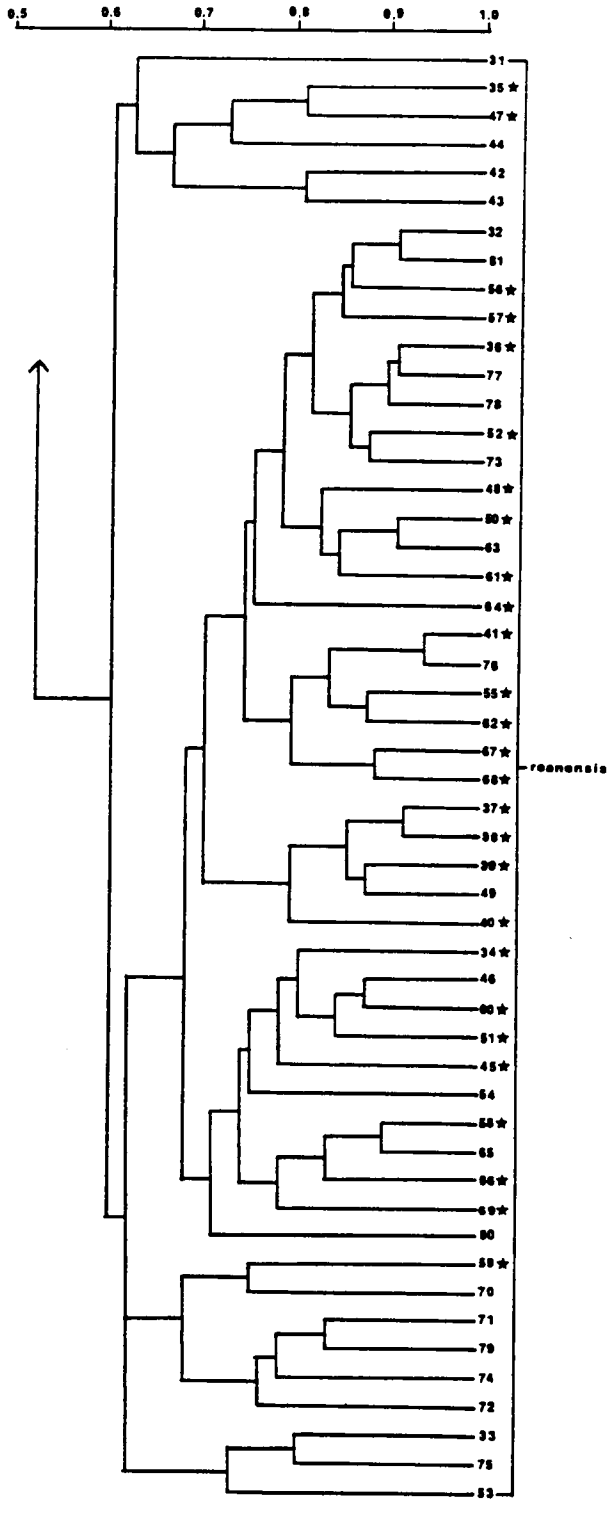


Figure V-1 (Continued)

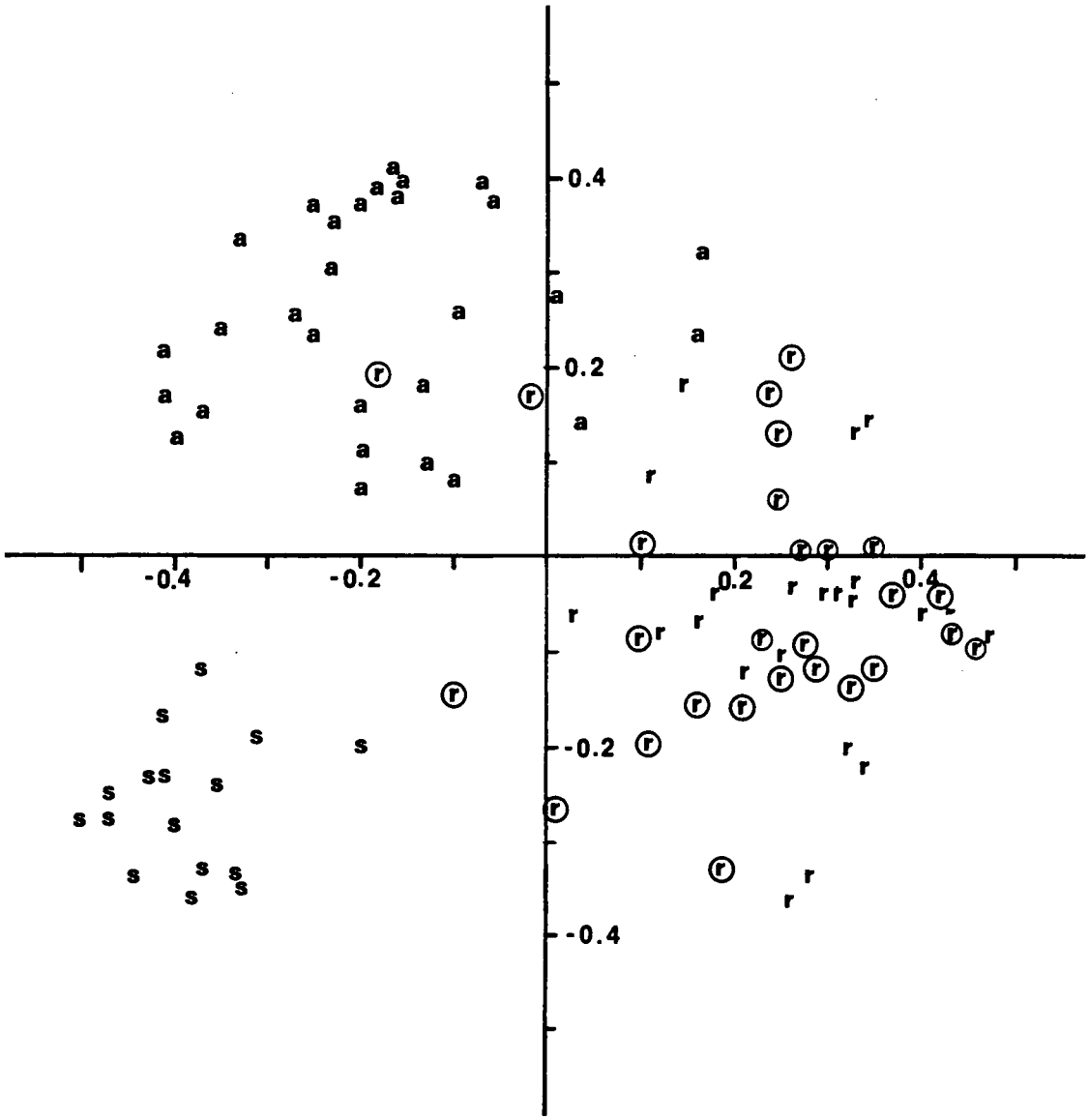


Figure V-2. Graph of 100 OTUs of Prenanthes altissima, P. roanensis and P. serpentaria on the First Two Axes Determined by Principal Coordinate Analysis. Species are denoted by first letter of their specific epithet as they were eventually recognized. Encircled r's denoted individuals that fit within the circumscription of P. cylindrica sensu Small (1933).

the same as from cluster analysis. Placement of OTUs in the graph corresponds well with taxa as eventually recognized; however, there is some overlap between the recognized species of P. roanensis and P. altissima. This partial overlap is probably due to the fact that P. roanensis is more variable in the number of florets and inner phyllaries per capitulum than the other recognized taxa. Both of these features along with phyllary pubescence have been shown by factor analysis to be important in the separation of OTUs along the first two principal axes. This overlap is also supportive of the hypothesis of Milstead (1964) that these taxa are the two most closely related species of the North American members of this polymorphic genus.

In both cluster analysis and ordination, the OTUs corresponding to P. serpentaria form a uniform, highly distinct morphological group (Figure V-1, V-2). These analyses show P. serpentaria to be distinct but differences between P. altissima and P. roanensis are still questionable. Prenanthes serpentaria is easily distinguished from the other taxa by the number of florets and inner phyllaries per capitulum, its corymbiform inflorescence and its hispid phyllaries.

Since P. serpentaria is distinct by these analyses, OTUs corresponding to it were removed from the analysis to see if their presence affected the clustering and ordination of the remaining 81 OTUs. From cluster analysis, the phenogram shown in Figure V-3 shows two large clusters that correspond to P. altissima and

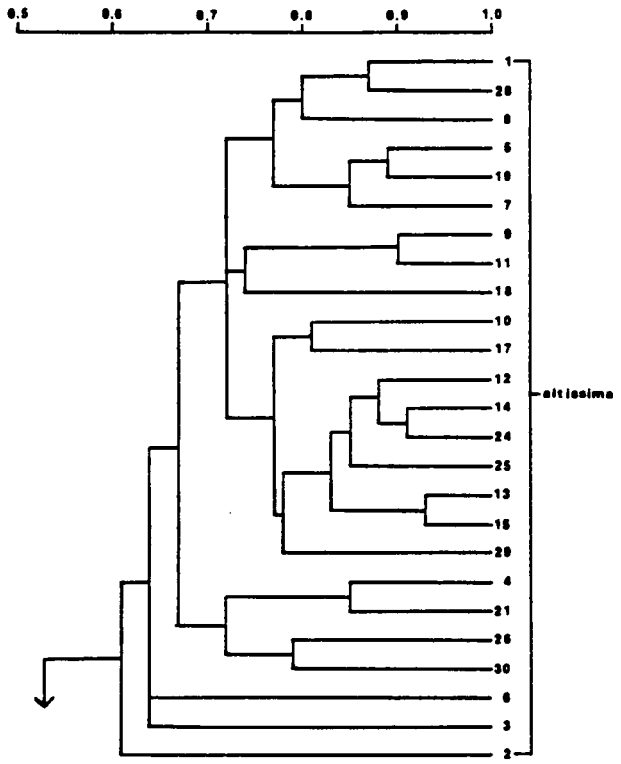


Figure V-3. Phenogram Derived from UPGMA Cluster Analysis of 81 OTUs of Prenanthes altissima and P. roanensis Using 18 Characters and Gower's Coefficient of Similarity. Species are denoted by first letter of their specific epithet as they were eventually recognized. Encircled r's denoted individuals that fit within the circumscription of P. cylindrica sensu Small (1933).

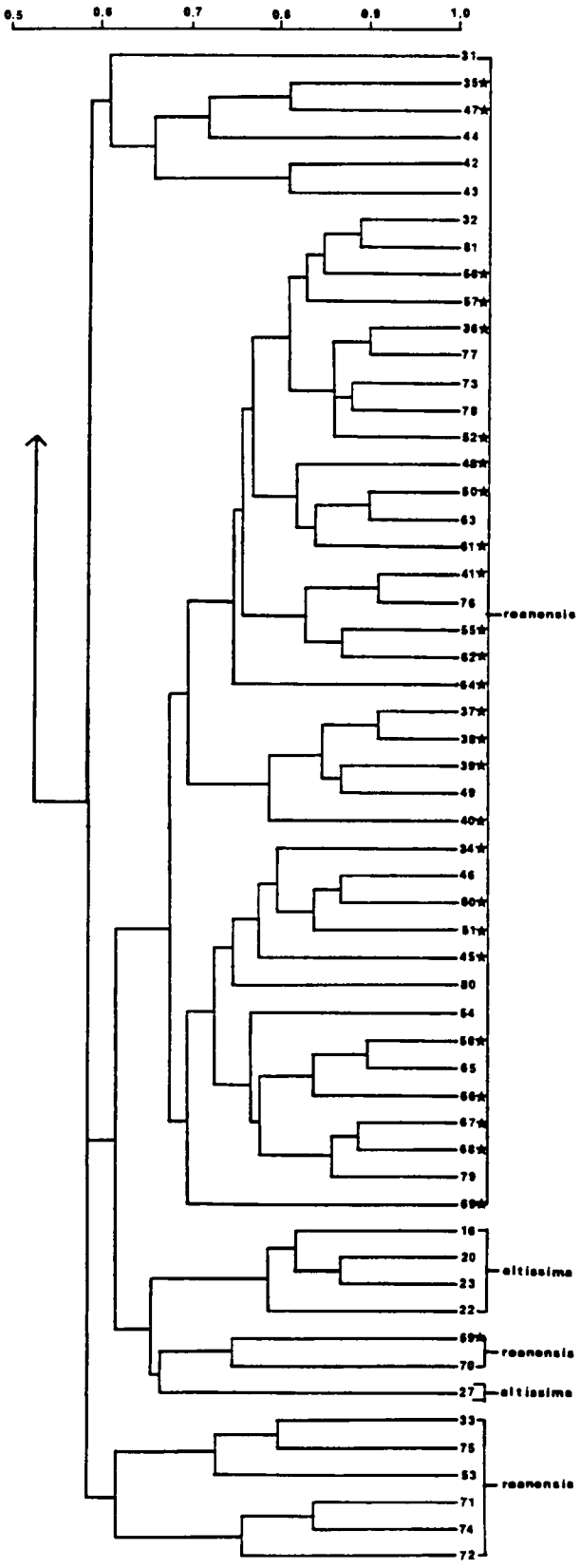


Figure V-3 (Continued)

P. roanensis. However, five OTUs (16,20,22,23,27) eventually identified as P. altissima cluster together within the large cluster of P. roanensis. These five OTUs had one character, a thyrsoid inflorescence, that was not present in any other specimen of P. altissima included in the numerical analysis. Inflorescence type has been shown by factor analysis to be important in the separation of OTUs. This feature is also more characteristic of P. roanensis. These samples were eventually identified as P. altissima because they had 4-5 glabrous inner phyllaries and 4-5 florets per capitulum that are more characteristic of this taxon than they are of P. roanensis.

The ordination results for these 81 OTUs, however, were more complex than the clustering results. The graph of the first two principal axes from PCA depicted in Figure V-4 shows, as in the full data set analyses, an overlapping appearance between the recognized taxa. Even though there is some overlap, a majority of OTUs corresponding to the recognized taxa are associated in similar areas of the graph. Thus, these analyses show that there are two closely related but separable taxa present.

In both cluster and principal coordinate analyses (Figures V-1, V-2, V-3, V-4), individuals that fit the circumscription of P. cylindrica sensu Small (1933) consistently clustered amongst individuals of P. roanensis. Thus these analyses support the contention of Milstead (1964) and Cronquist (1980) that P. cylindrica is conspecific with P. roanensis.

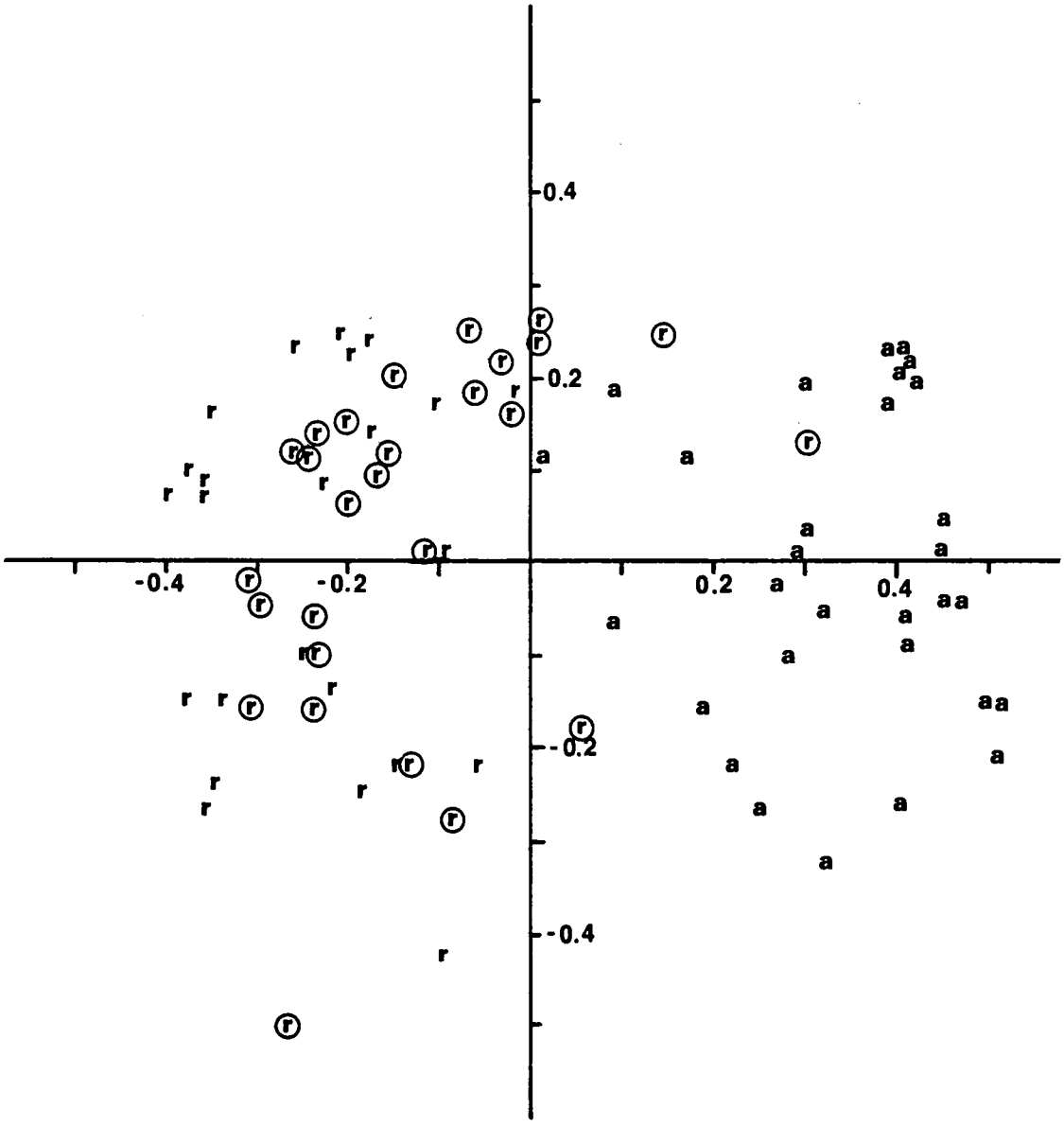


Figure V-4. Graph of 81 OTUs of Prenanthes altissima and P. roanensis on the First Two Axes Determined by Principal Coordinate Analysis. Species are denoted by first letter of their specific epithet as they were eventually recognized. Encircled r's denoted individuals that fit within the circumscription of P. cylindrica sensu Small (1933).

The results of the numerical analysis suggest that there are three morphological groups among the OTUs examined within the P. roanensis complex. The null hypothesis that these OTUs represent three taxa and their hybrids is thus rejected. Hybridization has been ruled out because of the consistency by which groups were formed in both cluster analysis and ordination. The results of UPGMA cluster analysis support the contention that three distinct groups are present that correspond to the eventually recognized taxa; these three groups, however, are less apparent in the ordination results. The ordination results suggest that the three groups are distinct but do overlap partially. It appears, therefore, that the P. roanensis complex contains three distinct but morphologically overlapping species.

In conclusion, these results, especially those derived from cluster analysis, support the recognition of three taxa that were suggested by traditional taxonomic techniques. These techniques revealed nothing that could not be discerned by traditional methodology. They did, however, provide a convenient and useful way to analyze a large volume of data.

CHAPTER VI

TAXONOMIC TREATMENT

Members of the Prenanthes roanensis complex belong to Prenanthes subsection Altissimae (sensu Milstead, 1964). Taxa of this subsection are found throughout the eastern portion of the United States and adjacent Canada and are characterized by paniculate to thyrsoid inflorescences, long petiolate leaves, yellow florets (except in P. alba with lavender to white florets), 4-14 florets per capitulum, 4-10 glabrous or pubescent inner phyllaries subtended by a series of shorter outer phyllaries. As defined by Milstead (1964), subsection Altissimae contains the following five species: P. alba L., P. altissima L., P. roanensis (Chickering) Chickering, P. serpentaria Pursh and P. trifoliolata (Cassini) Fernald.

The P. roanensis complex, as considered in this study, has no formal nomenclatural or taxonomic standing. Its definition is one based on convenience so that easy reference can be made to its members. It is considered to contain the following three taxa: P. altissima, P. roanensis and P. serpentaria. Even though it has no formal standing, its members are easily distinguished from the other members of subsection Altissimae by the following: 4-10 (rarely more in P. serpentaria) yellow (never white, red or lavender) florets per capitulum, 5-10 (never more than 10) glabrous or pubescent inner phyllaries per capitulum, 4-10 short lanceolate (sometimes

approaching triangular in P. serpentaria) outer phyllaries per capitulum and polymorphic leaves.

Although two members of the P. roanensis complex are widely distributed geographically, the key and descriptions that follow are for specimens of the southern Appalachians. Type specimens for each of the taxa were examined to develop nomenclatural consistency. Complete synonymies were not within the scope of the present study. These synonymies, however, may be found in Milstead's (1964) revision for North American species of Prenanthes. Care, however, must be taken regarding these synonymies especially in light of the nomenclatural confusion regarding P. altissima discussed below. All data were obtained from dried herbarium specimens. Within the descriptions, the following symbols are defined as: M=mode, $\bar{x}$=mean, s=standard deviation.

Key to the Prenanthes roanensis Complex

1. Inner phyllaries mostly 4-7 per capitulum; flowers 4-8 per capitulum; inflorescence paniculate or thyrsoid.
    2. Inner phyllaries glabrous, phyllary tips green; 4-6 (mostly 5) florets per capitulum; inflorescence commonly paniculate. . . . . .1. P. altissima
    2. Inner phyllaries with long pilose hairs along the midrib, phyllary tips usually black; 5-8 florets per capitulum; inflorescence thyrsoid or cylindric, sometimes with short branches. . . . . .2. P. roanensis

1. Inner phyllaries more than 7 per capitulum; flowers more than 8 per capitulum, inflorescence corymbiform. . . . . .3. P. serpentaria

1. Prenanthes altissima L. Sp. Pl. 2: 797. 1753.

TYPE: exact locality unknown (LECTOTYPE to be designated pl. 317, figure 2, p. 355 Plukenet, Almagestrum botanicum sive Phytographie Plukenetinae, 1696). See comments below for further discussion.

Stems 3.1-12.7 dm tall, erect arising from a tuberous-thickened taproot, stems green or green with purple areas, usually pubescent within theinflorescence. Leaves alternate, entire, chartaceous, ovate, cordate or palmately lobed. Capitula nodding, arranged commonly in a paniculate inflorescence, sometimes thyrsoid in smaller plants, occupying the upper third of the plant; florets 4-6 (M=5, $\bar{x}$=4.8, s=0.319) per capitulum; inner phyllaries 4-6 (M=5, $\bar{x}$=4.9, s=0.139) per capitulum, 1.38-2.20 mm in width, 8.45-12.5 mm in length, usually lanceolate to elliptic with green tips, essentially glabrous except rarely marginal ciliate hairs, subtended by 4-10 (M=5, $\bar{x}$=6.25, s=1.31) glabrous outer phyllaries per capitulum. Achenes elongate, glabrous and reddish brown, with irregular striations or ribs, pappus of tawny capillary bristles, 5.6-7.3 mm in length. n = 8.

FLOWERING TIME: Late August to early October.

HABITAT: Woodlands, woodland edges, roadsides.

DISTRIBUTION: Eastern United States and adjacent Canada, extending from Maine to South Carolina, Georgia, west to Louisiana, Arkansas, Missouri and Michigan (Figure VI-1). See Appendix B for specimens examined.

COMMENTS: Prenanthes altissima is probably the most common and easily recognized member of the genus in North America. However, it is nomenclaturally complex.

Prenanthes altissima was published in Species Plantarum 2:797 (1753) with the phrase name P. erecta; flosculis quinis, foliis trilobis. According to Stearn (1957), the phrase name is usually a good indication that Linnaeus had material in front of him at the time the description was made. Two synonyms are also listed. The first is to Valliant whose phrase name, Prenanthes canadensis altissima, foliis variis, flore luteolo, was apparently drawn from a specimen collected by Sarraz but which Linnaeus would not have seen (Jarvis, personal communication). The second synonymy, Sonchus elatus f. dendroides virginianus, ari in modum ariculatus foliis, ramosissimus, floribus luteis, parivis pentapetalis, is to an illustration in Leonard Plukenet's (1696) Almagestrum botanicum sive Phytographie Plukenetinae that is based on a specimen in the Sloane Herbarium (vol. 102, fol. 30, BM).

Milstead (1964), in his unpublished doctoral dissertation, suggests that the type of P. altissima is sheet 952.8 at the Linnaean Herbarium (LINN). This sheet contains two specimens. The information listed in Table VI-1 that was kindly provided by

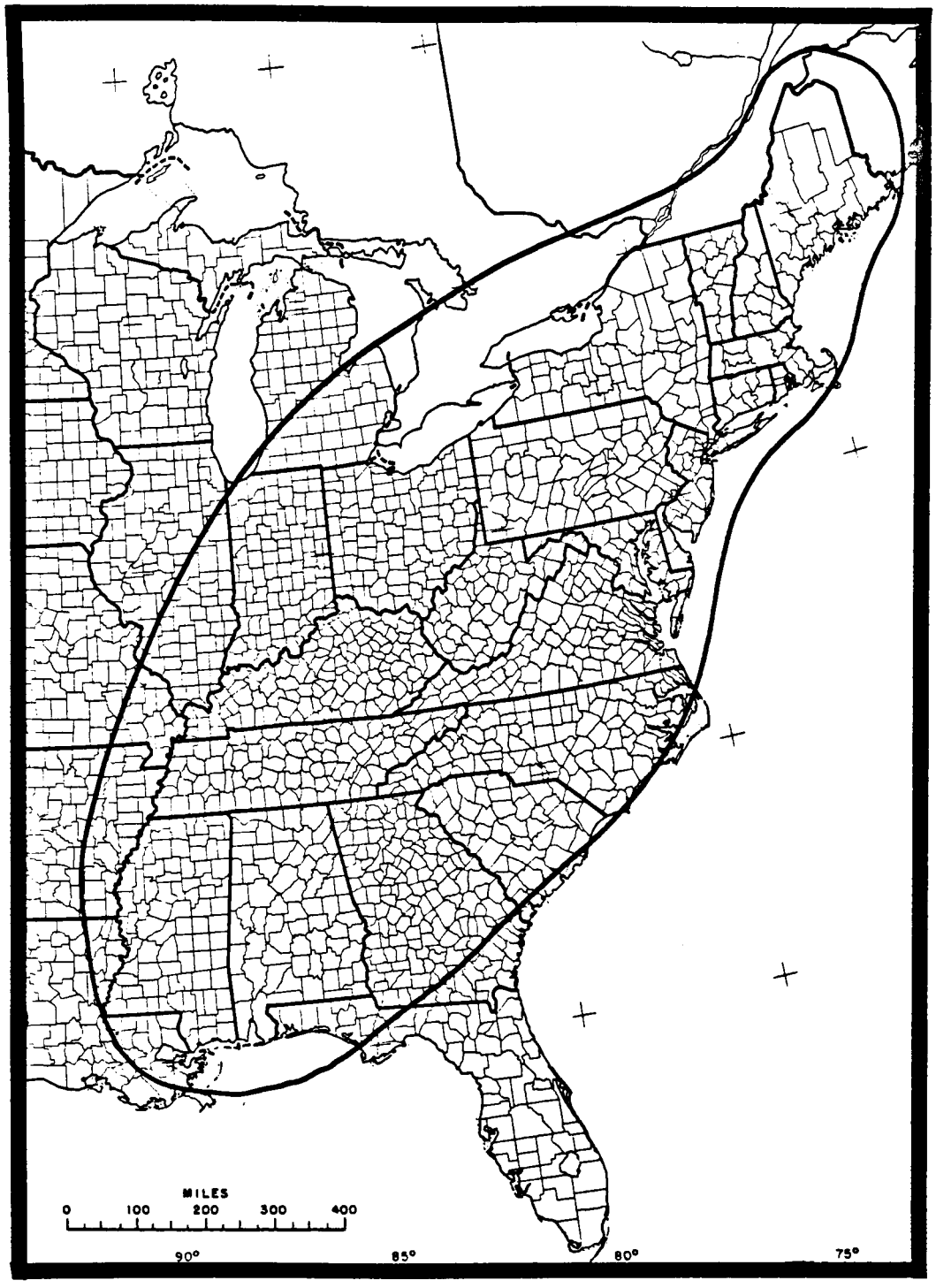


Figure VI-1. Distribution of Prenanthes altissima. Adapted from Milstead (1964).

Table VI-1. Morphological Information on Probable Prenanthes altissima Type Specimens.

Characters	Specimens			
	LINN 952.7[a]	LINN 952.8 Lefthand[a]	LINN 952.8 Righthand[a]	S 69.7[b]
Number of inner phyllaries per capitula 4-6 vs 8 or more	c. 8	4-6	c. 8	8, sometimes 7, difficult to count without damaging specimen.
Number of florets per capitula 4-6 vs. 8 or more	8 or more	c. 6	c. 8	c. 8 or more, difficult to count without damaging specimen.
Floret color (IF POSSIBLE) yellow vs. red/white	not possible	not possible	not possible	? The specimen is in fruit.
Pappus color tawny vs. reddish brown	tawny	tawny	reddish brown	tawny
Pubescence on the inner phyllaries absent vs. present IF PRESENT: TYPE: ciliate vs. long pilose LOCATION: margins, midrib or scattered	Tiny whitish glandular structures over abaxial surface, much more dense towards the apex. Adaxial surface glabrous.	Glabrous. Scattered, small whitish incrustations.	As in 952.7	A few rather coarse hairs dorsally on the midrib only.

Table VI-1 (Continued)

Characters	Specimens			
	LINN 952.7[a]	LINN 952.8 Lefthand[a]	LINN 952.8 Righthand[a]	S 69.7[b]
Pubescence on the outer phyllaries	Present, as in inner, scattered but mainly in midrib area.	Whitish incrustations, mainly in midrib	As in 952.7, mainly marginal, brownish	Apparently none, possibly occasional shorthairs.

[a]Information supplied by Dr. C. E. Jarvis of the British Museum (Natural History) of London (BM).

[b]Information supplied by Dr. Kare Bremer of the Swedish Museum of Natural History, Stockholm (S).

Dr. C. E. Jarvis (BM) strongly suggests that the specimen in the upper lefthand corner of the sheet is P. altissima, as we regard it. However, the specimen in the lower righthand corner of the sheet is probably P. alba as was likewise suggested by Milstead (1964). Although sheet 952.8 (LINN) bears "altissima" in Linnaeus' hand, it does not bear the Species Plantarum number (i.e., 5) that would certainly indicate it was in Linnaeus' possession in 1753. The sheet also bears two names, "floridanus" and "Sonchus 7," that are in Linnaeus' hand and were subsequently crossed out. Evidently, Linnaeus first referred to this specimen as Sonchus floridanus and later, after 1753, realizing his mistake, corrected the name to "altissima." The sheet 952.8 (LINN) thus cannot be regarded as the type of P. altissima, since Linnaeus, himself, did not regard it as belonging to that taxon in 1753.

Another probable type specimen for P. altissima is sheet 952.7 (LINN). It bears the number "5" and "alba" in Linnaeus' hand. As P. alba is species number 6, either the number or name is incorrect. From information listed in Table VI-1, the specimen is probably P. alba as we now consider it; thus the number is apparently wrong.

Lastly, there is another specimen in the Linnaean Herbarium at Stockholm (S 69.7) which, although fragmented, may serve as the type. The sheet bears "5," "altissima" and "K" in Linnaeus' hand. Detailed information that was kindly provided by Dr. Kare Bremer (S) concerning this specimen listed in Table VI-1, however, indicated

that the specimen is not P. altissima but more probably is P. serpentaria as we now regard it. Thus specimen 69.7 (S) cannot be regarded as the type.

Therefore, in order to maintain nomenclatural and taxonomic consistency with the Linnaean concept and our present concept of P. altissima, the illustration in Plukenet's (1696) Almagestrum botanicum sive Phytographie Plukenetinae should be designated as the lectotype of P. altissima.

2. Prenanthes roanensis (Chickering) Chickering. Bot. Gaz. 6: 191. 1881.

   Nabalus roanensis Chickering. Bot. Gaz. 5:155. 1880. TYPE: United States: North Carolina, Roan Mt., Prof. Chickering, 1878. (LECTOTYPE to be designated US-accession number 47904!).

   Nabalus cylindricus Small. Fl. S. E. U. S., p. 1312. 1903. TYPE: United States: Tennessee, Roan Mt., Dr. and Mrs. Britton s.n. (HOLOTYPE: NY!).

   Prenanthes cylindrica (Small) Braun. Annot. Cat. Spermatophyt. Ky. p. 153. 1943.

Stems 2.8-14.4 dm tall, erect arising from a tuberous-thickened taproot, stems green with purple areas or purple. Leaves alternate, subcoriaceous to coriaceous, sagittate to hastate or palmately lobed. Capitula nodding, arranged in a thyrsoid inflorescence, sometimes with short branches, occupying the upper two-thirds or half of the plant; florets 4-9 (M=6, $\bar{x}$=7, s=1.4) per capitulum; inner phyllaries

5-8 (M=5, $\bar{x}$=6.4, s=1.06) per capitulum, 1.4-3.1 mm in width, 5.7-11.8 mm in length, usually lanceolate, tips acute or rounded, green or black with black tips, with clear or dark pilose hairs on the midrib, subtended by 4-10 (M=6.2, $\bar{x}$=6.2, s-1.116) pubescent outer phyllaries per capitulum. Achenes elongate, glabrous and reddish brown, with irregular striations or ribs, pappus of tawny capillary bristles, 4.5-7.7 mm in length. n = 8.

FLOWERING TIME: August to September.

HABITAT: Grassy balds, woodlands and roadsides at high elevations (greater than 4000 feet).

DISTRIBUTION: Western North Carolina, east Tennessee and southwest Virginia (Figure VI-2). See Appendix B for specimens examined.

COMMENTS: Prenanthes roanensis has been listed on the rare plant lists of North Carolina (Cooper, Robinson and Funderburg, 1979), Tennessee (Committee for Tennessee Rare Plants, 1978) and Virginia (Porter, 1979) and has also been proposed for federal listing (U.S. Department of the Interior, 1980). Based on the report of Wofford and Smith (1981, Special Report, U.S. Fish and Wildlife Service) and field observations made during the present study, P. roanensis is not currently threatened. It is presently known from at least 20 populations that are primarily on federally protected public lands (e.g., Great Smoky Mountains National Park, Cherokee National Forest) and is abundant on natural sites (e.g., Roan Mt., Whitetop Mt.) and on the highway right-of-way throughout

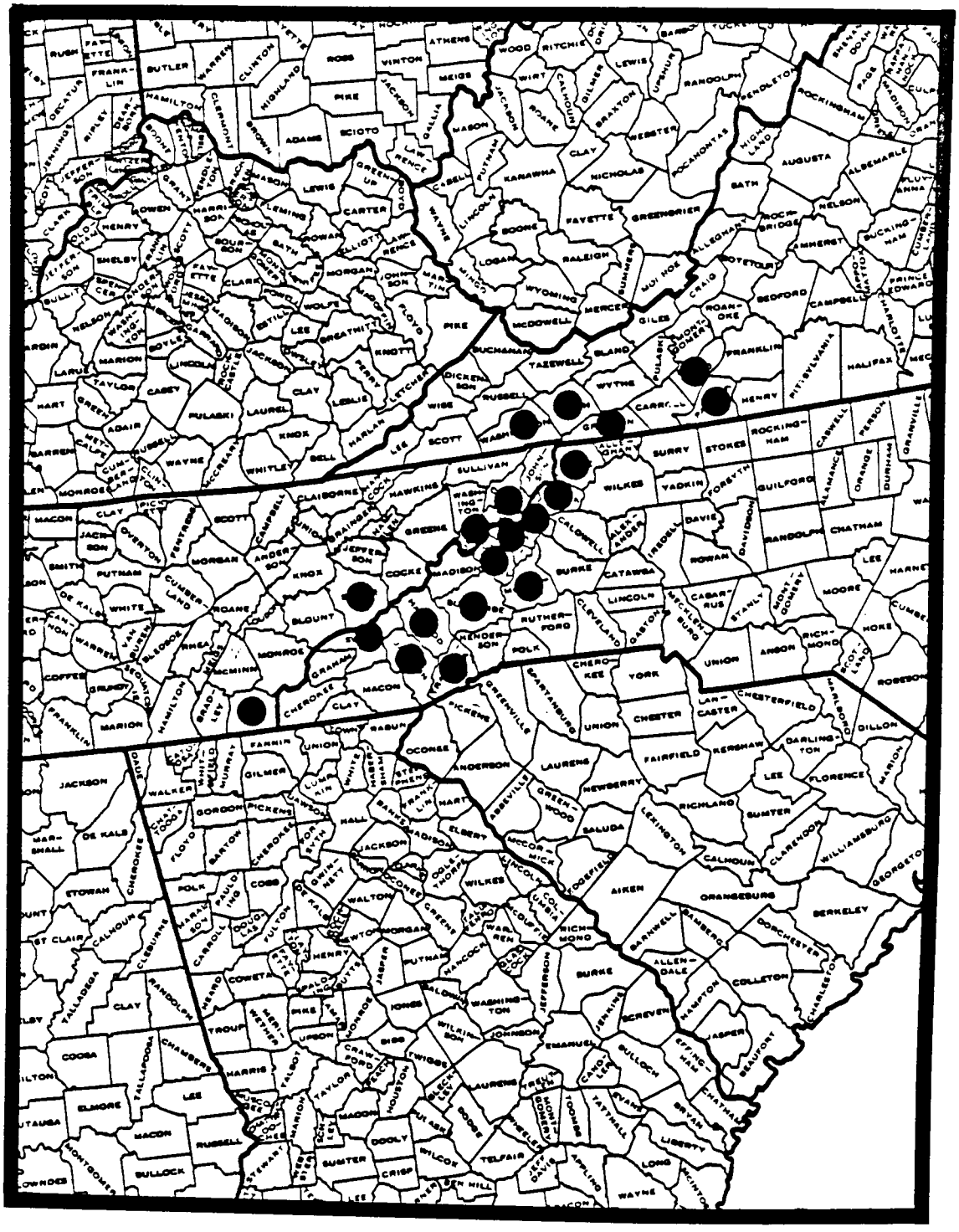


Figure VI-2. Distribution of Prenanthes roanensis.

the Blue Ridge Parkway. Therefore it is recommended that P. roanensis not be considered for federal listing; however, extant populations should be monitored by the individual states, particularly Tennessee and Virginia which have only 4 or 5 populations, to ensure the continued existence of this fascinating taxon.

3. Prenanthes serpentaria Pursh. Fl. Amer. Sept. 2:499. 1814.
TYPE: United States: Virginia and Carolinas, exact locality unknown (LECTOTYPE to be designated pl. 21, p. 499, Pursh, Fl. Amer. Sept. 1814). See comments below for further discussion.

Stems 5.4-16.5 dm tall, erect arising from a tuberous-thickened taproot, stems green or green with purple areas. Leaves alternate, coriaceous, pinnately lobed (sometimes simple f. simplicifolia Fernald). Capitula nodding, arranged in a corymbiform inflorescence, occupying the upper half to third of the plant; florets 7-12 (M=9, $\bar{x}$=9.8, s=0.97) per capitulum; inner phyllaries 6-10 (M=8, $\bar{x}$=7.7, s=0.46) per capitulum, usually lanceolate with acute tips, green occasionally with purple areas, usually scattered to scarce, hispid hairs on the midrib (rarely glabrous), subtended by 4-12 (M=9, $\bar{x}$=8.6, s=1.06) lanceolate approaching short triangular hispid outer phyllaries. Achenes elongate, glabrous and reddish brown, with irregular striations or ribs, pappus of tawny capillary bristles, 6.5-8.5 mm in length. n = 8.

FLOWERING TIME: Late August to early October.

HABITAT: Woodlands, thickets and roadsides.

DISTRIBUTION: Eastern United States from Massachusetts to northern Florida, west to Louisiana, Tennessee, Kentucky and southern Ohio (Figure VI-3). See Appendix B for specimens examined.

COMMENTS: Prenanthes serpentaria is easily distinguished from the other taxa examined by its larger capitula, its corymbiform inflorescence and its usually pinnately lobed leaves. One nomenclatural problem, however, does exist. Pursh (1814) in his description of this taxon, stated that it has pale purple flowers. This has led to confusion with another taxon, P. alba, and has resulted in a long synonym for both taxa (Milstead, 1964). A complete list of synonymy however was not within the scope of the present study. This information along with further comments, however, may be found in Milstead's (1964) revision of the North American species of Prenanthes. Fortunately, an illustration accompanies the description that clearly shows P. serpentaria, as we now consider it, with yellow flowers and its pinnately lobed leaves, thus resolving much of the confusion.

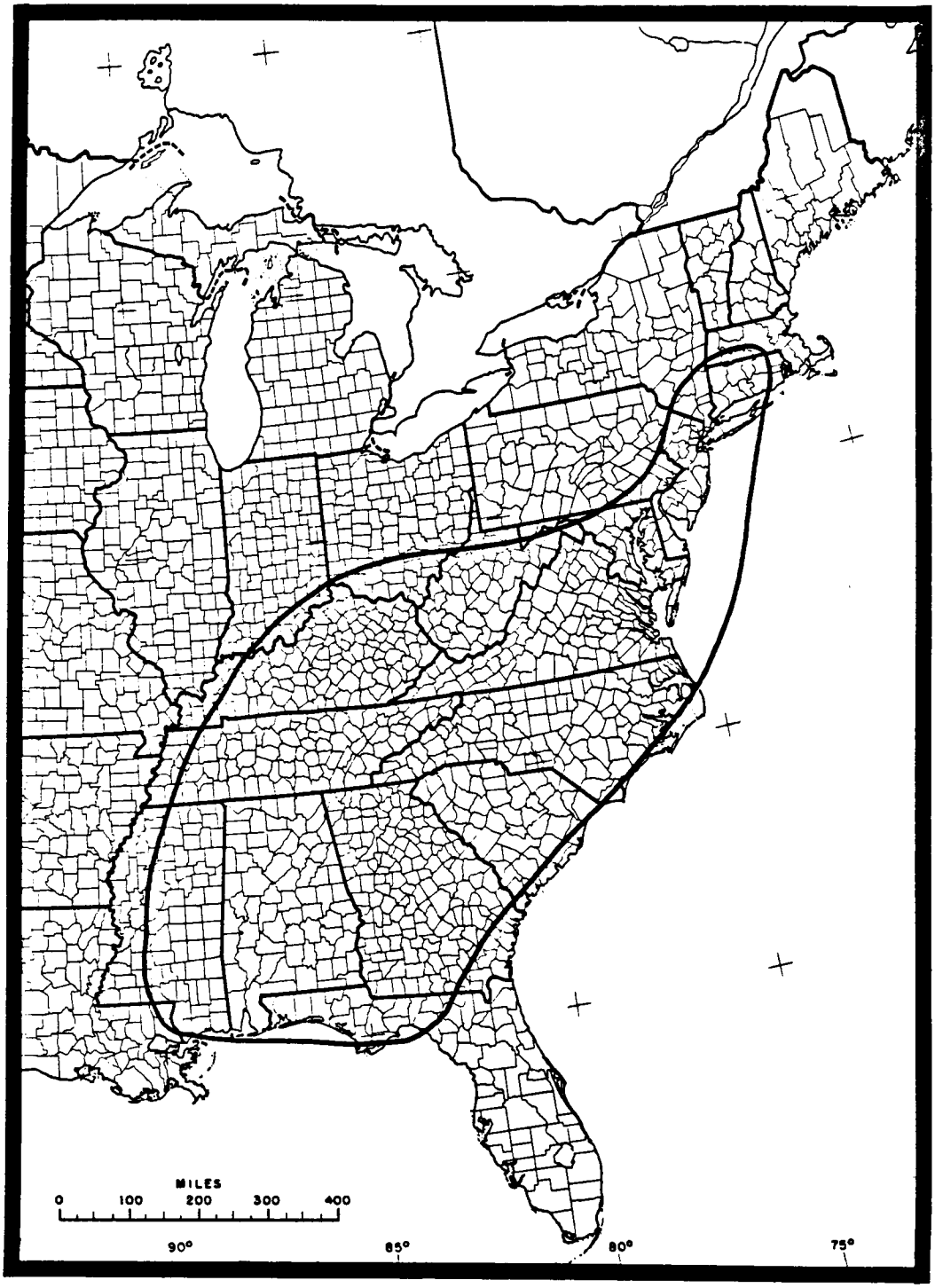


Figure VI-3. Distribution of Prenanthes serpentaria. Adapted from Milstead (1964).

CHAPTER VII

CONCLUSIONS

The purpose of the present study was threefold. First, the species validity, limits and relationships of members of the Prenanthes roanensis complex were evaluated using scanning electron microscopy, flavonoid chemistry, cytology, numerical taxonomy and traditional taxonomic techniques. Second, to determine if interspecific hybridization was occurring amongst members of the complex. Third, the endangered and threatened status of P. roanensis was assessed based on data obtained during the study.

At the present time, a conservative treatment of the P. roanensis complex is expounded (see Chapter VI). Traditional taxonomic techniques and numerical taxonomy suggest that the complex contains the following three morphologically distinct but overlapping species: P. altissima, P. roanensis and P. serpentaria. Results from cluster analyses support the recognition of three distinct taxa, while the ordination results of principal coordinate analyses show that the taxa do partially overlap. Results from flavonoid chemistry, cytological studies and scanning electron microscopy of pollen and ligules fail to reveal interspecific differences. These similarities indicate that these taxa are closely related to one another and that they form a natural coherent group of species.

Since flavonoid analysis, cytology and scanning electron microscopy of pollen and ligules did not reveal species-specific characters, the occurrence of natural interspecific hybridization could not be discerned from these results. Numerical analysis of gross morphological data also did not suggest the occurrence of interspecific hybridization. Although interspecific hybridization was not detected, its occurrence cannot be ruled out at this time to explain the morphological intermediacy.

Lastly, P. roanensis should not be considered federally endangered or threatened. It is presently known from 20 counties in North Carolina, Tennessee and Virginia (see Appendix B), and it is relatively abundant at most sites which are federally protected public lands.

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APPENDIXES

APPENDIX A

SPECIMENS EXAMINED IN NUMERICAL ANALYSES*

Prenanthes altissima L.

1. TN: Hardin Co., Jones 3204 (VDB); 2. VA: Montgomery Co., Kral 11540 (VDB); 3. NY: Albany Co., Russell 822531; 4. VT: Bennington Co., Knowlton s.n.; 5. NC: Orange Co., Stewart 31 (UNC); 6. NC: Haywood Co., Ahles and Duke 50345 (UNC); 7. VA: Giles Co., Massey s.n. (VPI); 8. NC: Haywood Co., Fusiak 243; 9. TN: Carter Co., Fusiak 160; 10. TN: Carter Co., Fusiak 174; 11. TN: Cocke Co., Fusiak 204; 12. TN: Cocke Co., Fusiak 208; 13. TN: Cocke Co., Fusiak 207; 14. TN: Cocke Co., Fusiak 214; 15. TN: Cocke Co., Fusiak 213; 16. NC: Haywood Co., Fusiak 227; 17. TN: Cocke Co., Fusiak 201; 18. TN: Cocke Co., Fusiak 205a; 19. TN: Carter Co., Fusiak 161; 20. NC: Haywood Co., Fusiak 224; 21. TN: Carter Co., Fusiak 163; 22. TN: Carter Co., Fusiak 174; 23. TN: Carter Co., Fusiak 171; 24. TN: Cocke Co., Fusiak 203; 25. TN: Cocke Co., Fusiak 205b; 26. NC: Buncombe Co., Fusiak 251; 27. NC: Buncombe Co., Fusiak 253; 28. NC: Buncombe Co., Fusiak 252; 29. NC: Avery Co., Fusiak 315; 30. NC: Avery Co., Fusiak 313.

Prenanthes roanensis (Chickering) Chickering

31. NC: McDowell Co., Bell 4432 (VDB); 32. VA: Montgomery Co., Kral s.n. (VDB); 33. VA: Washington Co., Croy 676 (VPI); 34. VA: Grayson Co., Patrick s.n.; 35. VA: Grayson Co., Patrick s.n.; 36. NC: Yancey Co., Fusiak 298; 37. NC: Avery Co., Fusiak 322; 38. NC: Avery Co., Fusiak 318; 39. NC: Avery Co., Fusiak 320; 40. NC: Avery Co., Fusiak 327; 41. VA: Grayson Co., Fusiak 329; 42. TN: Carter Co., Fusiak 600; 43. TN: Carter Co., Fusiak 601; 44. TN: Carter Co., Fusiak 602; 45. NC: Avery Co., Fusiak 154; 46. NC: Avery Co., Fusiak 145; 47. VA: Grayson Co., Patrick s.n.; 48. VA: Grayson Co., Fusiak 176; 49. NC: Yancey Co., Fusiak 295; 50. NC. Buncombe Co., Fusiak 280; 51. NC: McDowell Co., Fusiak 310; 52. NC: Haywood Co., Fusiak 237; 53. NC: Ashe Co., Fusiak 177; 54. NC: Avery Co., Fusiak 144; 55. NC: Jackson Co., Fusiak 230; 56. TN: Unicoi Co., Fusiak 625; 57. NC: Transylvania Co., Fusiak 239; 58. NC: Transylvania Co., Fusiak 250; 59. NC: Jackson Co., Fusiak 231; 60. NC: Buncombe Co., Fusiak 256;

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*Unless otherwise indicated, specimens are on deposit at TENN or will be distributed.

61. NC: Buncombe Co., Fusiak 273; 62. NC: Buncombe Co., Fusiak 258; 63. NC: Buncombe Co., Fusiak 290; 64. NC: Buncombe Co., Fusiak 292; 65. NC: Avery Co., Fusiak 316; 66. NC: Avery Co., Fusiak 317; 67. NC: Avery Co., Fusiak 325; 68. NC: Avery Co., Fusiak 326; 69. TN: Unicoi Co., Fusiak 13-2 (655); 70. NC: Yancey Co., Fusiak 309; 71. VA: Washington Co., Fusiak 342; 72. VA: Washington Co., Fusiak 344; 73. VA: Grayson Co., Fusiak 331; 74. VA: Grayson Co., Fusiak 328; 75. NC: Yancey Co., Fusiak 297; 76. NC: Yancey Co., Fusiak 300; 77. NC: Yancey Co., Fusiak 301; 78. NC: Yancey Co., Fusiak 302; 79. NC: Yancey Co., Fusiak 650; 80. NC: Yancey Co., Fusiak 304; 81. VA: Grayson Co., Fusiak 330.

Prenanthes serpentaria Pursh

82. TN: Cumberland Co., Somers 893 (VDB); 83. NC: Transylvania Co., Ware and White 3469 (VDB); 84. SC: Colleton Co., Ahles and Bell 21094 (UNC); 85. VA: Culpeper Co., Ahles and James 61594 (UNC); 86. VA: Augusta Co., Massey s.n. (VPI); 87. MS: Forrest Co., Rogers 4718 (VDB); 88. AL: Talladega Co., Kral 44610 (VDB); 89. TN: Sevier Co., Rogers 42438; 90. TN: Roane Co., Shanks 13794; 91. NC: McDowell Co., Bell 4523 (UNC); 92. NC: Montgomery Co., Radford 19474 (UNC); 93. VA: Prince Edward Co., Ahles and James 62672 (UNC); 94. SC: Calhoun Co., Ahles 35283 (UNC); 95. SC: Lee Co., Radford 29344 (UNC); 96. AL: Mobile Co., Rogers and Lelong 1852 (VDB); 97. AL: Washington Co., Kral 37226 (VDB); 98. VA: Buchanan Co., Kral 14227 (VDB); 99. FL: Leon Co., Kral 3612 (VDB); 100. VA: Dickenson Co., Kral 14253 (VDB).

APPENDIX B

SPECIMENS EXAMINED

Prenanthes altissima L.

ALABAMA: Cleburne Co., Kral 44163 (VDB); Colbert Co., Kral 44058 (VDB), McDaniel 12410 (VDB); Dekalb Co., Kral 37658 (VDB); Henry Co., Kral 37982 (VDB); Jackson Co., Kral 37695 (VDB); Lauderdale Co., Kral 62856 (VDB); Shelby Co., Kral 41498-2 sheets (VDB); St. Clair Co., Bostick 247-8 (UNC); Sumter Co., Kral 54640 (VDB); Winston Co., Kral 29458 (UNC, VDB).

ARKANSAS: Pike Co., Demaree 9551 (TENN); Stone Co., Demaree 29882 (UNC).

GEORGIA: Dade Co., Bozeman 8633 (UNC).

KENTUCKY: Bullitt Co., Gunn 857 (UNC); Caldwell Co., Athley 2970 (VDB); Calloway Co., Athley 2162 (UNC); Crittendon Co., Athley 3875 (VDB); Jessamine Co., Varner 11848 (TENN); Meade Co., Woodward 3881 (UNC); Nelson Co., Wharton 5763 (TENN); Powell Co., Higgins 2023 (UNC), Kral 59060 (VDB), Wharton 5215 (TENN).

LOUISIANA: Caldwell Parish, Marx 2775A (UNC, VDB).

MARYLAND: Frederick Co., Baltars 169 (UNC).

MASSACHUSETTS: Hampshire Co., Ahles 79941 (TENN).

MISSISSIPPI: George Co., Temple 2456 (UNC); Marshall Co., Temple 4316 (UNC); Tishmingo Co., Coleman 50227 (TENN), Rogers 45848 (TENN), 46226 (TENN), Temple 4389 (UNC).

MISSOURI: Ballinger Co., Thomas and Smith 17080 (TENN); Reynolds Co., Evans et al. 42563 (TENN).

NEW HAMPSHIRE: Coos Co., Ware 7132 (VDB).

NEW YORK: Albany Co., Russell R822531 (TENN).

NORTH CAROLINA: Ashe Co., Ashe s.n. (UNC), Radford 41196 (UNC); Avery Co., Barlett 2983 (UNC), Ahles and Duke 49532 (UNC); Buncombe Co., Bartlett 2964 (VDB, UNC); Burke Co., Bell 15124-1 (UNC); Catawba Co., McNeeley 634 (UNC); Cheatham Co., Ramseur and Hammond 2840 (UNC), Bannister and Mueller s.n. (UNC); Chowan Co., Ahles and Duke 51059 (UNC, TENN); Clay Co., Radford 16227 (UNC), Radford s.n. (UNC); Cumberland Co., Ahles 36696 (UNC); Davie Co., Radford 19909 (UNC); Forsyth Co., Schallert 4128 (UNC); Gaston Co., Ahles and Leisner 18868 (UNC); Granville Co., Ahles and Leisner 20054 (UNC); Halifax Co., Treiber and Nesom 707 (UNC); Haywood Co., Ahles and Duke 50345 (UNC); Iredell Co., Ahles and Haesloop 51924 (UNC); Jackson Co., Radford s.n. (UNC); Johnston Co., Radford 29183 (UNC); Iredell Co., Ahles and Haesloop 51924 (UNC); Jackson Co., Radford s.n. (UNC); Johnston Co., Radford 29183 (UNC); Madison Co., Ahles and Duke 50538 (UNC); Mecklenburg Co., Daggy 3867 (UNC); Mitchell Co., Ahles and Duke 49779 (UNC); Montgomery Co., Radford

19403 (UNC), Wickland 1652 (UNC); Moore Co., Houck 618 (UNC), Houck 773 (VPI), Radford 8790 (UNC); Nash Co., Ahles and Leisner 21326 (UNC); Orange Co., Gentry 35 (UNC), Ivey 65 (UNC), Parker 26 (UNC), Radford and Stewart 81a (UNC), Stewart 31 (UNC), Totten s.n. (UNC), Whigman 1777 (UNC), Ward s.n. (UNC), Wicker 38 (UNC); Person Co., Duke 0471 (UNC); Randolph Co., Bell 15667 (UNC); Richmond Co., Radford 19151 (UNC); Rockingham Co., Radford 18586 (UNC), 18597 (UNC); Rowan Co., Horton 408 (UNC); Stanly Co., Ahles and Leisner 19924 (UNC); Swain Co., Radford 17610 (UNC); Transylvania Co., Stewart s.n. (UNC), Ware and White 3277 (VDB); Union Co., Ahles 34124 (UNC); Vance Co., Ahles and Leisner 20454 (UNC); Wake Co., Sawyer 2127 (UNC); Warren Co., Ahles and Bell 21412 (UNC); Watauga Co., Ahles and Duke 49281 (UNC), 49320 (UNC), 49373 (UNC), Rohrer 2297 (UNC), 2298 (UNC), Smyth 2555 (VPI); Yadkin Co., Radford 19915 (UNC); Yancey Co., Ahles and Duke 50795 (UNC).

OHIO: Coshocton Co., Moldenke 14055 (VDB); Lake Co., Tandy 999 (VDB).

OKLAHOMA: Le Flore Co., Taylor and Taylor s.n. (VDB).

PENNSYLVANIA: Bucks Co., Morton 2317 (TENN); Verango Co., Baltzell 5-133 (VDB).

SOUTH CAROLINA: Abbeville Co., Radford 30816 (UNC); Cherokee Co., Freeman 56847 (UNC); Chesterfield Co., Radford 18824 (UNC); Edgefield Co., Radford 30314 (UNC); Greenville Co., Peattie TR117 (UNC); Kershaw Co., Radford 30118 (UNC); Lancaster Co., Ahles 24848 (UNC); Laurens Co., Bell 10195 (UNC, VDB); Marlboro Co., Radford 18990 (UNC); Oconee Co., Radford 17740 (UNC); Saluda Co., Radford 30516 (UNC); Spartanburg Co., Bell 10344 (UNC); Sumter Co., Radford 29654 (UNC); Union Co., Bell 10629 (UNC); York Co., Ahles 34551 (UNC).

TENNESSEE: Bledsoe Co., Kral 62819 (VDB); Blount Co., Hoss 18059 (TENN), Underwood 1419 (TENN), Thomas s.n. (TENN), Thomas and Sharp s.n. (TENN); Carter Co., Warden 306 (ETSU), 1337 (ETSU), Shanks 1156 (TENN); Cheatham Co., Tseng 11 (VDB), Kral 29328 (VDB); Claiborne Co., Drumke, Ramsey, Sharp and Sharp 30499 (TENN); Cocke Co., Sharp 27663 (TENN), Chapman s.n. (TENN); Davidson Co., Shaffer 40 (VDB), Kral 29249 (VDB), Yates 1 (VDB), Quarterman 692 (VDB); De Kalb Co., DeSelm 128 (TENN); Grundy Co., Phillippe and Webb 4605 (TENN), Kral 44527 (VDB); Hardin Co., Jones 3204 (VDB); Hawkins Co., Wolfe s.n. (TENN); Hickman Co., Lynn 32488 (TENN); Knox Co., Ruth 5 (TENN); Lewis Co., King 313 (VDB), Kral 43982 (VDB), 41159 (VDB), 44487 (VDB); McNairy Co., Shanks, Cooley and Woods 14667 (TENN); Monroe Co., Sharp, Underwood, Thomas, Chester and Rogers 40263 (TENN), 40269 (TENN); Montgomery Co., Wert s.n. (UNC), Chester 4304 (TENN); Morgan Co., Schmalzer 1444 (TENN), 1445 (TENN); Putnam Co., Kral 41478 (VDB); Rhea Co., Shanks, Cooley and Woods 8906 (TENN); Sevier Co., Rogers 42435 (TENN), Thomas s.n. (TENN), Sharp 5441 (TENN); Stewart Co., Ellis s.n. (UNC); Sumner Co., Alcorn 1058 (VDB); Unicoi Co., Sharp, Underwood, Pringle and Ramsey 29148

(TENN), Wofford and Smith 79-280 (TENN); Washington Co., Warden 1384 (ETSU); Wayne Co., Kral 43921 (VDB), 62859 (VDB); Williamson Co., Kral 41294 (VDB).

VERMONT: Bennington Co., Knowlton s.n. (TENN); Windham Co., Ahles 86242 (TENN).

VIRGINIA: Amherst Co., Freer 17262 (VPI), Stevens 8071 (UNC); Augusta Co., Freer 7350 (VDB, UNC), DeGarmo s.n. (VPI); Bland Co., Uttal 6764 (VPI), 6777 (VPI, TENN), 7699 (VPI); Botetourt Co., Freer 2896 (VPI); Brunswick Co., Lewis and Massey 3738 (VPI); Buchanan Co., Kral 14284 (VDB), 14231 (VDB), 14242 (VDB); Craig Co., Kral 11758 (VPI); Dickenson Co., Kral 14163 (VDB); Fauquier Co., Allard 3710 (VPI); Floyd Co., Uttal 12255 (VPI); Giles Co., Gentry 788 (UNC), Massey s.n. (VPI); Grayson Co., Ahles and Tucker 60304 (UNC); James City Co., Barans 658 (UNC); Loudon Co., Ahles and James 61364 (UNC); Lunenburg Co., Ahles and James 61990 (UNC); Montgomery Co., Uttal 8442 (UNC), Massey s.n. (VPI), Smyth 5435 (VPI), Kral 11540 (VDB); Nelson Co., Freer 17283 (UNC, VPI); Page Co., Hunnewell 16354 (VPI); Patrick Co., Ahles and Clark 60088 (UNC), Dale s.n. (VPI); Pittsylvania Co., Ahles and James 62376 (UNC); Prince Edward Co., Ahles and James 62881 (UNC); Roanoke Co., Harvill 17720 (UNC); Rockbridge Co., Freer 7272 (VPI, UNC), Roe 647 (UNC), Fosberg 17410 (UNC); Russell Co., Warden 517 (ETSU); Smyth Co., Kral 11610 (VDB), 11644 (VDB); Wise Co., Culbertson 1991 (VPI).

WEST VIRGINIA: Cabell Co., Gilbert 798 (TENN); Grant Co., Downs 7257 (UNC); Greenbrier Co., Martin 633 (UNC); Magnolia Co., Nicely and Yokum s.n. (UNC); Morgan Co., Downs 10256 (UNC); Randolph Co., Clarkson and Clarkson 724 (UNC), Radford 45976 (UNC); Wetzel Co., Haugh 7431 (TENN).

Prenanthes roanensis (Chickering) Chickering

NORTH CAROLINA: Ashe Co., Radford 41134 (UNC); Avery Co., Rohrer 2305 (UNC), 2244 (UNC); Buncombe Co., Bartlett 2934 (UNC), 2936 (VDB), 2937 (UNC); Haywood Co., Ramseur 1782 (UNC); McDowell Co., Bell 4432 (UNC); Mitchell Co., Ahles and Duke 49889 (UNC), Brown 115 (ETSU), Chickering s.n. (US), Smyth 3494 (VPI), Wood and Boufford 17699 (UNC); Swain Co., Bruhn 334 (UNC, TENN), Jennison 2899 (TENN), Pavlovic s.n. (TENN), Ramseur 1455 (UNC); Transylvania Co., Freeman 57775 (UNC), Stewart s.n. (UNC); Yancey Co., Ahles and Duke 50710 (UNC), 50741 (UNC), Ramseur 4237 (UNC).

TENNESSEE: Carter Co., Gonsoulin 3356 (VDB), Kral 56516 (VDB), Uttal 3030 (VPI), Warden 285 (ETSU), Wofford 79-271 (TENN), 79-272 (TENN); Polk Co., Fusiak 575 (TENN), Sevier Co., Cain 1011 (TENN), Rogers 4070-A (VDB), Sharp S 416 (TENN); Unicoi Co., Wofford 79-277 (TENN), 79-278 (TENN).

VIRGINIA: Floyd Co., Heath, Harrison and Rutledge 19 (VPI), Porter and Condit 3002 (VPI); Grayson Co., Croy 671 (VPI), 675 (VPI), Uttal 7628 (VPI); Montgomery Co., Kral s.n. (VDB); Patrick Co., Kral 11737 (VDB); Smyth Co., Kral 11678 (VDB), Croy 677 (VPI), 676 (VPI).

Prenanthes serpentaria Pursh

ALABAMA: Autauga Co., Kral 61110 (VDB), Kral 54384 (VDB); Bibb Co., Kral 41746 (VDB); Clarke Co., Kral 44919 (VDB); Cleburne Co., Kral 44163 (VDB); Covington Co., Kral 38118 (VDB); Crenshaw Co., Kral 41604 (VDB); Dallas Co., Kral 48827 (VDB); De Kalb Co., Kral 37648 (VDB); Etowah Co., Kral 37729 (VDB); Fayette Co., Kral 48653 (VDB); Greene Co., Kral 44443 (VDB); Houston Co., Kral 48958 (VDB); Jackson Co., Kral 37568 (VDB); Jefferson Co., Kral 33605 (VDB); Lamar Co., Kral 45021 (VDB); Lee Co., Boone 113 (VDB), Harper 021 (UNC), Kral 44257 (VDB), Moore 704-69 (VDB); Lowndes Co., Kral 44968 (VDB); Mobile Co., Lelong 4850 (UNC), 3907 (UNC), Rogers 1852-B (UNC), Rogers and Lelong 1825 (VDB), Wofford 10262 (TENN); Montgomery Co., Kral 54509 (VDB); Morgan Co., Kral 56540 (VDB); Shelby Co., Kral 41506 (VDB); St. Clair Co., Bostick 252-4 (UNC), Kral 37816 (VDB), 44624 (VDB); Talladega Co., Kral 37914 (VDB), 44610 (VDB); Washington Co., Kral 37226 (VDB), Kral 38272 (VDB); Winston Co., Kral 29464 (VDB), 41247 (VDB).

FLORIDA: Leon Co., Kral 3612 (VDB).

GEORGIA: Barton Co., Kral 44087 (VDB); Elbert Co., Coile 1625 (UNC), Coile and Coile 1435 (UNC); Greene Co., Jones and Jones 21466 (UNC); Harris Co., Jones 21652 (UNC), Jones 21514 (VDB); McDuffie Co., Bartlett 2442A (UNC); Oglethorpe Co., Montgomery 973 (VDB).

KENTUCKY: Bath Co., Wharton 5908a (TENN); Powell Co., Higgins 2001 (UNC).

MARYLAND: Montgomery Co., Barlett 2314 (UNC).

MISSISSIPPI: Forrest Co., Rogers 4718 (VDB); Perry Co., Rogers 1753 (UNC); Tishmingo Co., Ray 7583 (UNC), 7604 (UNC, VDB), Temple 4360 (UNC), Rogers 46214 (TENN).

NORTH CAROLINA: Alamance Co., Ramseur and Hammond 2010 (UNC); Alexander Co., Radford 18098 (UNC); Anson Co., Ahles and Leisner 19438 (UNC); Buncombe Co., Bartlett 2961 (UNC); Burke Co., Bell 15124 (UNC); Brunswick Co., Godfrey 10160 (UNC); Cabarrus Co., Ahles and Leisner 19585 (UNC); Caldwell Co., Radford 19745 (UNC); Catawba Co., Bell 14842 (UNC); Chatham Co., Ramseur and Hammond 2519 (UNC); Cherokee Co., Radford 17630 (UNC), 17324 (UNC); Chowan Co., Ahles and Duke 50996 (VDB); Clay Co., Radford 16298 (UNC); Cumberland Co., Ahles 36652 (UNC); Davidson Co., Ahles and Leisner 18515 (UNC); Davie Co., Radford 19801 (UNC); Durham Co., Ahles and Sears 59941 (UNC); Forsyth Co., Ahles and Haesloop 51690 (UNC); Franklin Co., Ahles and Leisner 20582 (UNC); Gason Co., Ahles and Leisner 18825 (UNC); Granville Co., Ahles and Leisner 20038 (UNC); Halifax Co., Ahles and Leisner 20765 (UNC); Hannett Co., Laing 518 (UNC); Haywood Co., Ahles and Duke 50339 (UNC); Iredell Co., Ahles and Haesloop 51925 (UNC); Jones Co., Radford 5921 (UNC); Lee Co., Stewart s.n. (UNC); Lincoln Co., Bell 15356 (UNC); Macon Co., Quarterman 1575 (VDB); Martin Co., Treiber and Nesom 617 (UNC); McDowell Co., Bell 4538 (UNC), Bell 4432 (VDB); Mecklenberg Co., Ahles and Leisner 49976

(UNC, VPI) ; Mitchell Co., Ahles and Duke 49722 (UNC); Montgomery Co., Radford 19407 (UNC), Wickland 1630 (UNC); Nash Co., Ahles and Leisner 21155 (UNC); Northampton Co., Ahles and Haesloop 52452 (UNC); Orange Co., Dorr and Nesom 1541 (UNC); Polk Co., Peattie TR47 (UNC); Randolph Co., Bell 15566 (UNC); Richmond Co., Radford 19145 (UNC); Rockingham Co., Radford 18411 (UNC); Rowan Co., Harton 589 (UNC); Rutherford Co., Melvin 5001 (UNC); Stanley Co., Ahles and Leisner 19965 (UNC); Stokes Co., Radford 41425 (UNC); Swain Co., Radford 17090 (UNC); Transylvania Co., Ware and White 3496 (VDB); Union Co., Ahles 34502 (UNC); Vance Co., Ahles and Leisner 20275 (UNC); Wake Co., Sawyer and Ahles 1717 (UNC); Warren Co., Ahles and Bell 21443 (UNC).

PENNSYLVANIA: Montgomery Co., Berkheimer 6708 (VDB).

SOUTH CAROLINA: Aiken Co., Ahles and Crutchfield 55513 (UNC); Bamberg Co., Ahles 37719 (UNC); Barnwell Co., Ahles and Crutchfield 55775 (UNC); Beaufort Co., Ahles and Bell 20941 (UNC); Calhoun Co., Ahles 35283 (UNC); Colleton Co., Ahles and Bell 21094 (UNC); Cherokee Co., Ahles 34337 (UNC); Chester Co., Bell 10046 (UNC); Darlington Co., Smith 483 (UNC), Coker s.n. (UNC); Dorchester Co., Ahles and Haesloop 37992 (UNC); Edgefield Co., Radford 30247 (UNC); Florence Co., Bell 10852 (UNC); Jasper Co., Bell 4973 (UNC, VDB); Lancaster Co., Ahles 34823 (UNC); Laurens Co., Bell 10227 (UNC); Lee Co., Radford 29344 (UNC); Lexington Co., Radford 29733 (UNC); Newberry Co., Bell 9839 (UNC); Oconee Co., Radford 17724 (UNC); Pickens Co., Radford 16693 (UNC); Spartanburg Co., Bell 10398 (UNC); Sumter Co., Radford 29585 (UNC); Williamsburg Co., Radford 31182 (UNC); York Co., Ahles 34463 (UNC).

TENNESSEE: Blount Co., Thomas and Rogers s.n. (TENN), Thomas s.n., Brown and Robinson 21556 (TENN); Coffee Co., Blum 3015 (VDB); Cumberland Co., Kral 41412 (VDB), Somers 893 (VDB), Shanks, Cooley and Woods 14148 (TENN); Fentress Co., Underwood, Shanks and Sharp 26070 (TENN), Kral 41490 (VDB), 54312 (VDB); Grundy Co., Phillippe and Webb 4048 (TENN), Kral 43755 (VDB), Wofford 77-301 (VDB); Marion Co., Kral 44517B (VDB); Monroe Co., Rogers 40434 (TENN), Rogers, Rogers and Rogers 40606 (TENN), Ford and Rusell 2030 (TENN); Morgan Co., Schmalzer 1448 (TENN); Pickett Co., Kral 62653 (VDB); Polk Co., Ford and Russell 23173 (TENN); Putnam Co., DeSelm 1945 (TENN), Kral 40474 (VDB); Roane Co., Shanks 13794 (TENN); Sevier Co., Rogers 42438 (TENN); Van Buren Co., Shanks 8917 (TENN), Ford and Rusell 1982 (TENN).

VIRGINIA: Amelia Co., Lewis 2128 (VPI); Augusta Co., Steele 113 (VPI), Garms s.n. (VPI), Massey s.n. (VPI); Buchanan Co., Kral 14227 (VDB); Charles City Co., Ware 3817 (VPI); Charlotte Co., Ahles and James 60891 (UNC); Craig Co., Kral 11757 (VDB), Uttal 8440 (VPI); Culpeper Co., Ahles and James 61594 (UNC); Dickenson Co., Kral 14253 (VDB); Fairfax Co., Bradley 5324 (UNC), Bradley and Causey 11409 (UNC), 113369 (VCU); Fauquier Co., Allard 7399 (VPI); Frederick Co., Hunnewell 16342 (VPI); Henrico Co., Johnson 4951 (VCU); James

City Co., Barans 712 (UNC); Lancaster Co., Ewing 1334 (UNC); Loudoun Co., Ahles and James 61526 (UNC); Lunenburg Co., Ahles and James 61804 (UNC); Montgomery Co., Uttal 6803 (UNC, VPI); Nelson Co., Uttal s.n. (VPI); Pittsylvania Co., Ahles and James 62196 (UNC); Prince Edward Co., Ahles and James 62672 (UNC); Prince William Co., Allard 5665 (VPI); Shenandoah Co., Hunnewell 12230 (VPI); Sussex Co., Kral 11365 (UNC, VDB).

VITA

Frank Fusiak was born July 31, 1958 in Jersey City, New Jersey. He attended Samuel A. Roberson elementary school in Bayonne, NJ and high school at Bayonne High School in Bayonne, NJ. In June 1980 he received the Bachelor of Arts degree with a major in Botany from Rutgers University, Newark College of Arts and Sciences. In September 1980 he entered The University of Tennessee, Knoxville, and received the Master of Science degree in June 1983 with a major in Botany.