Allozymic and Morphological Variation in Antennaria (Asteraceae: Inuleae) from the Low Arctic of Northwestern North America

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ABSTRACT. Twenty-five populations of *Antennaria* from the low arctic of northwestern North America, including individuals that have traditionally been referred to as *A. alaskana, A. angustata, A. compacta, A. densifolia, A. friesiana, A. monocephala, A. neoalaskana,* and *A. philonipha,* were analyzed to determine the degree of divergence among populations. Sixteen putative isozyme loci were assayed to assess the amount of allozyme divergence. Morphological variation among the populations was evaluated using 30 characters. Principal components and cluster analyses were used to quantify the morphological and allozymic differences among the populations. Results indicate that four distinct groups exist among the taxa including *A. densifolia, A. friesiana,* s.l., *A. monocephala* s.l., and *A. neoalaskana.* Both morphological and isozyme data were concordant in indicating that the taxa have diverged from each other to the extent that they can be recognized at the rank of species. Genetic structure of the populations is comparable to other previously studied species of *Antennaria* and is typical of perennial herbs in general. Asexual populations of *A. friesiana* and *A. monocephala* contain an average of 3.2 clones per population, which is generally characteristic of asexual populations of *Antennaria.* Comparisons of diploid vs. polyploid and sexual vs. asexual populations yielded no significant differences for genetic statistics such as A, P, and H_{obs} between the groups. *Antennaria friesiana* s.l. may contain a mixture of diploids, autopolyploids, and segmental allopolyploids.

The Alpinae are a group of *Antennaria* Gaertner occurring in a diversity of arctic and alpine habitats throughout the northern hemisphere (Bayer 1990a). The sexually reproducing taxa within the Alpinae, having both staminate and pistillate individuals in their populations, occur in western North America, except for a single species in northern Norway (Bayer and Stebbins 1987). Traditionally the Alpinae has been recognized as a section, but the formal taxonomic rank of Alpinae group is in question (Bayer 1990a). It is here viewed simply as a subgeneric group until the problem can be resolved. A monophyletic clade within *Antennaria,* most of its species are characterized by dark-colored phyllary tips, relatively short flowering stems, and cauline leaves bearing scarious appendages (flags) at their tips (Bayer 1990a). The sexual species of the Alpinae include *A. aromatica* Evert, *A. densifolia* Porsild, *A. friesiana* (Trautv.) Ekman subsp. *alaskana* (Malte) Hultén (=*A. alaskana* Malte), *A. monocephala* DC., *A. nordhagiana* Rune & Rönning, and *A. pulchella* Greene (Bayer 1990a). *Antennaria umbrinella* Rydb., a species of dubious affinities, has sometimes been included within the Alpinae (Bayer 1989a), but recent cladistic analyses (Bayer 1990a) indicate that it may belong to another clade.

These species are evolutionarily important because they have contributed to the complex reticulate patterns of evolution in *Antennaria.* They may be some of the sexual progenitors of both the *A. rosea* E. Greene (Bayer 1987a, 1990a) and *A. alpina* (L.) Gaertn. (Bayer 1987a) polyploid agamic complexes. The species that have contributed to the genomic composition of the *A. rosea* complex, viz. *A. aromatica, A. pulchella,* and *A. umbrinella* (Bayer 1989b, 1990b), all occur primarily in the Rocky Mountains of the western United States. A previous examination of their interspecific isozyme variation revealed less isozymatic divergence than might be expected for distinct species (Bayer 1989a), findings concordant with evidence (Bayer 1987b) showing that the species occasionally hybridize, intergrade morphologically, and have not diverged evolutionarily as much as other previously investigated species of *Antennaria* (e.g., Bayer and Crawford 1986; Bayer 1988).

Several species from the Alpinae occur in the North American arctic and subarctic; their classification has remained unsettled, ranging from

the conservative treatment of Hultén (1968) to those recognizing an immoderate number of microspecies (Malte 1934; Porsild 1950). In this paper, Hultén (1968) is followed with some notable modifications.

Hultén (1968) circumscribed *A. monocephala* as containing three subspecies. The sexual phase of *A. monocephala* [i.e., subsp. *monocephala* and subsp. *philonipha* (Porsild) Hultén] is confined to southern Alaska south of the Brooks Range, and to Yukon Territory and contiguous Northwest Territories (Hultén 1968; Bayer, pers. obs.). Hultén's (1968) key distinctions between subsp. *monocephala* and subsp. *philonipha* are obscure and seemingly arbitrary, therefore, herein subsp. *philonipha* has been subsumed into subsp. *monocephala*. Within his concept of *A. monocephala* s.l., Hultén circumscribed the apomictic form of the species as *A. monocephala* subsp. *angustata* (Greene) Hultén, thus extending the range of the species across the Canadian arctic into Greenland and down the western cordillera into Montana and Wyoming. *Antennaria monocephala* is most often found on the disturbed margins of solifluction lobes or on unstable, moist, gravelly sloping tundra.

Antennaria friesiana, like *A. monocephala*, has both sexual and apomictic phases that have distinct distributions. The sexual (*A. friesiana* subsp. *alaskana*) is restricted to Alaska and cordilleran areas of northern Yukon and adjacent Northwest Territories. The apomictic phase (*A. friesiana* subsp. *friesiana*) is almost circumpolar, occurring from the central and eastern Siberian plateau eastward across the North American arctic to Greenland. Hultén circumscribed a third subspecies within *A. friesiana* s.l., *A. friesiana* subsp. *compacta* (Malte) Hultén. After studying its morphology, both in the field and in the herbarium, it was apparent that Hultén's taxon contains at least three incongruous entities. These are possibly not at all related to the other two subspecies of *A. friesiana*. Hultén's circumscription of *A. friesiana* subsp. *compacta* included *A. densifolia*, *A. neoalaskana* Porsild, and *A. crymophila* Porsild as taxomonic synonyms. *Antennaria compacta* Malte and *A. crymophila* are perhaps hybrid apomicts derived from *A. densifolia* and one of the other sexual species of the Alpinae (Bayer 1989c). *Antennaria densifolia* is restricted to three mountain ranges in the Yukon Territory and Northwest Territories, Mackenzie Mountains, Ogilvie Mountains, and

southern Richardson Mountains (Bayer 1989c). One disjunct population of *A. densifolia* has recently been found in the Anaconda Range of Montana (Bayer 1989c). The species is an endemic of partially weathered limestone talus and usually occurs at or below alpine treeline or above arctic treeline in the case of the Richardson Mountains (Bayer 1989c; pers. obs.). *Antennaria neoalaskana* occurs from the eastern Brooks Range, Alaska, to the Richardson Mountains and into central MacKenzie Mountains, on the Yukon-Northwest Territories boundary. Its habitat is arctic fell fields or gravelly frost boils.

Both diploid and polyploid cytotypes are known to occur in *A. monocephala* s.l. and *A. friesiana* s.l. (Bayer and Stebbins 1987; current study), while *A. densifolia* has been reported only as diploid (Bayer 1989c; current study). *Antennaria neoalaskana* is disclosed as a tetraploid for the first time in the current study.

The purpose of this study is to evaluate the degree of isozymic and morphological divergence among populations of *A. densifolia* s. str., *A. friesiana* (sensu Hultén pro parte majore), *A. monocephala* (sensu Hultén), and *A. neoalaskana* s. str. The results of this study can then provide a more accurate assessment of the evolutionary relationships among these taxa. It will supplement the previous study of the Rocky Mountain species (Bayer 1989a) and complete an electrophoretic survey of North American sexual members of the Alpinae.

MATERIALS AND METHODS

The provenances of the 25 populations studied are presented in the appendix along with the elevation of the site, probable reproductive mode, and ploidy of populations. Populations were selected from throughout the entire specific ranges of each species in Alaska, Yukon, and Northwest Territories as limited road access would allow. Each of the 25 sites is mapped in figure 1.

Gender ratios from populations are reliable, although not infallible, indicators of the reproductive mode of populations (Bayer and Stebbins 1983; Bayer 1990c). The reproductive mode, i.e., whether the population was primarily sexually or asexually reproducing, was based on sex ratio determinations (Appendix). *Antennaria* plants form relatively small, spatially distinct
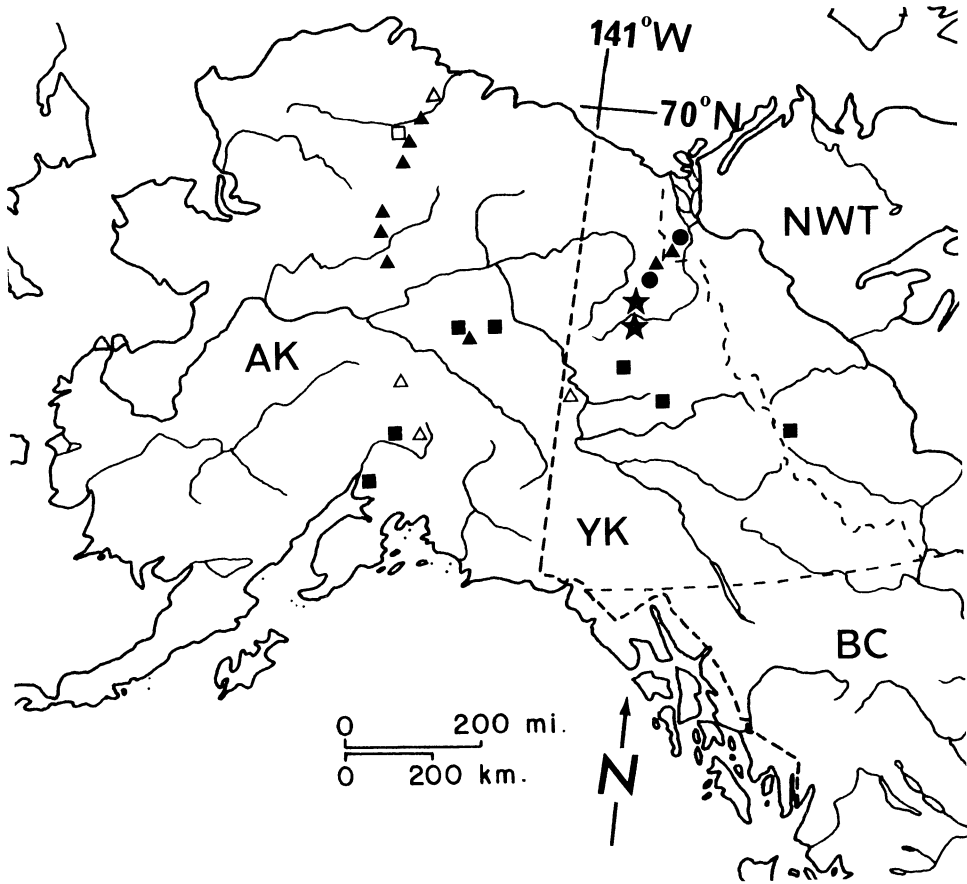
FIG. 1. Geographic positions of 25 populations used in the morphological and isozymic studies of *Antennaria* from Alaska (AK), Northwest Territories (NWT), and Yukon Territory (YK). Individual populations are labeled as follows: (★) *A. densifolia*, (▲) *A. friesiana* subsp. *alaskana* (sexual), (△) *A. friesiana* subsp. *friesiana* (asexual), (■) *A. monocephala* subsp. *monocephala*, (□) *A. monocephala* subsp. *angustata* (asexual), and (●) *A. neoalaskana*.

aggregations, so individuals can generally be identified confidently. Ramets were removed from up to 50 individual plants from each population because seeds are available only for a limited time before dispersal. Several meters separated consecutive plants sampled in large populations to insure that different individuals were being sampled. Ramets were transported to the phytotron of the University of Alberta for cultivation and subsequent analysis. The number of populations examined for each species was: *A. densifolia* (2), *A. friesiana* s.l. (13), *A. monocephala* (8), and *A. neoalaskana* (2). Chromosome numbers were obtained for the populations using techniques outlined previously (Bayer 1984; Bayer and Stebbins 1987).

Electrophoretic methodologies are similar to those used previously in *Antennaria* (Bayer 1988). Fresh pieces of actively growing leaf tissue from up to 35 individuals per population were assayed. Tissue was ground in ice-cold Tris-HCl extraction buffer: 0.1 M Tris-HCl, pH 7.5, 4.0 mM 2-mercaptoethanol, 1.0 mM EDTA (disodium salt), 0.2 M sucrose, 0.6% polyvinyl-polypyrrolidone (5:1 ratio of 40K:360K m.w.), 2.0% PEG (8K m.w.), 0.1% BSA, and 0.002 M ascorbic acid. Samples were stored at $-20.0°C$ overnight and electrophoresed the next morning. Supernatant was soaked onto filter paper wicks before being loaded into 12.5% starch gels. General protein (GP), glutamate dehydrogenase (GDH), leucine aminopeptidase (LAP), phosphoglucoisomerase (PGI), and triose-phosphate isomerase (TPI) were resolved on the following sys-

tem: gel buffer of 1 part 0.038 M lithium hydroxide·H₂O − 0.188 M boric acid (pH 8.3), and 9 parts 0.045 M Tris − 0.007 citric acid (pH 8.4) (Soltis et al. 1983); electrode buffer containing only the lithium borate constituent. Acid phosphatase (ACP), malate dehydrogenase ((NAD) MDH), phosphoglucomutase (PGM), and shikimic acid dehydrogenase (SKD) were visualized on another system: gel buffer of 0.016 M L-histidine (free base) and 0.002 M citric acid·H₂O (pH 6.5), and an electrode buffer of 0.065 M L-histidine (free base) - 0.007 M citric acid·H₂O (pH 6.5) (Cardy et al. 1981). Enzymatic assays followed Soltis et al. (1983), except for general protein (GP) in which the gel was stained with a solution of 5 parts methanol, 5 parts water, 1 part acetic acid, and 100 mg/100 ml Brilliant Blue G stain. After incubation for 30 min at 40.0°C the gel was destained with the same solution without the Brilliant Blue G. The locus specifying the most anodally migrating isozyme was designated as 1, the next 2, and so on. Similarly, the most anodal allozyme of a given gene was labeled A, etc.

Genetic variation was described by mean number of alleles per locus (A) (including monomorphic loci), proportion of loci polymorphic (P), observed and expected average heterozygosities (H_{obs} and H_{exp}, respectively), and mean fixation index (F_T). The observed and expected average heterozygosities were compared with chi-square tests to determine if the natural populations deviated from Hardy-Weinberg equilibrium expectations. The means for various subgroups of populations for the various genetic statistics were compared by t-tests (Norusis 1988). Gene diversity statistics and standard genetic distances and identities were calculated utilizing the methods of Nei (1972, 1973) implemented by the GENESTAT-PC program (Version 2.1, by Paul Lewis and Richard Whitkus; Whitkus 1988).

A matrix of genetic identities that included average identities for five low arctic taxa as well as three closely related species from the Alpinae from the Rockies (data from Bayer 1989a) was used to generate clustering phenograms. Clustering methods included the unweighted pair-group method using arithmetic averages (UPGMA), weighted pair-group method using arithmetic averages, weighted pair-group method using Spearman's average, weighted pair-group method using centroid average, sin-gle-linkage method, complete-linkage method, and flexible clustering and were executed using the SAHN subroutine of the NTSYS-pc (Rohlf 1987) program.

Principal components analysis (PCA) was used to ordinate the morphological and isozymic differences among the populations. For the morphological analysis, 30 vegetative and reproductive features were measured from each population. These characters include those listed in Bayer (1989c; table 1). Principal component analysis was also used to evaluate the phenetic interpopulational relationships based on allele frequency distributions. Only polymorphic loci were included in the analysis. A third data set combined the morphological and isozymic data into a single matrix. The original data matrices are available upon request from the author. The NTSYS-pc (Numerical taxonomy and multivariate analysis systems for IBM-PC microcomputer and compatibles; vers. 1.2; F. James Rohlf 1987) computed the PCA's. Two- and three-dimensional graphs projecting the populations onto the first two or three principal components were drawn with the MXPLOT and MOD-3DG subroutines of NTSYS-pc, respectively.

Results

The nine enzyme systems assayed here are believed to be coded by 16 loci. The genetic basis of the enzyme phenotypes was inferred from segregation patterns observed at putative loci in Antennaria plants from natural populations. ACP-1-(A and B), GDH-1-(A), GP-1-(A), LAP-1-(B, C, D), MDH-1-(A), MDH-2-(A), MDH-3-(A), MDH-4-(A and B), PGI-1-(A), PGI-2-(A), PGI-3-(C, D, E', and F), PGM-1-(A and B), SKD-1-(A and B), TPI-1-(A, B, C), TPI-2-(A), and TPI-3-(B, C, D, E, F) were the 16 putative isozymes and their allozymes studied. Details of the number of isozymes for each enzyme system, genetic interpretation of the banding patterns in diploids and polyploids, as well as the subcellular localization of several isozymes and the apparent gene duplication for the chloroplast forms of PGI and TPI in Antennaria, were discussed previously (Bayer 1988, 1989a, 1989b).

Some of the loci were monomorphic in all populations: Gdh-1, Mdh-1, Mdh-2, Mdh-3, Pgi-1, Pgi-2, and Tpi-2. Gp-1 consists of a single monomorphic band in all individuals and this

TABLE 1. Genetic variation in 25 populations of *Antennaria* from low arctic North America. Included are mean number of alleles per locus (A); proportion of polymorphic loci, with the most common allele's frequency less than 0.99 (P); observed average heterozygosity (H_{obs}); expected average heterozygosity (H_{exp}); and mean fixation index (F_T). Population designations are given in the appendix. Values of H_{obs} and H_{exp} are not significantly different at the 5% level.

Population designations	A	P	$H_{(obs)}$	$H_{(exp)}$	F_T
A. densifolia (sexual):					
n	1.50 ± 0.730	0.38	0.169	0.143	−0.182
o	1.44 ± 0.629	0.38	0.138	0.133	−0.038
Averages	1.47 ± 0.044	0.38	0.154	0.138	−0.110
(±s.d.)		(±0.00)	(±0.022)	(±0.007)	(±0.102)
A. friesiana subsp. *alaskana* (sexual):					
a	1.44 ± 0.727	0.31	0.103	—	—
b	1.38 ± 0.719	0.25	0.050	—	—
c	1.31 ± 0.602	0.25	0.079	0.074	−0.068
f	1.38 ± 0.719	0.25	0.115	0.116	0.009
g	1.38 ± 0.719	0.25	0.113	0.095	−0.189
h	1.27 ± 0.458	0.25	0.113	0.097	−0.165
j	1.31 ± 0.479	0.31	0.086	—	—
k	1.38 ± 0.619	0.31	0.113	0.091	−0.242
m	1.25 ± 0.447	0.25	0.083	—	—
Averages	1.34 ± 0.061	0.27	0.097	0.095	−0.131
(±s.d.)		(±0.03)	(±0.024)	(±0.02)	(±0.10)
A. friesiana subsp. *friesiana* (asexual):					
d	1.19 ± 0.403	0.19	0.167	—	—
e	1.31 ± 0.479	0.31	0.122	—	—
i	1.19 ± 0.403	0.19	0.028	—	—
l	1.13 ± 0.314	0.14	0.052	—	—
Averages	1.20 ± 0.078	0.21	0.097	—	—
(±s.d.)		(±0.073)	(±0.057)		
A. monocephala subsp. *monocephala* (sexual):					
p	1.31 ± 0.602	0.25	0.076	0.075	−0.013
q	1.38 ± 0.719	0.25	0.063	0.058	−0.082
r	1.31 ± 0.602	0.25	0.056	0.051	−0.098
s	1.38 ± 0.719	0.25	0.103	0.090	−0.144
u	1.13 ± 0.342	0.14	0.019	0.035	−0.543
v	1.31 ± 0.479	0.31	0.045	0.052	0.135
w	1.31 ± 0.602	0.25	0.061	0.049	−0.245
Averages	1.29 ± 0.089	0.29	0.061	0.058	−0.141
(±s.d.)		(±0.064)	(±0.024)	(±0.017)	(±0.21)
A. monocephala subsp. *angustata* (asexual):					
t	1.38 ± 0.544	0.118	0.063	—	—
A. neoalaskana (sexual):					
x	1.31 ± 0.479	0.31	0.063	—	—
y	1.31 ± 0.479	0.31	0.141	—	—
Averages	1.31 ± 0.000	0.31	0.102	—	—
(±s.d.)		(±0.000)	(±0.055)		
Grand averages:					
All diploids	1.34 ± 0.086	0.27	0.090	0.083	−0.133
(±s.d.)		(±0.061)	(±0.04)	(±0.033)	(±0.158)

TABLE 1. Continued.

Population designations	A	P	$H_{(obs)}$	$H_{(exp)}$	F_T
All polyploids	1.29 ± 0.094	0.24	0.087	—	—
(±s.d.)		(±0.074)	(±0.042)		
All sexuals	1.34 ± 0.079	0.28	0.084	—	—
(±s.d.)		(±0.054)	(±0.037)		
All asexuals	1.24 ± 0.046	0.19	0.086	—	—
(±s.d.)		(±0.074)	(±0.057)		
All	1.31 ± 0.091	0.25	0.089	—	—
(±s.d.)		(±0.066)	(±0.040)		

is interpreted as a single locus. These loci were generally monomorphic in other species of *Antennaria* as well (Bayer and Crawford 1986; Bayer 1988, 1989a, 1989b). The loci *Acp-1, Lap-1, Mdh-4, Pgi-3, Pgm-1, Skd-1, Tpi-1,* and *Tpi-3* were polymorphic in at least some populations. Polymorphism at *Mdh-4* was encountered for the first time in *Antennaria*, in the two populations of *A. densifolia*. The table of allelic frequencies is too large to be presented here, but will be supplied upon request from the author.

Measures of genetic structure were compared among populations and species (tables 1–4). The mean number of alleles per locus (A) ranges from 1.13 to 1.50 with an interpopulation mean of 1.30 (table 1). Values for P range from 0.118 to 0.38 (table 1) with an interpopulation mean of 0.26. The mean values of A and P for all diploid populations ($A = 1.34$; $P = 0.27$) and all polyploid populations ($A = 1.29$; $P = 0.24$) are not significantly different (table 1). The mean H_{obs} for all populations is 0.090, and ranges from 0.019 to 0.169. Sexual populations do not differ significantly from asexual ones with regard to A, P, and H_{obs} (table 1). The Hardy-Weinberg model assumes diploidy and sexual reproduction (Hartl 1980). Expected values of average heterozygosity (H_{exp}) were therefore calculated only for the diploids. The observed heterozygosities (H_{obs}; table 1) for the diploids were not significantly different from H_{exp} (table 1). The mean fixation index (F_T), useful in determining the degree of inbreeding in populations, has a mean value of −0.133 and range = −0.543 to 0.135 (table 1). One population (pop. v; table 1) has a value of F_T greater than 0.1 and a deficiency in the number of expected heterozygotes, so it is likely that some functional in-breeding is occurring in this population. The remainder of the populations have negative values of F_T (except population f, $F_T = 0.009$: table 1) indicating that random mating is occurring, i.e., their breeding system is primarily outcrossing. Asexual populations of *A. friesiana* and *A. monocephala* contain the following numbers of multilocus genotypes (d = 3, e = 2, i = 5, l = 3, t = 3; average = 3.2 clones per population).

Gene diversity statistics (Nei 1973) for individual and pooled taxa (table 2) are related: total gene diversity (H_T) = gene diversity within populations (H_s) + gene diversity between populations (D_{ST}). The coefficient of gene diversity $G_{ST} = D_{ST}/H_T$. *Lap-1, Skd-1, Tpi-3, Pgi-3,* and *Pgm-1* have the highest average levels of H_T, with a mean H_T for all taxa over all loci of 0.114 (range = 0.054–0.136; table 2). Highest levels of H_T with respect to all loci occur consistently in the sexual taxa, i.e., *A. densifolia, A. friesiana* subsp. *alaskana, A. monocephala* subsp. *monocephala,* and *A. neoalaskana*. The values of H_T are lower for the asexual taxa, *A. friesiana* subsp. *friesiana* and *A. monocephala* subsp. *angustata*. The most geographically restricted taxon, *A. densifolia*, has the highest total diversity (table 2). The average values of G_{ST} for individual taxa over all loci range from 0.017 to 0.268, indicating that most of the genetic diversity resides within (H_s) instead of among populations (D_{ST}).

Intraspecific mean genetic identities (I) and distances (D) (Nei 1972; table 3) range from 0.9733 to 0.9974 for values of I and 0.0026 to 0.0272 for D. The measure D ranges from 0.0031 to 0.0655 and I from 0.9366 to 0.9969 for the interspecific comparisons (table 4). Cluster anal-

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TABLE 2. Nei's genetic diversity statistics for individual and pooled taxa. Presented are gene diversities for individual polymorphic and pooled loci. Only taxa displaying polymorphism at a given locus are represented; monomorphic loci have gene diversity statistics values of 0.000. H_T = Total gene diversity within a taxon, H_S = gene diversity within populations of a taxon, D_{ST} = gene diversity between populations within a taxon, G_{ST} = coefficient of gene differentiation.

Loci	Taxa	H_T	H_S	D_{ST}	G_{ST}
Acp-1	A. neoalaskana	0.4676	0.4460	0.0216	0.0462
	All	0.0522	0.0000	0.0522	1.0000
Lap-1	A. densifolia	0.2994	0.3076	0.0000	0.0000
	A. friesiana subsp. alaskana	0.5933	0.4224	0.1709	0.2880
	A. friesiana subsp. friesiana	0.2976	0.2305	0.0671	0.2253
	A. monocephala subsp. monocephala	0.5489	0.3612	0.1877	0.3420
	A. neoalaskana	0.4345	0.3903	0.0442	0.1017
	All	0.5670	0.3540	0.2130	0.3757
Mdh-4	A. densifolia	0.4412	0.4532	0.0000	0.0000
	All	0.0628	0.0362	0.0266	0.4232
Pgi-3	A. densifolia	0.1855	0.1939	0.0000	0.0000
	A. friesiana subsp. alaskana	0.3625	0.3243	0.0382	0.1054
	A. friesiana subsp. friesiana	0.3334	0.1718	0.1616	0.4846
	A. monocephala subsp. angustata	0.3556	0.3556	—	—
	A. monocephala subsp. monocephala	0.1798	0.1612	0.0186	0.1032
	A. neoalaskana	0.1091	0.0991	0.0100	0.0917
	All	0.2716	0.2317	0.0399	0.1469
Pgm-1	A. densifolia	0.4214	0.3506	0.0708	0.1680
	A. friesiana subsp. alaskana	0.1060	0.1054	0.0006	0.0052
	A. friesiana subsp. friesiana	0.0968	0.0906	0.0062	0.0636
	A. monocephala subsp. monocephala	0.0360	0.0344	0.0016	0.0453
	A. neoalaskana	0.0545	0.0555	0.0000	0.0000
	All	0.1060	0.0950	0.0110	0.1041
Skd-1	A. densifolia	0.2753	0.2867	0.0000	0.0000
	A. friesiana subsp. alaskana	0.3890	0.3404	0.0486	0.1248
	A. friesiana subsp. friesiana	0.4198	0.3026	0.1172	0.2791
	A. monocephala subsp. monocephala	0.4909	0.4017	0.0892	0.1818
	A. neoalaskana	0.5000	0.2552	0.2448	0.4896
	All	0.4033	0.3291	0.0742	0.1839
Tpi-1	A. densifolia	0.5499	0.5433	0.0066	0.0120
	All	0.0708	0.0434	0.0274	0.3870
Tpi-3	A. friesiana subsp. alaskana	0.3901	0.3528	0.0373	0.0957
	A. friesiana subsp. friesiana	0.2916	0.2577	0.0339	0.1163
	A. monocephala subsp. angustata	0.5111	0.5111	—	—
	A. monocephala subsp. monocephala	0.2120	0.2006	0.0114	0.0539
	A. neoalaskana	0.1796	0.1688	0.0108	0.0601
	All	0.2893	0.2607	0.0286	0.0990
All loci:	A. densifolia	0.1358	0.1335	0.0023	0.0172
	A. friesiana subsp. alaskana	0.1151	0.0900	0.0251	0.2180
	A. friesiana subsp. friesiana	0.0900	0.0658	0.0241	0.2682
	A. monocephala subsp. angustata	0.0542	0.0542	—	—
	A. monocephala subsp. monocephala	0.0917	0.0724	0.0193	0.2102
	A. neoalaskana	0.1091	0.0884	0.0207	0.1893
	All	0.1139	0.0841	0.0299	0.2623

TABLE 3. Intraspecific mean genetic distances and mean genetic identities for four species of *Antennaria*.

Taxa	Mean identity (range)	Mean distance (range)
A. densifolia	0.9974 (0.9974–0.9974)	0.0026 (0.0026–0.0026)
A. friesiana subsp. *alaskana*	0.9733 (0.9313–0.9987)	0.0272 (0.0013–0.0711)
A. friesiana subsp. *friesiana*	0.9746 (0.9637–0.9835)	0.0258 (0.0167–0.0370)
A. monocephala subsp. *monocephala*	0.9794 (0.9529–0.9998)	0.0209 (0.0002–0.0483)
A. neoalaskana	0.9779 (0.9779–0.9779)	0.0224 (0.0224–0.0224)

yses of genetic identities of five arctic taxa (table 4) combined with the three species from the Rockies (data from Bayer 1989a) produced phenograms of identical topologies; the UPGMA cluster analysis had the highest cophenetic correlation coefficient (fig. 2). Similar topology among phenograms from within each analysis indicates that relationships portrayed in the phenograms are stable.

The PCA's demonstrate that *A. densifolia, A. friesiana* s.l., *A. monocephala*, and *A. neoalaskana* are distinct taxa (figs. 3–5). The first two axes from the PCA of allele frequencies of the polymorphic loci are displayed in figure 3. Highest character loadings are depicted directly on the figure. Some geographic trends in allele frequencies are evident along axis 2 among populations of *A. monocephala*, as the eastern populations from Yukon (u, v, and w) form one group, whereas the western populations from Alaska constitute another (fig. 3). A similar, weaker trend can be seen among the populations of *A. friesiana*. The first two axes of the PCA based on morphological data from 25 populations are represented by figure 4. The characters with high loadings along component 1 are primarily reproductive: pistillate and staminate involucre, corolla, and pappus heights, and number of heads per pistillate flowering stalk, as well as the vegetative characters of stolon length and lower cauline leaf width. The second component has the highest loadings pri-

marily for vegetative characters such as basal leaf length and amount of pubescence, stolon length, length of the lowermost cauline leaf, as well as some reproductive features such as achene length and number of heads in staminate capitulescences. Populations of *A. friesiana* s.l. with both staminate and pistillate individuals (*A. friesiana* subsp. *alaskana*; pops. a, b, c, f, g, h, j, k, and m) are not morphologically distinct from asexual ones (*A. friesiana* subsp. *friesiana*; pops. d, e, i, and l). The first three axes from the third PCA, combining morphological and allele frequency data, are presented in figure 5. The allozymes MDH-4-A and B, TPI-1-B and C, as well as presence of flags, height of the pistillate and staminate involucres and phyllaries, and staminate corolla length all have high loadings along axis 1. High loadings along component 2 include PGI-3-D and TPI-3-D, as well as basal leaf dimension characters, stolon length, staminate phyllary width, and staminate head number. The third component has highest loadings for ACP-1-B, maximum width and pubescence of the basal leaves, width of the lowermost cauline leaf, achene length and staminate head number.

DISCUSSION

Population Genetics. The results of the current investigation will be compared with those from previous studies of *Antennaria* (Bayer and

TABLE 4. Nei's genetic distances (lower triangle) and genetic identities (upper triangle) for all pairwise comparisons of populations within six taxa of *Antennaria*. Taxa are labeled with first four letters of their specific or subspecific epithets.

| Taxa	ALAS	FRIE	DENS	MONO	ANGU	NEOA
A. friesiana subsp. *alaskana*	******	0.9931	0.9629	0.9860	0.9878	0.9805
A. friesiana subsp. *friesiana*	0.0069	******	0.9699	0.9723	0.9969	0.9610
A. densifolia	0.0378	0.0305	******	0.9460	0.9654	0.9366
A. monocephala subsp. *monocephala*	0.0141	0.0281	0.0556	******	0.9592	0.9907
A. monocephala subsp. *angustata*	0.0123	0.0031	0.0352	0.0417	******	0.9524
A. neoalaskana	0.0197	0.0398	0.0655	0.0094	0.0487	******
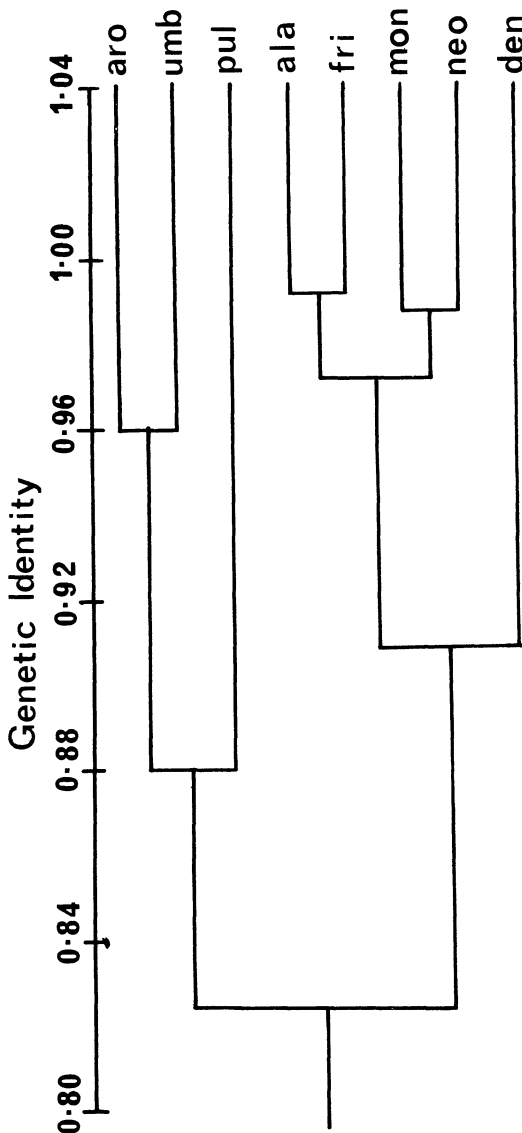
FIG. 2. Phenogram (UPGMA) derived from Nei's standard genetic identities of all pairwise comparisons of eight taxa of *Antennaria* including *A. aromatica*, *A. densifolia*, *A. friesiana* subsp. *alaskana* (sexual), *A. friesiana* subsp. *friesiana* (asexual), *A. monocephala* s.l., *A. neoalaskana*, *A. pulchella*, and *A. umbrinella*. Taxa are labeled with the first three letters of their specific or subspecific epithets. Cophenetic correlation coefficient is 0.8961.

structure of these arctic species seems to parallel that of other species of *Antennaria* (Bayer 1989a). Values of A are about the same as, and P slightly higher than, values obtained for other species of *Antennaria* ($A = 1.28$, $P = 0.22$ in Bayer and Crawford 1986; $A = 1.29 \pm 0.163$, $P = 0.19 \pm 0.103$ in Bayer 1988). Compared to values of H_{exp} for other amphimictic species of *Antennaria* (0.068 in Bayer and Crawford 1986; 0.065 in Bayer 1988) the values are somewhat higher, but still within the range of previously reported values. Mean observed heterozygosity for diploid population ($H_{obs} = 0.09$) is not significantly different from the mean for polyploid population ($H_{obs} = 0.087$; table 1). Diploid populations resemble polyploid populations in genetic statistics such as A, P and H_{obs}. A previous comparison (Bayer 1989a) showed that the H_{obs} of diploids and polyploids of the Alpinae were basically similar, but that they were significantly higher in polyploids. The mean H_T for all loci (0.114) is within the range of values found for nine other amphimictic species of *Antennaria* (0.062 to 1.36; Bayer and Crawford 1986; Bayer 1988). Values of A and P directly influence the magnitude of H_T. Consequently many of the individual taxon values of H_T (table 2) are slightly higher than other previously investigated sexual species of *Antennaria* because of the higher values for A and P (table 1).

The asexual populations (appendix; pops. d, e, i, l, and t) contain about as many clonal genotypes as were found in 63 populations of *A. rosea* (average = 3.5; Bayer 1990c). Asexual *A. rosea* populations had significantly higher values for A, P and H_{obs} than their amphimictic relatives (Bayer 1989b), but in this study sexual populations do not significantly differ from asexual ones with regard to these genetic statistics (table 1). This could suggest different origins for the two groups of apomicts. *Antennaria rosea* is of diverse hybrid origin with several (perhaps eight) sexual ancestors (Bayer 1989b, 1990b), whereas the asexual members of *A. friesiana* and *A. monocephala* complexes are either autopolyploid apomicts or perhaps segmental allopolyploids involving two or three sexual ancestors that have not diverged greatly genetically.

Interspecific populations are quite similar genetically as indicated by the high intraspecific values for D and I (table 2), values typical of most plant species (Crawford 1983). These spe-

Crawford 1986; Bayer 1988, 1989a, 1989b) to understand how the isozymatic profiles of these taxa relate to the overall isozyme variational pattern for *Antennaria*. The population genetic
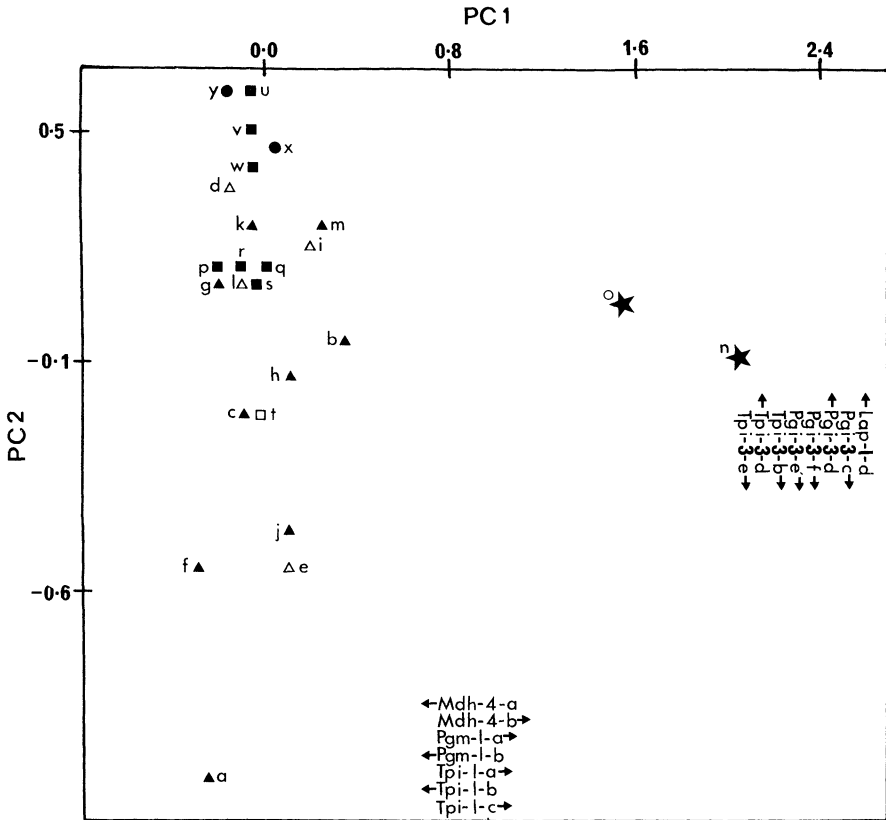
PC 1

FIG. 3. Principal components analysis of allelic frequencies from 25 populations of (★) *A. densifolia*, (▲) *A. friesiana* subsp. *alaskana* (sexual), (△) *A. friesiana* subsp. *friesiana* (asexual), (■) *A. monocephala* subsp. *monocephala*, (□) *A. monocephala* subsp. *angustata* (asexual), and (●) *A. neoalaskana*. Populations are labeled with the single letter population designations given in the appendix. Character loadings are indicated along the margins of the graph where negative coefficients are indicated by left pointing arrows and positive coefficients by right pointing arrows. The first two axes account for 44.9% of the total variation and the first eight factors have individual eigenvalues greater than 1.000.

cies of *Antennaria*, as well as others (Bayer and Crawford 1986; Bayer 1988, 1989a, 1989b), have a genetic structure typical of other herbaceous outcrossing perennials [see comprehensive reviews by Loveless and Hamrick (1984) and Hamrick et al. (1979)]. Isozyme studies on arctic vascular plants are apparently few and one study of clonal structure of dwarf arctic *Betula glandulosa* Michx. (Hermanutz et al. 1989) does not include statistics needed for a meaningful comparison to *Antennaria*.

Morphological Differentiation. Principal component analysis demonstrates that at least four morphologically distinct groups exist among the 25 populations, that can be circumscribed as four species (fig. 4). *Antennaria den-*

sifolia (populations n and o) is easily separable from the other three species by its glandless, cushion-growth form and relatively many small heads per synflorescence. On the other hand, *A. neoalaskana* (pops. x and y) is characterized by long stolons, many stalked glands on the stems and leaves, and relatively few, mostly large heads per synflorescence. *Antennaria monocephala* (pops. p, q, r, s, t, u, v and w) is distinguished by its long stolons, relatively small, adaxially glabrous, bright green leaves and large, solitary heads. One asexual population of *A. monocephala* (pop. t; =*A. monocephala* subsp. *angustata*) is somewhat intermediate to *A. monocephala* s. str. and the *A. friesiana* subsp. *friesiana* complex and may be of hybrid origin.
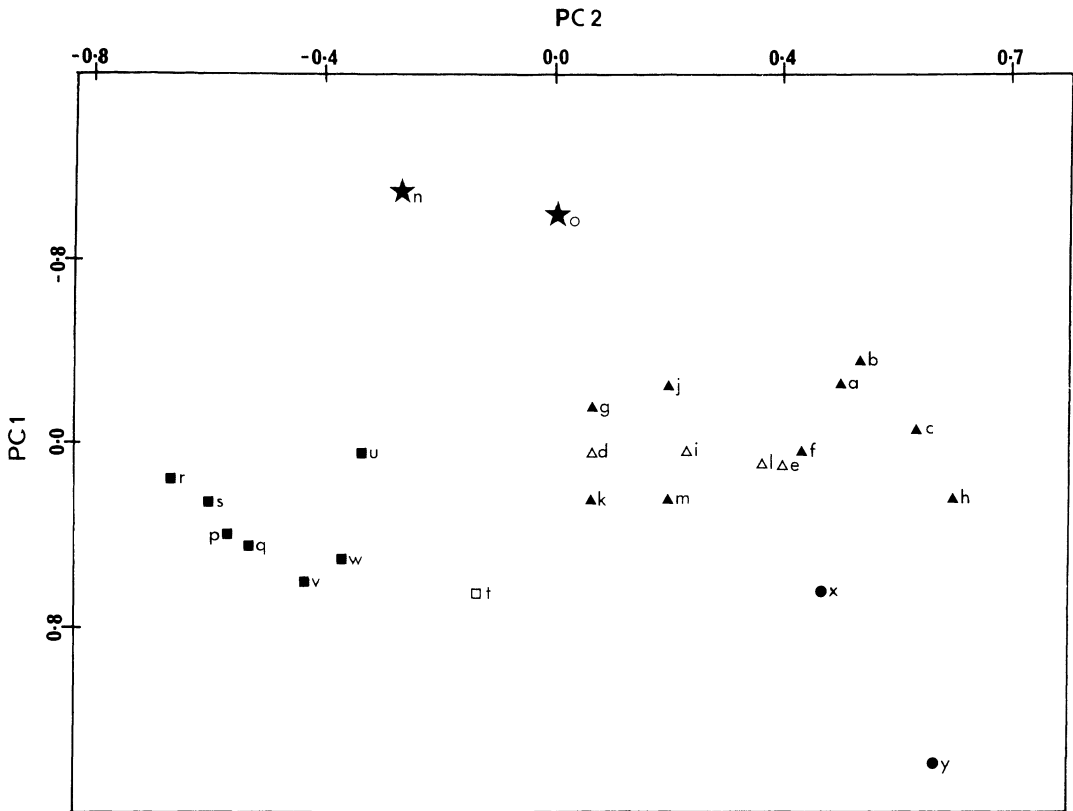
FIG. 4. Principal components analysis of morphological characters of individuals from 25 populations of (★) *A. densifolia*, (▲) *A. friesiana* subsp. *alaskana* (sexual), (△) *A. friesiana* subsp. *friesiana* (asexual), (■) *A. monocephala* subsp. *monocephala*, (□) *A. monocephala* subsp. *angustata* (asexual), and (●) *A. neoalaskana*. Populations are labeled with the single letter population designations given in the appendix. The first two axes account for 47.5% of the total variation and the first seven factors have individual eigenvalues greater than 1.000.

Antennaria friesiana is characterized by its long, linear leaves, very short stolons, and a relatively large pleiocephalous synflorescence. Two other groups are evident within *A. friesiana* (fig. 4). One group is composed of populations a, b, c, e, f, h, and l and the other is composed of pops. d, g, i, j, k, and m, and is somewhat intermediate to the first group and *A. monocephala*. The first group is characterized by longer, more linear, basal leaves and a synflorescence with many heads, whereas the other group has leaves that are much shorter and oblanceolate to spathulate. Further studies will be required to determine whether the two groups are worthy of taxomonic recognition.

Isozymic Differentiation. In general, the populations of *Antennaria* from the arctic are poorly differentiated isozymically. Interspecific differences are due primarily to divergence in frequencies of common alleles rather than the result of many unique alleles that have evolved independently in the separate evolutionary lineages. Principal component analysis demonstrates that the two *A. densifolia* populations (pops. n and o) are isozymically distinct primarily because of their high frequencies of MDH-4-B, PGM-1-A, and TPI-1-A and -C. The two populations of *A. neoalaskana* (pops. x and y) resemble several populations of *A. monocephala* (pops. u, v, and w) isozymically, but are characterized by having high frequencies of LAP-1-D, PGM-1-B, and TPI-1-B. The populations of *A. monocephala* (fig. 3) overlap with some populations of *A. friesiana*, but interestingly the latter (pops. d, g, i, k, and m) are morphologically intermediate between *A. monocephala* and
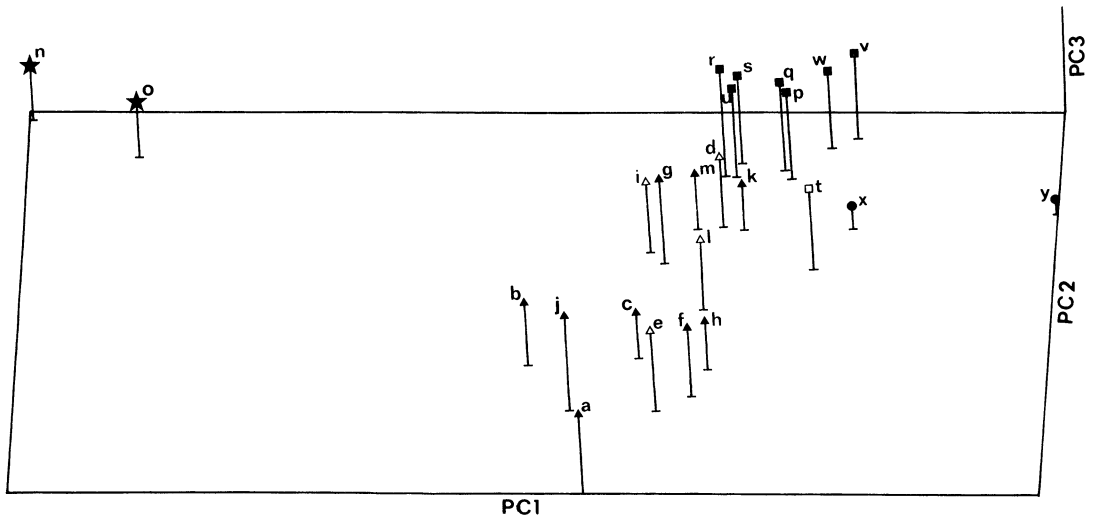
FIG. 5.   Principal components analysis of allelic frequencies and morphological characters from 25 populations of (★) *A. densifolia*, (▲) *A. friesiana* subsp. *alaskana* (sexual), (△) *A. friesiana* subsp. *friesiana* (asexual), (■) *A. monocephala* subsp. *monocephala*, (□) *A. monocephala* subsp. *angustata* (asexual), and (●) *A. neoalaskana*. Populations are labeled with the single letter population designations given in the appendix. The first three axes account for 51.0% of the total variation and the first 14 factors have individual eigenvalues greater than 1.000.

the more typical *A. friesiana*. The apparent differentiation of Alaskan from Yukon populations of *A. monocephala* could be the result of relatively long isolation of these groups of alpine tundra populations. Many *Antennaria* populations could have survived glacial episodes in situ because most of this area was not glaciated by continental icesheets during the Pleistocene (Prest 1984).

The populations of *A. friesiana* (a, b, c, e, f, and h), which are the most distinct morphologically (fig. 4), are also so isozymically (fig. 3). Both morphological and allele frequency characters have high loadings along all three axes of figure 5, indicating the taxonomic importance of both types of characters. In this analysis, *A. densifolia*, *A. friesiana* s.l., *A. monocephala*, and *A. neoalaskana* are clearly delimited. The one asexual population of *A. monocephala* (=subsp. *angustata*) is intermediate between sexual *A. monocephala* and *A. friesiana* subsp. *friesiana*, as are some populations of *A. friesiana* (pops. d, g, i, k, and m). These populations may represent either allopolyploids between *A. monocephala* and *A. friesiana* subsp. *alaskana* or, in the case of sexual diploids, populations of *A. friesiana* subsp. *alaskana* that have undergone introgression with *A. monocephala*.

The cluster analysis of the arctic *Antennaria* and members of the Alpinae from the Rocky Mountains (fig. 2) demonstrates that the southern species *A. aromatica*, *A. pulchella*, and *A. umbrinella* are isozymically similar and cluster into one group that is isozymically distinct from the arctic taxa. The relatively high degree of isozyme divergence between the two groups may indicate a relatively long period of isolation from each other. The cluster analysis suggests a close relationship of sexual *A. friesiana* subsp. *alaskana* and apomictic subsp. *friesiana*. Although *A. monocephala* and *A. neoalaskana* would appear to be closely allied isozymically (fig. 2), this is not the case with morphology (fig. 4). *Antennaria densifolia* is the most isozymically and morphologically distinct arctic species in the group studied (fig. 4).

Speciation and Taxonomy.   The low arctic species of *Antennaria* exhibit a pattern of speciation resembling that of other species of *Antennaria* (Bayer 1988, 1989a; Bayer and Crawford 1986). While the species tend to be morphologically well differentiated, isozymically they have not diverged to the same extent.

It seems best to recognize *A. densifolia*, *A. friesiana*, *A. monocephala*, and *A. neoalaskana* at the rank of species. The recognition of subspecies

in *A. friesiana* and *A. monocephala* may not be warranted in that the primary distinction among Hultén's (1968) subspecies is presence or absence of staminate plants. *Antennaria friesiana* subsp. *friesiana* is not morphologically distinct from subsp. *alaskana*; the former is simply the apomictic phase of the latter. From an evolutionary aspect, it may be desirable to recognize the distinction taxonomically because these two reproductive phases perhaps represent two different evolutionary lineages. From a practical aspect, the delineation of subspecies in *A. friesiana* may be unfeasible because determining reproductive mode from herbarium specimens may be difficult, especially in light of evidence that populations of *A. media* Greene containing both staminate and pistillate individuals may actually represent complex mixtures of sexual, facultatively apomictic, and obligately apomictic pistillate plants (Bayer et al. 1990). Additional morphological studies are needed to determine whether subspecific categories should be recognized within *A. friesiana* and *A. monocephala*.

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APPENDIX.   Locality data for 25 populations of *Antennaria densifolia, A. friesiana* s.l., *A. monocephala,* and *A. neoalaskana.* Presented are taxon names, population designations (a to y), Canadian territory/U.S. state, topographic quadrangle, latitude, longitude, elevation (meters) above sea level, collectors, collection number, reproductive mode and ploidy level [S = sexual; A = asexual; 28 = diploid ($2n = 28$); 56 = polyploid ($2n = 56$)]. Collectors are: BJMPU = R. J. Bayer, B. Jonsell, L. C. Marvin, B. G. Purdy, and K. Urbanska; BMP = Bayer, Marvin and Purdy. Voucher specimens are at ALTA.

Population designation/territory or state/ topographic quad name			Lat.	Long.	Elev.	Collectors and number (reproductive mode-ploidy level)
A. freisiana s.l.						
a	AK	Bettles	66°30'	150°45'	595	BJMPU *AK-89052* (S-56)
b	AK	Bettles	66°13'	150°20'	560	BJMPU *AK-89048* (S-56)
c	AK	Circle	65°25'	145°58'	995	BJMPU *AK-89138* (S-28)
d	AK	Healy	63°49'	148°40'	580	BJMPU *AK-89044* (A-56)
e	AK	Mt. Hayes	63°15'	145°40'	1040	BMP *AK-89162* (A-56)
f	AK	Philip Smith Mts.	68°06'	149°32'	1160	BJMPU *AK-89065* (S-28)
g	AK	Philip Smith Mts.	68°44'	149°01'	765	BJMPU *AK-89081* (S-28)
h	AK	Philip Smith Mts.	68°03'	149°38'	1100	BJMPU *AK-89057* (S-28)
i	AK	Sagavanirktok	69°28'	148°40'	370	BJMPU *AK-89086* (A-56)
j	AK	Wiseman	67°18'	150°05'	1165	BJMPU *AK-89056* (S-56)
k	NWT	Porcupine River	67°06'	136°09'	650	BMP *NWT-89027* (S-28)
l	YK	Klondike	64°05'	140°58'	1340	BMP *YK-89102* (A-56)
m	YK	Porcupine River	66°55'	136°15'	740	BMP *YK-89082* (S-56)
A. densifolia						
n	YK	Klondike	65°04'	138°13'	1045	BMP *YK-89081* (S-28)
o	YK	Klondike	65°24'	138°12'	610	BMP *YK-89091* (S-28)
A. monocephala s.l.						
p	AK	Anchorage	61°46'	149°16'	1285	BMP *AK-89032* (S-28)
q	AK	Circle	65°25'	145°58'	995	BJMPU *AK-89135* (S-28)
r	AK	Circle	65°28'	145°25'	1225	BJMPU *AK-89141* (S-28)
s	AK	Mt. Hayes	63°05'	145°41'	965	BMP *AK-89173* (S-28)
t	AK	Philip Smith Mts.	68°06'	149°32'	1160	BJMPU *AK-89066* (A-56)
u	NWT	Pelly River	63°15'	130°02'	1395	BMP *NWT-89023* (S-28)
v	YK	Klondike	64°29'	138°07'	1830	BMP *YK-89066* (S-28)
w	YK	Pelly River	63°56'	135°12'	1670	BMP *YK-89052* (S-28)
A. neoalaskana						
x	NWT	Porcupine River	67°04'	136°10'	850	BMP *NWT-89029* (S-56)
y	YK	Porcupine River	66°43'	136°20'	670	BMP *YK-89088* (S-56)