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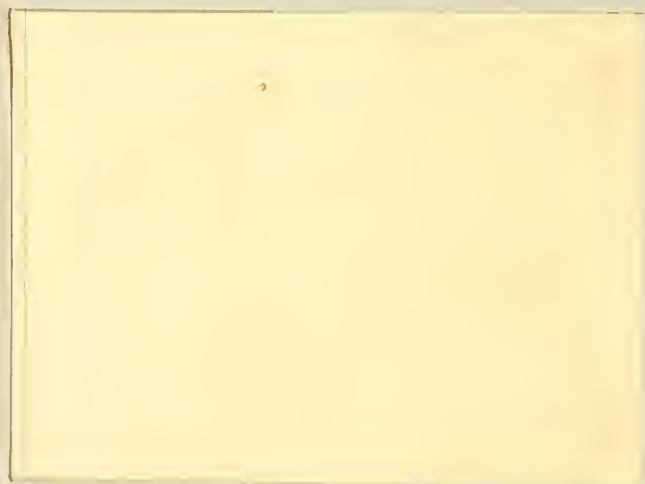
TECHNIQUES TO INCREASE SURVIVAL OF
NEW HIGHWAY PLANTINGS
(SEEDING PORTION)

P. L. Carpenter

J. B. Masiunas



PURDUE UNIVERSITY



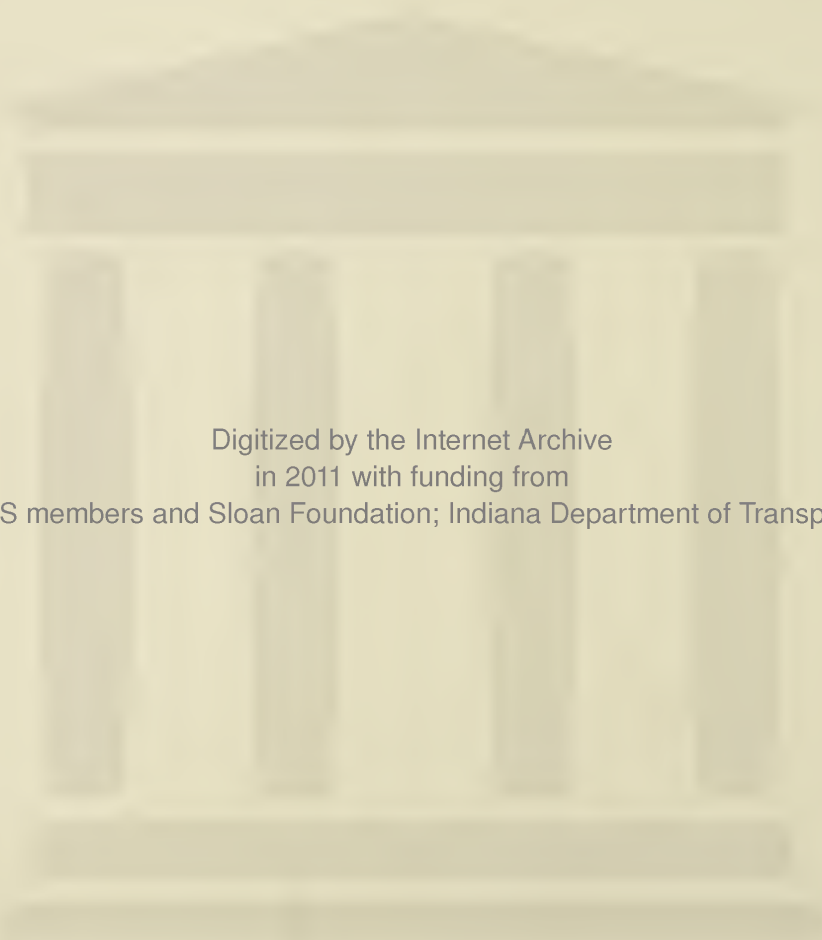
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FINAL REPORT
TECHNIQUES TO INCREASE SURVIVAL OF NEW HIGHWAY PLANTINGS
(SEEDING PORTION)

by

Philip L. Carpenter

and

John B. Masiunas

Department of Horticulture
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Joint Highway Research Project

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Prepared as Part of an Investigation

Conducted by

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in cooperation with the

Indiana Department of Highways

and the

U.S. Department of Transportation
Federal Highway Administration

The contents of this report reflect the views of the authors who are responsible for the facts and the accuracy of the data presented herein. The contents do not necessarily reflect the official views or policies of the Federal Highway Administration. The report does not constitute a standard, specification, or regulation.

Purdue University
West Lafayette, Indiana
October 26, 1982
Revised: May 17, 1983

Final Report

TECHNIQUES TO INCREASE SURVIVAL OF NEW HIGHWAY PLANTINGS
(SEEDING PORTION)

TO: Harold L. Michael, Director
Joint Highway Research Project

October 26, 1982
Revised: May 17, 1983
Project: C-36-48H

FROM: Philip L. Carpenter
Department of Horticulture

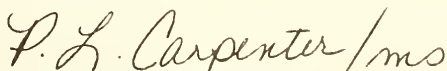
File: 9-5-8

Attached is the Final Report on the Seeding Portion of the HPR Part II Study "Techniques to Increase Survival of New Highway Plantings". The Report has been authored by myself as principal investigator and Mr. John B. Masiunas of our research staff in the Department of Horticulture.

Various seed mixtures were evaluated for effectiveness as to rapidity of establishment. Other factors evaluated were date of seeding and soil temperature at time of seeding and during establishment. The results are reported and provide good information for practical use by DOH. Mixtures containing warm season grasses and legumes performed the best - better than currently used highway mixes.

The report is submitted as a summary of the findings of the seeding portion of the study. Close contact by the investigators has been maintained with personnel of the roadside development area of DOH throughout the study and hence many of the results included in DOH roadside seeding plans for the future. The report is submitted for acceptance as fulfillment of the seeding portion of the study.

Sincerely,



Philip L. Carpenter
Department of Horticulture

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<p>16. Abstract Seeding studies were conducted on two roadsides and one farm site to evaluate the effectiveness of various seed mixtures on the rapidity of establishment. Fall seedings performed better than spring seedings with the latter having severe weed problems. In June seedings seed mixtures containing warm season grasses and legumes provided more rapid cover than cool season grass mixtures.</p> <p>A soil temperature study was conducted on north and south facing slopes using mulched and non-mulched plots. The maximum temperature observed was 51°C at 2 p.m. on July 13 on the south slope's surface. A straw mulch modified the temperature by an average of 5°C. The north facing slope averaged 5°C cooler than the southern exposure. Laboratory studies were conducted using pregerminated seed of 12 common herbaceous erosion control species. Negligible growth occurred at 47°C. Radicle growth was slight between 12 and 17°C. Optimal growth occurred at 22°C for <u>Lolium multiflorum Lam.</u> (annual ryegrass), at 27°C for <u>Bromus inermis Leyss.</u> (smooth brome grass), and at 32°C for <u>Panicum virgatum L.</u> (switchgrass), <u>Andropogon gerardii Vitman.</u> (blue bluestem), and <u>Lespedeza stipulacea Maxim.</u> (Korean lespedeza). In the other species optimal growth occurred over the 22-32°C range.</p> <p>The potential use of prairie vegetation was investigated. Influence of cold soaking treatments of seeds was compared for prairie species and standard highway species. In the second year after seeding, mixtures containing warm season grasses and legumes (especially <u>Medicago sativa</u> (alfalfa)) performed the best. The current and previously used highway mixes had very little growth, especially on plots which were not fertilized. Prairie forbs contributed only slightly to cover.</p>			
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HIGHLIGHT SUMMARY

Seeding studies were conducted on two roadsides and one farm site to evaluate the effectiveness of various seed mixtures on the rapidity of establishment. Fall seedings performed better than spring seedings with the latter having severe weed problems. In June seedings, seed mixtures containing warm season grasses and legumes provided more rapid cover than cool season grass mixtures. A presoaking treatment of the seed did not increase establishment. The effectiveness of mulches in improving seedling survival was also tested. Wheat straw performed best.

A soil temperature study was conducted on north and south facing slopes using mulched and non-mulched plots. The maximum temperature observed was 51°C at 2 pm on July 13 on the south slope's surface. Straw mulch decreased the temperature by an average of 5°C. The north facing slope averaged 5°C cooler than the southern exposure. Crownvetch regrowth after hoeing occurred only on the northern slopes.

Laboratory studies were conducted to investigate the effect of temperature on pregerminated seed of 12 common herbaceous erosion control species. In the experiments seedlings were grown for 48 hours, and the increase in radicle length was measured. In the first study, measurements were made at 5°C increments between 12 and 37°C and at 47°C. Negligible growth occurred at 47°C. Radicle growth was slight between 12 and 17°C. Optimal growth occurred at 22°C for annual ryegrass, at 27°C for smooth bromegrass, and at 32°C for switchgrass, blue bluestem, and Korean lespedeza. In the other species optimal growth occurred over the 22-32°C range. Radicle growth at 37°C was significant for C-4 warm season grasses, while for the C-3 cool season grasses no growth occurred.

Seedlings of annual ryegrass, Korean lespedeza and sideoats grama were grown in 12 hour alternating temperature regimes of 10/18°C, 18/26°C, 24/32°C, 30/38°C, and 36/44°C. The optimum radicle growth of annual ryegrass was at 24/32°C and 30/38°C, and for sideoats grama it was in the 30/38°C regime. Radicle growth of all species was stunted compared to that occurring at the constant temperatures of the previous experiment.

Growth of the same three species was also studied in 3 different concentrations of polyethylene glycol 20000 (4, 7 and 9 bars) at 22, 27, 32, and 37°C. Radicle growth rate decreased as the concentration of polyethylene glycol increased. Polyethylene glycol induced osmotic stress also caused a shift in temperature at which optimum growth occurred. Optimum temperature for radicle growth was increased from 22°C to 27°C for annual ryegrass and decreased for sideoats grama and Korean lespedeza from 27-37°C to 27-32°C.

A field study was conducted on a newly cut highway slope in Vermillion County, Indiana. The purpose of the study was to better understand the potential use of prairie vegetation as a method to reduce mowing, fertilization, and maintenance costs, as well as to achieve an aesthetically pleasing landscape.

Individual species and seeding mixtures were tested. Also the influence of cold soaking treatments of seeds was compared for prairie species and standard highway species.

Cold soaking of seeds improved early germination of seven prairie species and two highway grass species. Length of stratification significantly affected germination levels, with maximum germination for seven prairie species occurring after 8 to 16 weeks cold soaking.

Differences were seen in initial germination of three cold soaked test species containing legumes, rapid cover species, cool-season species, and

warm-season species. One week after seeding, numbers of seedlings per square meter were greatest for the two test mixtures containing cool-season species predominantly and either a tall prairie grass, switchgrass, or short prairie grasses, little bluestem and prairie dropseed. By the second and fourth week, numbers of seedlings per square meter of these two test mixes were not significantly different from the current and proposed highway mixes. However, germination of the prairie species test mix was lower than the other four mixes after two and four weeks. Fertilizer application at the time of seeding was detrimental to early germination of all five seed mixtures, but in the second year a significant increase in above ground biomass production was noted with the fertilizer addition.

In the second year after seeding, mixtures containing warm season grasses and legumes (especially alfalfa) performed the best. The current and proposed highway mixes had very little growth, especially on plots which were not fertilized. Prairie forbs contributed only slightly to cover. Overall weeping lovegrass, and the legumes alfalfa, Korean lespedeza, and birdsfoot trefoil looked very promising at this location after the second year.

INTRODUCTION

In establishing mixtures of herbaceous species along roadsides, seeding times do not always coincide with the optimal time for germination and growth (Duell, 1969). It is especially hard to successfully establish cover in late fall or in late spring to early summer. During both these periods germination may occur, but subsequent conditions are often unfavorable for continued growth. Temperatures are often extreme, especially for soil surface layers in which seeds actually germinate (Blaser et al., 1961). Moisture may also be limiting. Thus although conditions allowing germination may occur during late fall or late spring to early summer, environmental conditions must remain favorable for subsequent seedling growth (Curry, 1980).

Plants that will grow under favorable conditions should be selected for problem sites. As an example, there are two categories of grasses; cool and warm season, based on when their growth occurs. Warm season grasses could have promise during the critical late spring to early summer periods of seeding.

Inclusion of legumes in seeding mixtures is important for long term coverage and ease of maintenance. Commonly used legumes have a low maintenance requirement, but are also slow to establish (Wright et al., 1978). Thus, it is also important to select species whose growth is favored by the prevailing environmental conditions present.

Numerous studies have shown that seed mixtures result in more productive stands than cover produced from pure seeding of one species (Kulfiniski, 1957). The varied growth requirements of species in seed mixes due to differences in root quality, root position in the soil, periodicity of growth and environmental requirements may at least partially explain increased growth of mixtures. The better growth of grass-legume mixtures has been generally assumed to be a consequence of the ability of legumes to fix nitrogen, which is often the most limiting nutrient in soils along Indiana highways. Thus it has been recommended that nitrogen fixing species be included in seed mixtures (Carpenter and Hensley, 1977).

How rapid cover is achieved by a seed mixture depends on the germination and growth of individual species in the mixture (Henderlong, 1971). Germination in turn depends on such factors as genetic differences, seed size, pretreatment and environmental conditions after planting. Under similar conditions certain species such as ryegrass inherently show more rapid germination and seedling growth than other species such as creeping red fescue. A presoaking treatment of many cover species has been found to accelerate

germination. Environmental conditions will then determine the degree to which pretreatment will affect germination. Hydroseeding can provide initially favorable moisture particularly important for presoaked seed.

The objectives of these studies were to determine:

1. The ability of various mixtures of herbaceous species to establish during late fall or spring.
2. The effect of seed treatments, fertilization, and mulch on establishment of seeding mixtures.
3. The persistence of herbaceous vegetation along highways.
4. Temperature conditions present on north facing and south facing highway slopes during the summer in Indiana.
5. Radicle growth of common erosion control species in constant or alternating temperatures.
6. Effect of the interaction of temperature and polyethylene glycol-induced moisture stress on radicle growth of selected species.
7. To test the critical early establishment capabilities of new seed mixtures compared to formerly used and currently used mixtures of the Indiana State Highway Department.

FIELD STUDIES ON THE ESTABLISHMENT AND PERSISTENCE OF HERBACEOUS SPECIES

Abstract

Field studies were conducted at two sites to test the establishment and persistence of various seed mixtures when sown either in late spring or in the fall. Two seed treatments were used, dry seed and moist-chilled seed. In June seedlings, mixtures that contained warm season grasses and legumes (especially Medicago sativa (alfalfa)) performed the best. Above ground biomass production was better on the fertilized than the nonfertilized plots.

Fall seedings were better than spring seedings, with the latter having a severe weed problem. The seed treatment effect was not significant. The warm season-cool season grass mixture, when seeded in the fall, provided the least number of shoots the following spring; otherwise mixture effects were not consistent. Lolium multiflorum (annual ryegrass), Lolium perenne (perennial ryegrass), and Festuca arundinacea (tall fescue) or Festuca rubra (creeping red fescue) performed well in all mixtures. Above ground biomass peaked at the end of June, then declined.

A field study was conducted to determine the effect of mulch on seedling survival of late fall seeded mixtures. Straw increased survival in comparison with no mulch or to a hardwood mulch.

Materials and Methods

Experiment 1. The Establishment of Herbaceous Species

Mixtures During the Spring and Fall

Establishment experiments were conducted at two locations. The first was a research farm west of Purdue University, and the second was a west facing highway slope south of Linden in Montgomery County, on Indiana 231. The soil of the research farm was prepared during the beginning of October, whereas the

highway site was prepared by a contractor during the last week of October. At both sites the soil was harrowed, limed, and fertilized. Soil samples were taken after all seeding was completed.

The seed mixture components are listed in Table 1. One half of each mixture was stored dry inside a polyethylene bag at room temperature. The other half was placed in a beaker and mixed with .5 ml distilled water per gram of seed until thoroughly moistened. The moistened seed was placed inside a polyethylene bag and stored at approximately 5°C until seeding either 5 weeks or 1 week.

Seeding was conducted on October 25, November 5, and May 7, at Horticulture Farm, and November 1 and May 27 at Linden. The seed was hand-broadcasted, and then mulched with wheat straw at 3.3 t/ha. Erosion netting was applied on top of the straw mulch to hold it in place. Before the spring seeding at Linden, the soil surface was reworked with a rototiller due to slight gully and rill erosion.

The proposed methods of pelletizing seed to be able to force the seeds under the soil surface was deemed not practical due to the high cost of pelletizing the seed and the lack of equipment to force the pelletized seed into the soil. Also, the use of a "no-till" seeder was investigated. Such equipment has been used to rework highway median strips in Marion County and discussions with highway personnel indicated that frequent breakdown occurred and that they felt the use of such equipment on slopes was not practical. Therefore, in the experiments described, all seed was surface applied and covered with a mulch.

Table 1. Seed mixtures used in seeding at Horticulture Farm and Linden.

<u>Mixture 1: Legume-Cool Season Grass</u>	<u>kg/ha</u>	<u>% of mixture by seed number</u>
1. alfalfa	9.1	7.7
2. crownvetch	18.2	8.2
3. annual ryegrass	5.1	4.6
4. tall fescue	58.4	53.3
5. perennial ryegrass	<u>28.1</u>	26.2
	118.9	
<u>Mixture 2: Warm Season-Cool Season Grass</u>		
1. annual ryegrass	5.1	4.7
2. tall fescue	39.0	36.0
3. perennial ryegrass	11.2	10.6
4. Indiangrass	22.4	15.0
5. blue bluestem	22.4	16.3
6. switchgrass	11.2	11.9
7. prairie dropseed	<u>5.1</u>	5.5
	116.4	
<u>Mixture 3: Current Highway</u>		
1. annual ryegrass	11.2	2.3
2. creeping red fescue	28.1	11.3
3. Kentucky bluegrass	22.4	40.3
4. Kentucky bluegrass	22.4	40.3
5. perennial ryegrass	<u>28.1</u>	5.8
	112.2	
<u>Mixture 4: Former Highway</u>		
1. tall fescue	44.9	21.1
2. perennial ryegrass	28.1	13.5
3. Kentucky bluegrass	11.2	47.0
4. annual ryegrass	<u>39.3</u>	18.4
	123.5	

The fall seeded plots were checked on November 12 and no germination had occurred for either the November 1 seeding at Linden or the November 5 seeding at Horticulture farm. The number of seedlings with at least one leaf in an upright position within a randomly thrown $.1m^2$ quadrat were counted for the October 25 seeding at Horticulture farm. A final fall count for all seedlings was conducted on December 4. Counts were taken the following spring from the beginning of May through the beginning of June. Also, the May 7 seeding at Horticulture farm and the May 27 seeding at Linden were counted at 1, 2, and 4 weeks after seeding.

Once coverage reached a stage where counts were no longer feasible, fresh weights were used as a ranking method. Samples were taken by throwing an $.1m^2$ quadrat, then removing and weighing the above ground tissue. Sampling started on May 21 for the November and October seedings at Horticulture farm, and on June 4 at Linden for the November seeding. Shoot fresh weight were taken again on June 4 at Horticulture farm and at approximately monthly intervals thereafter until September. Fresh weights were obtained for the May 7 seeding at Horticulture farm on July 1 and August 23, but were discontinued because of substantial weed problems. For the May 27 seeding at Linden, shoot fresh weights were obtained on August 12 and September 12.

Because of weed problems in the spring seeding, and to simulate actual highway maintenance practices, the seedings at Horticulture farm were mowed to approximately 15 cm height during the last week of August. This mowing caused residual weed material to remain on top of the May 7 seeding.

Experiment 2. The Effects of Mulch Treatments on Late Fall

Performance of Seeding Mixtures

The experiment was conducted on a southwest facing slope at the Interstate 65 and Route 43 interchange immediately north of Lafayette. Four of the

mixtures were the same as those used at Linden and Horticulture farm. Two others used were suggested by the Indiana Highway Department for wildlife use (Table 2). The experiment was a randomized block design with three replications. Seeding was conducted October 7, 28 and November 6. Areas of the slope lacking vegetation were selected, and 1m^2 plots staked out. Preweighed fertilizer was applied at 880 kg/ha. After fertilizer application the seed mixtures were spread by hand over the surface of the plots. Then one of three treatments was applied: no mulch (control), hardwood bark ($.01\text{m}^3$ bark/ m^2), and straw (3.35 t/ha). Seedlings within a randomly tossed $.1\text{m}^2$ quadrat were counted at one week intervals until the first week of December.

Results and Discussion

Experiment 1. The Establishment of Herbaceous Species

Mixtures During the Spring and Fall

Horticulture Farm

Seed presoaking resulted in a negligible increase in the rate of germination and the number of seedlings obtained for either of the fall seedings. Presoaking also appeared to be detrimental to legume survival. Legumes germinated during late fall, but were killed by low temperature injury during the winter.

In November, the effect of seed treatment for the October seeding depended on the mixture (Table 2). The previously used highway mixture had more seedlings emerged than any other mixture. By December, no significant differences occurred between either mixtures or treatments. A large variability between replications occurred which may contribute to the finding of nonsignificance. Shoot counts for mixtures seeded in November were significantly different, with the previously used highway mixture containing the highest number of seedlings (Table 2). This was because it contained the highest percentage of annual ryegrass, a winter annual.

Table 2. Proposed seeding mixtures to be used on bridge projects for the benefit of wildlife.

<u>Mixture 6: Legume</u>	<u>kg/ha</u>	<u>% of mixture by seed number</u>
1. Korean lespedeza	8.9	1.8
2. Alsike clover	8.9	4.4
3. birdsfoot trefoil	8.9	3.7
4. Kentucky bluegrass	35.6	75.4
5. creeping red fescue	26.7	12.6
6. annual ryegrass	<u>9.0</u>	2.1
	98.0	

<u>Mixture 7: Cool Season Grass</u>	<u>kg/ha</u>	<u>% of mixture by seed number</u>
1. smooth bromegrass	13.4	1.7
2. orchard grass	8.9	4.8
3. Kentucky bluegrass	35.6	77.3
4. creeping red fescue	26.7	12.9
5. annual ryegrass	<u>13.4</u>	3.3
	98.0	

The percent establishment was higher for the October seeding than the November seeding (Table 3). In December the mixtures with the highest percentage establishment were those that contained the least amount of Poa Pratensis (Kentucky bluegrass). Kentucky bluegrass has a slower rate of germination than either ryegrass or Festuca spp (fescue). By May, percent establishment had decreased for the October seeding, due to the unsuccessful overwintering of many young seedlings. The November seeding had a low percentage of seedlings established by December, but an increase in the percent establishment occurred by the May 22 count as more seed germinated in early spring.

In spring, the number of seedlings from the dry seed treatment were significantly greater than for the presoaked seed treatment in the October seeding. Presoaking could have contributed to increased low temperature injury. Results from the mixtures seeded in November indicated that the warm season-cool season grass mixture had fewer seedling than any other mixture.

This was probably because it consisted of approximately half warm season grasses, whose germination due to low temperature is limited during early spring and late fall. The contribution of legumes to cover was negligible. In May, a further decline in the number of seedlings present from October seeding occurred, while the November seeding stabilized. An increase in the number of shoots for the October seeding occurred between May 22 and June 4. This appeared to result from mature plants tillering and not from an increase in the number of plants present.

Table 3. Percentage establishment of fall seedings on December 4 and May 22.

	<u>October seeding</u>		<u>November seeding</u>	
	Dec.	May	Dec.	May
Mixture 1	31*	10	2	13
Mixture 2	--**	--	1	6
Mixture 3	8	3	1	4
Mixture 4	22	9	4	7

* The percentage is the number of seedlings counted per the number of seed per square meter.

**Mixture 2 was not seeded in October.

Table 4. Mean shoot counts obtained at the Horticulture Farm seeding.

Sampling date**		<u>Seeding Date</u>											
		<u>10-25-79</u>			<u>11-5-79</u>				<u>5-7-80</u>				
		Mixture			Mixture				Mixture				
		1	3	4	1	2	3	4	1	2	3	4	
Nov. 12	wet	190a*	430ab	250a									
	dry	170a	75a	795b									
Dec. 4		1780	2055	2243	130a	70a	240a	455b					
May	wet	835a	1040ab	450a	740b	360a	960b	810b	295	150	180	245	
	dry	1030ab	2040b	1160ab									
June 6		1562	1712	1277	994b	599a	1130b	1005b	840	570	840	940	

** Where two means are given for a sampling date the treatment effect was significant. The May sampling dates were on May 8 for October's seeding, May 4 for November's seeding and May 15 for May's seeding.

* Means (shoot/m²) which are the average of three replications, within a sampling date and seeding date followed by a different letter are significantly different by the Duncan Multiple Range Test at the .05 level.

Table 5. Mean shoot fresh weight obtained from the Horticulture Farm seedings.

Sampling Date		<u>Seeding Date</u>											
		<u>10-25-79</u>			<u>11-5-79</u>				<u>5-7-80</u>				
		Mixture			Mixture				Mixture				
		1	3	4	1	2	3	4	1	2	3	4	
5-21		480a*	290a	625b	400	530	515	1445					
6-4		1075	700	1220	1220	870	950	1180					
7-1		1870	1990	2320	2050	2275	2840	2210	980	1420	1450	835	
8-12		950	1030	890	1240	1590	1790	1645	1785a	1345ab	920a	795a	
9-5		790	1250	790	1060	930	775	822					

* Means (g/m²) are an average of two seed treatments over three replications. Means within a seeding date and a sampling date followed by a different letter are significantly different using Duncan's Multiple Range Test at the .05 level.

Fresh weights of the shoots, shown in Table 5, were also used to determine mixture performance. On May 21, for the October seeding, the legume-cool season grass mixture and the previously used highway mixture had a greater shoot fresh weight than the current highway mixture. One explanation for this is that the current highway mixture is over eighty percent Kentucky bluegrass, which has a thinner blade than either tall fescue or ryegrass. Also, this mixture contained the least amount of annual ryegrass, which produced the most growth at this time. After May there were no statistically significant differences for either mixture. There was an increase in fresh weights with the peak in production occurring in early July. The only mixture containing warm season grasses was the warm season-cool season grass mixture whose performance was similar to the other mixtures.

Weed growth in the May seeded plots were extensive. The weeds, especially Chenopodium album (lambquarter) and Cirsium arvense (Canada thistle), competed detrimentally with the desired plants. Besides shading the young grass seedlings, the weeds also used nutrients and water thus making them unavailable to the desired species. Thus, weed growth contributed to a lower fresh weight production in the spring seeding compared to either of the fall seedings.

For the spring seeding on August 12, the shoot fresh weight of the legume-cool season grass mixture was significantly greater than for either the current highway mixture or the proposed highway mixture, although it was not significantly different than the warm season-cool season grass mixture. The only difference between the mixtures was that the legume-cool season grass mixture contained legumes while the others did not. However, legumes were not an appreciable part of coverage. Although spring is the most favorable time for seeding legumes, establishment is successful only if weeds are controlled.

The fall seedings were virtually weed free with a complete coverage of the soil surface. In September, when final fresh weight samples for the fall seedings were taken, those of the spring seeding were not obtained because of the extensive amount of dead weeds laying on its surface. Observations of these plots two years after seeding indicated that Coronilla varia (crownvetch) had established over a substantial proportion of the fall seeded plots.

Linden

At Linden, as for Horticulture farm, the fall seeding had better results than those obtained from the spring seeding. Most seedlings that established by the beginning of December were either annual ryegrass or perennial ryegrass. The contribution to cover by the legumes was negligible. Although alfalfa germinated, it was not present the following spring.

In December, the warm season-cool season grass mixture tended to have the smallest number of seedlings, probably because it contained about fifty percent warm season grasses. However, even this mixture prevented erosion. But, the fall and early winter of 1980 were unusually mild.

No significant differences in fresh weight occurred between mixtures in either the spring or summer. In the spring, there was an increase in the number of shoots because individual plants were larger. The percent establishment was similar to that of Horticulture farm's November seeding. In fresh weight samples the current highway mixture tended to be the lowest, probably because of the large amount of Kentucky bluegrass it contained. A peak in fresh weight production occurred on June 26, as tall fescue, perennial ryegrass, and annual ryegrass matured. Their inflorescences gave an unsightly appearance during the late sampling dates, but could easily be removed by mowing. Alfalfa had an increased contribution to coverage as the grass species matured.

Table 6. Mean shoot counts and shoot fresh weight production at the Linden highway seedings.

a. Shoot counts (shoots/m²).

Mixture	<u>November seeding</u> Sampling date			6-3	<u>May seeding</u> Sampling date	
	12-4	5-4	6-3		6-12	6-26
Mix 1	235*	585	475	290	2940	635
Mix 2	75	440	525	370	1580	720
Mix 3	245	545	505	165	1200	830
Mix 4	295	510	700	39	1965	850

b. Fresh weights (gm/m²).

Mixture	<u>November seeding</u> Sampling date				<u>May seeding</u> Sampling date	
	6-3	6-26	8-8	9-4	8-8	9-4
Mix 1	465*	655	540	500	430	660
Mix 2	575	955	550	580	315	440
Mix 3	330	550	420	550	385	615
Mix 4	510	870	370	360	460	475

*Means are the average value of two treatments over three replications.

No means within a sampling date were significantly different by Duncan's Multiple Range Test at the .05 percent level.

The results obtained from the spring seeding are also shown in Table 6. There were no significant differences for either treatments or mixtures for the first two sampling dates. The number of shoots peaked during the second week after seeding. By later sampling dates the temperature had increased, the soil was drier, and plant size was larger, causing competition to be greater, and the number of plants to decline. On June 26, the number of shoots present for

dry seed treatments were greater than those for moist treatments, possibly because either more dry seed germinating over a longer time period or from the wet seeded plots declining earlier.

A major weed problem occurred in the May seeding. The major weed species present were lambsquarter and Polygonum hydropiper (marshpepper smartweed). These weeds competed with the grasses and legumes, especially in the replications where soil fertility was higher.

THE MODIFICATION OF SLOPE SOIL TEMPERATURE BY
MULCH AND VEGETATIVE GROWTH

Abstract

A soil temperature study was conducted on north and south facing slopes along I-74 at Veedersburg, Indiana. Soil temperatures were taken at the surface and an 8-12 cm depth on bare soil and mulched plots. The maximum temperature, 51°C, was observed on July 13 on the surface of the south slope. A straw mulch modified the temperature. The temperature found at the 8-12 cm depth was less extreme than the surface, and the fluctuations were not as great. The north facing slope averaged 5°C cooler than the south facing slope, and smaller temperature fluctuations were observed. Differences between mulched and nonmulched plots on the north facing slope were not as great. Regrowth of crownvetch occurred on the north facing slope and not on the south facing slope. The regrowth occurred first on the mulched plots, then on the nonmulched plots. It was concluded that the only hope of successfully establishing vegetation on the south facing slope was to seed in early spring or late fall.

Materials and Methods

The experiment was established on north and south facing slopes along I-74 near Veedersburg, Indiana. The angle of incline on both slopes was between 20° and 25° from the horizontal. A section of each slope with similar amounts of crownvetch was selected. The existing vegetation was removed with a hoe leaving the underground portions of the plants intact. Plots 1m^2 were staked out in cleared areas and 10 cm sections of 12 mm conduit tubing were buried in the middle of each plot to allow for the measurement of soil temperature at an 8-12 cm depth. A 50 cm^2 piece of cardboard was then placed over the exposed opening of the conduit tubing.

A 12-12-12 (N,P,K) fertilizer was applied to each plot at the Indiana State Highway Department suggested rate of 880 kg/ha. The preweighed seeding mixture, shown in Table 7, and/or mulch (wheat straw at a rate of 3.3 t/ha) were applied to selected plots to give the following treatments: mulched, bare surfaced, bare surfaced that was seeded, and a mulched surface with seed.

Initial soil temperature readings were taken on July 3, on the surface and at the bottom of the piece of conduit tubing (8-12 cm) using a telethermometer and calibrated thermocouples. The readings at the bottom of the conduit tubing were compared to readings taken with thermocouples buried at an 8-12 cm depth, and a negligible difference was noted between the two. Temperature readings were conducted at 2 day intervals for July, then twice a week in August and once a week during September and October. The temperature readings were taken during the early afternoon between 1 and 3 pm, and air temperatures (30-40 cm) were also recorded. The experiment was terminated on October 7.

Results and Discussion

The seeding mixture, consisting of species which looked promising at Covington, failed to germinate. On the south facing plots temperatures of approximately 50°C were reported throughout the month of July. The high temperatures appeared to be the major explanation for the failure of the seeding mixture.

Table 7. The seeding mixture used at the Veedersburg experimental plots.

Species	kg/ha	% of seed in mixture
1. smooth brome grass	72	30
2. weeping lovegrass	12	30
3. Indiangrass	22	20
4. blue grama	24	20

Mulch modified the temperature on both slopes. This modification occurred at both the surface and the 8-12 cm depth. The reduced temperatures appeared to be from the mulch maintaining higher levels of surface moisture. Differences between the mulched and nonmulched treatments were not as great or nonexistent immediately after a rain such as on August 12. Differences between the mulched and nonmulched plots were also not as great later in the season when the mulch started to decompose.

For the 8-12 cm depth of both slopes the temperatures were lower and the extremes were not as great as those occurring on the soil surface. The soil temperature decreased later at the 8-12 cm depth than at the surface. Temperatures at 8-12 cm on both slopes in July were at levels which would be within the range of temperatures where growth of warm season grasses would occur.

Soil temperatures on the north facing slope were not as great as those on the southern exposure. Soil temperature fluctuations also were less on the northern exposure, due possibly to its higher level of soil moisture. The temperature found on the north facing slope average 5°C cooler than the midsummer temperatures recorded on southern exposures.

Another difference between the slope with a southern exposure and the one with a northern exposure was that the latter had regrowth of crownvetch. Regrowth was observed first on August 7 for the mulched plots and then on August 21 for the nonmulched plots. The growth of crownvetch appeared to modify the temperature found on the north slope. In July and August the average monthly temperature for the north bare surface was comparable to that for the south mulched surface. In September, the temperature on the bare surface of the north slope, where a significant amount of crownvetch regrowth had occurred, was similar to the mulched plots of the north slope.

Thus, the temperatures found on a southern or northern exposure during the month of July would not appear to allow growth of vegetation. Conditions existing on the south-facing slope would not allow vegetative growth during most of the study period. Temperatures encountered there were outside the normal range of temperatures for plant growth. For successful seeding on a south facing slope, an attempt would have to be made to seed earlier in the spring or later in the fall than a north facing slope.

THE RESPONSE OF RADICLE GROWTH TO FIXED TEMPERATURE

Abstract

Pregerminated seeds of Andropogon gerardii Vitman. (blue bluestem), Panicum virgatum (switchgrass), Sporobolus heterolepis Gray. (prairie dropseed), Bouteloua curtipendula (sideoats grama), Bouteloua gracilis (blue grama),

annual ryegrass, Lotus corniculatus (birdsfoot trefoil), Bromus inermis (smooth brome grass), Lespedeza stipulacea (Korean lespedeza), Trifolium hybridum (Alsike clover), alfalfa, and Kentucky bluegrass were placed between the glass of a test tube and a sheet of moistened filter paper. The initial root length was marked and the seeds were grown at a constant temperature for 48 hours. Then root growth was measured. The experiment was conducted at 5°C increments between 12°C and 37°C and at 47°C. A negligible amount of radicle growth occurred at 47°C for all species. The seminal root growth of all species remained low between 12°C and 17°C. Optimum growth occurred at 22°C for annual ryegrass, 27°C for smooth brome grass, and 32°C for switchgrass, blue bluestem, and Korean lespedeza. In the other species optimum growth occurred over a range of temperatures from 22°C to 32°C. Kentucky bluegrass, annual ryegrass, and smooth brome grass, cool season C-3 grasses, had negligible growth at 37°C, whereas sideoats grama, switchgrass, blue bluestem, and blue grama, warm season C-4 grasses, had significant radicle growth at that temperature.

Materials and Methods

Seed of blue bluestem, switchgrass, and prairie dropseed, were placed in a plastic 9 cm petri dish on top of two pieces of Whatman #1 9 cm filter paper, moistened with 2 ml distilled water. Two more pieces of filter paper also moistened with 2 ml distilled water were placed on top of the seed. The petri dishes were sealed inside polyethylene bags, which allowed limited gas exchange. They were stored in a refrigerator at 4°C. Seed of prairie dropseed, blue blustem, and switchgrass were stratified for 7, 8, and 9 weeks, respectively.

The stratified seed and untreated seed of sideoats grama, blue grama, annual ryegrass, birdsfoot trefoil, smooth brome grass, Korean lespedeza, Alsike clover, Kentucky bluegrass, and alfalfa were sized according to weight. The

sideoats grama seed were cleaned of chaff before weighing. A procedure similar to that for stratification was followed in germinating the seed. Fifty seeds per 9 cm petri dish were placed on top of two pieces of moistened filter paper, and two more moistened pieces of filter paper were used to cover the seed. The petri dishes were sealed inside polyethylene bags, placed in a laboratory drawer, and the seed allowed to germinate.

The petri dishes were opened when the radicle of the germinated seeds were approximately 5 mm long. The petri dishes were opened after 36 hours for sideoats grama, blue bluestem, switchgrass, and Korean lespedeza, 48 hours for blue grama, alfalfa, annual ryegrass, smooth bromegrass, and Alsike clover, 3 days for prairie dropseed, and 4 days for Kentucky bluegrass and birdsfoot trefoil. Selected seedlings were placed in test tubes, between the glass of the test tube and a 5.5 cm x 5.5 cm piece of rolled Whatman #1 filter paper moistened with .5 ml of distilled water. The test tubes were then sealed with rubber serum stoppers and the initial root length marked on the glass.

Test tubes containing seed were set in holes 1.8 cm in diameter by 6 cm deep drilled in a 91x21x8 cm solid aluminum bar, so that all but the top 12 mm of the tube were in the block. The desired temperature was maintained by passing deionized water heated or cooled by a Lauda/Brinkmann R-3 circulator (temperature range -20 to 100°C) through the circulation ports drilled in the bar. The bottom and sides of the bar were insulated with 3.5 cm and 5 cm of Styrofoam, respectively. The top was insulated with 2.5 cm of Styrofoam, into which holes had been drilled to allow placement and extraction of test tubes. A 1.5 cm thick covering of black cloth was then placed on top of the Styrofoam insulated bar to limit circulation of air into the holes and to ensure that the seedlings were in darkness. The bar was placed at a 22° angle from the horizontal.

The experiment was conducted at 5°C increments between 12°C and 37°C, and at 47°C. The bar temperature was monitored in two test tubes at approximately one-third and two-thirds the length of the bar using thermocouples with periodic readings of a telethermometer. The largest observed variation in temperature was ± 1.0 at 47°C. After 48 hours the test tubes were removed from the bar and the amount of radicle growth measured.

Results and Discussion

The temperature-species interaction was found to cause significant differences in radicle growth. The general trend was that the rate of radicle growth was negligible below a certain temperature, rose to a maximum, and then declined as temperature continued to increase.

Negligible growth for all species was observed at 47°C, as can be seen in Tables 7 and 8. At this temperature, the roots of the cool season species, smooth brome grass, annual ryegrass, and Kentucky bluegrass appeared brown compared to the normal white color of the other species.

Radicle growth was retarded until temperatures rose above 17°C. Poor root growth at low temperatures has been reported to limit the distribution of C-4 warm season grasses. Within the category of warm season grasses, some grow better than others at lower temperatures, as is shown in comparing the amount of radicle growth of sideoats grama to that of switchgrass in the 12°C to 17°C temperature range. Between 17°C and 22°C there was a significant increase in the amount of radicle growth observed after 48 hours for each species.

Table 8. Mean** radicle growth in 48 hours.

Species	12	17	Temperature (°C)				
			22	27	32	37	47
sideoats grama	1.3ab*	1.3a	24.5c	29.7d	39.2d	27d	0.5a
blue grama	1.9ab	3.6ab	9.5a	14.4b	13.9b	5.6b	0.1a
annual ryegrass	8.0c	8.6c	39.5d	32.9e	17.8c	1.0a	0a
birdsfoot trefoil	3.0b	5.1b	10.3a	11.3a	11.9b	5.6b	0.2a
alfalfa	8.1c	12.4d	20.5b	24.6c	17.5c	8.7c	0.1a
Kentucky bluegrass	1.1ab	3.0ab	7.8a	9.5a	6.9a	.2a	0a

Species	12	17	Temperature (°C)				
			22	27	32	37	47
blue bluestem	2.2abc*	6.0b	16.9b	28.4d	28.4c	19.5c	0a
smooth bromegrass	5.8d	9.8c	19.3b	30.3d	15.5b	1.2a	0a
Korean lespedeza	4.8cd	9.6c	23.3c	28.2a	35.5d	30.6d	0a
switchgrass	1.4ab	3.9ab	15.8b	22.9c	30.4c	22c	.1a
prairie dropseed	0.6a	1.9a	5.9a	8.1a	6.2a	2.8a	0a
Alsike clover	3.9bc	6.8b	14.9b	16.2b	19.9b	9.9b	.1a

**Means are the average value of 3 replications of 12 observations each.

* Means within columns (temperatures) followed by the same letter are not significantly different by the Duncan Multiple Range Test at the .05 significance level.

The importance of an increase in temperature from 17°C to 22°C can be observed in the substantial increases that took place in the growth of annual ryegrass and sideoats grama. The similarity in response of annual ryegrass and sideoats grama to the change from 17°C to 22°C is especially interesting in light of differences in their genotypes. Annual ryegrass is a cool season grass that has a C-3 photosynthetic pathway, while sideoats grama is a warm season grass with a C-4 photosynthetic pathway. The legumes used also showed a significant, although less dramatic response to the increase in temperature from 17°C to 22°C. The response of radicle growth tends to support the idea that the reason for the increase in the rate of radicle growth at 22°C over that at 12°C to 17°C was due to some general physical change occurring within the plant's cells.

At 22°C, annual ryegrass reaches its peak amount of radicle growth per 48 hour period. It corresponds to temperature encountered in the early spring and late fall. Also, at 22°C, birdsfoot trefoil, Kentucky bluegrass, Alsike clover, and prairie dropseed reached a plateau in the amount of seminal root growth.

It is interesting that the four species with the least amount of radicle growth observed, prairie dropseed, birdsfoot trefoil, Kentucky bluegrass, and blue grama are the hardest to establish. But with blue grama the rate of radicle growth is not related to drought tolerance, because it has an excellent ability to withstand droughts.

Above 22°C the rate of seminal root growth observed for annual ryegrass decreases rapidly, whereas that for the remaining species is either increasing or constant. At 27°C, smooth bromegrass had its optimum rate of radicle growth. Korean lespedeza had its optimum rate of radicle growth occurring at

32 and 37°C. The 32-37°C range was the highest optimum temperature noted for any of the species used, and could indicate that this species has promise for warm season seeding.

Alfalfa, Alsike clover, birdsfoot trefoil, blue grama, and Kentucky bluegrass did not exhibit the sharp peak in amount of radicle growth as was observed for smooth brome grass, annual ryegrass, and sideoats grama. The latter three species had a narrow range in which optimum radicle growth was observed. Thus, a change of 5°C for those species could cause a large change in the amount of radicle growth occurring in a 48-hour period. This type of temperature response may have implications in the establishment of smooth brome grass, annual ryegrass, and sideoats grama in either the early spring or late fall, when soil surface temperatures can fluctuate widely. The C-4 warm season grasses had optimal growth at higher temperatures than did the C-3 cool season species.

The difference between C-3 and the C-4 grasses was most evident at 37°C, where the amount of radicle growth for annual ryegrass, smooth brome grass, and Kentucky bluegrass was negligible, whereas a significant amount of growth was observed for blue bluestem, switchgrass, sideoats grama, and blue grama. Since seedlings were in the heterotrophic stage of growth, photosynthetic differences could not have occurred between the two categories of grasses, and definitely suggested the possibility of genetic differences leading to biochemical dissimilarities. The species specific response to high temperature appears to be different than the general response of all species to a change in temperature from 17°C to 22°C.

The response of a plant to temperature varies immensely. One factor of importance is the climatic adaptation of a species. Species such as sideoats grama, blue grama, blue bluestem, and switchgrass, all native to the American

plains, require adequate seminal root growth when the soil temperature is warm to keep up with the drying of the soil. The climatic adaptation of a species can be important in the use of popular turfgrasses like Kentucky bluegrass in areas where they are not normally adapted.

Seed size between species does not seem to be important in the initial rate of seminal root growth. This is evident in the amount of growth the small seeded sideoats grama had at its optimum temperature for radicle growth, which was 39.2 mm at 32°C. It was not significantly different than the amount annual ryegrass, a larger seeded grass, had its optimum temperature, which was 39.5 mm at 22°C. Thus, even though annual ryegrass has larger seeds the total length of its radicle was not significantly different from sideoats grama when growth at their respective optimum temperatures were compared. Seed size is probably more related to the amount of time over which growth can occur before reserves are exhausted.

RADICLE GROWTH OF SELECTED SPECIES UNDER VARYING

ENVIRONMENTAL CONDITIONS

Abstract

Seedlings of annual ryegrass, Korean lespedeza, and sideoats grama were grown in 12 hour alternating temperature cycles of 10/18°C, 18/26°C, 24/32°C, 30/38°C, and 36/44°C. annual ryegrass had its optimum radicle growth at 24/32°C, whereas the optimum growth of Korean lespedeza was at 24/32°C and 30/38°C and the optimum growth of sideoats grama was in the 30/38°C regime. Radicle growth of all species was stunted when compared to that occurring at similar constant temperatures.

Seedlings of the same three species were also grown at 22, 27, 32, and 37°C in three different concentrations of polyethylene glycol 20,000 (average

molecular weight 15,000-20,000): 20 g solute/100 ml distilled water, 25 g solute/100 ml distilled water, and 30 g solute/100 ml distilled water. Radicle growth rate decreased with increasing levels of polyethylene glycol. Radicle growth was less at 32°C and 37°C than at cooler temperatures for annual ryegrass and Korean lespedeza. Polyethylene glycol induced water stress also caused a shift in the temperature for optimal radicle growth in comparison to the optimal temperature in distilled water. The shift was from 27-37°C to 27-32°C for Korean lespedeza, from 22°C to 27°C for annual ryegrass, and from 32-37°C for sideoats grama.

Materials and Methods

Experiment 1. Effects of Alternating 12-hour Temperature Cycles on the Amount of Radicle Growth in a 48-hour Period.

Sized seeds of sideoats grama, annual ryegrass, and Korean lespedeza were germinated using a previously explained method. Two germinated seed were then selected for uniformity and placed inside a test tube which contained a rolled square of Whatman #1 filter paper, moistened with .5 ml distilled water. Test tubes were sealed with rubber serum stoppers and placed inside the aluminum temperature gradient bar.

To minimize fluctuations from temperature changes across the length of the bar, only the two-thirds of the bar furthest from the circulation ports was used. There were enough holes in this section of the bar to allow for two replications of each species with six test tubes per species (two seed per test tube). Each alternating temperature regime was then replicated twice over time. Thus the experiment was a split-split plot design.

The temperature of the bar was altered every 12-hours. The circulator gave temperature regimes of 10/18°C, 18/26°C, 24/32°C, 30/38°C, or 36/44°C. Temperatures were monitored continuously throughout the course of the

experiment using a thermocouple recorder with 5 calibrated thermocouples evenly spaced throughout the length of the bar. Periodic temperature readings also were taken using a telethermometer. The largest temperature variation found was $+ 2^{\circ}\text{C}$, obtained when the bar temperature setting was altered.

The germinated seeds were grown for 48 hours under an alternating temperature regime. After this period, the test tubes were removed from the bar and the increase in radicle growth was measured.

Experiment 2. The Interaction of Constant Temperatures and
Polyethylene Glycol Induced Water Stress on

The Amount of Radicle Growth in a 48-hour Period

Three solutions of polyethylene glycol (PEG) 20,000 (average molecular weight 15,000-20,000) were prepared. The concentrations used were 20 g PEG/100 ml distilled water, 25 g PEG/100 ml distilled water, and 30 g PEG/100 ml distilled water to give solutions with osmotic potentials of approximately 4, 7, and 9 bars respectively. These solutions were then used to moisten a 3 m^2 piece of Whatman #1 filter paper, which was rolled inside a 65 mm x 15 mm test tube.

Seeds of sideoats grama, annual ryegrass, and Korean lespedeza were germinated between pieces of moistened filter paper inside petri dishes sealed in polyethylene bags. Seedlings were then placed between the rolled piece of filter paper and the test tube wall. Tubes were placed in the bar at a constant temperature of either 22°C , 27°C , 32°C , or 37°C . Temperatures were monitored with three thermocouples placed at one-quarter intervals the length of the bar and periodically read with a telethermometer. The last two rows of holes closest to the ports for the circulation of water were not used in an effort to minimize the temperature variation observed over the length of the bar. The largest temperature variation observed was $\pm 1.2^{\circ}\text{C}$ at 37°C . The

section of the bar used was large enough to allow for ten seedlings (2 seedlings/test tube) of each species to be exposed to each concentration of PEG. There were three replications of each bar temperature over time. The experiment was a split plot design. The seedlings were allowed to grow for 48 hours and then the increase in radicle length was measured.

Results and Discussion

Experiment 1. The Effects of Alternating 12-Hour Temperature Cycles on the Amount of Radicle Growth in a 48-Hour Period

The interaction between species and alternating temperature regimes was found to be significant with the rate of radicle growth dependent on the alternating temperature regime. The general trend was that as the temperature regime increased, the amount of radicle growth in a 48-hour period increased until a certain temperature regime, then the amount decreased in higher temperature regimes (Table 9).

The rate of radicle growth for sideoats grama, Korean lespedeza, and annual ryegrass was similar in the 10/18°C temperature regime, with radicle growth appearing to be stunted by the low temperatures. In the 18/26°C temperature regime, the growth of annual ryegrass was significantly greater than that of either of the other two species. This temperature regime is similar to the 18/27°C regime which Watschke et al. (1970) found best for root production of Kentucky bluegrass another cool season C-3 grass.

The most radicle growth in a 48-hour period for both annual ryegrass and Korean lespedeza occurred in the 24/32°C regime. The rate of radicle growth for sideoats grama was still stunted at this temperature regime, and was not significantly greater than the amount of radicle growth in a 48-hour period observed in the 18/26°C temperature range.

In the 30/38°C temperature regime there was a substantial decrease in the amount of radicle growth of annual ryegrass, whereas the amount of radicle growth of the other two species remained the same or increased. The most root growth of sideoats grama occurred under this regime. In the highest temperature regime (36/44°C), sideoats grama performed best of the three species but the high temperatures had an inhibitory effect on the amount of seedling root growth.

Table 9. Radicle growth in a 48-hour period under five alternating temperature regimes.

Species	Alternating Temperature Regime (°C)				
	10/18	18/28	24/32	30/38	36/44
	mm				
sideoats grama	5a*	9ab	10b	16c	9ab
annual ryegrass	8b	21c	28d	8d	0a
Korean lespedeza	7a	15b	25c	24c	8a

* Means are the average value of four replications 12 observations per replication rounded to the nearest whole mm. Means within rows followed by the same letter are not significantly different by the Duncan Multiple Range Test at the .05 level.

The amount of radicle growth was less in the alternating regimes than that obtained in the constant temperature regimes. The differences were especially evident that the optimal temperatures for radicle growth of each species. In the constant temperature regime at their optimal temperature annual ryegrass and sideoats grama both had approximately 40 mm of radicle growth in a 48-hour period, whereas with alternating temperatures the greatest radicle growth was 28 mm for annual ryegrass and 16 mm for sideoats grama. Thus, alternating temperatures appeared to retard radicle growth. This could be due to some

residual effect of the extreme (or most unfavorable) temperature of the regime, or just from a slower growth rate occurring at the most unfavorable temperatures of the regime.

At the coldest alternating temperatures (10/18°C) the rate of seminal root growth was similar to that at either 12 or 17°C constant temperatures. This observation indicates that radicle response to temperature over this range is fairly constant. Also the temperature optimum is shifted in alternating temperatures. The optimal alternating temperature regime for radicle growth of annual ryegrass is higher, while the optimal regimes for root growth of Korean lespedeza is still in the same range as the best constant temperatures.

Thus the response of the selected species to alternating temperature was different than from the constant temperature regimes. Not only was the amount of radicle growth less in the alternating temperature regimes, but also for annual ryegrass the optimum temperature range for radicle growth was shifted.

Experiment 2. The Interaction of Constant Temperatures and
Polyethylene Glycol Induced Water Stress on the Amount
of Radicle Growth in a 48-Hour Period

The interaction of temperature and PEG induced water stress was found to have a significant effect on the rate of radicle growth in a 48-hour period. Increasing concentration of PEG caused radicle growth to decrease. The concentration of PEG also shifted the temperature at which optimal radicle growth occurred (Table 10).

The lowest concentration of PEG did not cause a significant decrease in radicle growth of Korean lespedeza compared to growth in distilled water. At 22°C and 27°C radicle growth in the 25 g/100 ml concentration of PEG was similar to growth in the distilled water control. The similarity in growth rates could be due to the higher temperature placing more of a moisture

requirement on the seedling or hampering its ability to obtain moisture, while at cooler temperatures the seedling was able to obtain an adequate level of moisture.

The results for sideoats grama were different. At 22°C, 27°C, and 32°C none of the PEG concentrations allowed as much growth as did distilled water. As the temperature increased, the amount of radicle growth for the 20 g/100 ml concentration of PEG improved in comparison to distilled water. Thus at 37°C it appears that temperature was more inhibitory to radicle growth than was low amounts of PEG induced moisture stress.

Table 10. Effect of temperature and polyethylene glycol induced water stress on radicle growth of three species.

Concentration*	Korean lespedeza Temperature ($^{\circ}$ C)			
	22	27	32	37
distilled water	22a**	28ab	35b	30b
20	16a	24b	36c	23ab
25	11a	24b	25b	13a
30	9a	13ab	17b	10ab

Concentration	sideoats grama Temperature ($^{\circ}$ C)			
	22	27	32	37
distilled water	26a	30a	40b	27a
20	10a	18b	20b	23b
25	10a	22b	13a	12a
30	6a	15bc	17c	11ab

Concentration	annual ryegrass Temperature ($^{\circ}$ C)			
	22	27	32	37
distilled water	40d	32c	18b	1a
20	20c	35d	10b	0a
25	14c	22d	7b	0a
30	10c	14c	5b	0a

* The concentration is in g polyethylene glycol/100 ml distilled water.

** Means are the average value of three replications, 10 observations rounded to the nearest mm. Means within rows followed by the same letter are not significantly different by the Duncan Multiple Range Test at the .05 level.

At 22° C and 27° C, the amount of radicle growth for annual ryegrass in the PEG solution of the lowest concentration is significantly greater than at the other two PEG concentrations. And at 27° C the amount of radicle growth in the 20 g/100 ml solution is not significantly different that it is in distilled

water. There are no significant differences in the amounts of radicle growth between any of the three PEG solutions at 32°C, and the amount of radicle growth in each is significantly less than in distilled water. Greater moisture requirement at the higher temperatures could have caused radicle growth of each PEG concentration to be similar.

Polyethylene glycol induced moisture stress also causes a shift in the temperature at which optimum radicle growth of a species occurs. As Table 10 shows, this shift tends to be toward lower temperatures for a species whose optimum radicle growth is at high temperatures and in the opposite direction for a species whose optimum growth is at a low temperature. Thus for Korean lespedeza the optimal temperature became 27-32°C as the concentration of PEG increased, while the optimum temperature for radicle growth of annual ryegrass shifted from 22°C to 27°C in the 20 and 25 g/100 ml concentrations of PEG.

Field Study on the Effects of Seed Presoaking and Seed

Mixtures on the Initial Establishment of Prairie and

Non-Prairie Species

Abstract

Field experiments were conducted on a west facing newly cut highway slope along Highway 63 just south of Interstate 74 in Vermillion county, Indiana. Three separate tests were conducted to evaluate individual species and seed mixtures. Individual species observations were only conducted the first year, while mixtures were observed for two years after establishment.

Tall, warm season-cool season and short, warm season-cool season mixtures appear to be promising seed mixtures for Indiana highway slopes where minimal slope preparation is mandated. These test mixtures showed greater germination levels, and hence better initial erosion control, than the currently used mixture, the proposed mixture, and the prairie mixture, but by 4 weeks cover

provided by these two test mixes was not higher than that found with the proposed and current highway mixes.

The use of presoaked seed did result in higher germination levels compared to dry seed after 1 week.

The effects of fertilizer in this study were generally detrimental to early establishment of all species tested, whether seed was dry or presoaked, but later fertilizer was very important in ensuring adequate coverage. Also, when seeding legumes, rates of fertilizer have to be controlled to favor them over the grasses in the mixture.

Prairie forbs did not contribute substantially to the cover obtained, and due to economic constraints they probably should not be used in mixtures except in special situations such as roadside rest areas, where an aesthetically pleasing landscape is important.

Materials and Methods

The site was brought to final grade the week of planting. No further slope preparation was made. Following slope preparation soil samples were taken to determine pH and fertilizer requirements. Three test seeding mixes containing varying combinations of nitrogen fixing legumes, temporary rapid cover species, cool-season grasses and warm-season grasses were made along with a state highway current mixture and the previously used highway mixture (Table 11). Seventeen species or varieties included in the five seed mixtures were individually tested at the same rates as in the mixtures. Ten additional species or varieties were also selected for individual species evaluation.

Cold soaking pretreatment was initiated on March 21 and 22, and April 18, 1979 respectively for the 18 nonprairie grasses and legumes, 7 prairie species and prairie forb Echinacea purpurea (purple coneflower) (Tables 11 & 12). Prior to pretreatment all seed but those of the prairie species were stored at

room temperature (approximately 20-23°C). Seed of the prairie species was airdried (15 to 18°C) for about 65 days and after December 21 stored dry at 4°C in one liter jars, with the exception of purple coneflower. On March 21 and 22, seed lots of the 25 species to be presoaked were placed in polyethylene bags and enough tap water added to allow for seed imbibition (a volume in milliliters equal to half the seed lot weight in grams). The presoaked seed lots were placed in dark storage at 4°C. The bags were rotated several times the first week to insure even imbibition of seed. Seed of coneflower was placed in polyethylene bags between folded halves of Whatman #1 9 cm filter paper moistened with 1.5 ml of tap water and stored dark at 4°C after April 18. During the week prior to planting, seed lots required for individual field plots were weighed. The lots of soaked seed were returned to 4°C dark storage.

Table 11. Five Trial Seed Mixtures and Seeding Dates (kg/ha).

	Seed ¹ <u>Pretreatment</u>	<u>kg/ha</u> ²
<u>Mixture 1: Tall Warm-Season Cool-Season Mixture</u>		
1. alfalfa	W	5.1
2. crownvetch	W	11.2
3. annual ryegrass	W	5.1
4. weeping lovegrass	W	5.1
5. Kentucky 31 tall fescue	W	39.3
6. perennial ryegrass	W	11.2
7. red top	W	16.8
8. switchgrass	W	<u>11.2</u>
		105.0
<u>Mixture 2: Short Warm-Season/Cool-Season Mixture</u>		
1. alfalfa	W	5.1
2. crownvetch	W	11.2
3. annual ryegrass	W	5.1
4. weeping lovegrass	W	5.1
5. Kentucky 31 tall fescue	W	39.3
6. perennial ryegrass	W	11.2
7. red top	W	16.8
8. little bluestem	W	22.4
9. prairie dropseed	W	<u>5.1</u>
		121.3
<u>Mixture 3: Current Highway Mixture</u>		
1. annual ryegrass	D	11.2
2. creeping red fescue	D	28.1
3. Kentucky bluegrass	D	22.4
4. Kentucky bluegrass	D	22.4
5. perennial ryegrass	D	<u>28.1</u>
		112.2
<u>Mixture 4: Previous Highway Mixture</u>		
1. Kentucky 31 tall fescue	D	44.9
2. perennial ryegrass	D	28.1
3. Kentucky bluegrass	D	11.2
4. annual ryegrass	D	<u>39.3</u>
		123.5

Mixture 5: Prairie Mixture

1. alfalfa	W	5.1
2. white prairie clover	W	7.6
3. prairie clover	W	7.6
4. lead plant	W	15.2
5. annual ryegrass	W	5.1
6. weeping lovegrass	W	22.4
7. perennial ryegrass	W	22.4
8. little bluestem	W	22.4
9. blue bluestem	W	22.4
10. Indiangrass	W	22.4
11. switchgrass	W	<u>3.4</u>
		138.7

¹W = stored wet at 4°C for 10 weeks prior to planting; D = stored dry at room temperature (approximately 20-23°C) until time of planting.

²Kilogram weight based on dry seed.

Table 12. Seeding Rates of Non-prairie species at Highway 63 Experimental Plots.

Species	Seeding Rate (kg/ha) ¹
<u>Grasses</u>	
red top*	16.8
smooth bromegrass	11.2
orchard grass	16.8
weeping lovegrass*	7.3
Kentucky 31 tall fescue*	39.3
creeping red fescue*	28.1
annual ryegrass*	11.2
perennial ryegrass*	11.2
perennial ryegrass*	28.1
reed canary grass	11.2
Kentucky bluegrass*	11.2
Kentucky bluegrass*	22.4
Kentucky bluegrass*	22.4
rough-stalk bluegrass	29.2
<u>Legumes</u>	
crownvetch	11.2
crownvetch*	11.2
Korean lespedeza	16.8
birdsfoot trefoil	5.1
alfalfa*	5.1
Alsike clover	5.1
red clover	5.1
winter vetch	16.3

¹Kilogram weight based on dry seed.

*Included in one or more trial seed mixtures.

In the first experiment the five test seed mixtures were randomly assigned to plots 4.6 x 1.5 meters, replicated four times. Randomly selected halves of each plot were fertilized. In the individual species experiment 14 grasses and 8 legumes were randomly assigned to 3.0 x 1.5 m fertilized plots. The treatments included stratified and dry seed of 18 species and dry seed of four additional species (Table 12). In a third experiment seed of four cold soaked warm-season prairie grasses, blue bluestem, Andropogon scoparius (little bluestem), Indiangrass, and switchgrass with 3.2 g of purple coneflower were sown on plots 1.5 x 1.5 m, replicated three times. No fertilizer was applied to these plots.

The planting for these three experiments was done on June 1, 1979. the surface soil was still moist from a rain 2 days earlier. A 12-12-12 fertilizer was applied to the plots to be fertilized at 800 pounds/acre (880 kg/ha). Seed was hand sown and lightly incorporated with a rake. All plots were straw mulched at two tons/acre (440 kg/ha) and covered with erosion control netting.

Rapidity of germination was evaluated approximately once a week for the remainder of June. Numbers of seedlings of test species were recorded in randomly thrown 0.1m^2 quadrats on all plots.

In the second year only the experiment involving seeding mixtures was continued, although, observations were periodically taken of the individual species plots. Road construction disturbed one replication, thus only the other three were used in the continuation of the study. Individual species plots which did not perform well were mowed and reseeded with the current highway mixture on June 20, 1980. They were remulched with wheat straw at 1.5 t/ha. During the beginning of August the highway department assumed normal maintenance procedures by mowing the plots.

Mixture performance was measured by taking fresh weights of above ground tissue. The samples were obtained on November 2, 1979, May 16, June 20, and September 6 of 1980, by randomly throwing a 0.1m^2 quadrat and trimming the tissue at approximately 1 cm above the soil surface.

Results and Discussion

An analysis of variance of tested seed mixtures showed time, mixes, and their interaction to be highly significant factors contributing to variations seen in numbers of seedlings between plots. The general, germination tended for all mixes to be slow the 1st week, increase dramatically by the 2nd week and level off by the 4th week. A mean separation test showed that week one germination rates were significantly less than those at weeks two and four while no significant change occurred between weeks two and four. The significance of the mix by time interaction indicates that timing of emergence was not the same for all species (Table 13). The tall warm-season/cool-season and short warm-season/cool-season mix had the highest emergence during week one. The prairie, current highway, and previously used highway mixes increased at a much higher rate than the other two mixtures between the 1st and 2nd weeks.

Initially, fertilizer was significant only at .10 level, this low significance level was due probably to the small number of replicates rather than lack of actual differences (Table 14). Seedling counts on fertilizer plots were consistently lower than on nonfertilized plots (Table 13). Probably the moisture remaining from a 1.4 cm rain 2 days earlier dissolved but did not initially leach the fertilizer. This high surface salt content could have then damaged emerging seedlings. Dramatic differences in seedling counts the 1st week between nonfertilized and fertilized plots offers support to this

explanation. Rains recorded at Covington, Indiana on June 8 (1.0 cm), 9 (0.4 cm), 10 (0.6 cm) and 12 (0.6 cm) decreased the surface salt concentration which allowed for germination increases on the fertilized plots. The significant interaction of mixtures and fertilizer indicated that the fertilizer effect was not the same for all mixtures.

Table 13. Number of Seedlings/m² After One, Two, and Four Weeks With and Without Fertilizer.

Seed Mix	Weeks					
	Ferti- lizer ¹	No Ferti- lizer	Ferti- lizer ²	No Ferti- lizer	Ferti- lizer ⁴	No Ferti- lizer
Mix 1	565	1113	2845	2979	2365	2790
Mix 2	354	1241	2018	3717	2687	3536
Mix 3	28	379	1617	3840	1935	3213
Mix 4	86	417	2433	3188	2581	2791
Mix 5	63.5	279	718	1033	1094	1065
Mean (wks) ¹	452 b		2439 a		2406 a	

¹Tukey, .05

Germination rates of individual species explain differences seen over time with different seed mixtures. Cold-soaked seed of Agrostis alba (red top) and Kentucky 31 tall fescue had the highest germination level after 1 week and were both contained in the two mixtures with the highest germination the 1st week, mixtures 1 and 2. The only other species at the tested seeding rates to have more than 50 seedlings/m² after 1 week were cold soaked seed of little bluestem contained in mixtures 2 and 5, dry seed of annual ryegrass contained in mixtures 3 and 4 and Poa trivialis (rough-stalk bluegrass) tested only on an individual species basis.

By the 2nd and 4th weeks only the germination level of mixture 5 was significantly different from the germination levels of the other four mixes. Again individual species responses provide an explanation. Annual ryegrass, little bluestem, red top, and Kentucky 31 tall fescue increased five times or more by the 2nd week. During the second week germination of perennial ryegrass, Kentucky bluegrass, and creeping red fescue contained in either the current or proposed highway mixes increased at least tenfold. In contrast germination levels of the three prairie species, blue bluestem, switchgrass, and Indiangrass increased less than sevenfold.

The effect of fertilizer on presoaked seed is also better understood by examining the response of individual species. Seedling numbers were either increased or similar the 1st week when presoaked seed was compared to dry seed of the same species. This suggests that it is advisable to plant presoaked seed if speed of germination is important (Appendix A). The only three exceptions were rough-stalk bluegrass, annual ryegrass, and alfalfa, where dry seed had higher emergence after 1 week than cold soaked seed of these species. However, the increased seedling numbers on the nonfertilized areas of seed mixture plots after 1 week suggests that the advantage of presoaking seed to increase germination was greater when fertilizer was not used.

Table 14. Number of Seedlings/m² With and Without Fertilizers (Averaged Over One, Two, and Four Weeks).

Seed Mixture	Fertilizer	No Fertilizer	Mean/Seed Mixture
Mix 1	1925	2294	2109 a*
Mix 2	1686	2831	2259 a
Mix 3	1193	2474	1834 a
Mix 4	1700	2132	1916 a
Mix 5	<u>625</u>	<u>793</u>	<u>709</u> b
Mean/fertilizer treatment	1426 a**	2105 b	

* Tukey, .05

**Tukey, .10

Mixtures containing legumes and warm season grasses had the greatest fresh weight production (Table 15). In November 1979, the two way interaction between mixture and treatment was significant. The tall, warm season-cool season grass mixture (mix 1), when fertilized, was better than any other of the mixture-fertilizer combinations. The difference between this mixture and the short, warm season-cool season grass mixture (mix 2) was that it contained switchgrass while the short, warm season-cool season grass mixture had little bluestem and prairie dropseed (Table 11). The short, warm season-cool season grass mixture and the prairie mixture (mix 5) were the next best. All three mixtures contained warm season grasses and legumes whereas the other mixtures only contained cool season grasses, which required a high fertility level.

When shoot fresh weights were sampled again the next spring, the mixture effect was significant, with the mixtures ranked in the same order as the previous fall. In the better performing mixtures, Eragrostis curvula (weeping lovegrass) dominated the fertilized plots. On the better performing nonfertilized plots alfalfa was the dominant species. Weeping lovegrass is a good nurse crop for legumes because its erect habit permits considerable light penetration and weeping lovegrass starts growth later in the spring. In June and September, all mixtures showed a similar response to fertilization with fertilized plots being significantly better than the nonfertilized plots. The tall warm season-cool season grass mixture, the short warm season-cool season grass mixture and the prairie mixture were the best performing mixtures. There was an increase in shoot fresh weights from the May to June samples. The increase was due to alfalfa flowering and weeping lovegrass starting to grow.

Table 15. Mean Shoot Fresh Weights at the Covington Highway Site.

		g/m^2			
		Sampling Date			
Mixture		11-2	5-16	6-20	9-6
Mix 1	Fertilized	660e*	350abc	830abc	990cd
	Unfertilized	60ab	270abc	260ab	520abc
Mix 2	Fertilized	440d	460c	1150bc	630bc
	Unfertilized	30ab	150ab	210a	420abc
Mix 3	Fertilized	140abc	40a	280ab	100ab
	Unfertilized	0a	10a	20a	30a
Mix 4	Fertilized	210bc	90ab	60a	100ab
	Unfertilized	0a	20a	30a	50a
Mix 5	Fertilized	300cd	160ab	1200c	800cd
	Unfertilized	20ab	370bc	450abc	1250d

* Means (grams/m^2) within a sampling date followed by the same letter are not significantly different using the Duncan Multiple Range Test at the .05 level.

In September only the mixture effect was statistically significant, with the prairie mixture (mix 5) better than either the tall warm season-cool season grass mixtures and those better than the current or proposed highway mixtures. The cover from the prairie mixture contained mainly alfalfa, although each replication contained a few plants of the Petalostemum spp. (prairie clover). The prairie mixture probably did the best because the prairie species included in it were less competitive with alfalfa. Besides containing prairie clover this mixture also included Amorpha canescens (lead plant), blue bluestem, and Indiangrass which no other mixture had.

Conclusions and Application

1. Fall seeding of cool season grasses will provide best cover. Seeding should be completed by mid-October except in extreme southern Indiana.
2. Although germination and emergence in the fall is slower than in the spring, seeding operations should not be delayed from late fall until early spring.
3. Less weed problems will result if fall seedings are used. Spring seedings may not provide as permanent cover due to competition from weeds.
4. Legumes seeded in the fall apparently germinate but freeze out in the winter. If legumes are to be used in mixtures they should be included only in spring-summer seedings.
5. Presoaking of seeds for fall seedings may speed germination, but overwintering of some seedlings is marginal. Presoaking should not be done for fall seedings.
6. The currently used highway seed mix does not produce as great biomass as the former mix but seedling numbers are very comparable and no over-effect on permanent cover should occur.

7. Prairie forbs did not contribute substantially to the cover obtained, and due to economic constraints they probably should not be used in mixtures except in special situations such as roadside rest areas, where an aesthetically pleasing landscape is important.

8. Seeding mixtures containing warm season grasses should not be used in late fall, because most of their growth occurs between June and September. But warm season species have a great potential for use in seeding during late spring and early summer. Weeping lovegrass was especially promising at the Covington Highway site.

9. In order to establish some grasses like Kentucky bluegrass, high levels of fertility and maintenance are required. Results indicated that Kentucky bluegrass containing mixtures are extremely wasteful of seed. In early establishment, these mixtures were dominated by fescue and ryegrass, which produce unsightly inflorescences in early June.

10. Adequate mulching with wheat straw appears to be the most promising treatment to allow the germination and survival of seedlings in the late fall. Thus a straw mulch with species adapted to environmental conditions present at seeding time appears to offer the most hope for establishment of grasses and legumes in late fall.

11. Radicle growth response to temperature varies depending on the species. Annual ryegrass reaches its peak rate of radicle growth in a 48-hour period at 22°C. Smooth brome grass has an optimum temperature for radicle growth at 27°C, and sideoats grama reaches its peak radicle growth at 32°C. All three species have a narrow range of temperatures at which optimum growth occurs. A change in temperature of as little as 5°C will cause a large change in seminal root growth thus restricting establishment.

12. Kentucky bluegrass, blue grama, birdsfoot trefoil, and prairie dropseed are hard to establish through seeding, and they also have the least amount of radicle growth. Thus, radicle growth may have an influence on the ease of establishment of a species.

13. Temperature conditions existing on the south facing slope in the summer are too extreme to allow for the growth of common herbaceous species. It is doubtful whether microclimate conditions on the south-facing slope could be modified to allow for successful seeding. Thus, to obtain successful vegetative cover requires seeding in early spring or late fall when favorable environmental conditions would occur.

14. On the northern exposure, conditions were not as extreme, and vegetative growth occurred. Seeding on this slope could be expected to be successful during August as long as some method to retain moisture was used.

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Appendix A. Common Names of some Herbaceous Species

<u>Latin Name</u>	<u>Common Name</u>
1. <u>Agrostis alba</u>	red top
2. <u>Andropogon scoparius</u>	little bluestem
3. <u>Bouteloua curtipendula</u>	sideoats grama
4. <u>Bouteloua gracilis</u>	blue grama
5. <u>Bromus inermis</u>	smooth bromegrass
6. <u>Coronilla varia</u>	crownvetch
7. <u>Cynodon dactylon</u>	Bermudagrass
8. <u>Dactylis glomerata</u>	orchard grass
9. <u>Eragrostis curvula</u>	weeping lovegrass
10. <u>Festuca arundinacea</u>	tall fescue
11. <u>Festuca elatior</u> `Arundinacea`	Kentucky 31 tall fescue
12. <u>Festuca rubra</u>	creeping red fescue
13. <u>Lespedeza stipulacea</u>	Korean lespedeza
14. <u>Lolium multiflorum</u>	annual ryegrass
15. <u>Lolium perenne</u>	perennial ryegrass
16. <u>Lotus corniculatus</u>	birdsfoot trefoil
17. <u>Medicago sativa</u>	alfalfa
18. <u>Panicum virgatum</u>	switchgrass
19. <u>Phleum pratense</u>	timothy
20. <u>Poa pratense</u>	Kentucky bluegrass
21. <u>Secale cereale</u> `Abruzzi`	Abruzzi rye
22. <u>Sorghastrum nutans</u>	Indiangrass
23. <u>Trifolium hybridum</u>	Alsike clover
24. <u>Trifolium repens</u>	red clover

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