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SURVEY AND MONITORING OF RUNNING BUFFALO CLOVER (Trifolium stoloniferum) IN OHIO

A Report for Region 3, U.S. Fish and Wildlife Service
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INTRODUCTION

Trifolium stoloniferum Muhl. ex A. Eaton (running buffalo clover) is a perennial, stoloniferous herb with a historic distribution throughout several states in the midwestern United States. Running buffalo clover is formerly known from Arkansas, Illinois, Indiana, Kansas, Kentucky, Missouri, Nebraska, Ohio and West Virginia. The species was believed to be extinct until 1983 when two small populations were found in West Virginia. Several other populations were later discovered in Indiana and Kentucky and by 1987 running buffalo clover was listed by the U.S. Fish and Wildlife Service as an endangered species. The species is currently known from over 65 populations across 4 states: Indiana, Kentucky, Ohio and West Virginia. There is also evidence of at least one natural population in Missouri as well as four restored populations. Many of the known populations of running buffalo clover are small (10 - 200 plants) but some of the recently discovered populations in Kentucky, Ohio and West Virginia are relatively large (> 1000 plants).

In Ohio, running buffalo clover is presently known from 11 populations in 2 southwestern counties (Figure 1). Nine populations, including the largest in the state, are located in Hamilton County. The remaining two natural populations occur in Clermont County, along with a transplanted population located at the Cincinnati Nature Center. Two populations had previously been recorded from Warren County but these populations have not been re-located since 1988-89. There are also historical records of running buffalo clover in several other Ohio counties including: Belmont, Butler, Clark, Clinton, Franklin, Greene, Miami and Montgomery (Figure 1).

Only one of the 11 extant Ohio populations is fully protected in a dedicated State Nature Preserve. Eight populations are under some protection in Park Districts and Historical

Society property. These sites are not legally protected, however, and may be subject to disturbance or destruction without notice. The remaining two populations are on private property. Although current landowners have shown an interest in these populations, part of one population has been destroyed and their long-term protection and management cannot be guaranteed.

The habitats of the Ohio populations of running buffalo clover are generally semi-shaded, moist areas with moderate levels of disturbance. Several populations are located near streams or other watercourses and most of the populations experience some sort of disturbance including mowing, trampling, grazing or any combination of these.

Due to its stoloniferous habit, running buffalo clover is a difficult species to census and monitor. Preliminary data collected by the Division of Natural Areas and Preserves (DNAP), show that populations fluctuate in number both during the growing season and from year to year. A simple census of all populations can aid in documenting the status of the species, but more complex demographic monitoring is needed to determine the stability and status of individual populations over time.

Little monitoring of this type has been conducted on natural populations and there are no standardized methods being consistently used to study the plant. This study was conducted to establish monitoring techniques for running buffalo clover, as well as to determine the status of the plant in Ohio. The objectives of the study were threefold: 1) to census known populations; 2) to survey sites in several Ohio counties where there are historical records of running buffalo clover; and 3) to conduct demographic monitoring of several populations, including the largest known population.

METHODS

A survey was conducted for running buffalo clover in 8 southwestern Ohio counties from May - October of 1993 by DNAP seasonal botanist, Sabina Sulgrove (Figure 1). Survey sites were selected on the basis of recent or historical records, or evidence of historical Native American or pioneer activity such as old cemeteries. Four sites were surveyed in Warren county, two of which supported populations of running buffalo clover as recently as 1988-89. One new site in both Hamilton and Clermont counties was searched and twenty-one sites, including several parks and cemeteries, were surveyed in Clark County. Eleven cemeteries in Clinton County and 14 cemeteries and parks in Greene County were surveyed. Finally, four cemeteries in Miami County and eight sites in Montgomery County were also searched for populations of running buffalo clover.

A census of all known populations, including the transplants, was conducted at 16 sites in Hamilton and Clermont Counties. Censuses, to estimate population size, were conducted by one of the following techniques: 1) the total number of plants was counted (Note: a plant was defined as a rooted crown); or 2) the total number of plants was estimated by counting the number of plants in one or more representative square-meter plots, measuring the total area occupied by the plants and extrapolating. The second technique was used in large populations where counting individual plants was not practical.

Two populations, Niehaus (the largest known population in the state) and Warder-Perkins, were selected for additional demographic studies. Demography is the quantitative study of population changes and their causes throughout the life cycle. This type of study is often more complex and time consuming but provides detailed information on the stability of individual populations by measuring reproduction, recruitment, and mortality of individual plants.

At each of these two sites, one permanent square-meter plot was established. Using a grid system within the square-meter frame, all plants (i.e. rooted crowns) were mapped and the following data recorded: 1) the number of stolons per plant (and their location); and 2) the number of flowering stalks (and their location). These data were collected twice (May and July) during the 1993 growing season for both plots, and once (May) in 1991 and 1992 for the Warder-Perkins plot.

Additional demographic data have been collected from 1991 - 1993 at three sites in the Hamilton County Park District (HCPD). Marjorie Becus, an independent researcher funded by DNAP research grants, established permanent square-meter plots at these sites and made at least three visits during each growing season to count plants. Becus has also censused these and four other HCPD populations annually from 1989/90 - 1993. Her data provide important information on the fluctuation of population size throughout the growing season and from year to year (Becus 1989-1993).

RESULTS AND DISCUSSION

A total of 64 new sites were surveyed in eight counties in southwestern Ohio. No running buffalo clover was found at surveyed sites in Clark, Clinton, Greene, Miami, Montgomery or Warren Counties. Only one of the surveyed sites, the Milford Area Historical Society Center in Clermont county, contained running buffalo clover. However, the 1993 search for running buffalo clover was not comprehensive. Other possible sites within these counties should be searched and several other counties with historical records (Belmont, Butler and Franklin) still remain to be surveyed. Given the tendency of the plant for disturbed habitats, historical sites and other appropriate areas should be searched periodically for recently established populations.

A total of 16 sites with recent populations of running buffalo clover were censused.

Only 12 of these populations, three in Clermont (including the transplants) and nine in Hamilton Counties, had plants in 1993. The census of the 11 natural populations revealed a wide range in population sizes (Table 1). Miami Whitewater Forest had the smallest population with only five plants, while the Niehaus population was the largest with an estimated 2500 plants. Plants occurred in areas as small as a few square feet to as large as an acre. Six populations (Bobcat Ridge, Cabin View, Fankhauser, Congress Green Cemetery, Milford Area Historical Society Center and Miami Fort) were found in picnic areas, lawns, cemeteries and other semi-open, grassy fields frequently disturbed by mowing. The remaining five populations (Warder-Perkins, Niehaus, Newberry, Mitchell Memorial Forest and Miami Whitewater Forest) were located within disturbed areas in woodlands such as deer trails, human trails and old logging roads. All of these populations should continue to be censused periodically in order to monitor changes in population size.

The transplanted population at the Cincinnati Nature Center in Clermont County was also censused. This population consists of plants removed from the Fankhauser population due to the construction of a garage. In 1990, 17 plants were transplanted to a seemingly appropriate area at the Cincinnati Nature Center. Sixteen plants remained in 1991; in 1993 only five plants were found. The continued monitoring of this population will aid in determining the long-term success and stability of transplanted and/or reintroduced populations.

Demographic data collected at the Warder-Perkins, Niehaus and Hamilton County Park District sites reveal important information about the fluctuation of population size throughout the growing season and from year to year. Through the beginning of the 1993 growing season (May - July) the number of rooted crowns increased in both the Warder-Perkins and the Niehaus square-meter plots (Table 2). The opposite was true, however, for the HCPD plots in 1993 (Table 3); the number of rooted crowns decreased from May to August in all

three plots. Sampling in August, which was extremely dry in 1993, rather than July may account for these differences among populations.

The HCPD plots were also monitored during the 1991 and 1992 growing seasons on a more frequent basis. During these two years the number of rooted crowns generally increased from May to July - August, and then began to decrease in September - October (Table 3). Based on these data, at least part of the life history of the species may be suggested here. In May, the number of plants (i.e. rooted crowns) is similar to the number recorded for the previous fall (Table 3). At this time, plants flower and/or produce stolons and "daughter crowns". The peak number of plants appears from July - August indicating the stolons and new crowns produced in May had rooted and were counted as new plants. The decline in the number of plants in September - October suggests that rooted crowns experience senescence or mortality during this period.

Exactly which crowns ("parent" or "daughter crowns") die and/or senesce is not known, but maps of the Warder-Perkins plot in May 1991-93 suggest that there may be an uneven distribution of mortality/senescence among rooted crowns. In May 1991, 13 rooted crowns and total of 36 stolons were mapped in the plot (Figure 2 and Table 4). If all of these stolons rooted and the original 13 parent crowns survived, the potential number of rooted crowns the following May would be 49. As seen in Table 4, the number of rooted crowns observed in May 1992 was well below this.

In May 1992, the plot was mapped again and only five of the possible 49 rooted crowns were found. All five crowns were found in new positions compared to the "parent crowns" found in May 1991 (Figure 2). These data suggest that the "parent crowns" may die after producing stolons and "daughter crowns", and that very few of the "daughter crowns" develop and survive as rooted crowns. Data collected from the same plot from 1992 to 1993

appear to confirm these conclusions (Figure 2 and Table 4).

The 1993 map of the Warder-Perkins plot in May and July also confirms, at least in part, the proposed life history (Figure 3). In May, six rooted crowns were counted in the plot. Two of these crowns had one stolon and one had two stolons. In July, although one rooted crown and its stolon had disappeared, the remaining stolons had rooted bringing the total number of rooted crowns in the plot to eight (Table 2). A similar increase in the number of rooted crowns from May to July was observed in the Niehaus plot in 1993 (Table 2). The fate of the "daughter crowns", stolons and "parent crowns" was not determined in the Warder-Perkins and Niehaus plots later in 1993. Based on the HCPD plots, however, (Table 3) one might expect to see a decrease in the number of rooted crowns late in the growing season due to the death or senescence of both "parent" and "daughter crowns". Repeating this study in 1994 and mapping the plants in these plots three to four times during the growing season (May, July, August and September) should confirm the growth pattern proposed here.

Variation in population size has been documented not only throughout the growing season but also from year to year. Monitoring of the Warder-Perkins square-meter plot over the past three years has revealed a general decrease in the number of rooted crowns, stolons and flowers (Table 4). Censuses of seven HCPD sites over the past five years have shown increases in population size at some sites and decreases in others (Table 5). Four of the HCPD populations have steadily increased in numbers while two are slowly declining in size and two have disappeared altogether. Due to the limited protection and variable sizes of the Ohio populations, censusing should continue in order to monitor population dynamics and to alert management agencies of deteriorating populations.

Based on the results of this study, several recommendations can be made for others involved in censusing and monitoring running buffalo clover. The first recommendation

involves the definition of a plant. Initially an effort was made to define a plant as a single rooted crown and all its rooted and unrooted connections. Counting plants in this manner proved to be difficult and time consuming. A more practical method of counting was established by defining a plant as a single rooted crown. For the purposes of comparing the size of populations this definition is sufficient when used consistently across populations.

This definition is not sufficient, however, when investigating the genetic size or diversity (i.e. the number of genetic individuals) of a population. Because of the clonal growth form of this plant, the number of rooted crowns in the population is not necessarily equal to the number of genetic individuals in the population. Information on the number of rooted crowns provides a measurement that is comparable among populations. Ohio populations have been measured in this way. Genetic diversity, however, is important when estimating the long-term stability of populations. For example, low genetic diversity may have many potential consequences for plant populations, including possible negative effects on long-term stability. Some information on the genetic diversity of populations of running buffalo clover is available (Hickey, Vincent and Guttman 1991) but much work remains to be done. In addition, little work has been done to investigate sexual reproduction in running buffalo clover, although seeds and seedlings have been found in Ohio populations.

This study has resulted in recommendations on not only how to count plants but also when to count plants. Population size fluctuates throughout the growing season and therefore, when censusing populations from year to year, counts should be made at the same time during the growing season. For example, counts made in May of one year and August the next may give false estimates on the increase or decrease of a population. Counts made in May or September of each year should give better estimates of true variation in population size. Any reports of population size should include how and when plants were counted.

Although this study begins to answer several questions concerning the life history of running buffalo clover, many other questions still remain unanswered. This study has established practical techniques for censusing and monitoring the plant. However, these techniques are only beginning to reveal the complex life history and population dynamics of the species. The limited data that have been collected here, however, provide important information for the recovery of the species. Recovery plan goals include knowledge of "population size, dynamics and other aspects of life history necessary for a population to be self-maintaining in perpetuity" (Bartgis 1989). Although preliminary, this study begins to provide some of this knowledge.

Because of abundant vegetative reproduction, it has been suggested that there may be a relatively small number of genets (genetically distinct individuals) in wild populations of running buffalo clover (Bartgis 1989 and Hickey and Vincent 1991). Data on genetic diversity and sexual reproduction will be critical elements in the recovery of the species. Genetic work, both electrophoresis and random amplified polymorphic DNA techniques, is being conducted for populations in all four states. This genetic work and the continued efforts to census and monitor populations will aid in providing the needed information to recover the species.

LITERATURE CITED

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Table 1. Location, size and owners of Ohio populations of running buffalo clover as of the 1993 census. (Note: the number of plants at Newberry Wildlife Sanctuary was approximated from a count of the original population in 1992 and a count of a new colony found in 1993.)

SITE	COUNTY	NUMBER OF PLANTS	DATE OF CENSUS	OWNER
1. Shawnee Lookout Park (Blue Jacket Trail)	Hamilton	0	May 1993	HCPD
2. Shawnee Lookout Park (Bobcat Ridge)	Hamilton	125	May 1993	HCPD
3. Shawnee Lookout Park (Cabin View)	Hamilton	97	May 1993	HCPD
4. Shawnee Lookout Park (Miami Fort)	Hamilton	500	Aug. 1993	HCPD
5. Shawnee Lookout Park (Trailside)	Hamilton	0	May 1993	HCPD
6. Niehaus Property	Hamilton	2500	May 1993	Private
7. Warder-Perkins State Nature Preserve	Hamilton	20	May 1993	ASO
8. Congress Green Cemetery	Hamilton	138	May 1993	OHS
9. Mitchell Memorial Forest	Hamilton	235	1992	HCPD
10. Newberry Wildlife Sanctuary	Hamilton	~182	Oct. 1993	HCPD
11. Miami Whitewater Forest	Hamilton	5	Sept. 1992	HCPD
12. Fankhauser Property	Clermont	104	May 1993	Private
13. Milford Area Historical Society Center*	Clermont	18	May 1993	MAHS
14. Hall's Creek Woods	Warren	0	June 1993	ODNR
15. Morrison Property	Warren	0	May 1993	Private

*Discovered in 1993 survey

OHS - Ohio Historical Society
HCPD - Hamilton County Park District
MAHS - Milford Area Historical Society
ASO - Audubon Society of Ohio

Table 2. Number of rooted crowns, stolons, and flowers in the permanent square-meter plots at Warder-Perkins and Niehaus in May and July of 1993 (NC=not counted).

SITE	# ROOTED CROWNS		# STOLONS		# FLOWERING STEMS
	MAY	JULY	MAY	JULY	MAY
Warder-Perkins	6	8	4	3	0
Niehaus	71	94	67	NC	6

Table 3. Number of rooted crowns at the Hamilton County Park District permanent square-meter plots in May, July/August and September from 1991-1993.

SITE	1991			1992			1993	
	MAY	AUGUST	SEPT.	MAY	JULY	SEPT.	MAY	AUGUST
Bobcat Ridge	21	41	38	35	48	49	40	6
Cabin View	61	228	201	203	207	140	50	0
Miami Fort	22	103	99	97	128	134	74	62

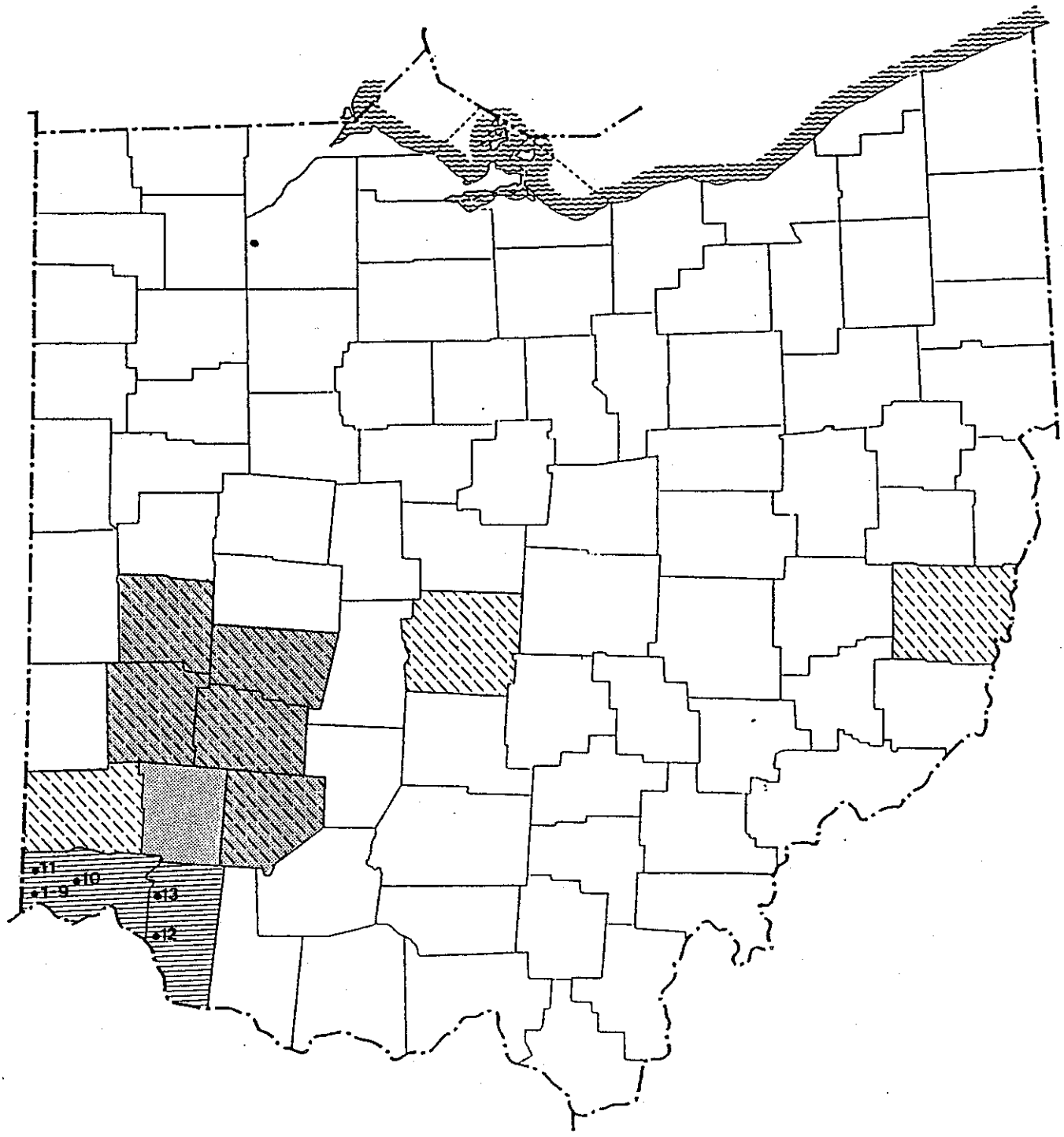
Table 4. Number of rooted crowns, stolons and flowering stems in the Warder-Perkins square-meter plot in May 1991, 1992 and 1993.

YEAR	# ROOTED CROWNS	# STOLONS	# FLOWERING STEMS
1991	13	36	1
1992	5	10	1
1993	6	4	0

Table 5. Number of plants and the number flowering plants at the Hamilton County Park District sites from 1989-1993. Number in upper right hand corner indicates month censused (NC = not counted, * = data not available).

SITE	1989		1990		1991		1992		1993	
	Plts.	Flws.	Plts.	Flws.	Plts.	Flws.	Plts.	Flws.	Plts.	Flws.
Miami Fort	103	6 [5]	104	27 [5]	188	76 [5]	427	111 [7]	500	263 [8]
Bobcat Ridge	82	16 [5]	109	20 [5]	115	NC [5]	115	NC *	125	NC [5]
Cabin View	NC	NC	148	NC [6]	246	NC [5]	246	NC *	97	NC [5]
Trailside	9	2 [5]	2	0 [5]	0	0 [5]	0	0 [5]	0	0 [5]
Blue Jacket	12	0 [5]	15	0 [5]	0	0 [5]	0	0 *	0	0 *
Mitchell Memorial Forest	135	1 [8]	168	43 [5]	198	12 [5]	235	NC *	NC	NC
Miami Whitewater Forest	NC	NC	10	2 [5]	11	2 [5]	5	NC [9]	2	NC [6]
Newberry	NC	NC	14	1 [5]	35	2 [5]	162	26 [5]	182	NC [10]

Figure 1. Ohio counties with known or historic populations of running buffalo clover, and counties which were surveyed in 1993. Numbers correspond to populations listed in table 1.

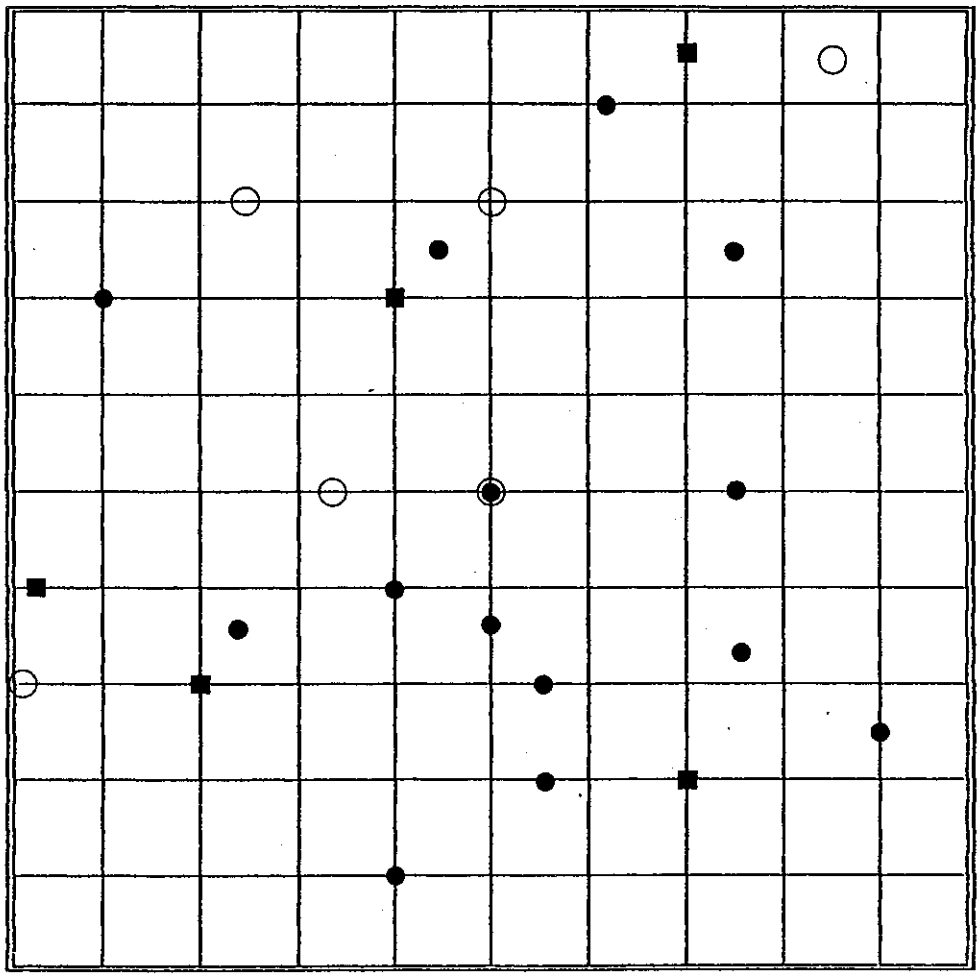


Counties with known populations

Counties with historic populations

Counties surveyed

Figure 2. Location of rooted crowns in the Warder-Perkins square-meter plot in May of 1991, 1992 and 1993.

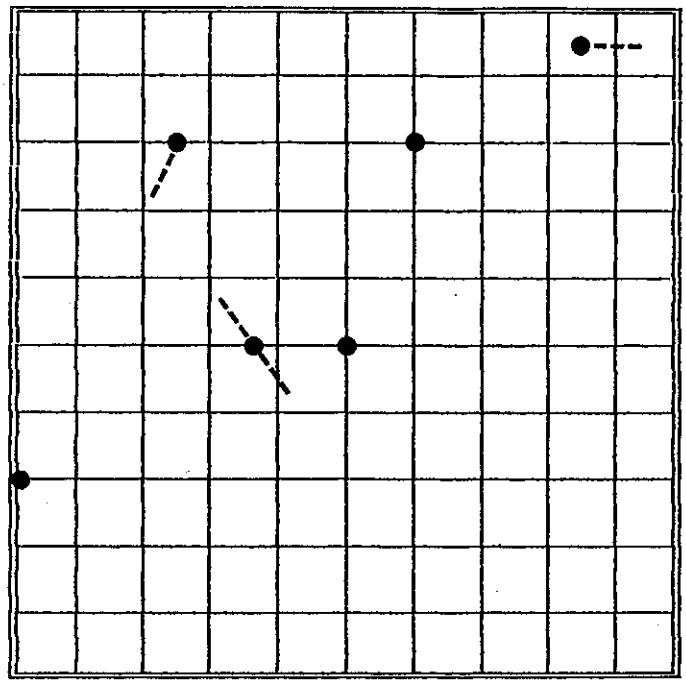


● 1991

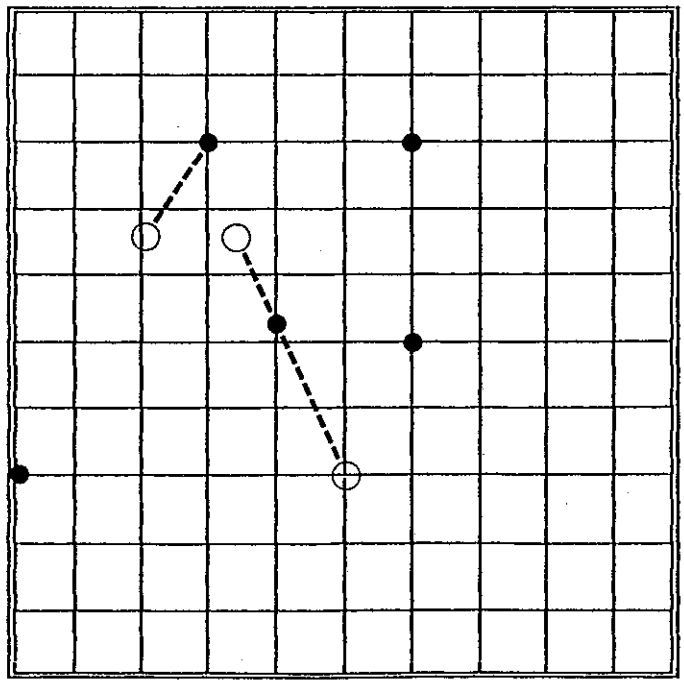
■ 1992

○ 1993

Figure 3. Location of rooted crowns in the Warder-Perkins square-meter plot in May and July 1993.



MAY



JULY

● "parent crown"

---- stolon

○ "daughter crown"