Distribution, Habitat, and Reproductive Biology of *Phlox oklahomensis* Wherry (Polemoniaceae)

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Although initially considered a candidate for federal designation as a threatened plant species, three years of observations indicate that *Phlox oklahomensis* is not threatened and its current designation as a Category 3C species is appropriate. *P. oklahomensis* is endemic to tallgrass and midgrass prairies of Butler, Chautauqua, Comanche, Cowley, and Elk counties of Kansas, and Woods and Woodward counties of Oklahoma. The species comprises populations of a few scattered individuals to several hundred plants occurring on Tertiary deposits in western Oklahoma and adjacent Kansas, and limestone deposits in the Flint Hills of south-central Kansas. Plants are primarily autogamous and seed are dispersed by an explosive fruit mechanism. Seed dormancy is induced by high temperatures and broken by moist, cold temperatures. Field observations suggest that *P. oklahomensis* is not threatened in northwestern Oklahoma and adjacent Kansas; however, populations may be declining in the southern Flint Hills of Kansas due to the current range management practice of spring burning. Future monitoring is appropriate.

INTRODUCTION

Phlox oklahomensis, a perennial herb, was first described by E. T. Wherry in 1944 (1) and initially was known only from Woods and Woodward counties in Oklahoma. Later, it was reported in Kansas (2,3) and in Texas (4). Reports of *P. oklahomensis* occurring in northwestern Arkansas (5) were later shown to be incorrect in a biosystematic study of the relationship between *P. oklahomensis* and *P. bifida* (6). Because of its restricted geographical distribution, *P. oklahomensis* was designated a threatened plant species by Ayensu and DeFilipps (7). While this listing did not constitute an official or legal designation, it indicated a need for additional information about the taxon. Although field surveys and status evaluations had been conducted (8-10), a detailed study of the taxon's biology seemed appropriate. The objectives of this investigation were to: 1) determine the extent of its geographic range; 2) describe its habitat; and 3) describe its reproductive biology.

MATERIALS AND METHODS

Field studies were initiated in 1980 with observations of the species' distribution and habitats. Detailed investigations of selected populations were conducted during the 1981 and 1982 growing seasons and included observations of habitat, vegetative and floral morphology, and reproductive biology. The last observation encompassed a determination of chromosome number, pollen size, pollen viability, phenology, reproductive system, seed size, and requirements for seed germination.

The geographical range of *P. oklahomensis* was determined by field observations and examination of herbarium specimens deposited at F, GH, KANU, KSC, LL, OKL, OKLA, PH, NY and TEX. Aspects of the habitat and phenological patterns of the plants were observed as populations were encountered. Soil nitrates and pH were determined by the Oklahoma State University Soil Testing Laboratory.

Chromosome numbers were obtained from root tips of germinating seed. Pre-treatments and fixation methods were modifications of those used by Meyer (11). Pollen viability (stainability) and diameter were determined from plants at two sites on April 5 and 18, 1982. At each site on each date, intact flowers from five plants were collected. Pollen grains were teased from the anthers onto a watchglass and dispersed in deionized water with the aid of a Coulter Type II-A cationic dispersing agent. Pollen samples

were counted and sized with a Coulter counter model Z_{BI} and Coulter channelyzer unit which were calibrated using a 22.5-μm reference particle (12). Approximately forty thousand grains per population and date were observed.

Phlox oklahomensis' capacity for autogamy, allogamy, and agamospermy was tested using standard techniques (13). Because of its floral morphology, flowers were not tested for anemophily.

Seed size and seed-size distribution were determined for samples from two sites. Seed were separated by density into five seed-size classes with a South Dakota seed blower model B using air valve openings of 25, 30, 35, 40, and 45 degrees.

Experiments to determine requirements for germination were conducted on seed of the 35-degree seed class harvested in 1981. Seed were germinated in a 20 °C constant dark environment on a substrate moistened with either distilled water or 0.2% KNO_3 solution. The latter has been found to enhance germination of dormant seed in some *Phlox* species (14); however, no significant enhancement of germination in *P. oklahomensis* was observed. Data obtained with KNO_3, therefore, are omitted in Results and Discussion. One experiment involved drying (heat treatment) seed at 65 °C for 0, 8, 24, 48, or 96 hr. A second experiment consisted of subjecting seed to a moist prechill environment at 0 - 5 °C on a substrate moistened with distilled water for 0, 1, 2, or 4 weeks prior to germination at 20 °C. In addition, studies were conducted to determine the relationship of seed size to germination. This study utilized four seed classes, a control (bulk seed not separated into size classes), and the 30-, 35-, and 40- degree seed classes. Seed collected from both field sites in 1981 and 1982 were studied. The germination conditions were the same as in the preliminary studies except that prior to germination, seed were subjected to a moist prechill at 0-5 °C for two weeks in distilled water. All germination experiments were conducted as a randomized complete block design with four replications of each treatment.

RESULTS AND DISCUSSION

Geographic Distribution

Phlox oklahomensis is restricted to Oklahoma and Kansas. In northwestern Oklahoma, populations occur in Woods and Woodward counties. They are also found in adjacent Comanche County, Kansas. Populations also occur in the southern Flint Hills of Kansas (Butler, Chautauqua, Cowley, and Elk counties). Prior to this investigation, the species was represented by approximately 40 herbarium sheets. This paucity of specimens indicates a lack of early-spring collecting in the region rather than rarity of occurrence. Numerous populations were discovered in census areas (12), but plants were not found in adjoining counties with similar habitats. Wherry (5) suggested that the populations of the Flint Hills region were once continuous with those in northwestern Oklahoma, but that farming, ranching, and improper land management had eliminated the intervening populations.

Phlox oklahomensis was reported also to occur in Dallas County, Texas, a report based on one herbarium specimen (4). Subsequent examination of the specimen revealed that the plant is *P. bifida* var. *induta* Shinners, not *P. oklahomensis* (15).

Habitat

Phlox oklahomensis is adapted to tallgrass and midgrass prairies and populations thrive in areas of low to moderate grazing. In open areas on gently rolling hills, it is associated primarily with *Andropogon gerardii, Schizachyrium scoparium, Bouteloua curtipendula, B. gracilis*, and *B. hirsuta*. In canyons and drainage basins, it is associated with these grasses and *Juniperus virginiana, Prunus angustifolia, Ulmus americana*, and *Yucca glauca*. Other species occasionally encountered with *P. oklahomensis* compose the late winter and early spring flora of the area and are cited in Bruner (16). In late winter and early spring, when *P. oklahomensis* is actively growing, flowering, and producing seed, most of the native plants of the area are dormant.

Throughout this *Phlox*'s range in northwestern Oklahoma and adjacent Kansas, the land is dissected by many canyons and drainage basins. Populations occur both on the gently rolling uplands (0-15% slopes) and on steeper slopes of the canyons. Plants were observed on all exposures, but were more abundant on the cooler north-facing slopes. Similar habitats occur in the southern Flint Hills region of Kansas; however, in this region the slopes are interrupted by limestone outcrops. In the Flint Hills, populations generally occur near these outcrops. The restriction of plants to such outcrop areas possibly is related to the increasing frequency of prescribed spring burning. Fire intensity is lower around the outcrops because there is less vegetation. Burning usually occurs in April when *P. oklahomensis* is flowering most profusely; thus, yearly burning interferes with vegetative growth and seed production. In northwestern Oklahoma and adjacent Kansas, burning is not routinely used as a range management practice.

Throughout its geographic range, *P. oklahomensis* occurs on well-drained grassland soils that have weathered from calcareous shales. Although plants were observed growing in clay and silt loam soils, they appeared to be more common in very fine sandy loams. These soils were low in nitrates and had a pH near neutral.

The taxon's association with specific geologic formations is quite apparent. In northwestern Oklahoma and adjacent Kansas, the distribution of *P. oklahomensis* closely coincides with that of Tertiary deposits (Ogallala formation). Plants also occur on exposed Rush Springs and Marlow formations directly below the Ogallala (17). The Ogallala formation is characterized by sandstone and siltstone, the Rush Springs formation by sandstone, shale, gypsum, and dolomite, and the Marlow formation by sandstone. In the Flint Hills region of Kansas, *P. oklahomensis* is associated with limestone formations of the Chase and Council Grove Groups (18).

Reproductive Biology

Flower bud formation in *P. oklahomensis* begins in late February and continues through March. The three or four flower buds produced by each fertile shoot are protected from the winter environment by appressed bracts and leaves. Flowering begins in late March and continues through May. As the flower opens, the contorted corolla lobes untwist and separate until the lobes are fully extended. Flowers usually open at night and once open remain open. The anthers dehisce longitudinally in the morning after the corolla opens. Capsules mature three to four weeks after fertilization. Upon drying, the capsule quickly separates along three sutures and catapults its seed outward, occasionally up to 2 m. This phenomenon has also been reported for *P.drummondii* (19). As the capsule splits, the sound is similar to that produced by a kernel of popcorn popping.

Intra-populational variation in the size, shape, and color of the corolla lobes was observed. Some lobes were one-half the normal length of 7.4 - 8.1 mm, or were pointed rather than rounded. The corolla was typically white; however, shades of blue and lavender were observed. The pair of deep-hued striae reported to occur occasionally at the lobe bases was not seen (5). Insect visitation is rare. During three years of day and night field observations, only one insect was observed visiting *P. oklahomensis.* At approximately 1500 hours CST, a clear-winged sphinx moth, *Hemaris diffinis* (Boisduval), was observed. It was found to be carrying pollen of *P. oklahomensis.* The lack of insect visitation is possibly due to unfavorable temperature. The mildness of spring days and considerable coolness of nights are not conducive to insect activity.

All plants examined produced pollen in abundance. The grains are spherical, 18-33 μm in diameter, and are sculptured with echinate spines. The populations examined exhibited similar distributions of pollen size (Fig. 1). Slight differences in pollen size were found in each population over time, with larger pollen produced later in the season. Viable (stained) pollen averaged 81.3 percent.

Fruit and seed set indicate that *P. oklahomensis* is both autogamous and allogamous (Table 1). Natural fruit set by plants not

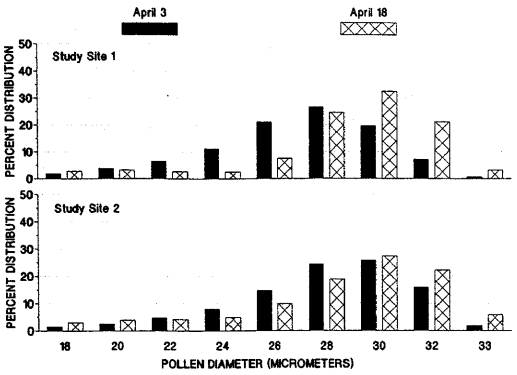

FIGURE 1. Percent pollen-size distribution of *Phlox oklahomensis* on April 3 and 18, 1982.

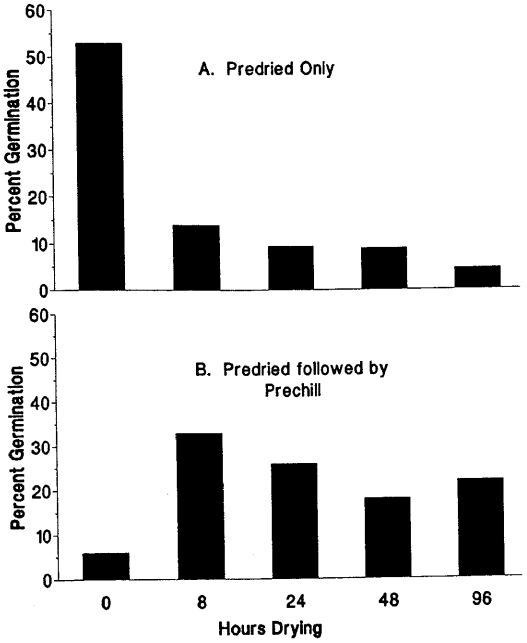

FIGURE 2. Effect of drying on seed germination of *Phlox oklahomensis*. A. Percent germination of dried seed. B. Percent germination of seed dried and then subjected to a moist prechill.

manipulated or caged was 81.4% and seed set averaged 4.3 seed per capsule. Plants artificially self-pollinated exhibited 37.5% fruit set and averaged 4.1 seed per capsule. Fruits and seed failed to form when plants were caged and flowers left undisturbed. The insect-exclusion cages are believed to have reduced wind velocity necessary for flower agitation to induce self-pollination. The anthers are positioned at three levels within the narrow-throated corolla tube. At anthesis, the lowest anther is approximately 1.5 to 2 mm above the stigmas and when the flower is disturbed, pollen is shed onto the stigmas. Agitation of flowers by wind or insect visitors to effect selfing has been described in other taxa (20). Manually cross-pollinated plants, in the test for allogamy, exhibited 43.5% fruit set and averaged 2.3 seed per capsule. Plants tested for agamospermy did not set fruit. Counts of root-tip chromosomes revealed a mitotic number of 14, which is characteristic of the genus (21).

Seed sizes and distribution percentages of seed classes are given in Table 2. *P. oklahomensis* seed weight ranges from 50 to 150 mg per 100 seed. The percent distribution by seed-size class was highly variable for sites

TABLE 1. Fruit and seed set in reproductive systems studied for *Phlox oklahomensis*.

Reproductive mode tested	Flowers tested (no.)	Fruits recovered (no.)	Fruit set (%)	Seed set per fruit set (avg. no.)
Control (open pollination)	27	22	81.4	4.3 (1–10)
Autogamy				
Natural (caged)	30	0	0.0	–
Artificial (hand-pollinated)	24	9	37.5	4.1 (1–6)
Allogamy (hand-pollinated)	23	10	43.5	2.3 (1–6)
Agamospermy	24	0	0.0	–

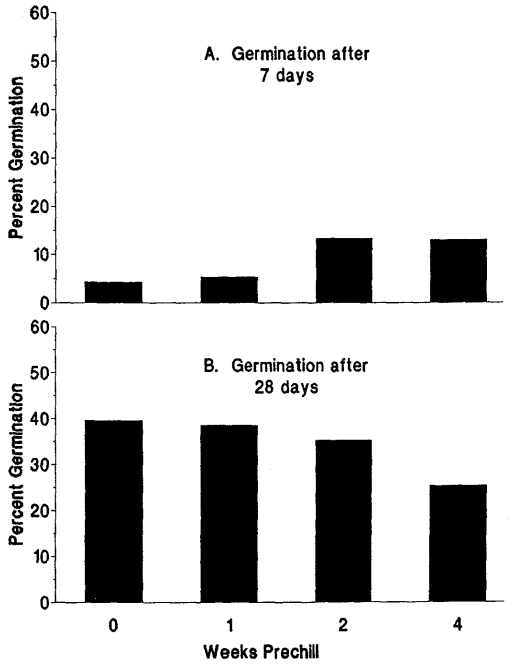

FIGURE 3. Effects of moist prechill on seed germination of *Phlox oklahomensis*. A. Percent germination after 7 days. B. Percent germination after 28 days.

and years, and displays a unimodal distribution.

Seed of *P. oklahomensis* showed a significant ($P < 0.05$) decrease in germination following drying treatments at 65 °C (Fig. 2A). At the end of the 28-day germination period, the remaining ungerminated seed of the previous treatment were subjected to a 2-week moist prechill at 0-5 °C, and placed at 20 °C in a constant dark environment. After 7 days, a significant increase in germination was observed for all but the zero-hour drying treatment (Fig. 2B). Approximately fifty percent of the seed in this treatment had previously germinated prior to subjecting them to the moist prechill. The maximum observed germination in any test of *P. oklahomensis* was 60%. The remaining ungerminated seed in this treatment are possibly 'hard' seed. Hard seed have a thickened seedcoat and given enough time will soften, imbibe water, and germinate. These results suggest that the longer the drying the greater the seed dormancy. In a separate germination experiment, a significant ($P < 0.05$) increase in germination after 7 days occurred following a 2- and 4-week moist prechill at 0-5 °C (Fig. 3A). At the end of the 28-day germination period, seed subjected to the 4-week moist prechill showed a significantly lower total germination (Fig. 3B). These data suggest that *P. oklahomensis* requires a minimum of 2 weeks moist prechill to promote early seed germination, and that extended moist cold conditions reduce seed germination. Similar germination results have been reported for seed of *Gilia capitata* (22). Climatic conditions in northwestern Oklahoma are seasonal with respect to temperature and precipitation. The average January temperature is 2 °C and the average July temperature is

TABLE 2. Seed size and distribution percentages (based on seed numbers) of *Phlox oklahomensis* by seed class for sites and years.

Seed class	Range (mg/100 seed)	Distribution			
		1981		1982	
		Site 1 (%)	Site 2 (%)	Site 1 (%)	Site 2 (%)
25	49.2–62.9	4.46	3.71	32.59	12.84
30	69.0–77.7	24.08	15.64	34.19	27.21
35	92.2–96.3	47.48	42.21	23.25	35.39
40	114.0–116.5	22.86	36.11	8.75	33.33
45	137.0–152.2	1.18	2.33	1.22	3.14
Bulk Lot (Control)	73.1–93.3				

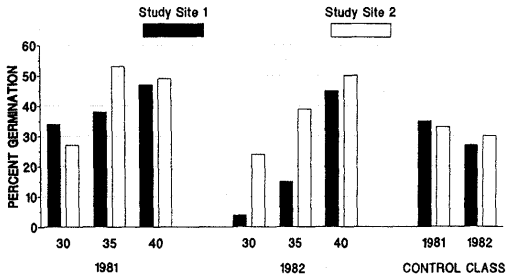

FIGURE 4. Effects of seed size on germination of *Phlox oklahomensis*.

28 °C. Extreme temperatures from a low of -27 °C to a high of 49 °C have occurred in the region. The mean average precipitation is 72.1 cm. Approximately 37% of the precipitation occurs during the spring (April-June), 32% during the summer (July -September), 18% during the fall (October -December), and 13% during the winter (January -March) (23).

The studies to determine how germination is influenced by seed size revealed a significant three-factor interaction between sites, years, and seed classes (Fig. 4). Germination differences among seed-size classes were highly significant ($P < 0.01$). The trend among seed classes is that larger seed germinate better than smaller. In general, germination of 40-degree class seed is twice that of the 30-degree class. Although germination differences were significant between sites, germination of the control seed classes showed no differences. Germination in the control class, combined over years, showed 31.0 and 31.5% for sites 1 and 2, respectively. Seed dormancy and age of seed at the time of germination may account for the differences found between years. The data suggest that a 2 week moist prechill prior to germination was not sufficient to break seed dormancy for seed harvested in 1982. In 1981 and 1982, germination at the end of a 21-day period was 39.5 and 29.3%, respectively. The hard seed remaining at the end of this same period was 36.9 and 49.6% for 1981 and 1982, respectively. Thus, the theoretical total viable seed, i.e., actual germination plus hard seed remaining, was 76.4 and 78.9% for 1981 and 1982, respectively.

These tests suggest that *P. oklahomensis* has evolved with respect to its environment and seed dormancy induced by hot dry temperatures can be broken by a moist prechill. Also, it appears that the seed produced will not germinate completely from year to year, so that seed remains in the soil's seed bank.

Status

Although initially considered a candidate for designation as a threatened species in 1978 by Ayensu and DeFilipps (7) under the guidelines of the 1973 Threatened and Endangered Species Act (PL-93-205), the studies reported above, based on three years of field observations, indicate that *P. oklahomensis* is neither threatened nor endangered. Its current Federal designation as a Category 3C species (not recommended for listing or more abundant or widespread than previously believed, 1980 FR 45:82557) is appropriate. Populations, ranging in size from a few scattered individuals to several hundred, are stable and comprise reproductively immature and mature plants. Present land use practices in northwestern Oklahoma and adjacent Kansas do not threaten the existence of the species. In the Flint Hills of Kansas, it is suspected that populations are declining due to the current practice of spring burning, but future monitoring will be necessary to detect any decline.

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