APPROACHES TO WILDFLOWER MEADOW ESTABLISHMENT

by

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ABSTRACT

Several aspects of wildflower meadow establishment using native Virginia herbaceous perennial species were investigated. The initial task lay in identifying the truly native species from those European introductions which have successfully naturalized in the state. Germination testing was conducted in accordance with procedures deemed feasible for use by the Virginia Department of Transportation. Direct seeding in the field, either in spring or fall, failed to produce acceptable levels of emergent plants. Under two treatments conducted in the greenhouse many species germinated well.

Weed control is the most critical factor in successful wildflower meadow establishment. Dacthal®, Pennant®, and Treflan® all suppressed germination and/or retarded development of at least some of the five species tested. Use of these three herbicides is not recommended in conjunction with seeding of Virginia wildflowers.

Direct seeding a nurse crop of non-persistent species in the spring which would be followed by installation of the native mix in the fall was investigated. Soil analysis

and subsequent development of the Virginia wildflower mix failed to differentiate any soil quality influence of the nurse crop as compared to the other two treatments.

Direct seeding was compared to the use of the Virginia Tech Transplanted Meadow (VTTM) technique developed by Lyons for fall establishment. High levels of winter survival, strong competitive ability against weeds, and good spring growth by the wildflower species indicate that the VTTM technique is a promising method for establishing wildflower meadows. Further research is needed to determine the long term performance of a native Virginia wildflower meadow.

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I am grateful for the encouragement and support of John Hinton, who has held down the fort over the years while I have been at Tech.

DEDICATION

For my parents, who gave up their dreams so that mine might come true.

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Approaches to Wildflower Meadow Establishment: An Overview

Wildflower gardening is popular among Americans ranging from rural homeowners seeking to attract wildlife to urban communities in the Bronx, New York (Martin, 1986). Meadows provide an alternative to lawns for large areas of land (Art, 1987). Apart from planted meadow sites, " the use of native plants in American gardens has increased dramatically" (Phillips, 1985, p.3). During the colonization of the American continent by Europeans, native plants were important to settlers for food and medicinal reasons as well as for their ornamental value (Martin, 1986). Now, the emphasis on conserving native plants is directed more towards preserving the habitat of other creatures that depend on specific types of vegetation (Stevenson, 1985). Prior to European settlement, the Northeast region of the United States was dominated by deciduous forests; even so, these were punctuated by openings which contained eastern meadow species adapted to full sunlight and periodic drought stress (Art, 1987).

Much conflicting information exists in the literature with regard to the definitions of terms used in the discussion of meadow establishment. Confusion has particularly arisen over the use of the word "wildflower." In its very broadest sense, a wildflower is one which exists somewhere in the world having arisen without human intervention. The National Wildflower Research Center defines wildflowers in a narrower sense: "Flowering plants, native to a specific geographical area or habitat, capable of growing in unimproved habitats without the assistance of humans...Wildflowers can include naturalized species...not aggressively competitive or invasive" (Paulson, 1989, p.3). Still more restrictive is the definition that a wildflower is a native, herbaceous flowering plant which may also include grasses and many aquatic plants (Jones and Foote, 1990).

"Meadow" is another term assuming various context-related meanings. Colloquially, as "meadow-in-a-can," the term generally refers to a mix of colorful annual and perennial, native and exotic flowers (Bartels, 1992). A true meadow, i.e. one that could have occurred in nature without human intervention, is composed primarily of grasses interspersed with flowers whether it arises in the Northeast, where it is frequently temporary, or the Midwest prairies (Bartels, 1992; Jones and Foote, 1990). A "prairie" is simply the French language equivalent of meadow, as French explorers were the first to describe the vegetation found in the central plains of the United States (Smith and Smith, 1980).

The American movement for the preservation of wildflowers along roadways began in Texas through the work of garden clubs in the 1930s (Feltwell, 1992). The Highway Beautification Act of 1965 was the legislation which provided the spark for the development of large-scale public projects using native wildflowers along the nation's roadsides (Gould, 1988). Virginia currently receives approximately $75 million in Federal Highway funds which are allocated for Surface Transportation Projects; of this amount 10% is targeted for enhancement categories, one of which is landscaping uses (Cassada, pers. comm., 1995). Much of the impetus in using American species for highway beautification originated with Ladybird Johnson (Paulson, 1989). On the occasion of the former First Lady's seventieth birthday in 1982, Mrs. Johnson donated 60 acres and $125,000 for the establishment of the National Wildflower Research Center [NWRC] in Austin, TX (Gould, 1988). As a consequence, species most common in early wildflower mixes were those native to the southwestern portions of the United States. *Lupinus texensis* (Texas bluebonnets), *Coreopsis tinctoria* (plains coreopsis), and *Castilleja indivisa* (Texas paintbrush) are a few of the species which perform well in the Southwest but will not become established in Virginia (Martin, 1986). Bonnie Harper-Lore, a landscape architect with the Federal Highway Administration "commented on the progress (or lack thereof) that has been made in the use of wildflowers along highways since the effort was launched in the 1970s", citing the substitution of exotic species such as *Cosmos bipinnatus, C. sulphureus*, and *Cyanus centaurea* for those species which are truly native (Sorrells, 1995, p.3).

Unlike most of their counterparts, some seed companies that specify wildflower mixes by region, as in Park's Wildflower Mix No. 5 for the Southeastern U. S., do not list the species composition of the mix (Park Seed, 1995). Johnny's Catalogue (Johnny's Selected Seeds, 1995) offers two wildflower mixes; one is composed of annuals and the other of perennial species. Both incorporate naturalized species such as *Hesperis matronalis* (Dame's rocket) and *Centaurea cyanus* (bachelor's buttons), along with many other species not native to Virginia. Of the 18 species included in Applewood Seed Company's Northeastern mixture used for a research project in Virginia (Sabre, 1994), only five are truly native in the state (Harvill et al., 1992).

What are some reasons for using native species? Mixes are often formulated for such broad geographic ranges that many species may not be adapted for the specific sites where they are planted (Art, 1987). Where seeded species fail to become established, the outcome will be the entry of invading weeds among the desired species that were successful (Sabre, 1994), leading to a planting characterized as a "midsummer weed patch" by Lyons (Laurent, 1994, p.18). Other research has demonstrated that reliable bloom and seed set are best achieved by locally adapted species and ecotypes (Beckwith, 1991; Jones and Foote, 1990). Introduced species such as *Chrysanthemum maximum, Cosmos bipinnatus*, and *Daucus carota* were found to be invasive when combined with other species in a mix during research trials in Georgia (Corley and Smith, 1990). Other invasive species found in seed mixes tested by the New England Wildflower Society included *Hesperis matronalis, Cichorium intybus*, and *Lythrum salicaria* (Art, 1987). Research conducted by Lindgren and Clay (1993) demonstrated that even supposedly sterile cultivars of purple loosestrife (*L. salicaria*) are interfertile with the wild type which has displaced numerous wetland species and continues to endanger ecosystem diversity. Several states, including Minnesota, Illinois, Indiana, Ohio, Washington, and Wisconsin, have listed purple loosestrife as a noxious weed (Anderson and Ascher, 1993); therefore, its continued inclusion in wildflower mixes must be deemed unconscionable and irresponsible. Native wildflowers by definition are adapted to the area in which they originate. While highway medians often possess severely damaged and/or non-native soils, the Virginia wildflowers will be adapted to local rainfall, temperature, and photoperiod conditions. Whereas exotic annual plantings require reseeding each year in order to maintain visual impact (Bartels, 1992; Jones and Foote, 1990), a meadow composed of Virginia natives is expected to be self-perpetuating.

Identifying native Virginia species was the first step in formulating a natives-only seed mix to be planted along roadsides. Primary references used in this research were *Wildflowers of the Shenandoah Valley and Blue Ridge Mountains* (Gupton and Swope, 1979), *A Field Guide to Wildflowers: Northeastern/Northcentral North America* (Peterson and McKenny, 1968), *Guide to the Vascular Plants of the Blue Ridge* (Wofford, 1989), and *Atlas of the Virginia Flora III* (Harvill et al., 1992). The majority of "ornamental" herbaceous Virginia natives are perennial; thus, no annual species were selected for inclusion in trial mixes.

The primary consideration in selecting species for inclusion in a native wildflower mix was to provide for a maximum period of continuous bloom. Height was considered such that early blooming species benefitting from some shading would be planted with taller, later flowering species. This strategy would additionally serve to disguise weather-beaten foliage and seed pods of spring blooming species. Whenever possible, canopy bloom heights were staggered to yield a greater depth of color. Species were also selected on the basis of family diversity, primarily to minimize competition for nutrients and maximize resistance to any one pest or pathogen. Monoculture plantings have been blamed for increasing plant losses to insect and disease invasion (Peirce, 1987). Color harmony within each mix was considered despite its subjective nature. Accordingly, this was given the least weight in the selection process.

Other selection criteria included tolerance for full sun, drought, low soil fertility, heavy-textured soil, and visibility at interstate highway speeds. The species selected also occurred throughout Virginia or over large areas encompassing the geographic subdivisions of Tidewater, Piedmont, and Mountain regions.

Commercial seed sources for each species selected were located. Earlier research indicated that seed produced at similar latitudes to the area in which the species were to be grown would be more reliable with regard to hardiness, blooming, and reproductive success (Beckwith, 1991; Jones and Foote, 1990; Martin, 1986). Of the 25 species initially chosen for this thesis research, 14 proved to be commercially unavailable from 46 supply sources. After further research, 14 suitable species were obtained from Environmental Seed Producers (CA), Lafayette Home Nursery, Inc. (IL), and Prairie Moon Nursery (MN) (Appendix I). *Asclepias tuberosa* was donated by Ms. Nicky Staunton of the Virginia Native Plant Society.

Germination characteristics for the selected species were compiled from the literature. Information regarding germination rates and procedures for certain species was not available from the sources consulted (Association of Official Seed Analysts, 1989; Deno, 1993; Hartmann and Kester, 1983; Jones and Foote, 1990; Martin, 1986; Paulson, 1989; Phillips, 1985; Smith and Smith, 1980).

Recent research has identified pretreatment strategies for some wildflower species. Stratification improves germinability of several species (Bratcher et al., 1993; Martin, 1986). Growth regulators and storage temperature were found to control germination response for *Coreopsis lanceolata* (Carpenter and Ostmark, 1992). Samfield et al. (1991) focused on seed priming as a means of improving uniformity of germination for selected wildflower species. Conditioning techniques may well prove beneficial to commercial producers of wildflower seed; however, the present study was intended to develop pragmatic procedures useful to the Virginia Department of Transportation (VDOT) in the establishment of roadside plantings. For this reason, germination trials were conducted without any chemical conditioning treatment or particular stratification regimes.

Light and temperature cycles favoring germination of many native herbaceous perennials have been reported by Deno (1993). "Temperature is, perhaps, the most important environmental factor that regulates germination and controls subsequent seedling growth" (Hartmann and Kester, 1983, p.146). "It appears that seeds of many wild species or those with a short domestication history have a relative dormancy that is responsive to temperature alternation" (Gardner et al., 1985, p.226). For this reason, temperature was allowed to fluctuate diurnally during the germination testing of the selected wildflower species in this experiment. As early as 1926, Kinzel classified several hundred species on the basis of light response; i. e., whether germination was favored by light, by dark, or was unaffected by light and dark (Gardner et al., 1985). Where information was available, the light/dark requirements of selected wildflower species were provided for in this experiment. As *Teucrium canadense* (germander) was the only species in this work postulated to require dark for germination (Deno, 1993), seven other species with no documented photoperiod germination requirements were also subjected to dark treatment to examine their suitability for inclusion in a mix with germander.

The next phase for the present research was to investigate the use of pre-emergence herbicides to inhibit weed competition and enhance the success of direct-seeded wildflowers. Weed control in wildflower plantings is the most critical factor in determining successful establishment (Laurent, 1994). A primary source for weed emergence in landscape plantings is the reservoir of dormant weed seeds already present in the soil known as the "seed bank" (Gallitano et al., 1993). The persistence and ubiquity of weeds in disturbed soils is due to the ability of weeds to produce numerous and long-lived seeds which are easily transported (Ross and Lembi, 1985). The key in weed management is to prevent one or two aggressive species from gaining dominance in a wildflower planting (Martin, 1986). The longevity of weed seeds, in combination with the sheer numbers produced, guarantees that a given weed will continue to persist almost indefinitely once established (Ross and Lembi, 1985).

Several strategies may be used in an effort to control weed pressures. Fumigants such as methyl bromide are highly effective soil sterilants when applied prior to installation of wildflowers (Derr, 1994); however, severe toxicity, great expense, and labor intensiveness are several disadvantages of these compounds (Ross and Lembi, 1985). Instead, site preparation for meadow plantings frequently combines tillage with preplant, nonselective, contact or systemic herbicide treatment (Corley and Smith, 1990; Laurent, 1994). Two methods in common use exemplify this approach: the site may be treated with a systemic, nonselective herbicide such as glyphosate; the bed is then rototilled approximately two weeks later to expose buried weed seeds; finally, regrowth is treated again with herbicide and not tilled (Laurent, 1994). Alternatively, the site may be mowed initially prior to application of a nonselective contact herbicide such as paraquat (Ross and Lembi, 1985) after regrowth; this is followed by tillage of the area to various depths (Corley and Smith, 1990). As continued weed control is often either necessary or desired during the establishment phase of the wildflower planting, some research has been conducted on the response of wildflowers to a variety of pre-emergence and post-emergence herbicides (Corley and Smith, 1990; Derr, 1993, 1994).

Corley and Smith (1990) first determined that deeper tillage resulted in greater weed control problems; thus, tilling shallowly following pre-plant herbicide treatment is recommended. The graminicides Poast® (sethoxydim) and Fusilade® (fluazifop) were found to control grassy weeds in wildflower plantings with no damage to desirable species; Surflan® (oryzalin), a pre-emergence herbicide, resulted in some depression of wildflower survival; Treflan® 10G (trifluralin) resulted in minimal death to desirable species (Corley and Smith, 1990). Broadleaf weeds were particularly difficult to control, requiring hand-pulling, spot spraying, or wicking with a contact herbicide (Corley and Smith, 1990).

The experiments conducted by Derr (1993, 1994) on transplanted wildflowers focused on the use of pre-emergence herbicides in container production. In 1993, Derr reported that metolachlor was not found to affect survival of *Coreopsis lanceolata* while it did provide acceptable control of *Cyperus esculentus* (yellow nutsedge). In 1994, *Rudbeckia hirta* and *Coreopsis lanceolata*, two Virginia native wildflowers, were largely unaffected by DCPA (Dacthal®) four weeks after herbicide application. Both species did demonstrate damage from dinitroaniline herbicides, with the greatest injury resulting from oryzalin (Surflan®) application. Three months after herbicide application, however, neither of these species showed significant differences in survival from the control.

The impact of EPTC/Treflan pre-emergence herbicide combination on direct-seeded wildflowers was investigated by Erusha et al. (1991). Among the wildflower species included in the study were *Coreopsis lanceolata* and *Rudbeckia hirta*. The herbicide combination had no impact on stand establishment for *R. hirta*. *Coreopsis lanceolata* did not differ from the control at the Nebraska location; however, stand establishment in Oregon was only 35% for the control, while the herbicide treated plot demonstrated an 80% establishment. No other species of the 47 tested in this experiment had better stand establishment for herbicide treated plots compared to the controls, and no hypothesis for the anomaly was provided by the authors.

Another objective of the current study was to determine whether a nurse crop might be a viable method of site preparation. A nurse crop has been defined as a fast growing crop planted with the intended crop, but which does not persist within the crop for a long period of time (Ross and Lembi, 1985). A slight alteration of this idea was used in the present study, where the nurse crop was to be planted ahead of the intended crop. The purpose was to investigate whether a spring-seeded nurse crop could improve soil conditions for a subsequent wildflower planting. By selecting species for the nurse crop with certain properties, some scientifically documented and others not, it was hypothesized that soil structure and nutrient levels might be enhanced to facilitate autumn establishment of a wildflower planting. In addition to providing tangible benefits, the nurse crop would also display a high level of ornamental value, as it would need to comply with VDOT's mandate of highway beautification. Finally, nonpersistent species must comprise the nurse crop such that no competition with the wildflower crop would occur.

The final phase of this research project was to determine a method for the field establishment of native Virginia wildflowers which would be appropriate for use by VDOT or, by extension, other groups interested in large-scale meadow installation. Field establishment of extensive wildflower meadows is analogous to the production of agronomic stands in many respects. Horticulture is ordinarily concerned with the intensive management of relatively high value crops, whereas agronomy is usually directed towards the large-scale production of crops such as hay, corn, wheat, and soybeans. One measure of successful establishment is leaf area index (LAI), which is a ratio of the area of photosynthesizing surface divided by the soil area occupied by the crop (Mengel and Kirkby, 1987). Broadleaved crops are said to be maximizing available solar irradiation when LAI values range between 3.0 and 5.0; that is, maximum dry matter production occurs at this level of stand density (Gardner et al., 1985). The species used in the present study all possess leaves which are oriented in a horizontal plane; thus, a minimum LAI value of 3.0 was a criterion for successful stand establishment.

Failure to obtain vigorous stands from direct-seeded methods may result from competition by more aggressive weed species, improper seeding times, and/or an inability to break dormancy (Salac et al., 1982). Another complication with direct-seeding on a large scale can be the prohibitive cost of seed for some species (Airhart et al., 1983). In the case of wildflower mixes, proper seeding dates may be especially troublesome to pinpoint as research by Kaspar and McWilliams (1982) indicates variability in germination response to temperature among the four species that were tested. Little information is available for the germination requirements of many wildflowers, and the majority of data extant are derived from greenhouse or laboratory experiments (Salac et al., 1982). Methods for extensive field establishment of many wildflower species used in this study are largely unavailable (Airhart et al., 1983, Association of Official Seed Analysts, 1989; Deno, 1993; Hartmann and Kester, 1983; Jones and Foote, 1990; Martin, 1986; Paulson, 1989; Phillips, 1985; Smith and Smith, 1980).

Airhart et al. (1983) investigated the use of wildflower sods as a means of assuring better stand establishment. Of the 13 species tested in this experiment, only two were native Virginia perennials: *Asclepias tuberosa* and *Liatris spicata*. Sods were produced by species; no mixes were used. Suitable sods for the Virginia native wildflowers could only be achieved by seeding a soilless mix at twenty times the seed manufacturer's recommended rate. After one year in the field, no *A. tuberosa* or *L. spicata* survived.

A more promising establishment technique, the Virginia Tech Transplanted Meadow (VTTM), was developed by Lyons working with annual species (Laurent, 1994). The method utilizes up to 12 species combined in equal weights to form a mix. Sand is added to act as a dispersal agent. Flats filled with a soilless mix are then seeded by hand broadcasting. The wildflower mix is grown in the greenhouse for one month before transplanting to the field site. Preparation consists of applying glyphosate (Roundup®) in April, rototilling 7 to 10 days later, and a final glyphosate application again in late May. Plugs are transplanted into the field in late May or early June spaced on a minimum of 30 cm centers. The transplanted meadow mix is then mulched to a depth of 7.5 cm with hardwood bark.

The species selected for research in the present study are listed in Appendix II with brief descriptions of appearance, germination procedures, and site preferences.

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Appendix I

Commercial Sources for Wildflower Seeds Used for This Thesis Research

Environmental Seed Producers
P. O. Box 2709
Lompoc CA 93428
species obtained from this source:
Lupinus perennis

Lafayette Home Nursery, Inc.
Rural Route 1, Box 1A
Lafayette IL 61449
species obtained from this source:
Coreopsis lanceolata
Tephrosia virginiana
Veronicastrum virginicum

Prairie Moon Nursery
Route 3, Box 163
Winona MN 55987
species obtained from this source:
Aquilegia canadensis
Aster laevis
Aster novae-angliae
Baptisia australis
Euphorbia corollata
Liatris spicata
Monarda fistulosa
Rudbeckia triloba
Solidago speciosa
Teucrium canadense

Appendix II

Description of species selected for native Virginia perennial seed mixes. Heading information for each mix is provided in the following format:

Genus species	common name	Family
height	flower color	bloom span

1. TALL MIX

Asclepias tuberosa	butterflyweed	Asclepiadaceae
30-60 cm	orange	July-August

Butterflyweed is native from New England south to Florida, west to Texas, and north to Colorado to Minnesota in the United States (Niering and Olmstead, 1995). Bright orange flowers occur in umbels on multiple flowering stems. The most critical factor in site selection is that the soil be well-drained. Full sun is also preferred. The deep taproot inhibits successful transplanting of mature specimens (Martin, 1986), but contributes to its long life (Smith and Smith, 1980). Seed propagation is easily accomplished under a regime of 40^0 F (4^0C) for three months followed by 70^0 F (21^0C) using either fresh seed or seed kept in dry storage at either 40^0 F (4^0C) or 70^0F (21^0C) for six months; light has no impact on germination (Deno, 1993). Blooming can be expected in the second year from plants grown from seed (Smith and Smith, 1980); plants may bloom in the first year (Stevenson, 1985).

Aster novae-angliae	New England aster	Asteraceae
90-150 cm	blue-violet	August-October

New England aster is native in the U. S. east of the Rocky Mountains. Blue-violet ray flowers surround a gold disk. Soil must be well-drained, although sufficient moisture is considered necessary for successful culture (Martin, 1986). Smith and Smith (1980) maintain that New England aster readily adjusts to dry soils. Full sun is preferred. Conflicting information exists concerning seed propagation. Art (1987) and Martin (1986) state that seeds benefit from a period of moist, cold stratification. Phillips (1985) states that germination percentage is low, while Art (1987) suggests that stratification triples the germination rate. Deno (1993) prescribes germinating *A. novae-angliae* at 70^0 F (21^0C) using either fresh seed or seed kept under dry storage for six months.

Baptisia australis	blue false indigo	Fabaceae
30-90 cm	dark blue	May-June

The blue-green foliage of blue false indigo is enhanced by the appearance of blue flowers on upright racemes. Well-drained soil is necessary for optimum growth (Martin, 1986). *Baptisia australis* is suited to dry or mesic sites, and is an excellent soil builder, fixing nitrogen as do many of the legumes (Smith and Smith, 1980). Full sun is preferred. Seed propagation is easily accomplished at 70^0 F (21^0 C) using either fresh seed or seed stored under dry conditions for six months (Deno, 1993).

Liatris spicata	blazing star	Asteraceae
30-90 cm	pink-lilac	July-September

Blazing star flowers are rose or lilac colored and occur as thistle-like puffs along wands above grassy foliage (Peterson and McKenny, 1968), opening first from the top of the spike and proceeding downwards (Phillips, 1985). Well-drained soil is required; mesic conditions are considered necessary for optimum performance (Martin, 1986), with plants becoming shorter and almost stocky in dry soils (Phillips, 1985). Germination occurs most rapidly under 70^0 F (21^0 C) (Deno, 1993; Phillips, 1985); light has no impact (Deno, 1993).

Rudbeckia triloba	thin-leaved coneflower	Asteraceae
60-150 cm	gold-yellow	June-October

This species of coneflower is the only one used in this study which is known to be apomictic (Harkess and Lyons, 1994). It is considered to be a short-lived perennial (Stevenson, 1985). The thin-leaved coneflower may be distinguished from *R. hirta* by the three-lobed lower leaves and by smaller, more numerous flowers with fewer ray florets (Peterson and McKenny, 1968). The gold ray florets surround a dark brown disk, giving rise to other common names such as black-eyed or brown-eyed Susan. This species is tolerant of both drought and shade (Phillips, 1985) and will bloom in the first season from seed which is abundantly produced (Stevenson, 1985). No published germination information was obtained for *R. triloba*.

Solidago speciosa	showy goldenrod	Asteraceae
60-180 cm	yellow	August-October

This species is native to woodland openings, fields, and prairies from Michigan, New York, and Massachusetts to the southern limits of the United States (Peterson and McKenny, 1968). Martin (1986) and Phillips (1985) note that poor germination is characteristic of the goldenrods. No published germination information for this species was found.

Teucrium canadense	germander	Lamiaceae
30-90 cm	pink	July-September

Germander is native to all but four counties in Virginia (Harvill et al., 1992). The pink flowers occur as spikes and are shaped much as other members of the mint family (Peterson and McKenny, 1968). Deno (1993) describes germination procedures only for two Turkish species; as one of these germinated in 10-14 days under a 70^0F (21^0C) dark treatment, the same technique was employed for this study.

Veronicastrum virginicum	culver's root	Scrophulariaceae
60-210 cm	white	June-September

Culver's root occurs naturally in Manitoba, southern Ontario, and from Massachusetts to the southern limits of the United States (Peterson and McKenny, 1968) in meadows and woods (Dobelis, 1986) . The white flowers occur in terminal spike-like racemes 15-20 cm long (DeWolf, 1986). Leaves are lance-shaped, encircling the stem in whorls of three or more (Dobelis, 1986). Smith and Smith (1980) state that seed is difficult to germinate, but provide no information for treatment.

2. MODERATE HEIGHT MIX

Aquilegia canadensis	columbine	Ranunculaceae
30-60 cm	red/yellow	April-June

Columbine has a red and yellow bicolored flower. It occurs naturally on rocky slopes and dry, open areas in full or partial sun (Martin, 1986) from New England to Georgia, then west to Tennessee and Wisconsin in the United States (Niering and Olmstead, 1995). Although tolerant of dry conditions once mature, sufficient moisture is required during seedling establishment (Art, 1987). As the plants do not divide or transplant well when mature, seed propagation is recommended (Martin, 1986). Deno (1993) stipulates that dry storage at 70^0F (21^0C) is required to remove the requirement for gibberellic acid treatment unless certain (unspecified) outdoor treatments are used. Light is not listed as a germination factor for this species by Deno (1993). Smith and Smith (1980) state that seed propagation is easily accomplished and do not specify any required treatment, while Phillips (1985) states that cool, moist stratification and light are germination promoting treatments for *A. canadensis*.

Aster laevis	smooth aster	Asteraceae
30-90 cm	blue-violet	August-October

The smooth aster occurs in dry soil along wood edges from Saskatchewan to Maine and to the southern limits of the United States (Peterson and McKenny, 1968). Martin (1986) also includes moist prairies as a natural habitat. This species was cited as one aster which does not require stratification for germination; *A. laevis* will often bloom in the first season from a spring sowing (Martin, 1986).

Coreopsis lanceolata	tickseed	Asteraceae
30-60 cm	yellow	May-August

The yellow flowers of tickseed are typical of the Asteraceae, including ray and disk flowers (Martin, 1986). This species occurs naturally on poor soils and along roadsides from Ontario south (Peterson and McKenny, 1968), being native to approximately two-thirds of the counties in Virginia (Harvill et al., 1992). Martin (1986) states that seeds have a high germination rate within one week when exposed to light; Deno (1993) prescribes six months of dry storage before germinating at 70^0 F (21^0C), with no mention of a light requirement.

Euphorbia corollata	flowering spurge	Euphorbiaceae
30-90 cm	white	June-October

Flowering spurge is found from New York to the southern limits of the United States in fields, open woods, and along roadsides (Peterson and McKenny, 1968). White inflorescences are in terminal cymes (DeWolf, 1986); the "petals" are actually five white bracts surrounding minute flowers (Peterson and McKenny, 1968). Deno (1993) reports that several alternating temperature cycles are required for germination of other *Euphorbia* species. Smith and Smith (1980) recommend moist stratification for seed to be sown in spring, with flowers produced the following year.

Lupinus perennis	wild lupine	Fabaceae
30-60 cm	light blue	April-July

Wild lupines are found in dry soils and clearings from New York and southern Maine to the southern limits of the United States (Peterson and McKenny, 1968). Typical of the Fabaceae, the blue pea-like flowers occur on an upright stem above seven to nine leaflet, palmately divided leaves (Martin, 1986). Impervious seed coats are mentioned by Deno (1993) to require scarification in order to yield high germination rates. Martin (1986) states that seeds may alternatively be soaked for up to 24 hours prior to sowing.

Monarda fistulosa	wild bergamot	Lamiaceae
60-90 cm	lavender-pink	July-August

Bergamot has pink or lavender flowers which occur as tubular clusters in heads subtended by bracts similar in color (Martin, 1986). This species is native to dry soils in clearings and thickets from Minnesota and western New England to the southern limits of the United States (Peterson and McKenny, 1968), but is adaptable to wet, mesic, or dry sites (Smith and Smith, 1980). Propagation is easily accomplished from seed (Martin, 1986), and dry storage treatment is recommended by Deno (1993). Deno (1993) further states that germination is promoted by light, Art (1987) recommends chilling, and Smith and Smith (1980) consider moist stratification to enhance germination.

Tephrosia virginiana	goat's rue	Fabaceae
30-60 cm	yellow/pink	May-August

Goat's rue has a yellow and pink bicolored flower with the typical pea-like form of the Fabaceae which is found in sandy soils from New York and central New England south (Peterson and McKenny, 1968). In Virginia it occurs along roadsides, in open woods, and fields (Gupton and Swope, 1979), thriving on very dry sites (Smith and Smith, 1980). High germination rates result from scarification or from sowing fresh seed, but dry storage increases the impervious qualities of the seed coat (Deno, 1993). The plants will not flower until the third year (Smith and Smith, 1980).

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Chapter 2

Germination of Selected Native Virginia Herbaceous Perennials

Abstract. Fifteen species of native Virginia herbaceous perennials were studied for germination response to four treatments, two of which were conducted in greenhouses and the other two in the field. The field germination tests were conducted in late April and early June; only *Asclepias tuberosa* germinated at acceptable levels. The two greenhouse treatments consisted of intermittent mist or hand irrigation. Performance of several species was acceptable under either or both of these conditions. Given the relatively high cost of native Virginia wildflower seed, greenhouse production of transplants is recommended for meadow establishment.

Native Virginia species suitable for incorporation into a mix to be planted along roadsides were identified using *Wildflowers of the Shenandoah Valley and Blue Ridge Mountains* (Gupton and Swope, 1979), *A Field Guide to Wildflowers: Northeastern/Northcentral North America* (Peterson and McKenny, 1968), *Guide to the Vascular Plants of the Blue Ridge* (Wofford, 1989), and *Atlas of the Virginia Flora III* (Harvill et al., 1992). The majority of "ornamental" herbaceous Virginia natives are perennial; thus, no annual species were selected for inclusion in trial mixes.

The primary consideration in selecting species for inclusion in a native wildflower mix was to provide for a maximum period of continuous bloom (Figure 2.1). Height was considered such that early blooming species benefitting from some shading would be planted with taller, later flowering species. This strategy would additionally serve to disguise weather-beaten foliage and seed pods of spring blooming species. Whenever possible, canopy bloom heights were staggered to yield a greater depth of color. Species were also selected on the basis of family diversity, primarily to minimize competition for nutrients and maximize resistance to any one pest or pathogen. Monoculture plantings have been blamed for increasing plant losses to insect and disease invasion (Peirce, 1987). Color harmony within each mix was considered despite its subjective nature. Accordingly, this was given the least weight in the selection process.

Other selection criteria included the tolerance for full sun, drought, low soil fertility, heavy-textured soil, and visibility at interstate highway speeds. The species selected also occurred throughout Virginia or over large areas encompassing the geographic subdivisions of Tidewater, Piedmont, and Mountain regions.

Commercial seed sources for each species selected were located. Earlier research indicated that seed produced at similar latitudes to the area in which the species were to be grown would be more reliable with regard to hardiness, blooming, and reproductive success (Beckwith, 1991; Jones and Foote, 1990; Martin, 1986). Of the 25 species initially chosen for this thesis research, 14 proved to be commercially unavailable from 46 supply sources. After further research, 14 suitable species were obtained from Environmental Seed Producers (CA), Lafayette Home Nursery, Inc. (IL), and Prairie Nursery (WI). Virginia latitude spans roughly 37^0N to 39^0N; the seed suppliers are located from about 40^0N to 45^0N. *Asclepias tuberosa* was donated by Ms. Nicky Staunton of the Virginia Native Plant Society.

Germination characteristics for the selected species were compiled from the literature. Information regarding germination rates and procedures for certain species was not available from the sources consulted (Association of Official Seed Analysts, 1989; Deno, 1993; Hartmann and Kester, 1983; Jones and Foote, 1990; Martin, 1986; Paulson, 1989; Phillips, 1985). Recent research has identified pretreatment strategies for some wildflower species. Stratification improves germinability of several species (Bratcher et al., 1993; Martin, 1986). Growth regulators and specific storage temperatures were found to control germination response for *Coreopsis lanceolata* (Carpenter and Ostmark, 1992). Samfield et al. (1991) focused on seed priming as a means of improving uniformity of germination for selected wildflower species.

Conditioning techniques may well prove beneficial to commercial producers of wildflower seed; however, the present study was intended to develop pragmatic procedures useful to the Virginia Department of Transportation (VDOT) in the establishment of roadside plantings. For this reason, germination trials were conducted without any chemical conditioning treatment or particular stratification regimes.

Light and temperature cycles favoring germination of many native herbaceous perennials have been reported by Deno (1993). "Temperature is, perhaps, the most important environmental factor that regulates germination and controls subsequent seedling growth" (Hartmann and Kester, 1983, p.146). "It appears that seeds of many wild species or those with a short domestication history have a relative dormancy that is responsive to temperature alternation" (Gardner et al., 1985, p.226). For this reason, temperature was allowed to fluctuate diurnally during the germination testing of the selected wildflower species in this experiment. "The optimum germination temperature is usually about 25^0 to 30^0C" for plants native to temperate zones (Hartmann and Kester, 1983, p.146). Other sources also reported temperature to be critical for germination (Deno, 1993; Kaspar and McWilliams, 1982; Mengel and Kirkby, 1987; Phillips, 1985; Salac et al., 1982). As early as 1926, Kinzel classified several hundred species on the basis of light response; i. e., whether germination was favored by light, by dark, or was unaffected by light and dark (Gardner et al., 1985). Where information was available, the light/dark requirements of selected wildflower species were provided for in this experiment. As *Teucrium canadense* (germander) was the only species in this work postulated to require dark for germination (Deno, 1993), seven other species with no documented germination requirements were also subjected to dark treatment to examine their suitability for inclusion in a mix with germander.

Materials and Methods

Seeds of *Baptisia australis, Liatris spicata, Rudbeckia triloba, Solidago speciosa, Tephrosia virginiana, Teucrium canadense*, and *Veronicastrum virginicum* were provided with dark conditions by covering the seeds with 1 to 2 cm of medium. Seeds were sown on top of the medium to provide light germination conditions for *Aquilegia canadensis, Asclepias tuberosa, Aster laevis, A. novae-angliae, Coreopsis lanceolata, Euphorbia corollata, Lupinus perennis*, and *Monarda fistulosa.* All treatments were replicated five times in a completely randomized design. Data were analyzed by repeated measures analysis (Littell et al., 1991).

Treatment 1: Germination using automatic mist irrigation.

Twenty-five seeds of each species were sown into individual #801 plastic cells (Kord Products, Toronto, Ontario, Canada) measuring 15 cm per side containing Sunshine Mix® #1 (3 peat: 1 perlite: 1 vermiculite/ v:v:v) (Fisons Horticulture Inc., Vancouver, British Columbia, Canada). Eight cells, each containing a different species, were placed into plastic flats to constitute one replication. The flats were randomly arranged on benches equipped with an intermittent mist irrigation system which delivered water for 30 sec at eight minute intervals from 800 hrs to 1800 hrs in a fiberglass greenhouse. The temperature was constantly controlled between 21^0 and 26^0 C. The treatment was begun 11 April 1994; seedling emergence was recorded for 30 days.

Treatment 2: Germination using daily hand mist irrigation.

Twenty-five seeds of each species were sown as described in Treatment 1 in the same replication manner. The flats were randomly arranged on benches in a glass greenhouse. Irrigation was delivered once daily, such that flats were watered to saturation, between 900 and 1200 hrs using a coarse-mist head attached to a garden hose. The temperature was constantly controlled between 21^0 and 26^0 C. The treatment was begun 11 April 1994; seedling emergence was recorded for 30 days.

Treatment 3: Germination following spring field sowing.

A one m^2 plot per replication in the Virginia Tech Horticulture Gardens was rototilled to a depth of approximately 20 cm and hand-raked to ensure a smooth seedbed. Each plot was then subdivided into squares of 15 cm per side. On 25 April 1994, 25 seeds of each species were sown into individual squares; each square was labeled. Soil temperature at a 4 cm depth was recorded at 1400 hrs in each plot; temperatures were then averaged and recorded. Seeds were watered in using a breaker and garden hose to ensure good seed-soil contact. No subsequent irrigation was supplied except as provided by rainfall. Seedling emergence was recorded for 30 days.

Treatment 4: Germination following late spring field sowing.

Soil temperatures were recorded as in Treatment 3. When temperatures above 20^0 C were recorded for three consecutive days, a one m^2 plot per replication was hand-raked to ensure a smooth seedbed. Each plot was then subdivided into squares of 15 cm per side. On 2 June 1994, 25 seeds of each species were sown into individual squares and subsequently handled as in Treatment 3.

Results

Species exhibiting a moderate to high emergence level were shown to have a normal distribution according to univariate analysis for normality. Only those species showing low or no germination were lacking in a highly significant Day * Treatment response as well as demonstrating non-normal distribution. The three species failing to germinate during the 30 day duration of data collection were *Solidago speciosa, Teucrium canadense*, and *Veronicastrum virginicum. Tephrosia virginiana* germinated poorly, producing only 6 seedlings across all treatments and replications. As shown in Table 2.1, the combined effect of all treatments was a highly significant determinant of germination response for all species except *T. virginiana*, where this was only marginally significant.

Table 2.2 illustrates the differing emergence patterns among species. *Aquilegia canadensis* failed to germinate until after day 10. For *Aster novae-angliae* and *Monarda fistulosa,* no further germination occurred after day 20; no significant response was obtained after day 15. *Rudbeckia triloba* exhibited much the same pattern; although germination continued through the end of the experiment, it was insignificant beyond day 20. *Baptisia australis* demonstrated a steady, significant germination response over the

30 day data collection period. *Aster laevis* also continued to germinate throughout the experiment, although the contrasts were not as marked as seen for *B. australis*. The most erratic germination pattern was observed for *Euphorbia corollata*; significant response was noted between days 5 and 10, response became insignificant between days 20 and 25, and then germination contrasts were highly significant between days 25 and 30. Germination contrasts over time were not significant for *T. virginiana*.

The automatic mist of Treatment 1 produced a mean germination of 141 ±0.26 out of 375 seeds sown across all species at the close of the data collection period. Treatment 2, hand irrigation on a daily basis, resulted in a mean germination of 150 ±1.76 after 30 days over all species. For Treatments 3 and 4, early and late field sowings, germination response was poor across all species over the 30 day experiment period, with mean germination values of 29 ±0.85 and 33 ±2.70, respectively. Individual species varied widely in response to the different treatments (Table 2.3). All species showed a marked decrease from maximum greenhouse germination levels to maximum field emergence. Species germinated under dark conditions in the greenhouse treatments had lower germination percentages in the fiberglass greenhouse used for Treatment 1 as compared to germination percentages in the glass greenhouse used for Treatment 2. *Rudbeckia triloba* germinated as well in the field under the warmer soil conditions of the 6 June 1994 sowing as it did under intermittent mist. For species germinated under light conditions, *A. canadensis*, *A. tuberosa*, and *L. perennis* produced higher germination levels under intermittent mist than under daily hand irrigation treatments. The most dramatic difference in germination response was observed for *A. novae-angliae* and *M. fistulosa*; these two species germinated at 90% levels under at least one greenhouse treatment, but failed to germinate in the field.

Qualitative observations were also made during the course of the experiment. For the majority of species, marked differences in growth and development of the seedlings were observed to occur between the two greenhouse treatments. As evidenced by photographic records (Fig. 2.2), seedlings were in some cases many times larger when grown under high light provided by the glass greenhouse than when grown under light limited conditions found in the fiberglass greenhouse. This contrast was most evident for the asters, bergamot, and tickseed.

Discussion

Three factors are generally considered to be of significance in germination response where no dormancy exists: water, temperature, and oxygen; some species are additionally responsive to light or dark conditions (Gardner et al., 1985). For the purposes of this experiment, oxygen levels were assumed to be appropriate for germination. The four treatments were designed to test the germination response where the moisture, temperature, and light levels varied to determine which of these factors might be most critical for germination of native Virginia wildflowers. Moisture was amply provided under the automatic intermittent mist system used for Treatment 1; however, light levels were probably attenuated in this older fiberglass greenhouse. Light transmission is roughly equivalent for new fiberglass panels as compared to glass, but decreases with age (Nelson, 1985). Higher light conditions were likely available in the glass greenhouse, but water was less available. Temperatures between the two greenhouses were controlled within the same range. The field experiments were designed to determine how higher soil moisture accompanied by colder temperatures and shorter days at an April sowing date compared to drier soil conditions under warmer soil temperatures and longer days at a June sowing.

Most obvious from our data is that the extremely poor germination of the field treatments combined with the relatively high cost of Virginia native wildflower seed makes direct-seeding in the spring an unacceptable means of establishing meadow plantings. While three species used in these experiments failed to germinate under any treatment, seven of the remaining twelve that germinated in the greenhouse failed to do so in the field. In all cases, field germination response fell far enough below maximum greenhouse response that direct-seeding of these perennial species in the spring should be viewed as a waste of time and money.

Multivariate tests for the hypothesis of no day effect demonstrated significant Day * Treatment effects on germination. The responses of certain species to the different treatments provide an indication of which germination factor may be the most critical for successful seed propagation. *Aquilegia canadensis* germination has been improved by GA_3 application (Deno,1993), cold stratification (Martin, 1986), and dry storage of the seeds prior to germinating at 70^0 F (21^0 C) (Deno, 1993). Deno (1993) states that scarification produces no attendant germination increase and concludes that hard seed coats are not responsible for delayed germination. The results of this experiment demonstrated that germination occurred sooner and produced more seedlings at the end of 30 days when the seeds were placed under a mist system. This may indicate that substances are present in the seed coat which must be leached before germination can proceed. Further experiments would be needed to confirm this hypothesis. That the contrast in germination response is highly significant between days 25 and 30 indicates that maximum germination levels were probably not obtained for columbine in this experiment.

While soil moisture levels were not monitored during the course of this experiment, it was readily observable that the field soil was dry to a depth of about 3 cm at the time of the April sowing and dry down to about 6 cm at the beginning of June 1994. Moisture was less available for seed imbibition under field conditions than in the greenhouse. Water would appear to be the critical factor in successful germination of *A. novae-angliae* and *M. fistulosa,* since both reached 90% germination levels under at least one greenhouse treatment and failed to germinate in the field. That *A. novae-angliae* germinated significantly better in the glass greenhouse may indicate that high light levels are also necessary for optimum response, or that excess moisture depresses germination. *Monarda fistulosa* demonstrated virtually no difference in germination between greenhouse treatments.

Under mist irrigation, *A. tuberosa* produced a mean germination percentage of

94%, the highest recorded for any species under any treatment. Hand irrigation reduced this level by 10%, while the June field sowing resulted in a 67% germination mean, which was higher than that demonstrated in the greenhouses by six other species that did germinate in this experiment. Higher temperature seems to promote germination for this species, and germination appears to reach maximum levels when warmth is accompanied by high moisture levels. One of the surprises in this experiment was that butterflyweed sown on 2 June 1994 in the field was in full bloom by 26 August 1994 despite weed competition and the lack of any irrigation or fertilizer supplements. Further research needs to be conducted using different seed lots of *A. tuberosa*; of all species used in this experiment, only those of butterflyweed were obtained from a Virginia source.

No indication for light/dark preference was found in the literature consulted for the species used in our Tall Mix; thus, these species were germinated under dark conditions as required for *Teucrium canadense*. The contrast in germination response for these species between the fiberglass greenhouse treatment and the glass greenhouse indicates that high light may, in fact, enhance the germination of these wildflowers. There might also be some other factor such as oxygen abundance in the medium pore space which was present in the glass greenhouse (irrigation supplied once a day) but reduced in the fiberglass greenhouse (intermittent mist irrigation). The one exception would probably be *T. virginiana,* which germinated at a mean level of only 2% in both greenhouse treatments. For *T. virginiana,* Treatment was only marginally significant since there is only a slight difference between germination counts of 0 and 6 out of 625 seeds sown. Further research is needed to substantiate this conclusion.

Germination levels of 50% or greater combined with excellent field survival are probably necessary for economic justification in retaining a species in a wildflower mix destined for use by VDOT. Of the species tested in this experiment, *A. canadensis, A. tuberosa, A. novae-angliae*, *B. australis*, *L. perennis, and M. fistulosa* have met the germination criterion. *Coreopsis lanceolata* seed is comparatively inexpensive and has been included in commercial mixes, possibly because of its proven field performance. *Rudbeckia triloba* may also prove to be an effective performer, particularly if germinated under light conditions. Based on the results of this experiment, *A. laevis, E. corollata, L. spicata, S. odora, T. virginiana, T. canadense*, and *V. virginicum* would not be considered as viable species for inclusion in a wildflower mix appropriate for use by VDOT.

No commercial source for native Virginia wildflower seed could be located in the Mid-Atlantic region. The one locally obtained species, *A. tuberosa*, came as a gift from a member of the Virginia Native Plant Society. There would appear to be a profitable niche available on the east coast for a producer of native wildflower seed. At present, several species which met initial selection criteria are not commercially available. Some of the species tested in this experiment were second choices and many of those are the species which failed to perform acceptably. Further research and consumer interest are needed to elevate the commercial status of Virginia native wildflowers in the Commonwealth.

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Table 2.1 Statistical significance of germination responses of 12 native Virginia wildflowers to four combined treatments.

Species	Pr > F[z]
Aquilegia canadensis	0.0001
Asclepias tuberosa	0.0001
Aster laevis	0.0001
Aster novae-angliae	0.0001
Baptisia australis	0.0001
Coreopsis lanceolata	0.0004
Euphorbia corollata	0.0001
Liatris spicata	0.0001
Lupinus perennis	0.0001
Monarda fistulosa	0.0001
Rudbeckia triloba	0.0001
Tephrosia virginiana	0.0470

[z] Probabilities at 95% confidence level.

Table 2.2 Difference in mean germination percentages for 12 native Virginia wildflowers over a 30 day period.

	Change in % Germination from Day a:b				
Species	Day 5:10	Day 10:15	Day 15:20	Day 20:25	Day 25:30
Aquilegia canadensis	.[z]	1.0[y]*	10.2***	6.0***	7.8***
Asclepias tuberosa	21.6***	20.2***	17.0*	7.0*	2.9*
Aster laevis	2.0*	2.6*	3.4***	1.6^{NS}	0.8*
Aster novae-angliae	17.6***	3.8**	2.2^{NS}	.	.
Baptisia australis	2.6**	11.8**	15.8***	9.8*	9.2***
Coreopsis lanceolata	7.0***	11.4*	7.4**	3.6^{NS}	0.9*
Euphorbia corollata	0.8*	2.2^{NS}	2.0***	1.2^{NS}	3.2***
Liatris spicata	2.2**	10.4***	1.8^{NS}	1.0^{NS}	0.2^{NS}
Lupinus perennis	9.4**	6.0*	5.6**	3.8*	1.6^{NS}
Monarda fistulosa	31.2***	4.6**	0.6^{NS}	.	.
Rudbeckia triloba	6.2***	2.6**	1.2**	1.6^{NS}	1.6^{NS}
Tephrosia virginiana	0.2^{NS}	0.2^{NS}	0.6^{NS}	.	.

[z] No germination at day 10 for *A. canadensis*; no further germination after day 20 for *A. novae- angliae, M. fistulosa* or *T. virginiana*.

[y] *=significant; **=very significant; ***= highly significant; ^{NS}= not significant when analyzed by GLM repeated measures analysis at a 95% confidence level.

Table 2.3 Mean germination percentages after a 30 day period for 12 native Virginia wildflower species in response to four treatments.

	Mean Germination Percentage			
Species	Treatment 1 [z]	Treatment 2	Treatment 3	Treatment 4
Aquilegia canadensis	63	37	0	0
Asclepias tuberosa	94	84	49	67
Aster laevis	12	30	0	0
Aster novae-angliae	78	90	0	0
Baptisia australis	66	86	12	34
Coreopsis lanceolata	36	49	29	13
Euphorbia corollata	19	18	0	0
Liatris spicata	26	37	0	0
Lupinus perennis	71	40	20	10
Monarda fistulosa	92	90	0	0
Rudbeckia triloba	6	37	4	6
Tephrosia virginiana	2	2	0	0

[z] Treatment 1: automatic mist; Treatment 2: hand irrigation; Treatment 3: early field sown; Treatment 4: late field sown.

Species - Moderate Height	Apr	May	Jun	Jul	Aug	Sep	Oct
Aquilegia canadensis	***	***	***				
Asclepias tuberosa				***	***		
Aster laevis					***	***	***
Coreopsis lanceolata		*	***	***	***		
Euphorbia corollata			***	***	***	***	***
Lupinus perennis	***	***	***	***			
Monarda fistulosa				***	***		

Species - Tall	Apr	May	Jun	Jul	Aug	Sep	Oct
Aster novae-angliae					***	***	***
Baptisia australis		***	***				
Liatris spicata				***	***	***	
Rudbeckia triloba			***	***	***	***	***
Solidago speciosa					***	***	***
Tephrosia virginiana		***	***	***	***		
Teucrium canadense				***	***	***	
Veronicastrum virginicum			***	***	***	***	

Figure 2.1 Bloom sequence for "Moderate Height" and "Tall" native Virginia wildflower mixes.

Figure 2.2 Seedling growth variation between two greenhouse treatments.

Chapter 3

Germination and Growth Response of Five Native Virginia Wildflower Species as Affected by Three Pre-emergence Herbicides

Abstract. The treatments consisted of both field and greenhouse trials. Poor germination of the desired species accompanied by severe weed pressures in the field yielded no significant results and led to the conclusion that application of Dacthal® 2G (DCPA), Pennant® 5G (metolachlor), or Treflan® 5G (trifluralin) at seeding is not a viable strategy for weed control in meadow establishment. Greenhouse data indicated that germination suppression and/or developmental retardation occurred for at least some species treated with each of the herbicides. The germination reduction combined with relatively high injury levels to *Aquilegia canadensis, Asclepias tuberosa, Aster novae-angliae, Lupinus perennis*, and *Monarda fistulosa* preclude the use of the aforementioned herbicides at seeding for these wildflower species.

Weed control in wildflower plantings is the most critical factor in determining successful establishment (Laurent, 1994). A primary source for weeds in landscape plantings is the reservoir of dormant weed seeds already present in the soil known as the "seed bank" (Gallitano et al., 1993). The persistence and ubiquity of weeds in disturbed soils is due to the ability of weeds to produce numerous, long-lived seeds which are easily transported by wind, water, and animals to new sites (Ross and Lembi, 1985). The longevity of weed seeds in combination with their sheer numbers guarantees that a given weed will continue to persist almost indefinitely once established (Ross and Lembi, 1985). The key in weed management is to prevent one or two aggressive species from gaining dominance in a wildflower planting (Martin, 1986) and several strategies exist to accomplish this.

Fumigants such as methyl bromide can be applied to soil prior to installation of wildflowers (Derr, 1994). Severe toxicity to humans, labor-intensive application, and great expense are several disadvantages of these compounds (Ross and Lembi, 1985). Methyl bromide is a restricted use chemical which requires specialized handling (Derr, 1994) and may not be commercially available after the year 2000; other fumigants are likewise hazardous and cumbersome to apply (Ross and Lembi, 1985). Site preparation for meadow plantings frequently combines tillage with preplant nonselective herbicide treatment (Corley and Smith, 1990; Laurent, 1994). The site is first treated with a systemic, nonselective herbicide such as glyphosate. It is then shallowly tilled approximately two weeks later to expose buried weed seeds and aerate the soil surface. A final herbicide treatment is made to eradicate regrowth (Laurent, 1994). Alternatively, the site may be mowed initially prior to application of a nonselective contact herbicide such as paraquat after regrowth (Ross and Lembi, 1985); this is followed by tillage of the area to various depths (Corley and Smith, 1990).

As continued weed control is often either necessary or desired during the establishment phase of the wildflower planting, some research has been examined on the response of wildflowers to a variety of pre-emergence and post-emergence herbicides. Corley and Smith (1990) reported that tilling shallowly following pre-plant herbicide treatment is recommended. The graminicides Poast® (sethoxydim) and Fusilade® (fluazifop) were found to control grassy weeds in wildflower plantings with no damage to desirable species. Surflan® (oryzalin) resulted in some depression of wildflower survival when applied pre-emergence, while Treflan® 10G (trifluralin) resulted in minimal death to desirable species. Broadleaf weeds were particularly difficult to control, requiring hand-pulling, spot spraying, or wicking with a contact herbicide. Derr (1993, 1994) focused on the use of pre-emergence herbicides on transplanted wildflowers in container production. *Rudbeckia hirta* var. *pulcherrima* and *Coreopsis lanceolata*, two native Virginia wildflowers, were largely unaffected by DCPA (Dacthal®) four weeks after herbicide application. Both species did demonstrate damage from dinitroaniline herbicides, with the greatest injury resulting from oryzalin application. Three months after herbicide application, however, neither of these species showed significant differences in survival from the control (Derr, 1994). Metolachlor did not affect survival of *C. lanceolata,* while it did provide acceptable control of *Cyperus esculentus* (yellow nutsedge) (Derr, 1993).

The influence of EPTC/Treflan pre-emergence herbicide combination on direct-seeded wildflowers was investigated by Erusha et al. (1991). Among the wildflower species included in the study were *Coreopsis lanceolata* and *Rudbeckia hirta*. The experiment was conducted on two sites: a silty-clay loam in Nebraska, and a sandy loam in Oregon. The herbicide combination had no impact on stand establishment for *R. hirta* as compared to the control at either site. The response of *C. lanceolata* did not differ from the control at the Nebraska location; however, stand establishment in Oregon was only 35% for the control, while the herbicide treated plot demonstrated an 80% establishment. No other species of the 47 examined in this experiment had better stand establishment for herbicide treated plots compared to the controls, and no hypothesis for the anomaly was provided by the authors.

Assessing the impact of pre-emergence herbicides on native Virginia species selected from the Asclepiadaceae, Asteraceae, Fabaceae, Lamiaceae, and Ranunculaceae was the objective of this experiment. Herbicides were selected from three different classes: chloroacetamide (Pennant® 5G), dinitroaniline (Treflan® 5G), and phthalic acid (Dacthal® 2G). Chloroacetamides primarily control annual grasses including *Digitaria* spp. (crabgrass), *Echinochloa crus-galli* (barnyardgrass), and *Panicum dichotomiflorum* (fall panicum), with some broadleaf control of species such as *Mollugo verticillata* (carpetweed), *Solanum* spp. (nightshades), *Amaranthus* spp. (pigweeds), and *Portulaca* *oleracea* (common purslane). Metolachlor (Pennant) controls yellow nutsedge, as well as suppressing *Chenopodium album* (common lambsquarters) and *Ambrosia artemisiifolia* (common ragweed). Dinitroaniline herbicides severely inhibit the root development of susceptible species. Trifluralin (Treflan) controls barnyardgrass, crabgrasses, foxtails (*Setaria* spp.), and fall panicum, as well as broadleaved species such as carpetweed, common lambsquarters, pigweeds, and common purslane. The phthalic acid DCPA (Dacthal) is a mitotic inhibitor that disrupts both root and shoot development of sensitive species, particularly crabgrasses, foxtails, common lambsquarters, and common purslane (Ross and Lembi, 1985).

Materials and Methods

The five Virginia wildflower species chosen for study were *Aquilegia canadensis* (Ranunculaceae), *Asclepias tuberosa* (Asclepiadaceae), *Aster novae-angliae* (Asteraceae), *Lupinus perennis* (Fabaceae), and *Monarda fistulosa* (Lamiaceae). Also included in the experiments was large crabgrass, *Digitaria sanguinalis* (Poaceae), since it is susceptible to all three herbicides used (Ross and Lembi, 1985). Herbicide activity could thus be monitored by recording the emergence of *D. sanguinalis*. Two experiments were designed to study the impact of pre-emergence herbicides on emergence and early development of the selected wildflowers; one was conducted in the field, while the other was performed in the greenhouse.

Field Experiment

The site was an area near the Virginia Tech Horticulture Gardens in Blacksburg, VA which had been planted with annual flowers for the past several growing seasons. The area was rototilled in April 1994; a 2% solution of glyphosate (Round-up®) was applied at five weeks and again two weeks prior to wildflower seeding. Forty plots of 0.5 m^2 were marked off for eight treatments (Fig. 3.1). For each treatment, 25 seeds of each of the five wildflower species were sown in rows; 0.5 g of *D. sanguinalis* were sown. All herbicide formulations were pre-emergence granules, incorporated with sand as an inert carrier to ensure even distribution, and applied using a shaker can. Each treatment was replicated five times. Irrigation was supplied by overhead sprinkler following seed sowing and herbicide application. Plots were irrigated throughout the experiment when rainfall was less than 2.5 cm for any seven day period, as occurred on 1 July and 18 July, in order to maintain activation of the herbicides as well as to prevent germination failure due to lack of soil moisture.

Plots were seeded on 17 June 1994 and herbicide was applied the following day. Seedling emergence was counted twice a week for 45 days. Weed pressures mandated termination of count data after 29 July 1994. On 18 August 1994, all weed species were harvested by species, by plot and then bagged and labeled. Wildflowers were harvested in a similar manner on 22 August 1994. All vegetation was oven dried and weighed. Repeated measures analysis was performed on the count data collected in accordance with current statistical practices (Littell et al., 1991).

Treatment 1: Dacthal® 2G was applied at the manufacturer's recommended rate of 10.66 g per plot.

Treatment 2: Dacthal® 2G was applied at 21.32 g per plot, twice the manufacturer's recommended rate .

Treatment 3: Pennant® 5G was applied at the manufacturer's recommended rate of 1.60 g per plot.

Treatment 4: Pennant® 5G was applied at 3.20 g per plot, twice the manufacturer's recommended rate.

Treatment 5: Treflan® 5G was applied at the manufacturer's recommended rate of 1.07 g per plot.

Treatment 6: Treflan® 5G was applied at 2.14 g per plot, twice the manufacturer's recommended rate.

Treatment 7: No herbicide was applied. Plots were hand-weeded such that the impact of weed pressure on germination and growth of the wildflowers could be assessed.

Treatment 8: No herbicide was applied.

Greenhouse Experiment

The same wildflower species were tested for germination and growth responses to the same herbicides used for the field experiment. Twenty-five seeds of each wildflower species and crabgrass were sown separately into 10 cm diameter plastic pots filled with Sunshine® Mix #1 (3 peat: 1 perlite: 1 vermiculite/ v:v:v) (Fisons Horticultural Inc., Vancouver, B. C., Canada). Pots were sown and herbicide was applied on 15 February 1995; pots were randomly placed on greenhouse benches. Each treatment was replicated five times. Supplemental illumination at 3-4 $\mu mol \cdot m^{-2} \cdot sec$ was supplied daily from 1730 to 2100 hours by overhead incandescent lighting in order to provide long day conditions that encourage vegetative growth of all species. Seedling emergence was recorded twice weekly through 18 April 1995. Repeated measures analysis was performed on the germination data collected in accordance with current statistical practices (Littell et al., 1991).

Treatment 1: Dacthal® 2G was applied at the manufacturer's recommended rate of 0.278 g per pot.

Treatment 2: Dacthal® 2G was applied at 0.555 g per pot, twice the manufacturer's recommended rate.

Treatment 3: Pennant® 5G was applied at the manufacturer's recommended rate of 0.042 g per pot.

Treatment 4: Pennant® 5G was applied at 0.084 g per pot, twice the manufacturer's recommended rate.

Treatment 5: Treflan® 5G was applied at the manufacturer's recommended rate of 0.028 g per pot.

Treatment 6: Treflan® 5G was applied at 0.056 g per pot, twice the manufacturer's recommended rate.

Treatment 7: No herbicide was applied.

Results

Field

Few significant results were obtained from the field study. Two main weed species, *Chenopodium album* and *Solanum ptycanthum*, became the dominant vegetation in the majority of plots. *Amaranthus hybridus* was also prevalent. The total dry weight for *S. ptycanthum* was less in plots treated with metolachlor than for other treatments, but there was no statistical significance in differences among herbicide treatments. Mean dry weights of *A. hybridus, C. album*, and *S. ptycanthum* are listed in Table 3.1. Weed pressures forced termination of data collection after 29 July 1994 .

Germination response of the wildflower species was likewise variable. *M. fistulosa* demonstrated very poor emergence, germinating in only three of the five hand-weeded plots and in only one of the control plots. Neither *A. canadensis* nor *A. novae-angliae* germinated during the data collection period. One obvious qualitative impact on *A. tuberosa* was noted in the plots treated with metolachlor. The plants emerged and developed a twisted, curled appearance within one week after germination. No injury to wild lupine was observed.

Greenhouse

Herbicide treatment had a significant effect on germination (Table 3.2). *Digitaria sanguinalis* was controlled by all three herbicides (data not shown). There were obvious qualitative differences in the germination and growth response of the individual wildflower species to the three herbicides. One consistent response across species to

DCPA was retarded and/or no shoot elongation, and was most pronounced for *A. tuberosa* (Fig. 3.2). *Aster novae-angliae* was also visibly affected by DCPA (Fig. 3.3). Metolachlor suppressed or prevented germination in all species. *L. perennis* was much less affected than other species; by 18 March 1995, there was no longer a significant herbicide effect for this species (Table 3.3). There were qualitative effects of metolachlor on development of lupine; leaf-seal was evident on a few plants which is consistent with symptoms resulting from the use of this herbicide (Hatzios, pers. comm., 1994).

Trifluralin produced stunted roots and lower stem swelling on all species tested in this experiment. This form of damage is characteristic for species sensitive to the herbicide (Ross and Lembi, 1985). The symptoms were particularly pronounced for *A. tuberosa* and *L. perennis* (Fig. 3.4). By the conclusion of the experiment, it was noted that those plants which were not killed by trifluralin began to recover from the effects of the herbicide.

Discussion

The three herbicides tested are considered most effective in controlling annual grassy weeds (Ross and Lembi, 1985). While metolachlor is reported to suppress black nightshade, and DCPA, metolachlor, and trifluralin to control common lambsquarters (Ross and Lembi, 1985), the statistical analysis for this experiment does not bear out the assertions. When considering the use of any herbicide treatment, the initial step lies in site assessment of the weeds that are dominant (Ross and Lembi, 1985). In all, 14 weed species were harvested from plots used in this study. Even though berms and ditches were constructed for each plot to minimize the possibility of runoff on this sloping site, several heavy thunderstorms occurred during the course of the experiment. The possibilty exists that some of the variability in weed density was a result of lost herbicide in some plots. Should this be the case, the conclusion remains that herbicides which could be translocated in this manner from the intended site of action may not be the most appropriate means of weed control in meadow plantings.

Given the relatively high cost of native Virginia wildflower seed, combined with inherently low germination rates for some species, any herbicide which depresses germination and subsequent development of seedlings must be deemed unacceptable for use in establishing wildflower plantings. Of the three herbicides tested, all resulted in major injury to one or more of the species included in this study. DCPA, metolachlor, and trifluralin are therefore unsuitable as pre-emergence treatments in meadows composed of native perennial species that are direct-seeded. Further research is needed to determine whether it may be appropriate to use these herbicides in established plantings for weed control. Another avenue of inquiry would be in assessing the effectiveness in applying these herbicides at the time of transplant installation in conjuction with the Virginia Tech Transplanted Meadow system (Laurent, 1994).

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Table 3.1 Mean dry weights of the three most prevalent weeds in field plots treated with pre-emergence herbicides.

Treatment	Mean Dry Weight of Weeds in Grams (SE)		
	Amaranthus hybridus	*Chenopodium album*	*Solanum ptycanthum*
Dacthal 1X	53.7 (±53.7)	79.8 (±47.1)	173.9 (±58.2)
Dacthal 2X	10.8 (±10.8)	53.9 (±53.9)	112.8 (±42.3)
Pennant 1X	5.3 (±3.9)	137.6 (±103.5)	67.0 (±25.7)
Pennant 2X	0.0 (±0.0)	106.0 (±49.1)	32.4 (±21.2)
Treflan 1X	17.1 (±13.6)	130.3 (±83.3)	153.3 (±61.8)
Treflan 2X	3.9 (± 3.9)	58.2 (±32.9)	307.2 (±164.0)
Control	66.6 (±66.6)	167.5 (±50.9)	99.0 (±18.9)

Table 3.2 Germination of five native Virginia wildflowers as affected by Dacthal® 2G, Pennant® 5G, and Treflan® 5G in a greenhouse study.

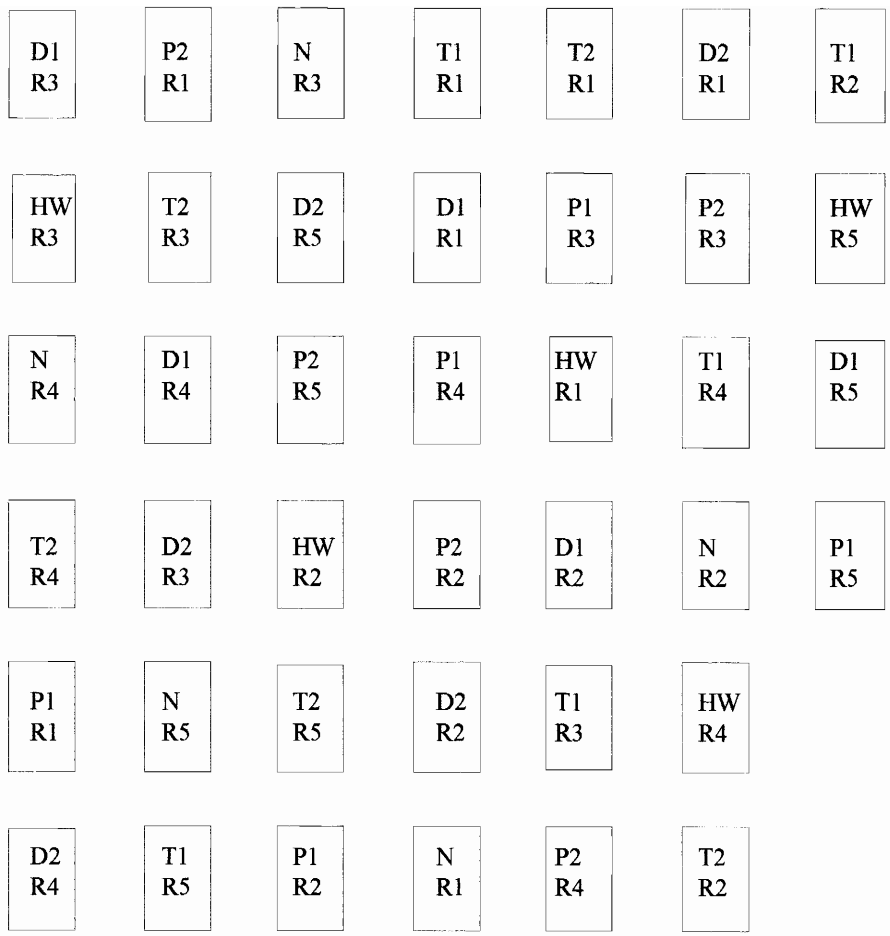
Species	Treatment, Pr > F[z]
Aquilegia canadensis	0.0001
Asclepias tuberosa	0.0001
Aster novae-angliae	0.0001
Lupinus perennis	0.0253
Monarda fistulosa	0.0001

[z] GLM repeated measurements analysis of variance at a 95% confidence level for tests of hypotheses for between subjects effects, where Treatment is the dependent variable (Littell et al., 1991).

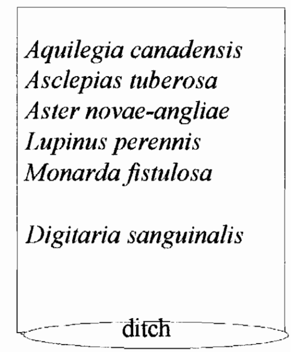
Table 3.3 Germination contrasts between time intervals of five native Virginia wildflowers as affected by Dacthal® 2G, Pennant® 5G, and Treflan® 5G in a greenhouse study.

	Germination Response Between Days; Pr > F[z]				
	25 F[y]:4 M	4 M:11 M	11 M:18 M	18 M:25 M	25 M:1 A
Aquilegia canadensis	.	0.0001	0.0094	0.0011	0.0031
Asclepias tuberosa	0.0023	0.7649	0.0001	0.1178	0.0001
Aster novae-angliae	0.0062	0.2878	0.0006	0.5936	0.7368
Lupinus perennis	0.1982	0.4270	0.5938	0.0147	0.0001
Monarda fistulosa	0.0003	0.1641	0.2396	0.1676	0.8578

[z]Day is the contrast variable with regard to Treatment in GLM procedure repeated measures analysis of variance (Littell et al., 1991).
[y]F=February; M=March; A=April



Detail of individual plots:



Key to plot designations:
D1: dacthal, 1X rate
D2: dacthal, 2X rate
P1: pennant, 1X rate
P2: pennant, 2X rate
T1: treflan, 1X rate
T2: treflan, 2X rate
HW: hand-weeded
N: no treatment

Figure 3.1 Plot layout for field herbicide experiment.

Figure 3.2 Influence of Dacthal® 2G on shoot elongation for *Asclepias tuberosa* (top) as compared to control (bottom).

Figure 3.3 Influence of Dacthal® 2G on shoot elongation for *Aster novae-angliae* (top) as compared to control (bottom).

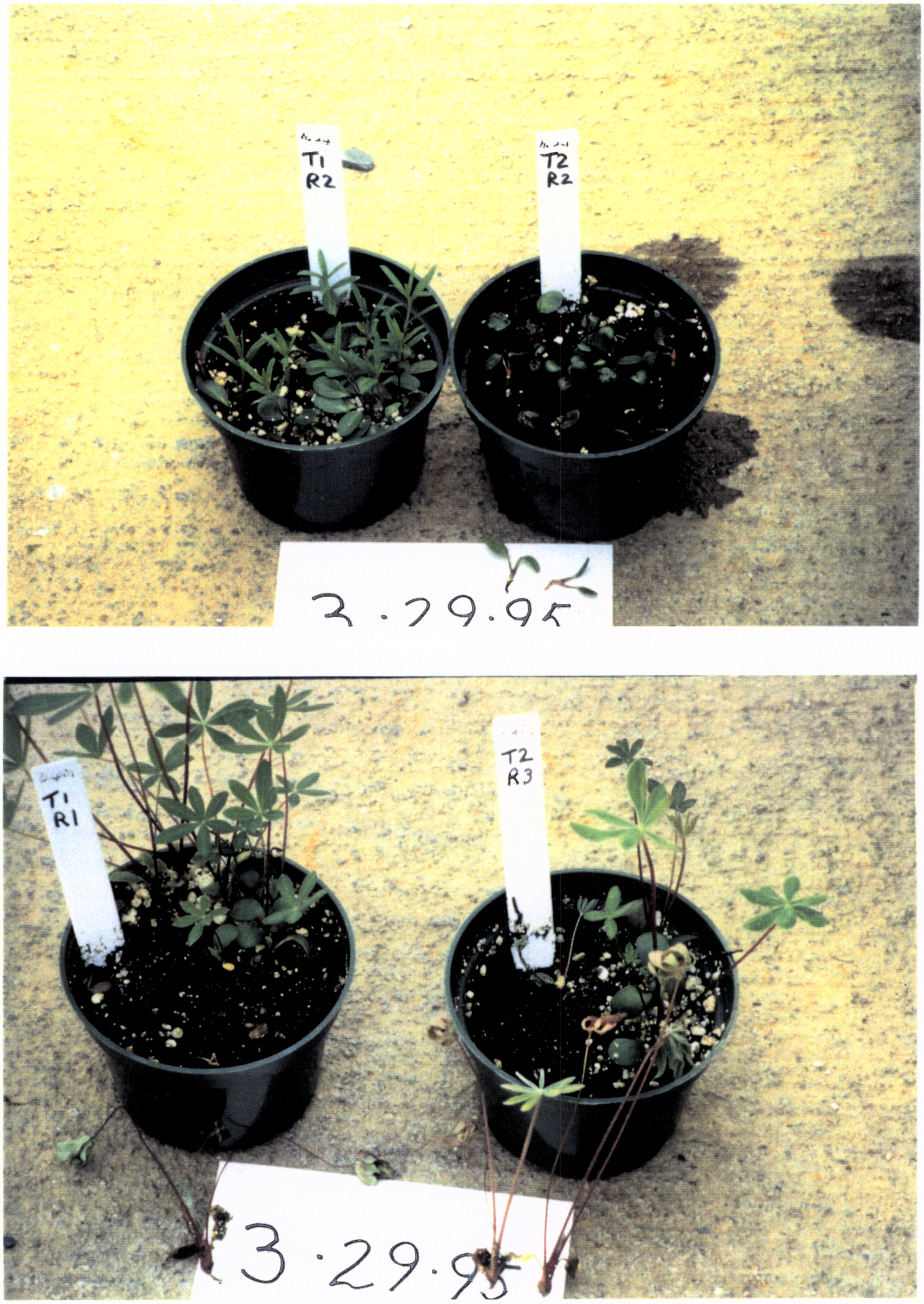


Figure 3.4 Influence of Treflan® 5G on root inhibition for *Asclepias tuberosa* and *Lupinus perennis*.

Chapter 4

Spring-Seeded Nurse Crop

Abstract. Direct seeding of native Virginia wildflowers in the spring is not a viable means for stand establishment. This study was conducted to evaluate the possibility of using a spring seeded nurse crop to improve soil conditions for subsequent installation of a wildflower meadow. The nurse crop was successfully established as determined by leaf area index values higher than 3.0 within 30 days of seeding. No weed species were found within the nurse crop plots. Soil analysis and quantitative evaluation of the fall planted wildflowers failed to differentiate the influence of the nurse crop on growth from the fallow and weedy treatments. In addition, the nurse crop was found to be lacking in ornamental value. The approach may yet warrant further investigation, but the results of this study indicate that the nurse crop used did not perform as desired.

Direct seeding of native Virginia perennial wildflowers in the spring, unaccompanied by supplemental irrigation or fertilization, is an unacceptable method for achieving acceptable stand establishment (Tuttle, 1995, Ch.2, p.35), as demonstrated in field germination tests. A nurse crop has been defined as a fast growing crop planted with the intended crop, but which does not persist within the crop for a long period of time (Ross and Lembi, 1985). A slight alteration of this idea was used in the present study, where the nurse crop was planted ahead of the intended crop. The purpose of this study was to investigate whether a spring-seeded nurse crop could improve soil conditions for a subsequent wildflower planting. By selecting species for the nurse crop with certain properties (Table 4.1), some scientifically documented and others not, it was hypothesized that soil structure and nutrient levels might be enhanced to facilitate autumn establishment of a wildflower planting. In addition to providing tangible benefits, the nurse crop would also display a high level of ornamental value. Finally, nonpersistent species must comprise the nurse crop such that no competition with the wildflower crop would occur in the future.

Materials and Methods

A site was selected near the Virginia Tech Horticulture Gardens which lacked permanent vegetative cover. The area was rototilled in April 1994, treated with a 2% solution of glyphosate (Roundup®) on 17 May 1994, and subdivided into plots measuring 1 m^2. A composite soil sample was collected from the site on 25 May 1994 and sent to the Virginia Tech Soils Laboratory for analysis. On 9 September 1994, composite soil samples were collected from each treatment for analysis. Photographic records were taken periodically throughout the experiment (Figure 4.1). On 25 September 1994, plots were mowed to a height of 8 cm prior to installation of the wildflower crop according to the Virginia Tech Transplanted Meadow method (VTTM) (Laurent, 1994). Leaf area index (LAI) measurements were recorded for each treatment on 9 June and 7 July 1995. Date of first flower for each wildflower species was recorded for each treatment. All treatments were replicated four times.

Treatment 1: Proportions for each species of the nurse crop were determined according to recommendations of the National Wildflower Research Center (Paulson, 1989), and the seeds were weighed out and mixed together (Table 4.2). The seed mix was combined with sand which acted as an inert carrier to facilitate even distribution during sowing. The nurse crop was hand sown and lightly watered to ensure good seed-soil contact on 8 June 1994. No supplemental irrigation or fertilization was supplied throughout the growing season. LAI measurements were taken on a weekly basis from 7 July 1994 until 25 August 1994 using the LAI-2000 Canopy Analyzer (Licor, Lincoln, NE). Successful establishment of the nurse crop was defined as having an LAI of 3.0 or greater (Gardner et al., 1985) within 30 days after sowing. Date of first flower was recorded for each species in the mix. Weed species identities were recorded.

Treatment 2: Weed species were allowed to grow through the season. Periodic cutting back of the weeds was performed to prevent further loading of the weed seed bank. Weed species identities were again recorded.

Treatment 3: Glyphosate was applied as needed to maintain the plots in a fallow condition throughout the growing season.

Results

The successful establishment of the nurse crop was evidenced by initial LAI values greater than 4.0, surpassing the minimum value stated by Gardner et al. (1985); LAI reached a maximum on 4 August 1994. Shortly thereafter, a strong thunderstorm caused lodging of the nurse crop plants. While density did not reach previous maximum levels for the remainder of the growing season, LAI values remained above 3.0 (Table 4.3). The first species to bloom was *Brassica napa* on 12 July, followed by *Amaranthus tricolor* on 19 July . These two species were dominant throughout the experiment. Not until 2 August did *Cassia fasciculata* flower, followed by *Tagetes erecta* 'Crackerjack' on 10 August. *Nicotiana alata* produced only three blooms, the first of which opened on 22 August. *Foeniculum vulgare* failed to bloom. No weeds were found at any point in the growing season where the nurse crop was planted. *Brassica napa* has been found to reseed itself, but has appeared only in unmulched areas adjacent to the wildflower plantings and not within the plots.While visual interest is a subjective parameter, the nurse crop lacked any high degree of ornamental value.

Many weed species were present in plots maintained only through periodic cutting back. *Chenopodium album* was dominant, reaching a canopy height of over 2 m. Other species present in approximate order of abundance were *Solanum ptycanthum, Amaranthus hybridus, Portulaca oleracea, Galinsoga ciliata*, and *Datura stramonium* (Derr, pers. comm., 1994). Virtually no differences in nutrient levels or organic matter were detected among the three treatments by routine soils analysis (Table 4.4).

Visual inspection of the wildflowers installed among the three treatment areas yielded no discernable differences throughout the winter and early spring (Figure 4.2). The first species to flower, *Aquilegia canadensis* on 13 May 1995, did so only in one replicate where the nurse crop had preceded. By this time, the wildflowers in the nurse crop plots were taller and denser than the other two plantings as judged by visual inspection. The wildflowers in the area that had been maintained in a fallow state through the previous growing season appeared less green and sparse. In the weedy treatment area, the weeds competed with the wildflowers such that the wildflowers were being overtopped by 25 May. *Coreopsis lanceolata* first bloomed on 24 May 1995, followed by *Monarda fistulosa* on 25 June; both species first bloomed in plots preceded by the nurse crop. LAI values on 9 June and 7 July 1995 indicated no difference among treatments; however, LAI data do not differentiate between desired and invading species (Table 4.5).

Discussion

The enhanced visual appearance of wildflowers found within the plots treated by installation of a nurse crop suggests that this form of site preparation may be worth researching further. The particular mix used in this experiment was lacking in at least two respects. With the exception of one odd *A. tricolor* plant which did display ornamental value (Figure 4.3), the nurse crop plantings were in fact, unattractive. The more ornamental species such as marigold and flowering tobacco were almost completely dominated by amaranth and rape. Thus, the proportions of the species used would need to be altered. This imbalance in species distribution was the second problem, as the nurse crop failed to achieve the appearance of a flowering mix which was the desired result for this experiment. While nurse crops are employed for turf and cropping situations, their use in ornamental landscaping is an area which needs exploration.

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Table 4.1 Non-persistent species tested in a mix as a nurse crop for native Virginia wildflower mixes.

Genus species height	common name -attribute	family
Amaranthus tricolor 60 cm	Amaranth - deep rooted	Amaranthaceae
Brassica napa 60-90 cm	Rape - deep rooted; bee crop	Brassicaceae
Cassia fasciculata 45 cm	Partridge pea - nitrogen fixation	Fabaceae
Foeniculum vulgare 60-120 cm	Bronze fennel - beneficial insects; mycorrhizal	Apiaceae
Nicotiana alata 90 cm	Flowering tobacco -beneficial insects; mycorrhizal	Solanaceae
Tagetes erecta 'Crackerjack' 45-90 cm	African marigold - nematode repellent	Asteraceae

Table 4.2 Species and proportions used in formulating the nurse crop which preceded the native Virginia wildflowers planting.

Species	Proportion	Seeding rate ($g \cdot m^{-2}$)
Amaranthus tricolor	25%	4.0
Brassica napa	25%	4.0
Cassia fasciculata	10%	8.0
Foeniculum vulgare	10%	2.0
Nicotiana alata	10%	0.5
Tagetes erecta	20%	2.0

Table 4.3 Mean Leaf Area Index (LAI) values recorded for the nurse crop, 1994 growing season.

Date	LAI
7 July	4.52
14 July	4.76
21 July	5.27
28 July	5.77
4 August	5.85
11 August	4.10
18 August	3.35
25 August	3.55

Table 4.4 Soil analysis data from samples taken 25 May 1994 and 3 October 1994 from site of nurse crop experiment.

		ppm		%				
Collection date	pH	P	K	Ca	Mg	Zn	Mn	OM [z]
Initial/ 25 May	6.1	60	157	1200	120	6.1	8.5	3.9
Nurse Crop/ 8 Oct	6.1	60	157	1200	117	6.1	6.9	4.1
Fallow/ 8 Oct	6.2	60	157	1200	116	6.1	6.8	4.1
Weedy/ 8 Oct	6.4	60	157	1200	120	6.1	6.1	4.1

[z] Organic matter.

Table 4.5 Influence of Nurse Crop, Fallow, or Weeds on Leaf Area Index (LAI) values of subsequently planted native Virginia wildflower mix. [z]

Source	F-value	Pr > F [y]
Treatment	1.32	0.29
Month	313.71	0.0001
Treatment*Month	0.72	0.50

[z] Treatment: Nurse Crop, Fallow, Weeds; Month: 7 June, 9 July 1995.
[y]Probability at 95% confidence level.

Figure 4.1 Appearance of the nurse crop in June (top) and August (bottom) 1994.

Figure 4.2 Appearance of wildflowers after installation on 6 October 1994 (top) and on 24 April 1995 (bottom).

Figure 4.3 Lack of nurse crop ornamental quality aside from a single *Amaranthus tricolor* plant.

Chapter 5

Field Establishment of Native Virginia Wildflowers

Abstract. Wildflower meadow establishment in the fall using native Virginia perennial species was studied for four treatments. Direct-seeded plots failed to produce more than a few plants of a couple of species. Transplants installed in the field using the Virginia Tech Transplanted Meadow (VTTM) technique demonstrated acceptable levels of winter survival, competition against weed species, and sufficient canopy growth the following spring. Not all species included in two wildflower mixes are evident in the field, leading to the conclusion that a more productive mix might be formulated by combining the most vigorous species from the two mixes into a single one. The VTTM method for wildflower stand establishment of nativeVirginia perennial species is a successful strategy for meadow production.

Field establishment of extensive wildflower meadows is analogous to the production of agronomic stands in many respects. Horticulture is ordinarily concerned with the intensive management of relatively high value crops per area, whereas agronomy is usually directed towards the large-scale production of crops such as hay, corn, wheat, and soybeans. One measure of successful crop establishment is leaf area index (LAI), which is a ratio of the area of photosynthesizing surface divided by the soil area occupied by the crop (Mengel and Kirkby, 1987). Broadleaved crops are said to be maximizing available solar irradiation when LAI values range between 3.0 and 5.0; that is, maximum dry matter production occurs at this level of stand density (Gardner et al., 1985). The species used in the present study all possess leaves which are oriented in a horizontal plane; thus, a minimum LAI value of 3.0 was a criterion for successful stand establishment.

The common technique for wildflower meadow establishment is direct-seeding . Failure to obtain vigorous stands may result from competition by more aggressive weed species, improper seeding times, and/or persistent seed dormancy (Salac et al., 1982). Another problem with direct-seeded wildflowers on a large scale is the prohibitive cost of seed for some species (Airhart et al., 1983). In the case of wildflower mixes, proper seeding dates may be especially troublesome to pinpoint due to variability in the germination response to temperature (Kaspar and McWilliams, 1982). Little information is available for the germination requirements of many wildflowers, and the majority of data extant are derived from greenhouse or laboratory experiments (Salac et al., 1982).

Methods for wildflower establishment are largely unavailable for several native Virginia perennial species such as *Rudbeckia triloba, Solidago speciosa, Teucrium canadense,* and *Veronicastrum virginicum.* (Airhart et al., 1983; Martin, 1986, Paulson, 1989; Smith and Smith, 1980). Art (1987) discusses direct seeding, as well as plug and sod production, but no information is provided concerning which method is most appropriate for any given species.

Airhart et al. (1983) investigated the use of wildflower sods as a means of assuring better stand establishment. Of the 13 species tested in this experiment, only two were native Virginia perennials: *Asclepias tuberosa* and *Liatris spicata.* Single species sods were produced and no mixes were used. Suitable sods for the two Virginia native wildflowers could only be achieved by seeding a soilless mix at twenty times the seed manufacturer's recommended rate. After one year in the field, no *A. tuberosa* or *L. spicata* survived.

A more promising establishment technique, the Virginia Tech Transplanted Meadow (VTTM), was developed at Virginia Polytechnic Institute and State University working with annual species (Laurent, 1994). The method utilizes up to 12 species combined together in equal weights, to which sand is added to facilitate even seed dispersal. Flats filled with a soilless medium are then seeded by hand broadcasting. The wildflower mix is grown in the greenhouse for a maximum of one month before transplanting to the field. Preparation of the field site consists of applying glyphosate (Roundup®) in April, rototilling 7 to 10 days later, and making a final application of glyphosate again in late May, since lack of preplant weed control has been found to result in "a midsummer weed patch" (Laurent, 1994, p.18). Plugs are transplanted into the field in late May or early June and spaced on a minimum of 30 cm centers. The transplants are then mulched to a depth of 7.5 cm with hardwood bark to inhibit additional weed growth, as well as to provide an aesthetically pleasing early season appearance.

Materials and Methods

The native Virginia wildflowers used in this study are all perennial. Data from previous germination trials and guidelines for formulating wildflower seed mixes published by the National Wildflower Research Center (NWRC) (Paulson, 1989) were utilized in formulating two mixes for field establishment testing. Formulas and mix components are listed in the Addendum and Table 5.1, respectively. The mixes are site interchangable; one is a moderate height composition, while the other is composed of tall species. Figure 5.1 lists the bloom sequence of individual species through the growing season and provides an indication of overall bloom period for each mix.

Direct- seeding in the spring was judged to be an ineffective technique for stand establishment for the present research (Tuttle, 1995, Ch. 2, p. 35). Two primary approaches were taken for this experiment: direct-seeding in the fall, and fall production of VTTM plugs. Several species which were not noted to have a photoperiod requirement (Deno, 1993) were germinated under dark conditions in an earlier experiment (Tuttle, 1995, Ch. 2, p. 29). The data indicated an improved germination response by these species under the higher light conditions available in the glass greenhouse as compared to their response in the fiberglass greenhouse. Whether the improved germination resulted from light or some other factor is not known. Thus, both the Moderate Height and Tall Mixes were sown on top of the medium. Species which failed to germinate under any previous treatments were nonetheless included in the field establishment trials as it was considered that winter stratification might serve to break dormancy and produce germination the following spring.

Four treatments of four replications were tested on a site at the Virginia Tech Horticulture Gardens lacking permanent vegetative cover. Site preparation consisted of mowing existing vegetation in the third week of September followed by rototilling to a depth of 15 cm. Thirty-two plots of 4 m^2 were marked off and lightly raked to smooth the soil surface. All treatments were installed on 6 October 1994; plots were labelled and a diagram of the layout was recorded (Fig. 5.2). Plots were watered using overhead irrigation for three hours immediately following installation. Data were recorded with regard to species present in each mix at installation and following winter dormancy. Invading weed species were identified and recorded. Date of first flower for each wildflower species was recorded. Leaf area index (LAI) was recorded on 9 June 1995 and again on 7 July 1995. Photographic records were taken periodically.

Treatment 1: Mixes were formulated according to the previously mentioned guidelines and sown into #801 cells (Kord Products, Toronto, Ontario, Canada) filled with Sunshine® #1 Mix (3 peat: 1 perlite: 1 vermiculite/ v:v:v) (Fisons Horticulture, Inc., Vancouver, B. C., Canada) in accordance with VTTM specifications on 29 July 1994. Because of the relatively slower development of perennial species as compared with annuals, the cell pack growing time was projected to continue for two months in contrast to the usual VTTM guidelines of 30 days. The two mixes were sown into separate flats and placed in a fiberglass greenhouse equipped with an automatic intermittent mist system at the Virginia Tech Greenhouse Range. After 10 days, the flats were moved on 8 August 1994 to another fiberglass greenhouse where irrigation was provided by hand on an as-needed basis. Flats were fertilized with 400 ppm N/330 ppm P/330 ppm K on 15 September, 22 September, and 6 October 1994. In order for the transplants to acclimate before installation into the field, flats were moved to an outside structure covered with 33% shade cloth on 12 September 1994, where irrigation continued to be supplied using a hose and breaker on an as-needed basis.

Transplant cells were installed in the field on 6 October 1994 using 30 cm center spacing as prescribed by the VTTM method. Plots were completely randomized. Hardwood bark mulch was applied to a depth of approximately 7.5 cm.

Treatment 2: All procedures for treatment 1 were duplicated except that no mulch was applied to the plots.

Treatment 3: Native Virginia wildflower mixes were direct-seeded by hand sowing and mulched with hardwood bark to a depth of approximately 7.5 cm.

Treament 4: Native Virginia wildflower mixes were direct-seeded by hand sowing; no mulch was applied.

Results

Species emergent in the greenhouse on 8 August 1994 for the Tall Mix (#1) were *Asclepias tuberosa, Aster novae-angliae, Baptisia australis, Liatris spicata*, and *Rudbeckia triloba*. No additional species were evident through the remainder of 1994. On the same date, *Coreopsis lanceolata, Lupinus perennis*, and *Monarda fistulosa* had emerged in the Moderate Height Mix (#2). *Aquilegia canadensis* and *Aster laevis* were evident by 3 September 1994; no further species emerged through the remainder of 1994.

Transplant survival in the fall of 1994 was deemed excellent, with no plant death detectable from photographic records and visual inspection prior to the first frost date. The official temperature reading nearest to Blacksburg was 29^0F recorded on 11 November 1994 in Christiansburg (Wooge, 1994). Seedling emergence in direct-seeded plots was virtually non-existent; one seedling of *Lupinus perennis* was found in three separate Treatment 4 plots on 20 October 1994. No seedling emergence was detected through 15 May 1995 in Treatment 3 or 4 plots. Survival of transplants through the winter was judged to be excellent according to visual inspection and photographic records which were compared to those taken in the fall of 1994 (Fig. 5.3). *Aquilegia canadensis* was the first species to bloom; this was noted on 8 May 1995 in plot 2TMR4 (Fig. 5.4). The first dates of bloom for *Coreopsis lanceolata, Monarda fistulosa, Rudbeckia triloba,* and *Aster novae-angliae* were 26 May, 22 June, 27 June, and 3 July 1995, respectively (Fig. 5.4 and 5.5). No other species had emerged as of 12 July 1995.

Weed species apparent through the winter and spring of 1995 were *Barbarea* *vulgaris, Cerastium vulgatum, Glechoma hederacea, Lamium amplexicaule, L. purpureum,* and *Stellaria media.* No suppression of wildflower growth and development could be detected by the presence of these weeds. Wildflower canopy height overtopped the weed cover by 11 April 1995 in all transplanted plots. As of 12 July 1995, the following additional weed species were or had been present in the research plots: *Acalypha virginica, Allium canadense, Ambrosia artemisiifolia, Chenopodium album, Chrysanthemum leaucanthemum, Cichorium intybus, Cirsium arvense, Conyza canadensis, Cyperus esculentus, Daucus carota, Digitaria sanguinalis, Echinochloa crus-galli, Erodium cicutarium, Galium aparine, Hibiscus trionum, Ipomoea purpurea, Lactuca serriola, Oxalis stricta, Plantago lanceolata, P. rugelli, Polygonum pensylvanicum, Rumex crispus, R. obtusifolius, Silene cucubalus, Taraxacum officinale,* and *Trifolium repens.* The research plot area was planted with annual meadow species in 1994; *Cosmos sulphureus, Coreopsis tinctoria,* and *Papaver rhoeas* were widely present among the perennial meadow species.

LAI data were recorded on 9 June and 7 July 1995 (Table 5.2). No difference was found between either the Moderate Height versus the Tall Mix or between the use of mulch versus the abscence of mulch when Seed Mix and Treatment were analyzed. The Tall Mix LAI values were significantly greater than the Moderate Height Mix when Seed Mix and Month of data recordation were analyzed.

Discussion

The two mixes formulated and transplanted into the field have performed well up to a point. The month of April was unusually dry in 1995, with no apparent damage to the wildflowers. The end of May and beginning of June have had more than the normal amounts of precipitation, again with no apparent negative impact on the wildflowers. Overall, there was no significant difference between the two wildflower mixes with regard to LAI values. However, when analyzed by means separation with regard to Month, the Tall Mix demonstrates significantly higher canopy density in July than the Moderate Height Mix . The explanation is that *Rudbeckia triloba* is the dominant species in the Tall Mix and did not attain its greatest vegetative growth until after the middle of June.The LAI values are indicative of successful stand establishment by 7 July 1995; however, LAI readings do not differentiate between desired species productivity and that of the weeds. Qualitative judgements suggest that the plantings that were mulched have resisted weed invasion better than those plots that were left bare. No qualitative growth or blooming suppression appears to have resulted from the presence of the annual meadow species as compared to plots where the annual species are less evident. While heaving of the transplanted plugs in the bare plots was observed through the winter, no plant losses or subsequent retarded growth could be detected as compared to the mulched plots. The area in which the trials are being conducted supports a large rabbit population. *Lupinus perennis* is evidently subject to herbivory, as many plants will flush a set of leaves that are not there at the next observation visit. By 7 July, no *L. perennis* plants could be found in any plots and there was no bloom produced in 1995. No insect or disease damage was noted through June 1995; Japanese beetle damage was evident but not severe by 7 July.

Survival and competitive ability of some species used in the native Virginia wildflower mixes is already apparent. The dominant species thus far are *Coreopsis lanceolata*, *Monarda fistulosa*, and *Rudbeckia triloba*. *Aster novae-angliae* has appeared primarily along the edges of the plots, suggesting that the taller *R. triloba* may be outcompeting the aster for sunlight (Fig. 5.6). Only limited conclusions can be drawn from the results of the field establishment experiment to date. Since several species have yet to make an appearance in the field, one can view the results in two ways. First, a more productive mix might be formulated by combining the best performing species from the two mixes into a single mix and discarding the remaining species. On the other hand, another season's worth of observation might be warranted before reaching a hasty conclusion. The literature mentions that *Tephrosia virginiana* will not bloom until the third season (Smith and Smith, 1980); there may be other species that may remain dormant for a year or more under field conditions. Two species that were evident at planting and up until frost 1994 have not emerged in 1995. *Asclepias tuberosa* and *Liatris spicata* both possess underground storage structures that may be constricted during the growth period in #801 cells and result in the eventual death of the plants over the winter. This would preclude the use of these species in a VTTM establishment procedure. Further research is necessary to determine whether deeper germination cells would improve survival. *Lupinus perennis* appears to have succumbed to herbivory.

Another approach that seems to merit further study is to formulate a mix of native Virginia perennials in combination with either American or exotic annuals that can be documented as non-persistent. One of the drawbacks to the mixes formulated for this research is that in the first year of establishment, early in the season, the visual interest depends solely on *Coreopsis lanceolata*. The addition of colorful annuals would provide a high level of ornamental value from the first season. As the perennial components mature and produce more flowers, the annuals would be disappearing from the mix.

The success of establishing a demand for native Virginia wildflower meadows depends on continued research and evaluation on a long term basis. No attempt was made during the course of this study to analyze costs of native species meadows versus those composed of naturalized and exotic species. That information could encourage VDOT or commercial landscapers to commit funds towards the establishment of native Virginia wildflowers. Another area of study that warrants research is the investigation into the mycorrhizal status of native species. One study located during the course of this research was conducted for *Baptisia australis* and *Liatris aspera* (Zajicek et al., 1987). The authors reported enhanced growth and drought tolerance for both species when inoculated by mycorrhizal fungi than when grown without the symbiosis. This could be of particular importance for wildflowers that are to be established on sub-optimal sites such as highway medians, landfills, or mine reclamation areas.

Native Virginia wildflowers show promise as a viable alternative to meadow establishment using exotic or naturalized species. Continued research will depend, in large measure, on the importance attached to encouraging the use of native species in public plantings by legislators and university educators in the Commonwealth. Focused public support for the incorporation of native species into highway beautification projects should be a primary mechanism to secure the funding that Virginia's wildflowers deserve.

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Table 5.1 Seeding rates for native Virginia wildflower mixes, derived from guidelines given in the National Wildflower Research Center Handbook (Paulson, 1989).

1. Tall Mix

Species	Proportion	Seeding Rate ($g \cdot m^{-2}$)
Asclepias tuberosa	20%	0.160
Aster novae-angliae	10%	0.020
Baptisia australis	20%	0.406
Liatris spicata	15%	0.179
Rudbeckia triloba	20%	0.160
Solidago speciosa	5%	0.030
Teucrium canadense	5%	0.025
Veronicastrum virginicum	5%	0.013

2. Moderate Height Mix

Species	Proportion	Seeding Rate ($g \cdot m^{-2}$)
Aquilegia canadensis	10%	0.095
Aster laevis	5%	0.016
Coreopsis lanceolata	25%	0.315
Euphorbia corollata	10%	0.158
Lupinus perennis	20%	0.504
Monarda fistulosa	15%	0.047
Tephrosia virginiana	10%	0.257

Table 5.2 Impact of Seed Mix, Treatment, and Month of data recordation on Leaf Area Index (LAI) values for native Virginia wildflower mixes [z].

Source	F-value	Pr > F[y]
Seed Mix	1.38	0.25
Treatment	0.64	0.43
Seed Mix*Treatment	0.17	0.68
Seed Mix	4.33	0.05
Month	1.50	0.0001
Seed Mix*Month	0.85	0.36

[z] Seed Mix: Tall vs. Moderate Height; Treatment: Mulched vs. Unmulched plots; Month: 9 June vs. 7 July 1995.
[y] Probability at 95% confidence level.

1. Tall

Species	Apr	May	Jun	Jul	Aug	Sep	Oct
Asclepias tuberosa				***	***		
Aster novae-angliae					***	***	***
Baptisia australis		***	***				
Liatris spicata				***	***	***	
Rudbeckia triloba			***	***	***	***	***
Solidago speciosa					***	***	***
Teucrium canadense				***	***	***	
Veronicastrum virginicum			***	***	***	***	

2. Moderate Height

Species	Apr	May	Jun	Jul	Aug	Sep	Oct
Aquilegia canadensis	***	***	***				
Aster laevis					***	***	***
Coreopsis lanceolata		*	***	***	***		
Euphorbia corollata			***	***	***	***	***
Lupinus perennis	***	***	***	***			
Monarda fistulosa				***	***		
Tephrosia virginiana		***	***	***	***		

Figure 5.1 Bloom sequence for "Moderate Height" and "Tall" native Virginia wildflower mixes.

1SMR4	2SBR2	1TMR3	2TMR4	1SBR2	1TBR3	2TMR1
2SMR1	1SMR1	2TBR1	1SBR3	1TBR1	2TBR4	2SBR3
fallow	1SBR1	1SMR3	1TBR2	2TMR3	fallow	1TMR4
1SMR2	2SBR4	2TBR3	2SBR1	1TMR2	2SMR2	2TBR2
2SMR3	1SBR4	1TMR1	2TMR2	2SMR4	1TBR4	fallow

Key to Plot Designations:
1: Tall Mix; 2: Moderate Height Mix
S: seeded; T: transplanted
B: bare; M: mulched
Rx: replication

Figure 5.2 Plot layout of field establishment experiment.

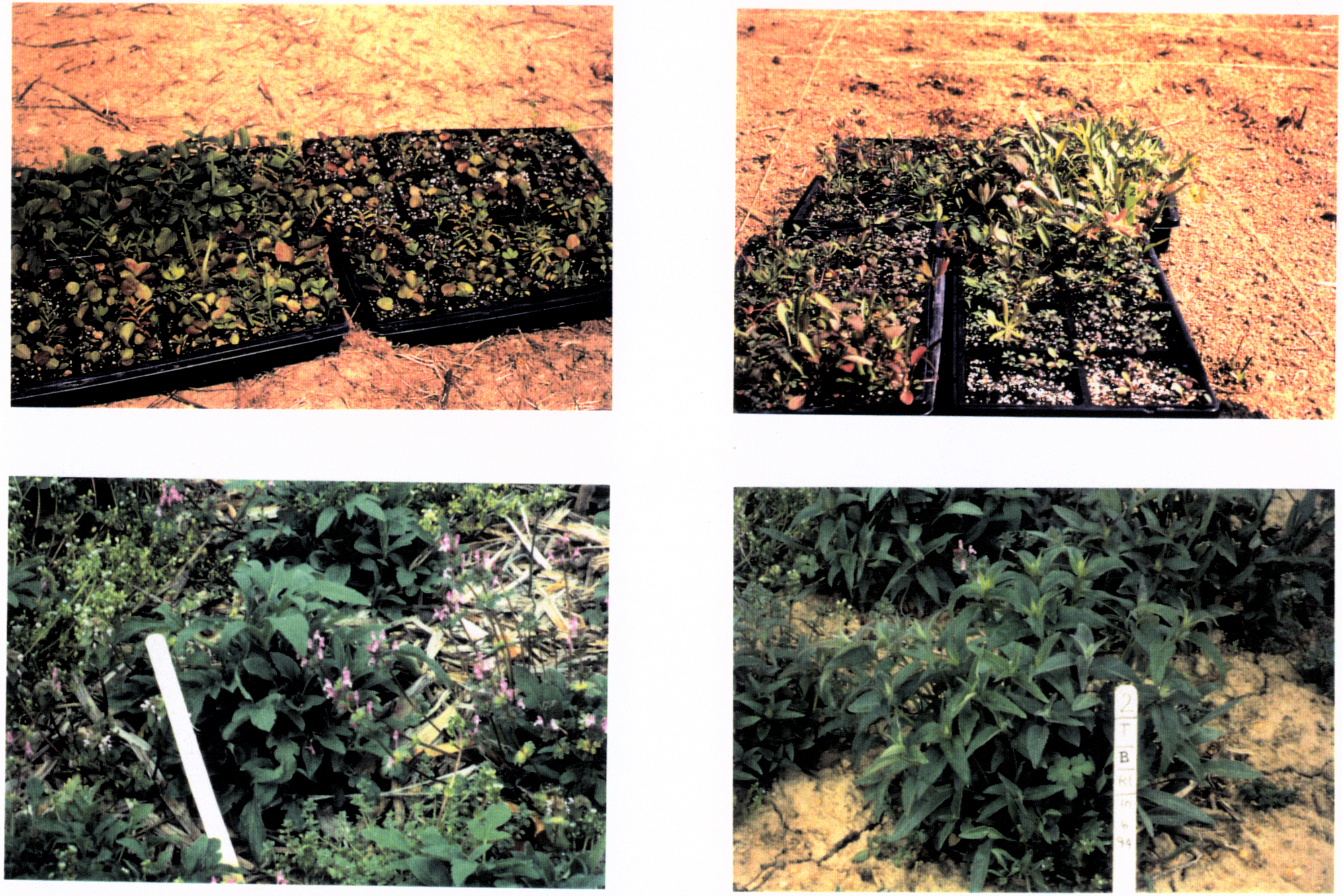

Figure 5.3 Appearance of wildflower mixes at installation on 6 October 1994 and on 24 April 1995.

Figure 5.4 First blooms for *Aquilegia canadensis, Coreopsis lanceolata,* and *Monarda fistulosa*.

Figure 5.5 First blooms for *Rudbeckia triloba* and *Aster novae-angliae*.

Figure 5.6 Appearance of *Aster novae-angliae* along plot margins.

Addendum

Seeding rate formulas for Virginia native wildflower mixes taken from the National Wildflower Research Center's Handbook (Paulson, 1989).

1. Calculate the number of seeds to plant per unit area as if that were the only species to be planted.

A (commercially recommended seeding rate) X B (number of seeds per gram) = C (number of seeds per unit area).

2. Add the total number of seeds per unit area obtained.

3. Divide the total number of seeds by the number of species being used.

C / #species = T (total number of seeds per unit area for this mixture)

4. Determine the percentage of the mix to be represented by each species.

% X T = # of seeds (for that species)

5. Convert the number of seeds per unit area for each species into weight per unit area. This is the seeding rate for each species in the mix.

(# seeds/area) / (# seeds/g) = g/area

6. Add the seeding rates obtained above to yield the total seeding rate for the mix.

VITA

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