Creating new populations of *Acanthomintha duttonii*. I. Preliminary Laboratory and Field Studies.

Bruce M. Pavlik and Erin K. Espeland
Department of Biology
Mills College
Oakland, CA 94613

Prepared for

Endangered Plant Program
California Department of Fish and Game
1416 Ninth Street, Room 1225
Sacramento, CA 95814

Funded by

U.S. Fish and Wildlife Service Section 6 Funds
California Endangered Species Tax Check-Off Funds
Contract No. FG-9517

July 1991

Pavlik, B.M. and E.K. Espeland. 1991. Creating new populations of *Acanthomintha duttonii*. I. Preliminary Laboratory and Field Studies. State of California, Department of Fish and Game, Endangered Plant Program, Sacramento, CA.

Abstract

The San Mateo thornmint, *Acanthomintha duttonii* (= *A. obovata* ssp. *duttonii*), is state and federally -listed as an endangered plant. Of the four known historical occurrences of this distinctive species in San Mateo County, California, three have been extirpated. The only remaining natural population occurs in Edgewood Park, where there is still a great potential for extinction. In order for the species to recover, populations must now be created in appropriate habitat within historic range. The current project represents the initial phase of an effort to create the new populations and to determine the demographic, ecologic, and genetic factors that limit their growth and long-term stability. The objectives of this study included developing basic information on 1) seed production *in situ* , 2) seedling density and survivorship to reproduction *in situ* , 3) laboratory germination, and 4) the phenotypic responses of three *Acanthomintha* species (*A. duttonii, A. lanceolata,* and *A. ilicifolia*)to native and non-native soils when grown under greenhouse conditions.

Individual plants of *Acanthomintha duttonii* were found to produce large numbers of nutlets *in situ*, and this was readily quantified using one of several correlations with non-destructive measures of plant size (e.g. stem lengths, number of glomerules per plant). Both of these findings are critical to recovery efforts in that they allow for the creation and demographic monitoring of new populations.

The total population size of *A. duttonii* at Edgewood Park was large (9,000 and 13,000 plants during 1991 and 1990, respectively), producing a nutlet rain of up to 21,000 nutlets/m^2 . In 1991, survivorship to reproduction was relatively high, with 56% of the plants recorded in February present as flowering individuals in June. Given the limited and apparently fixed area of population, the lens of suitable serpentine clay habitat at Edgewood Park may be saturated with *A. duttonii* . This strongly suggests that large numbers of nutlets (several thousand) could be removed from the site during favorable years (such as 1990 and 1991) without significantly reducing population size. These "excess" nutlets should be used to recover the species by creating new populations in appropriate, protected habitat within historic range.

Germination rates of *A. duttonii* nutlets were high in the lab (87%) and greenhouse (35% on native soil), but six months were required for post- production dormancy to be broken. The endogenous control of dormancy observed in *A. duttonii*, and to a lesser extent *A. ilicifolia*, has few references in the published literature and requires further investigation. Germination rates, combined with survivorship rates *in situ*, will be used to design a new population to be established on the Pulgas Ridge site in 1991-1992.

A. duttonii was the only species of the three tested that was able to germinate, grow and flower on serpentine soil. Although *A. lanceolata* is often described as growing on serpentine, these results suggest that serpentine tolerance may be found in certain populations but not in others. The lack of any apparent germination and growth of *A. lanceolata* plants from the Iron Spring population indicates intolerance because all other soils (native soil, *A. ilicifolia* soil and potting soil) produced good numbers of healthy *A. lanceolata* seedlings. Similarly, it is highly unlikely that the absence of *A. ilicifolia* seedlings on *A. duttonii* soil was by chance alone. The physiological distinctiveness of the three *Acanthomintha* species was matched morphologically, even when grown on the same soil. Leaf shape, color, and margins were obviously different, even for germinules and seedlings. Furthermore, the patterns of dry matter allocation were distinctive, with *A. duttonii* plants having a much higher proportion of leaves than the other two taxa.

Acknowledgements

We gratefully acknowledge the assistance and support of Frances Whittman (Mills College), Toni Corelli (California Native Plant Society), Jack Kuhn (Edgewood Park), David Christy (County of San Mateo, Parks and Recreation), James Jokerst (student of *Acanthomintha*), Orlando Mistretta (Rancho Santa Ana Botanic Garden), Brad Burkhart (ERC Environmental and Services Company), Corky Mathews (California Native Plant Society), Vern Yadon (Pacific Grove Natural History Museum) and Jack and Zera Varian (Varian Ranch). Ann Howald and Ken Berg of the Endangered Plant Progam made the research possible and contributed many helpful suggestions.

Creating new populations of *Acanthomintha duttonii*. I. Preliminary Laboratory and Field Studies

Bruce M. Pavlik and Erin K. Espeland
Department of Biology
Mills College, Oakland, CA 94613
(415) 430-2158

Introduction

The San Mateo thornmint, *Acanthomintha duttonii* (Abrams) Jokerst & B.D. Tanowitz (= *A. obovata* Jepson ssp. *duttonii* Abrams), is state and federally -listed as an endangered plant. Of the four known historical occurrences of this distinctive species in San Mateo County, California, three have been extirpated by residential development (York, 1987). The only remaining natural population* occurs in Edgewood Park, which is administered by the San Mateo County Department of Environmental Management (Parks and Recreation Department). Although the site (see Sommers 1984 for a description) is now protected by the County, there is still a great potential for extinction. Significant changes in upslope drainage patterns have already taken place due to house and road construction. In addition, fire, vandalism (including off-road vehicles), and accidental disturbance will probably occur with increasing frequency as the adjacent human population grows.

To protect the San Mateo Thornmint, the risk of extinction needs to be spread among several populations instead of being concentrated on a single population. In order for the species to recover, populations must be created in appropriate habitat within historic range and afforded adequate protection and management. The current project represents the initial phase of an effort to create the new populations and to determine the demographic, ecologic, and genetic factors that limit their growth and long-term stability. The objectives of this study included developing basic information on 1) seed production *in situ*, 2) seedling density and survivorship to reproduction *in situ*, 3) laboratory germination, and 4) the phenotypic responses of three *Acanthomintha* species to native and non-native soils when grown under greenhouse conditions.

*One additional, very small population is known to occur at the "Triangle", just south of highway 280. It may have been introduced to the site only a few years ago. Due to its small size and questionable origin, the triangle population will not be considered here.

Review of Relevant Demographic, Ecologic, Genetic and Taxonomic Information in the Literature

Other than being a rare plant restricted to serpentine grasslands, relatively little is known about the demography, ecology, and genetics of *A . duttonii.* No demographic monitoring (sensu Pavlik 1987 and Pavlik and Barbour 1988) has been conducted on this or any other species of *Acanthomintha.* Surveys of *A . duttonii* at Edgewood Park (EP) have recorded large fluctuations in population size. In 1984, fewer than 500 individuals were found, while estimates in 1981 and 1986 were closer to 3,000 (NDDB 1989). An apparent high of 6,000 during 1985 was reported by McCarten (1986). Although such fluctuations are to be expected in populations of annual plants, the responsible factors have yet to be identified and related to management of this endangered species.

Ecologically, *A . duttonii* is associated with mesic serpentine grasslands that probably receive 45-55 cm of precipitation per year. Mean annual temperature is 12-15^{o} C and the mean annual range of temperature is 11.1^{o} C (all climate data from NOAA). Frosts are rare, with freezing temperatures occurring in less than 0.5% of the hours of the year (estimated from a Bailey nomogram). McCarten (1986) conducted detailed surveys of actual and potential habitat of the species in San Mateo County. He did extensive soil sampling and found that the deep serpentine clay of the EP site was moist, chemically unique and rather uncommon in the County. Using these data and a wealth of field experience, McCarten and others (especially Toni Corelli and Ken Himes) have mapped several possible sites for creating new populations along Pulgas Ridge. The ridge, largely composed of serpentine clay, lies along the eastern edge of the San Andreas Rift Zone.

Nothing is known about the genetic structure of the EP population. As an annual with showy, insect-pollinated flowers, *A . duttonii* is likely to be an outcrossing species and probably had localized genetic neighborhoods. Interpopulation variability would have been high prior to extirpation, but intrapopulation variation relatively low. This pattern is typical of habitat specialists whose populations are isolated from each other by significant barriers to gene flow. Land development has precluded any knowledge of metapopulation genetic structure, but the EP population should be electrophoretically characterized in order to determine the sampling methods needed for gene conservation.

The most recent taxonomic treatment of the genus has been done by James Jokerst for the Jepson Manual (to be published in 1993). He recognizes four species (*A. ilicifolia, A. obovata, A. duttonii* , and *A. lanceolata*) on the basis of style morphology, corolla morphology, stamen fertility, leaf morphology, geographic distribution, and substrate preference. Our greenhouse studies on three of those taxa (*A. ilicifolia, A. duttonii* and *A. lanceolata*), which consisted of reciprocal soil transplants in a common garden, substantiated Jokerst's delineation of taxa.

With respect to distribution and substrate, two species appear to be serpentine tolerant - *A. duttonii* and *A . lanceolata* . *A . duttonii* is found on low elevation serpentine grasslands in San Mateo County, California. It appears to have the most northernly distribution of all four species. Geographically, its nearest congener is *A . lanceolata* , which prefers rocky slopes, sometimes of serpentine, in Santa Clara County and further south into the coast ranges. Evolutionarily, its closest relative is *A . obovata* , which is found at non-serpentine sites in the South Coast Range. Finally, *A . ilicifolia* is restricted to clay mesas and vernal depressions in San Diego County. Therefore, a gross correlation exists between serpentine tolerance and geographic distribution in *Acanthomintha* , with taxa becoming more serpentine-restricted to the north.

Of the four known species, only *A. ilicifolia* has been studied under controlled conditions in the laboratory and garden (Mistretta and Burkhart 1990). Nutlets were found to have high germinability (>90%) and resultant plants grew well and flowered profusely in the garden. Hand pollinations demonstrated that *A. ilicifolia* is an obligate outcrosser. The authors noted that the density of plants in the garden may influence pollinator activity and subsequent nutlet production. Also documented was a significant decline in the germinability of nutlets produced in the garden (95% for wild nutlets vs. 45% for garden nutlets). The decline was attributed to a loss of genetic variability under cultivation, but other factors, including strong endogenous dormancy, might be responsible.

Methods and Materials

<u>Nutlet Production *in situ*</u>

During the peak flowering period of 1990, whole plants of *A . duttonii* were collected at the Edgewood Park (EP) site. The plants were closely inspected before cutting to insure they were near maximum size for the year and that a majority of nutlets were ripe (i.e. dark colored rather than green). Each plant was cut at soil surface and placed in its own zip-lock bag. These were returned to the lab, allowed to air dry at room temperature, and then dissected to determine 1) the total number of nutlets produced, 2) the number of flowers and glomerules, and 3) the sum of the stem lengths for each plant. Since each flower produces exactly four ovules, an estimate of ovule output was possible. Stem length was measured from the clipped point (at soil surface) to the base of the lowest glomerule. Nutlets were removed by shaking the whole plant or crushing the dry calyxes, and placed in paper envelopes. The envelopes were stored in an air-tight plastic container and refrigerated at a constant 5^{o} C. Thirty-one plants were collected on the first visit to the EP site on 18 May 1990. A second trip on 12 June was required to collect 12 larger plants that had green, unripe nutlets in May.

Correlations between various measures of plant size (stem length, # of flowers, etc.) and reproductive output were then calculated using methods developed during studies of other endangered plants (Pavlik and Barbour 1988, Pavlik et al. 1988, Pavlik 1991). The same procedures were followed in order to characterize nutlet production of field-grown plants of *A . lanceolata* from southern Monterey County. However, only 10 individuals were collected from the small, cryptic population (n = 82 plants) near Iron Spring Creek on 30 May 1990. The exact location (T. 22S R. 14E, section 16 of the Curry Mountain 7.5 U.S.G.S. topographic map) was along the north side of an intermittent stream that flows from the spring at 2960' elevation, about 200 feet below the Parkfield Grade road. The plants grew in the scree of a large outcrop of gray-green rock among a dense blue oak woodland community. Attempts were also made to find populations of *A. obovata* from this region, but these were unsuccessful.

It was not possible to determine the plant size-nutlet production correlation for *A . ilicifolia* during 1990 because plants had already senesced and fragmented by the time the field site in San Diego was visited in June. Approximately 1500 nutlets (1989 crop) for studies of germination and growth were obtained from Orlando Mistretta at Rancho Santa Ana Botanical Gardens.

Seedling Density and Survivorship to Reproduction *in situ*

Estimates of adult plant densities at Edgewood Park were made in May 1990 and June 1991. A total of thirteen 0.125 m^2 circular quadrats were used to map the population and to record the densities of other species (e.g., *Lolium multiflorum* and *Avena fatua*) on and off the serpentine clays at EP. Five of the 13 plots were randomly positioned using measuring tapes as axes and a random numbers table. These five were used to determine the mean density/m^2 and estimate the total size of the population (when multiplied by the area of the population, about 42 m^2).

The permanent plots were also used to estimate survivorship to reproduction during the 1991 growing season. Instead of sampling for plant density, 50 seedlings of *A. duttonii* were marked within each plot on 15 February. The plots were revisited on 12 June and the number of marked plants that were alive and flowering were tallied.

Laboratory Germination

After they were counted, the dried, field-grown nutlets of *A . duttonii* and *A . lanceolata* , and the garden-grown nutlets of *A . ilicifolia* were stored in a refrigerator (5^o C) prior to testing. Standard germination trials were begun in July 1990 using triplicate dishes of 25 nutlets each of *A . duttonii* and *A . ilicifolia* . Sterile petri dishes and filter paper served as the substrate, wetted with distilled water every 2-3 days. The standard conditions consisted of complete darkness at room temperature (20-22^o C day, 7-10^o C night). Due to the small number of available nutlets, *A . lanceolata* was not tested.

Additional trials on *A . duttonii* and *A . ilicifolia* were run in August, November, and December of 1990 and June 1991 because of strong, endogenous dormancy. Several factors affecting germination were also investigated during these trials, including short-term stratification (10 days @ 0^o C), long-term stratification (30 days @ 0^o C), pericarp scarification, exposure to fire and charcoal, exposure to red light (30 min), and exposure to two wet-dry cycles.

Growth Responses of *Acanthomintha* to Native and Non-native Soils

Large samples of native soils were collected when populations of the three *Acanthomintha* species were surveyed during 1990. These soils included serpentine clay from the EP population of *A . duttonii* (AD soil), serpentine (?) gravels from the Iron Spring population of *A . lanceolata* (AL soil), and mesa clays from the San Diego population of *A . ilicifolia* (AI soil) . Native AD soil for *A . duttonii* was collected in May from two places on the northwest edge of the population. Shallow pits were dug, roughly 30 cm across, and two five gallon buckets filled with soil (-10 to -25 cm depth). Five gallons of native AL soil for *A . lanceolata* were taken from a similar pit at the Iron Spring site in late May. Two pits were again used to obtain native AI soil for *A. ilicifolia* on the northeast and northwest edges of the population. At the lab, all soils were put through a 1/4 inch sieve and had a subsample of ~3000 cm^3 removed and stored in a zip-lock bag for analysis by the Soil Laboratory at UC Davis.

These native soils were used to set-up a reciprocal growth experiment conducted in a common greenhouse. Ten replicate pots of AD, ten of AI and ten of standard UC mix were planted with four nutlets each of *A . duttonii* on 8 January 1991, about the same time that this species germinates *in situ* . Five replicate pots of AD, five of AL, five of AI and five of standard UC mix were planted with four nutlets each of *A . lanceolata* . Ten replicate pots of AD, ten of AI and ten of standard UC mix were planted with four nutlets each of *A . ilicifolia* . These were moved to the Mills College greenhouse, spatially randomized, and automatically watered with a misting sprinkler system. Serpentine clay from the "triangle" site and from a proposed reintroduction site on Pulgas Ridge were also sown with nutlets of *A . duttonii* . The responses of germination, height growth, and total leaf area of these three species to soil type were recorded at monthly or bi-monthly intervals. Final harvests to determine dry weight accumulation and allocation were made in June when plants were at least 150 days old.

Results and Discussion

Nutlet Production *in situ*

The output of nutlets by *Acanthomintha duttonii* plants at Edgewood Park was linearly related to the sum of the stem lengths per plant (Table 1, Figure 1). An average-sized plant (Σ stem lengths = 10 cm) produced about 50 nutlets by late May 1990. One especially large individual (Σ = 202 cm, excluded from the analysis) produced 439 nutlets from 203 flowers. The number of flowers and the number of glomerules per plant (Figure 2) also positively correlated with nutlet output. It is clear that estimates of nutlet production by the population can be made readily made non-destructive measurements of plant size (e.g. stem lengths or number of glomerules).

Ovule output of *A . duttonii* was also related to the sum of the stem lengths per plant (Figure 3), and larger plants were significantly less efficient than smaller ones in converting ovules into nutlets (Figure 4, Table 1). The average reproductive efficiency (nutlet/ovule ratio) was 0.56, much higher than reported for other endangered annual species (Pavlik 1991). A few small plants had efficiencies as high as 1.00 (or more due to a counting error), and virtually every flower produced at least one nutlet (average of 2.2 nutlets/flower). It is unlikely, therefore, that reproduction of *Acanthomintha duttonii* is limited by inbreeding depression, pollinator availability or low levels of critical resources.

Table 1. Linear correlations between various measures of plant size and nutlet output, ovule output or reproductive efficiency per *Acanthomintha* individual, May 1990. **Bold type** indicates the relationship shown in Figures 1-4 . ns = not significant, repro eff = reproductive efficiency

n	X	Y	slope	intercept	r	P
***A. duttonii* - Edgewood Park 1990**						
40	**Σ of stem lengths (cm)**	**# nutlets**	**2.83**	**21.11**	**.71**	**<0.01**
40	$LOG_{10}\Sigma$ of stem lengths (cm)	# nutlets	68.63	-10.81	.71	<0.01
40	# flowers/plant	# nutlets	0.99	22.98	.72	<0.01
40	**# glomerules/plant**	**# nutlets**	**12.68**	**11.72**	**.80**	**<0.01**
40	**Σ of stem lengths (cm)**	**# ovules**	**9.52**	**11.25**	**.82**	**<0.01**
40	**Σ of stem lengths (cm)**	**repro eff**	**-0.012**	**0.68**	**.46**	**< 0.05**
***A. lanceolata* - Iron Springs 1990**						
10	Σ of stem lengths (cm)	# nutlets	0.14	6.15	.10	ns
10	# flowers/plant	# nutlets	0.13	5.65	.73	<0.05
10	# glomerules/plant	# nutlets	2.52	3.92	.70	<0.05
10	Σ of stem lengths (cm)	# ovules	12.52	-45.19	.41	ns
10	Σ of stem lengths (cm)	repro eff	-0.045	0.634	.68	< 0.05

Relationships between plant size and nutlet output by *Acanthomintha lanceolata* were weaker than those observed for *A. duttonii* (Table 1). No significant correlations between nutlet or ovule output and stem length were found. Those calculated for the number of flowers or glomerules, although statistically significant, had shallow or negligable slopes. This was largely due to the small sizes and low nutlet outputs of these plants overall. An average-sized plant (Σ stem lengths = 7 cm) produced only 6 nutlets by late May 1990. The average reproductive efficiency (nutlet/ovule ratio) was 0.31, and a flower was likely to produce only 1.2 nutlets. There was also a significant decline in reproductive efficiency with plant size much like that observed for *A. duttonii*. These data would indicate that pollinator availability or low levels of critical resources limit the reproductive output of *A. lanceolata* at the Iron Springs site.

Seedling Density and Survivorship to Reproduction *in situ*

The population of *A. duttonii* at Edgewood Park covers an area of about 42 m^2, with densities ranging from 64 plants/m^2 to 960 plants/m^2 during the months of May and June (Figure 5). Mean plant density reflected that large range, especially in 1990 (Table 2). Survivorship to reproduction was relatively high, with 56% of the plants recorded in February present as flowering individuals in June. The total population sizes were larger

Table 2. Density and survivorship of *Acanthomintha duttonii* at Edgewood Park, 1990 and 1991. n = 5 permanent quadrats. No data = " ---."

year	mean density (# plants/m^2)	mean survivorship (%)	estimated total repro population size
1990	302 ± 294	---	12,864
1991	230 ± 78	54.8 ± 14.9	9,660

than had been reported by McCarten (1986), although the population area was about the same. This indicates that the lens of suitable serpentine clay habitat at Edgewood Park may be saturated with *A. duttonii*. Indeed, if the relationship between the number of glomerules/plant and nutlet output is used to calculate the total nutlet production per unit area of habitat (the seed rain), the total number of nutlets per m^2 can range between 2,000 and 20,800 in a single year.

Laboratory Germination

Nutlets of *A. duttonii* completely resisted any attempt to induce germination during the July to December period of 1990 (Figure 6). None of the experimental treatments released a single nutlet from its dormancy during this period, including stratification, scarification (with fire and a razor blade), and exposure to light, red light, charcoal, and exposure to wet-dry cycles. At the end of December another set of trials was run that roughly coincided with the period of germination *in situ*. Within four days most of the nutlets had germinated, achieving an average of 87% by the end of the ten day trial. Nutlets that had been stratified also germinated, but at significantly lower rates than untreated controls (Table 3). In June the nutlets had entered another period of dormancy, and required 16 days to achieve 9 % germination (only 5 % after the standard 10 days).

Table 3. Effects of stratification (10 day, 30 day) on germination (%) of nutlets of *Acanthomintha* under laboratory conditions. All trials were begun on 31 December 1990 after the stratification period. Means ± SD (n = 3) in a column followed by different letters are different at $P < 0.01$ (ANOVA, arcsine transformed data).

	A. duttonii	*A. ilicifolia*
control	86.7 ± 7.5^a	78.7 ± 8.2^a
10 day stratification	52.0 ± 8.6^b	20.0 ± 22.9^b
30 day stratification	20.0 ± 8.6^c	38.7 ± 6.8^b

These year-old nutlets were still viable, however, because after 24 days 60 % had germinated. It is clear that some endogenous control was being exerted over the ability of *A. duttonii* nutlets to germinate. This unidentified control mechanism allowed breaking of dormancy only after a certain amount of time had passed, even when the nutlets had been stored in a "clueless" environment (dark, constant-temperature refrigerator). *A. duttonii* germinates later in the year than many grassland annuals, perhaps because of this endogenous timing mechanism. There is little in the published literature pertaining to such endogenous timing in seeds, and further trials are required to establish the cyclical nature of dormancy patterns in *A. duttonii*.

A weaker pattern of dormancy and germination was observed in the nutlets of *A. ilicifolia*, but it was none-the-less apparent (Figure 7). Germination rose from 8% during the July trial to 79% in the late December trial. By June germination declined slightly but significantly ($P < 0.05$, ANOVA) to 61%. It must be noted, however, that nutlets of *A. ilicifolia* were one year older than those of *A. duttonii* and may have lost some endogenous control by the time they were tested.

Responses of *Acanthomintha* to Native and Non-native Soils

A. duttonii was the only species to germinate in all of the tested soils, with a range of 25 to 42% (Table 4). *A. lanceolata* and *A. ilicifolia* did not germinate at all on the serpentine soil of *A. duttonii* (referred to as AD soil), despite having relatively high

Table 4. Germination of *Acanthomintha* nutlets in native and non-native soils. Means ± SD (n = 5) tallied two months after sowing. **Bold type** indicates response to native soil for that particular species. No trial = "--".

	germination (% of sown)			
	AD soil	AL soil	AI soil	potting soil
A. duttonii (AD)	**35.0 ± 9.4**	--	25.0 ± 15.8	42.5 ± 15.0
A. lanceolata (AL)	0.0 ± 0.0	**33.0 ± 29.5**	59.6 ± 25.0	39.6 ± 13.2
A. ilicifolia (AI)	0.0 ± 0.0	--	**52.5 ± 9.4**	47.5 ± 14.6

germination (33 - 60%) on all other soils. These data suggest that nutlets of these two species were either inhibited by the chemical properties of AD soil and did not come out of dormancy or they, in fact, did germinate but died soon after radicle emergence. In either case, *A. duttonii* nutlets were unique in their physiological response to serpentine soil during germination.

All three species grew well for the first 90 days after germination (vegetative growth period), although the effects of soil type were quite apparent. Plants in their respective native soils were robust, branched, and leafy. *A. duttonii* and *A. lanceolata* were able to grow equally well on the *A. ilicifolia* soil (AI soil), attaining the same leaf area (Table 5) and height (data not shown) as they did on their native soils. None of the species, however, were able to grow well in potting soil, remaining small, unbranched, and with a few, chlorotic leaves. The effect of potting soil on leaf area was especially pronounced. Evidently, *Acanthomintha* plants were more susceptible to nutrient deficiency on potting soil, even though it is commonly used to raise a wide variety of species under greenhouse conditions. Subsequent additions of Hoagland's solution to some of these pots allowed plants to grow more vigorously but not to the same degree as on the other soils.

Only AI soil allowed for comparisons of leaf morphology between the three species under the same environmental conditions because it was the only soil in which all species grew. The distinctive features of these three species were maintained in this "common garden". Leaves of *A. duttonii* were obovoid to rhomboid, light green with whitish veins, and margins entire or vaguely crenulate. Those of *A. lanceolata* were lanceolate-oblong, dark green with red veins, and margins crenulate. The leaves of *A. ilicifolia* were lanceolate-ovate, light green with white veins, and margins deeply crenate-serrate. Furthermore, stems and hypocotyles of *A. duttonii* and *A. lanceolata* had an obvious red or pink pigmentation, while those of *A. ilicifolia* did not. These and other differences were maintained across soils and time, allowing easy discrimination between the youngest of seedlings.

The plants were more difficult to grow as they entered their reproductive phase. This was due to several factors, including uneven drainage in the pots (especially those with AD soil) and a few days with unusually high temperatures in the greenhouse. Repeated watering caused the clayey soils to aggregate and draw away from the walls of the pots. The gap thus created allowed water to flow around the outside of the soil instead of permeating it evenly, and also leached roots of nutrients at the bottom of the pots.

Table 5. Total leaf area of 72 day old *Acanthomintha* plants grown in native and non-native soils. Means ± SD in a row followed by different letters are significantly different (P<0.05, ANOVA). **Bold type** indicates native soil for that particular species. No trial = "--".

	leaf area (cm^2/plant)			
	AD soil	AL soil	AI soil	potting soil
A. duttonii (AD)	**23.2 ± 19.2**^{a}	--	31.2 ± 29.4^a	0.7 ± 0.4^b
A. lanceolata (AL)	0.0 ± 0.0*	**20.0 ± 9.4**^{a}	14.4 ± 13.5^a	1.1 ± 0.5^b
A. ilicifolia (AI)	0.0 ± 0.0*	--	**30.2 ± 12.1**^{a}	0.8 ± 0.3^b

* no nutlets germinated

As a result, a number of older plants of all three species died and others appeared stressed and nutrient deficient by the time of harvest (150 days post-germination). Some *A. duttonii* and all of the *A. lanceolata* plants remained in the vegetative state while other *A. duttonii* and all of the *A. ilicifolia* plants flowered. Smith (1984) discussed the problem of drainage when growing other species in serpentine soil.

In the absence of harvestable plants from all soil types, comparisons of total dry weight between species do not provide additional insight into growth responses (Table 6). Reproductive and vegetative individuals of *A. duttonii* grew best on native soil and worst on potting mix. Vegetative growth of the species on serpentine clay from the proposed reintroduction site at Pulgas Ridge (mean dry weight = 81.6 mg/plant) was not statistically different from growth on native soil (107.4 mg/plant), but no plants were reproductive at the time of harvest. These data suggest that the Pulgas Ridge site is appropriate but not optimal habitat for *A. duttonii*. Vegetative individuals of *A. lanceolata* grew equally well on either AL or AI soil, but did especially well in potting soil after fertilization with Hoagland's solution. *A. ilicifolia* grew and flowered profusely on its native soil, but did poorly on potting mix even after fertilization.

Table 6. Total dry weight of 150 day-old *Acanthomintha* plants grown in native and non-native soils. Means ± SD are shown (n = 4). **Bold type** indicates native soil for that particular species. No trial = "--".

	dry weight (mg/plant)			
	AD soil	AL soil	AI soil	potting soil
A. duttonii (AD)				
vegetative	**107.4 ± 29.2**	--	77.3 ± 64.6	24.7 ± 17.0
reproductive	**346.9 ± 106.3**	--	130.3 ± 7.3	110.5 ± 10.8
A. lanceolata (AL)				
vegetative	0.0 ± 0.0*	**148.2 ± 51.2**	100.3 ± 34.0	262.3 ± 100.8
A. ilicifolia (AI)				
reproductive	0.0 ± 0.0*	--	**1079.2 ± 602.3**	died

* no nutlets germinated

Patterns of dry matter allocation were distinctive for the three species regardless of phenological stage (Table 7). *A. duttonii* allocated significantly more of its dry matter to leaves and less to roots than either *A. lanceolata* or *A. ilicifolia*. As a result, *A. duttonii* had a significantly lower root/shoot ratio than the other species. The R/S ratios of *A. lanceolata* and *A. ilicifolia* , however, were statistically equivalent, indicating that a shift in allocation to shoots instead of shoots may be an adaptive characteristic on serpentine. Although an extensive search of the recent literature has not been done, there has been very little work on the comparative physiology of plants restricted to serpentine (Kruckeberg 1984).

Table 7. Dry weight allocation of 150 day old *Acanthomintha* plants grown on native soils. Means ± SD (n=4) in a column followed by different letters are significantly different ($P<0.05$, ANOVA). Mean root to shoot ratio (R/S) excluding flower weight is shown and also compared by ANOVA.

	dry weight (% of total)				
	leaves	stems	roots	flowers	R/S
A. duttonii					
vegetative	71.1 ± 3.0[a]	17.7 ± 2.4[a]	11.3 ± 2.5[a]	---	0.13[a]
reproductive	68.8 ± 10.1[a]	18.8 ± 5.6[a]	9.6 ± 5.4[a]	3.0 ± 3.6[a]	0.12[a]
A. lanceolata					
vegetative	43.5 ± 4.3[b]	26.1 ± 7.0[b]	24.5 ± 7.7[b]	---	0.37[b]
A. ilicifolia					
reproductive	20.8 ± 7.6[c]	23.6 ± 2.3[a]	21.2 ± 1.6[b]	34.3 ± 5.5[b]	0.46[b]

Conclusions and Management Recommendations

1) Individual plants of *Acanthomintha duttonii* can produce large numbers of nutlets *in situ*, and this can be readily quantified using one of several correlations with non-destructive measures of plant size (e.g. stem lengths, number of glomerules per plant). Both of these findings are critical to recovery efforts in that they allow for the creation and demographic monitoring of new populations.

2) The total population size at Edgewood Park was large (9,000 and 13,000 plants during 1991 and 1990, respectively), producing a nutlet rain of up to 21,000 nutlets/m^2. In 1991, survivorship to reproduction was relatively high, with 56% of the plants recorded

in February present as flowering individuals in June. Given the limited and apparently fixed area of population, the lens of suitable serpentine clay habitat at Edgewood Park may be saturated with *A. duttonii*. This strongly suggests that large numbers of nutlets (several thousand) could be removed from the site during a favorable years (such as 1990 and 1991) without significantly impacting the population. These "excess" nutlets should be used to recover the species by creating new populations in appropriate, protected habitat within historic range. Temporary cold (4^{o} C) storage of 5,000 ripe nutlets is recommended, therefore, as an interim measure to prevent extinction of the species. These could be collected from 80-100 individual plants representing 1% of the total population in 1991 and less than 6% of the total nutlet rain (gross estimate based on a minimum rain of 2000 nutlets/m^2 X 42 m^2).

3) Germination rates of *A. duttonii* nutlets were high in the lab (87%) and greenhouse (35% on native soil), but six months were required for post- production dormancy to be broken. The endogenous control of dormancy observed in *A. duttonii*, and to a lesser extent *A. ilicifolia*, has few references in the published literature and requires further investigation. Germination rates, combined with survivorship rates *in situ*, will be used to design a new population to be established on the Pulgas Ridge site in 1991-1992.

4) *A. duttonii* was the only one of the three species tested able to germinate, grow and flower on serpentine soil. Although *A. lanceolata* is often mentioned as occurring on serpentine, these results suggest that tolerance may be found in certain populations but not in others. The lack of any apparent germination and growth of plants from the Iron Spring population indicates intolerance because all other soils (native, AI and potting) produced good numbers of *A. lanceolata* seedlings. Similarly, it is highly unlikely that the absence of *A. ilicifolia* seedlings on AD soil was by chance alone. The physiological distinctiveness of these three species was matched morphologically, even when grown on the same soil. Leaf shape, color, and margins were obviously different, even for germinules and seedlings. Furthermore, the patterns of dry matter allocation were distinctive, with *A. duttonii* plants having a much higher proportion of leaves and a lower proportion of roots than the other two taxa. Genetically-based shifts in dry matter allocation from roots to shoots may be adaptive on serpentine soils.

5) Additional studies are needed to locate populations of *A. obovata*, collect seed, and to conduct a comparative growth experiment with its close relative *A. duttonii*. This will allow further evaluation of systematic relations in the genus and test addtional hypotheses related to germination and growth adaptations to serpentine soil.

6) The yearly census of the Edgewood Park population should be continued using the permanent and transient quadrats sampled during this study. This will standardize the estimates and allow for better comparisons between different years.

7) Attempts to create new populations of *A. duttonii* should be made immediately in order to spread the risk of extinction among several populations. There are no inherent limitations imposed by nutlet germination, plant survivorship, or nutlet output by mature plants. Furthermore, the soil at Pulgas Ridge allowed greenhouse-grown plants of *A. duttonii* to grow and mature in a similar manner as plants grown on soil from Edgewood Park. Pulgas Ridge should, therefore, be the site of the first attempt to create a new population.

Literature Cited

Kruckeberg, Arthur .R. 1984. California serpentines: Flora, vegetation, geology, soils and management problems. University of California Publications in Botany 78, 180 pp.

McCarten, Niall F. 1986. A study of the ecological aspects related to the reintroduction of *Acanthomintha obovata* ssp. *duttonii* . State of California, Department Fish and Game, Endangered Plant Program, Sacramento, CA 36 pp.

Mistretta, Orlando and Brad Burkhart. 1990. San Diego thornmint. Propagation, cultivation provides clues to ecology of endangered species. Restoration and Management Notes 8, 50.

NDDB (California Natural Diversity Data Base). 1989. File information on *Acanthomintha obovata* ssp. *duttonii* . State of California, Department Fish and Game, Natural Heritage Program, Sacramento, CA.

NOAA (National Oceanic and Atmospheric Administration). 1974. Climatic summary, California. U.S. Department of Commerce, Washington, D.C.

Pavlik, Bruce M. 1987. Autecological Monitoring of Endangered Plants. In: T. Elias (ed.)*Conservation and Management of Rare and Endangered Plants.* California Native Plant Society, Sacramento. pp. 385-390.

Pavlik, Bruce M. and Michael G. Barbour. 1988. Demographic monitoring of endemic sand dune plants, Eureka Valley, California. Biological Conservation 46, 217-242.

Pavlik, Bruce M., N. Ferguson, E. Manning, M. Nelson 1988. Demographic Studies of Endemic Plants at the Antioch Dunes National Wildlife Refuge. II. Seedling Demography, Seed Production and Seed Bank Dynamics. State of California, Department Fish and Game, Endangered Plant Program, Sacramento, CA 107 pp.

Pavlik, Bruce M. 1990. Reintroduction of *Amsinckia grandiflora* to Stewartville. State of California, Department of Fish and Game, Endangered Plant Program, Sacramento, Ca. 59 pp.

Pavlik, Bruce M. 1991. Reintroduction of *Amsinckia grandiflora* to three sites across historic range. State of California, Department of Fish and Game, Endangered Plant Program, Sacramento, Ca. 45 pp.

Smith, N. 1984. Growing natives: Plants of serpentine. Fremontia 11:5, 31-33.

Sommers, Susan. 1984. Edgewood Park. Fremontia 11:5, 19-20.

York, Richard P. 1987. California's Most Endangered Plants. In: T. Elias (ed.) *Conservation and Management of Rare and Endangered Plants.* California Native Plant Society, Sacramento. pp. 109 -120.

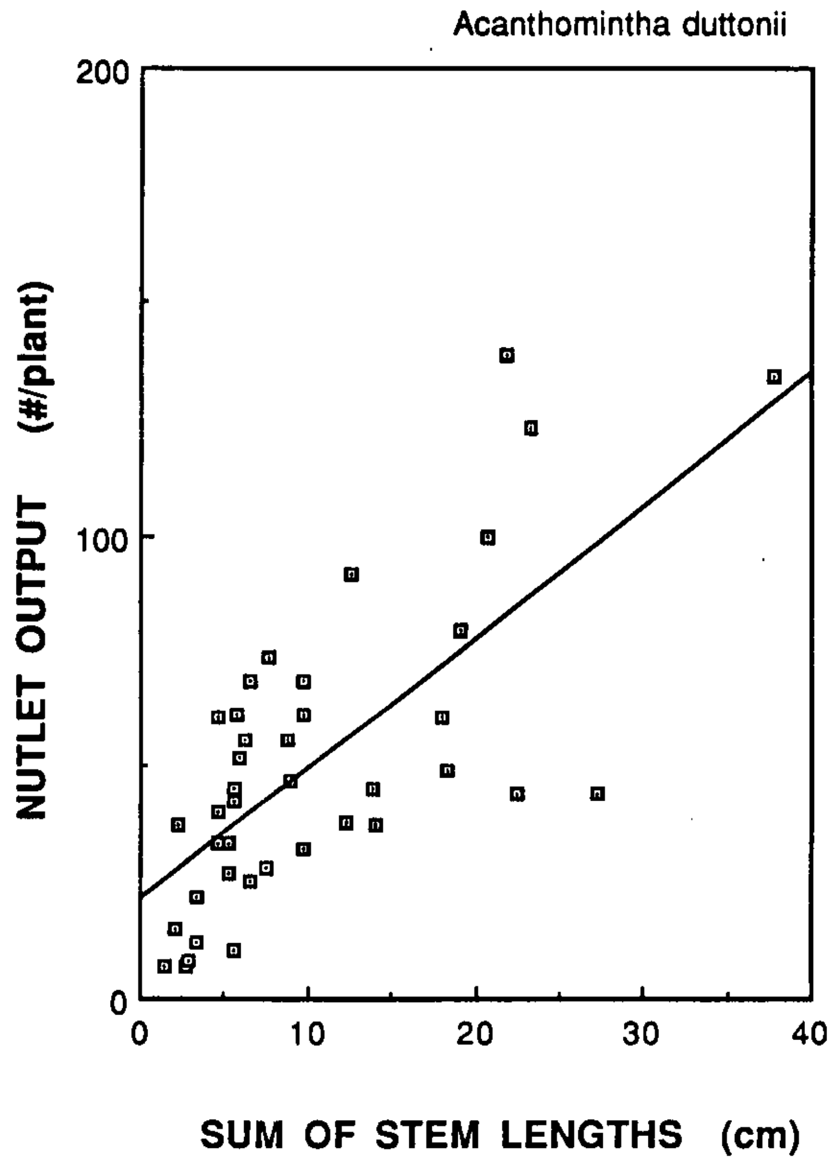

Figure 1. Nutlet output as a function of the sum of the stem lengths for *Acanthomintha duttonii* at Edgewood Park, May 1990. See Table 1 for line equation.

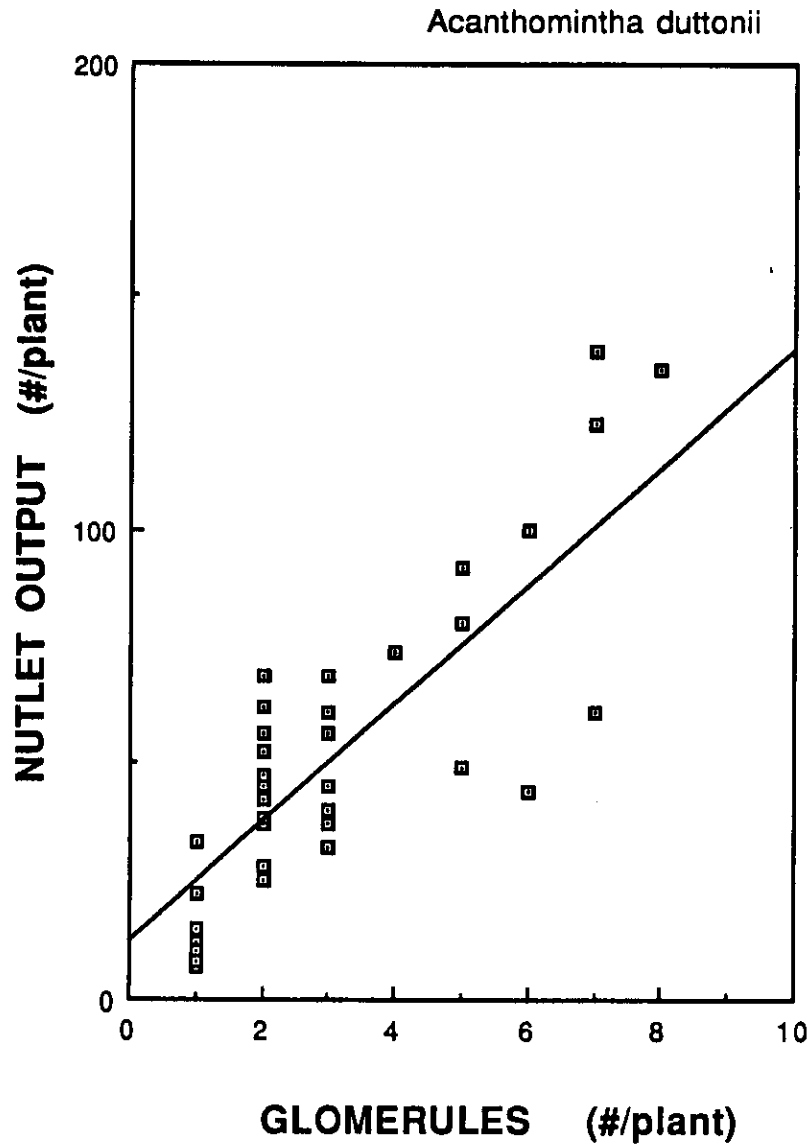

Figure 2. Nutlet output as a function of the number of glomerules of Acanthomintha duttonii at Edgewood Park, May 1990. See Table 1 for line equation.

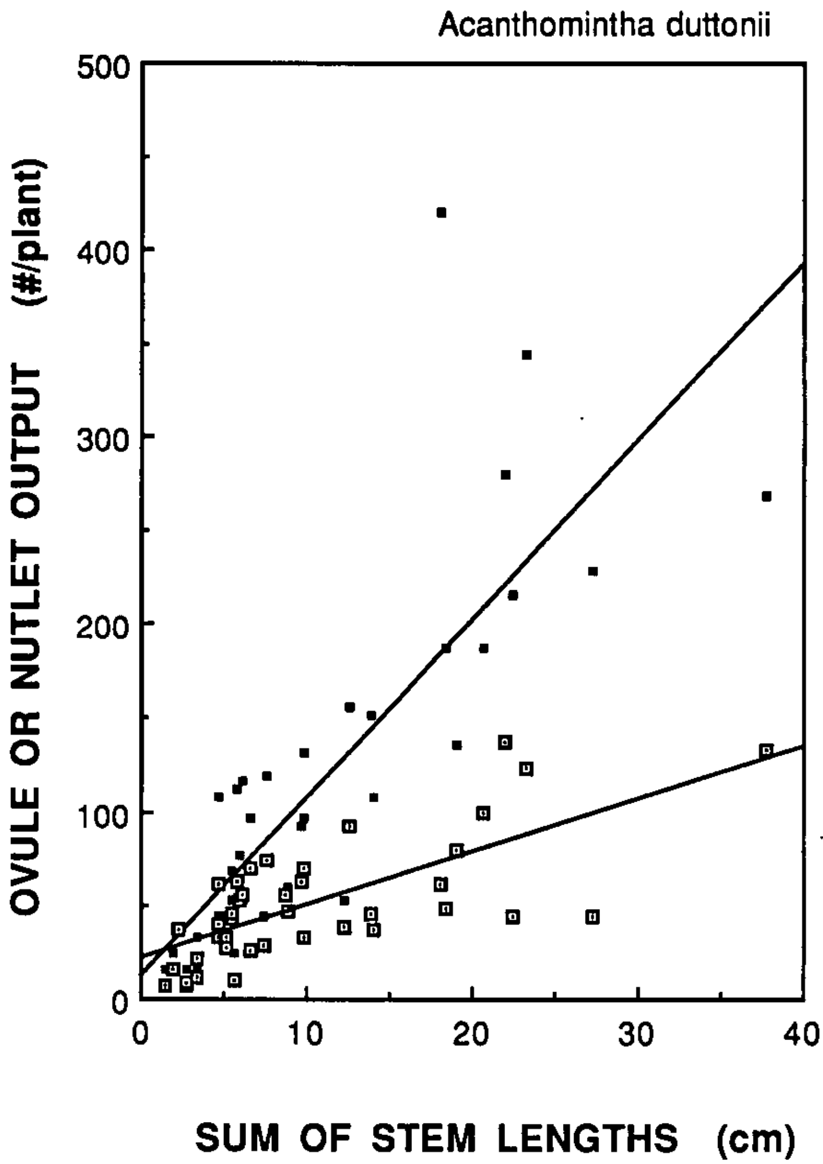

Figure 3. Ovule (closed squares) and nutlet (open-dot squares) output as functions of the sum of the stem lengths for <u>Acanthomintha duttonii</u> at Edgewood Park, May 1990. See Table 1 for line equations.

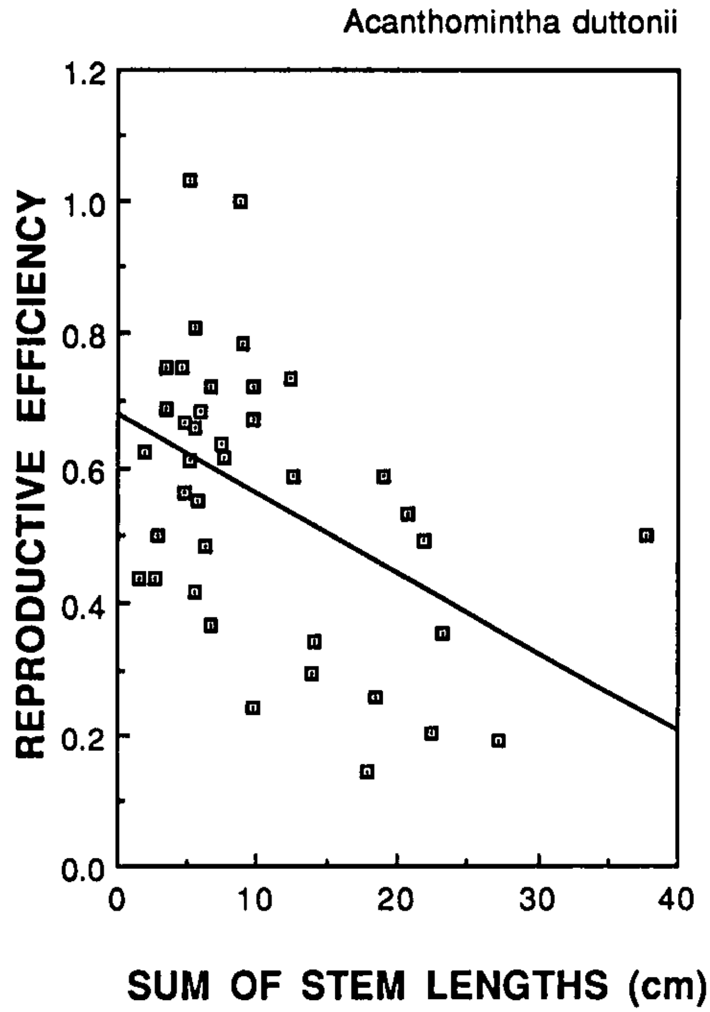

Figure 4. Reproductive efficiency (nutlet/ovule ratio) as a function of the sum of the stem lengths of Acanthomintha duttonii at Edgewood Park, May 1990. See Table 1 for line equation.

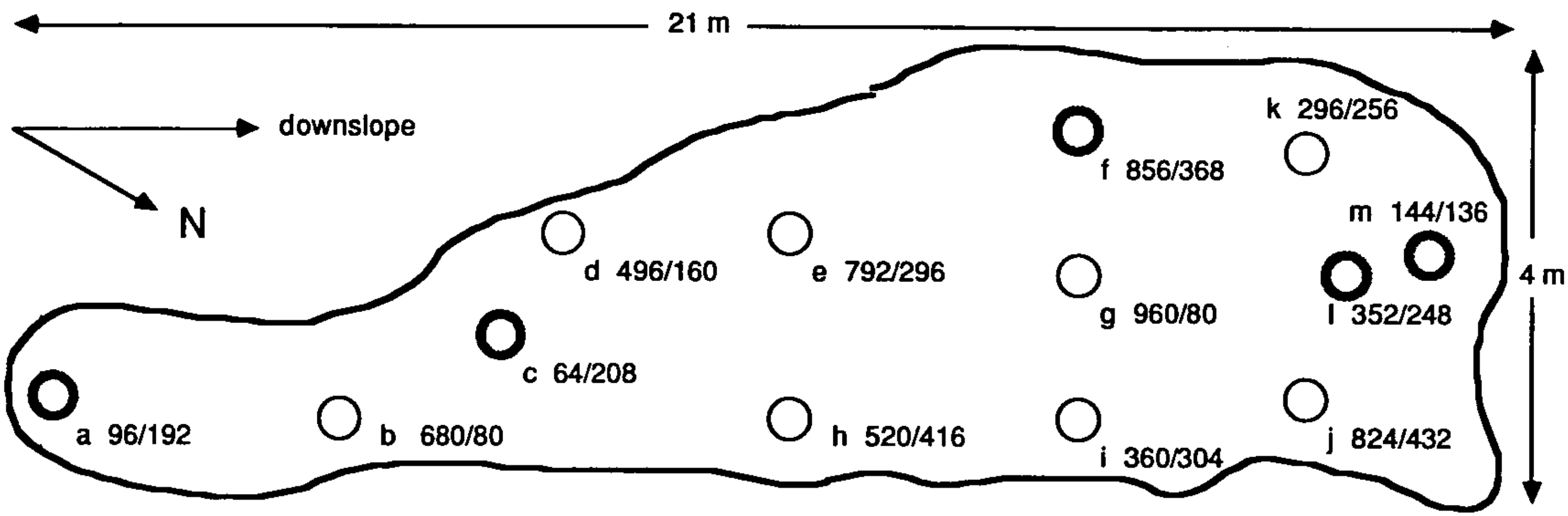

Figure 5. Map of the Edgewood Park population of _Acanthomintha duttonii_. Dark circles indicate positions of 5 randomly-placed permanant quadrats (0.125 m2). Light circles indicate positions of 8 non-random, transient quadrats (0.125 m2). Numbers indicate plant densities (#/m2) in May 1990/June 1991.

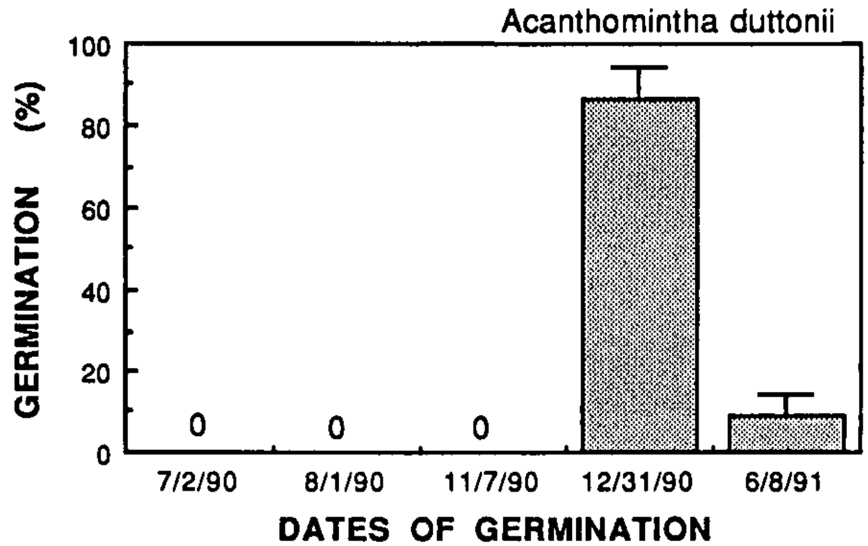

Figure 6. Laboratory germination of A. duttonii nutlets collected in May 1990. Mean ± SD (n=3) shown.

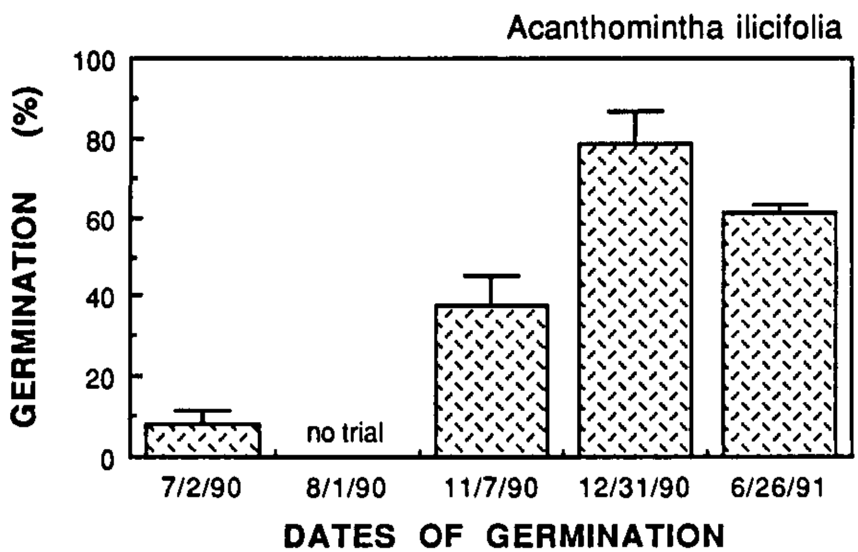

Figure 7. Laboratory germination of A. ilicifolia nutlets collected in 1989. Mean ± SD (n=3) shown.