Drought Effects on Adventitious Root Development in Blue Grama Seedlings

D.D. BRISKE and A.M. WILSON

Abstract

Crowns of blue grama (Bouteloua gracilis) seedlings of three ages were exposed to drought treatments for 2 days, in constant humidity environments, and were then planted in moist soil for a 10-day growth test at 25°C. Percentage survival of crowns decreased with a decrease in water potential during the temporary drought treatment and with a decrease in crown age at time of treatment. The percentage survival rates of 21-, 28-, and 35-day-old crowns treated at -180 bars were 5, 54, and 83, respectively. Crowns exposed to the 2-day drought treatment subsequently produced shorter adventitious roots than untreated crowns. Thus, a drought-induced inhibition of adventitious root growth may reduce the probability of successful seedling establishment.

Establishment of blue grama (Bouteloua gracilis) seedlings begins with germination and extension of the seminal primary root, which originates from the lower portion of the embryo (Gould 1968). After approximately 2 weeks of favorable environmental conditions following emergence, seedlings develop the capacity for adventitious root development from crowns of the primary shoot and tillers located at or very near the soil surface (Hyder et al. 1971; van der Sluijs and Hyder 1974). If seedlings do not initiate and develop adventitious roots within 6 to 10 weeks after emergence, they often die. Seedling death at this time does not result from an inherent limit to longevity of the seminal primary root; rather, seedlings expand leaf area to a maximum that can be supported by water from the seminal root and then succumb to an increase in transpiration stress (Wilson et al. 1976). Adventitious roots are effective in seedling establishment because they are longer-lived and have a greater capacity for water uptake than the seminal primary root. Thus, successful adventitious root establishment provides seedlings with an effective means of drought avoidance.

In the more arid portions of the Central Plains, blue grama plantings often fail because the environmental requirements for development of adventitious roots are not met. These requirements include a moist soil profile at the time of planting, temperatures between 15 and 30°C, and a moisture for 2 to 4 days—one period for emergence and one for development of adventitious roots (Briske and Wilson 1977, 1978; Fults 1944; Olmsted 1941, 1942; Riegel 1941; Wilson and Briske 1979). In addition to the direct effects of environment, morphological stage and physiological condition may influence the capacity of seedlings to initiate and extend adventitious roots. On the Central Plains, seedlings surviving on only the seminal root are often exposed to periods of drought, which may adversely affect their physiological condition. The objective of this research was to determine the capacity of seedlings to produce adventitious roots as affected by age and exposure to temporary drought. This capacity may determine whether or not seedlings will become successfully established once the rather restrictive environmental requirements for adventitious root initiation and elongation are met.

Methods

Blue grama accession PM-K-1483 is a synthetic blend of seed from 12 accessions originating in southern Kansas and Texas. Seed for this study was harvested in 1974 at the Plant Materials Center, Manhattan, Kansas.

Seeds were planted at a depth of 2 cm in plastic pots (15 cm diameter by 15 cm deep) filled with autoclaved sandy loam soil. Planting of seeds was scheduled so that seedling crowns of three age classes (21, 28, and 35 days) would be ready for temporary drought treatment on the same date. During initial seedling growth in the greenhouse, the pots were subirrigated to promote growth of the seminal primary root and to maintain a 1.5-cm layer of dry soil that would prevent the initiation of adventitious roots. After emergence, seedlings were thinned to 12 per pot. At 21, 28, and 35 days after planting, the seminal root system and subcoleoptile internode were removed just below the coleoptilar node. All leaf tissue was removed 1 cm above the coleoptilar node to simulate a drought-induced die-back of all exerted leaf tissue and to facilitate placement of crowns within the constant humidity tray. These procedures were accomplished to accurately control crown drought stress. Uniform stress generally is not achieved through drying soil in the entire rooting zone of the seminal root. Therefore, only the seedling crown (lowest 1-cm section of stem base) was retained to evaluate drought tolerance.

Seedlings were exposed to temporary drought for 2 days in constant humidity trays (Wilson 1971). Water potentials of -30, -60, -90, -120, -150, and -180 bars were maintained in the trays with various concentrations of NaCl solution (Lang 1967; Robinson and Stokes 1949). Seedling crowns lost water vapor to the constant humidity air but did not touch the NaCl solutions. The capillary flow of NaCl solution above and below the constant humidity tray was sufficient to compensate for evaporation from the moist paper.

After 2 days of exposure to drought, seedling crowns were planted at a depth of about 0.7 cm in plastic pots (15 cm diameter by 15 cm deep) filled with autoclaved sandy loam soil. Thus, the upper portion of the crowns extended about 0.3 cm above the soil surface. The soil was covered with a 0.5-cm layer of fine gravel to reduce evaporation from the soil surface. The soil was irrigated daily by surface application to maintain favorable moisture conditions (about -0.3 bars) for the growth of leaves and adventitious roots.
roots from treated crowns. After the planting of crowns, pots were placed in a growth chamber at a constant temperature of 25°C, daylength of 15 hours, relative humidity of 50%, and photosynthetic photon flux density of 480 microeinsts m⁻² sec⁻¹. The experiment also included a control consisting of 21-, 28-, and 35-day-old crowns that had not been exposed to temporary drought (water stress). The seminal root system and leaves were removed just as in the treated crowns, but they were then immediately planted on the same date as the treated crowns.

After a 10-day growth performance test, all pots were removed from the growth chamber, soil was washed from the roots with a fine sprayer of water, and seedlings were placed in a 10% solution of acetic acid and stored at 5°C until measurements of growth could be made. The number of shoots (primary shoot and all tillers) that had developed before the drought treatment, the number of shoots that produced regrowth during the 10-day test, and the number of adventitious roots were counted. The lengths of all leaf blades and adventitious roots were also measured. New shoots, leaf blades, and adventitious roots were counted and measured if they had reached lengths of 0.5, 0.5, and 0.1 cm, or more respectively. Percentage survival of treated and untreated crowns was based on the number of crowns in which one or more shoots exhibited leaf growth of at least 0.5 cm during the 10-day test under favorable temperature and moisture conditions. The number of surviving 21-day-old crowns in the -180 bar treatment was inadequate for evaluation of drought effects on selected growth measurements. Consequently, only crown survival was compared for all three age classes at the -180 bar drought treatment.

A randomized complete block experimental design with six replications was used. The six replications were six sequential repetitions of the experiment. Each value (within a replication) for seedling survival was based on a sample of 15 treated or untreated crowns planted in an individual pot. Each value for the other growth criteria was based on the average of all surviving seedlings from one pot. Analysis of variance, linear regression, and polynomial regression were used to evaluate relationships among age classes, drought treatments, and growth responses. All observations were included in statistical analyses, but only means were reported.

Additional seedlings (21, 28, and 35 days of age) were grown for measurement of xylem water potential with a pressure chamber (Waring and Cleary 1967). The main shoot and large tillers were used for these measurements. The objective was to determine xylem water potentials for 35-day-old blue grama seedlings for each of the six drought treatments after a 2-day equilibration period in constant humidity trays. Because water potentials represent average values for individual crowns, the possibility of minor water potential gradients within the crown has not been eliminated. Shoot apices and adventitious root primordia of large 35-day-old crowns might have been protected from dehydration more than the apices and primordia of comparatively small 21-day-old crowns. However, the close agreement between solution osmotic potential and average crown water potential suggests that differences among age classes in crown injury and survival resulted mainly from differences in drought tolerance rather than drought avoidance (Levitt 1964).

Table 1. Osmotic potentials of controlling solutions (bars) and water potentials for 35-day-old crowns for each of the six drought treatments after a 2-day equilibration period in constant humidity trays.

<table>
<thead>
<tr>
<th>Solution osmotic potentials</th>
<th>Crown water potentials</th>
</tr>
</thead>
<tbody>
<tr>
<td>-30</td>
<td>-30±2.1†</td>
</tr>
<tr>
<td>-60</td>
<td>-62±1.5</td>
</tr>
<tr>
<td>-90</td>
<td>-87±7.9</td>
</tr>
<tr>
<td>-120</td>
<td>-116±7.5</td>
</tr>
<tr>
<td>-150</td>
<td>-152±7.5</td>
</tr>
<tr>
<td>-180</td>
<td>-180±9.2</td>
</tr>
</tbody>
</table>

†Indicates standard deviation of the sample.

**Results**

**Seedling and Crown Water Potential**

Before harvest and treatment of crowns, 21-, 28-, and 35-day-old blue grama seedlings exhibited average xylem water potentials of -9.5, -14.5, and -17.0 bars, respectively. Thus, crown water potentials in the least severe drought treatment were about 13 to 20 bars lower than xylem water potentials of seedlings before harvest.

Water potential of soil in which the seminal root was growing was kept at about -0.3 bars. Therefore, differences in seedling water potential were not due to differences in soil moisture conditions, but were probably due to greater leaf area and transpirational water loss in older seedlings than in younger seedlings. Wilson et al. (1976) observed that an increase in leaf area at a time when there was little or no increase in capacity for water uptake created a condition of seedling drought, even though the seminal root was growing in moist soil. Apparently, water uptake was restricted by a threadlike nature of the subcoleoptile internode connecting the seminal primary root and the shoot.

Thermocouple psychrometer measurements indicated that seedling crowns had reached equilibrium with constant humidity environments during the 2-day treatment (Table 1). Because water potentials represent average values for individual crowns, the possibility of minor water potential gradients within the crown has not been eliminated. Shoot apices and adventitious root primordia of large 35-day-old crowns might have been protected from dehydration more than the apices and primordia of comparatively small 21-day-old crowns. However, the close agreement between solution osmotic potential and average crown water potential suggests that differences among age classes in crown injury and survival resulted mainly from differences in drought tolerance rather than drought avoidance (Levitt 1964).

![Fig. 1. Effects of age and temporary drought treatments on the percentage of blue grama crowns that produced regrowth during a 10-day growth test under favorable soil moisture conditions. Correlation coefficients (**) are significant at the 0.01 level. SEE = standard error of estimate.](image-url)
Table 2. Number of shoots per crown of 35-, 28-, and 21-day-old blue grama seedlings, before temporary drought treatments; and number of shoots per crown producing regrowth during a 10-day growth test following temporary drought treatments.

<table>
<thead>
<tr>
<th>Growth and criteria and crown age class</th>
<th>Temporary drought treatments (bars)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th>Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of shoots/crown before treatment</td>
<td>-180</td>
<td>-150</td>
<td>-120</td>
<td>-90</td>
<td>-60</td>
<td>-30</td>
</tr>
<tr>
<td>35 days</td>
<td>6.3*</td>
<td>6.1</td>
<td>6.3</td>
<td>6.6</td>
<td>6.9</td>
<td>6.4</td>
</tr>
<tr>
<td>28 days</td>
<td>3.9</td>
<td>4.5</td>
<td>4.2</td>
<td>3.9</td>
<td>4.5</td>
<td>4.2</td>
</tr>
<tr>
<td>21 days</td>
<td>2.6</td>
<td>3.1</td>
<td>2.7</td>
<td>2.8</td>
<td>2.8</td>
<td>2.8</td>
</tr>
<tr>
<td>Treatment Mean</td>
<td>4.3a</td>
<td>4.6a</td>
<td>4.4a</td>
<td>4.4a</td>
<td>4.7a</td>
<td>4.5a</td>
</tr>
<tr>
<td>Number of shoots/crown producing regrowth</td>
<td>35 days</td>
<td>3.5</td>
<td>4.0</td>
<td>4.2</td>
<td>4.8</td>
<td>5.0</td>
</tr>
<tr>
<td>28 days</td>
<td>2.1</td>
<td>2.6</td>
<td>2.5</td>
<td>2.8</td>
<td>3.1</td>
<td>3.0</td>
</tr>
<tr>
<td>21 days</td>
<td>1.0</td>
<td>1.6</td>
<td>1.2</td>
<td>1.6</td>
<td>1.7</td>
<td>2.0</td>
</tr>
<tr>
<td>Treatment Mean</td>
<td>2.2a</td>
<td>2.7c</td>
<td>2.6b</td>
<td>3.1cde</td>
<td>3.3de</td>
<td>3.3se</td>
</tr>
</tbody>
</table>

*Values represent an average of six replications. Means in columns or rows, within each growth criterion, labeled with the same letter are not significantly different at the 0.01 level.

Effects of Temporary Drought on Survival

Percentage survival of crowns exhibited a significant ($P<0.01$) age times drought interaction, indicating that crowns of younger seedlings were more susceptible to drought injury than crowns of older seedlings (Fig. 1). Percentage survival rates of treated (-180 bars) and untreated crowns were 5 and 86 for 21-day-old crowns, 54 and 95 for 28-day-old crowns, and 83 and 99 for 35-day-old crowns, respectively. Drought tolerance of 28-day-old crowns approached that of 35-day-old crowns but had not yet developed sufficiently for a high percentage survival in the severe drought treatments. Twenty-one-day-old crowns had developed neither the capacity for regrowth (control treatment) nor the drought tolerance exhibited by the 28- and 35-day-old crowns.

Seedlings randomly assigned to various treatments did not differ in number of shoots per crown before harvest and treatment (Table 2). The number of shoots that survived the drought treatments and produced regrowth during the 10-day test decreased with a decrease in water potential during temporary drought treatments.

Effects of Temporary Drought on Growth

Leaf Development and Growth

Leaf growth resulted mainly from the expansion of immature leaf blades that had been clipped at the time crowns were harvested; it resulted, to a lesser degree, from the initiation of new tillers from axillary buds. Twenty-six percent of the crowns produced one new tiller, and only rarely did crowns produce two new tillers. Data indicated that both leaf intercalary meristems and axillary buds possessed a high degree of drought tolerance.

Total leaf length produced during the 10-day test decreased with a decrease in water potential during the 2-day drought treatment (Fig. 2). Total leaf length produced by 21-, 28-, and 35-day-old crowns was reduced 56, 35, and 30%, respectively, as a result of treatment at -150 bars. Number of surviving shoots per crowns was positively associated with total leaf length produced per crown ($r = 0.96$).

Adventitious Root Development and Growth

Adventitious roots were not initiated in crowns of any age class until several centimeters of leaf had developed. Thus, leaf tissue was the first regrowth observed. These results agree with those of Hyder et al. (1976), who found that the capacity for root growth in mature blue grama crowns was more sensitive to drying than the capacity for shoot growth. Only 3% of the crowns produced leaf growth but no root growth in this study.

The number of roots per crown produced during the 10-day test decreased with a decrease in age of crowns at time of treatment (Fig. 3). The large number of roots produced by older crowns was mainly a result of the large number of surviving shoots from which adventitious roots could develop.

Number of roots per crown also tended to decrease with a decrease in water potential during the temporary drought treatments. In relation to the control treatment, the number of roots in 21-, 28-, and 35-day-old crowns was reduced 24, 8, and 8%, respectively, as a result of the -150 bar treatment. Only in the 21-day-old crowns was the reduction in root numbers significant ($P<0.05$).

Total length of adventitious roots per crown decreased with a decrease in age of crowns at the time of drought treatment (Fig. 4 and 5). Greater total length of roots in the older crowns can be attributed both to a greater number of
shoots from which adventitious roots could be produced and to a greater length per root.

Total length of adventitious roots per crown also decreased with a decrease in water potential during the temporary drought treatments (Fig. 4). Total root length produced from 21-, 28-, and 35-day-old crowns was reduced 52, 34, and 27%, respectively, by the -150 bar treatment. Number of live shoots per crown was positively associated with total length of roots per crown (r = 0.94).

The length of the longest root per crown decreased with a decrease in age of crowns at the time of treatment (Fig 6). The maximum rate of root elongation (averaged over the entire 10-day growth test) was about 1.0 cm per day and occurred in 35-day-old, untreated crowns. This is a low rate of elongation, compared with an elongation rate of 2.3 cm per day found in a previous experiment (Briske and Wilson 1977). The low rate of root elongation was probably a result of the long period (2 to 6 days) required by seedling crowns to produce photosynthetic tissue. Crowns required several centimeters of leaf length before the initiation and development of adventitious roots.

The length of the longest root per crown decreased with a decrease in water potential during the temporary drought treatment (Fig. 6). A significant \( P<0.01 \) age times drought interaction indicated that drought inhibited root elongation in younger crowns more than in older crowns. The length of the longest root produced by 21-, 28-, and 35-day-old crowns was reduced 44, 25, and 21%, respectively, as a result of the drought treatment.
of the −150 bar drought treatment. The relatively small percentage decrease in length of roots produced by 35-day-old crowns is similar to the results of Hyder et al. (1976), who found that root lengths of mature blue grama crowns were not affected by drying treatments. The crowns of mature blue grama plants and of older seedlings apparently are more drought tolerant than crowns of younger seedlings.

Discussion

Drought may affect the capacity for root development from blue grama seedling crowns in two ways: (1) by reduction of seedling leaf area, and (2) by direct injury of root primordia and other crown tissues. This study simulated the conditions in which drought causes die-back of exserted leaf tissue and, in addition, causes dessication of crowns.

The combined clipping and mild drought treatments probably injured seedlings more than a similar degree of drought (without clipping) in the field. For example, one would expect that only portions of leaves would die back at a water potential of −30 to −60 bars (Wilson and Sarles 1978). The retention of leaves by seedlings at such water potentials would probably result in a greater development of roots than in the present experiment, in which all exserted leaf tissue was removed. However, under severe drought conditions in the field, all exserted portions of leaves would die back; the effects of drought in that respect, would be similar to the effects observed in this study.

In relation to soil moisture requirements for development of adventitious roots, the test conditions following temporary drought were less severe than conditions generally found in the field. Favorable moisture conditions during the 10-day growth test were adequate for development of leaves and roots from many of the crowns, in spite of drought injury. On the Central Plains, periods of 2 or more days with a continuously moist soil surface are rare (Briske and Wilson 1977), and seedlings often fail to develop adventitious roots because of rapid drying of the soil surface. Even under conditions in which seedling drought is not severe, the initial rate of root extension is sometimes just sufficient to keep the root tip ahead of the drying soil front (Wilson and Briske 1979). Thus, a drought-induced delay of root initiation and an inhibition of root growth may reduce the probability of successful seedling establishment. Injury and death of seedlings might be reduced by conserving soil moisture and by planting when the soil profile is moist and the probability of precipitation and favorable temperatures is high.

On the basis of leaf and root growth following temporary drought, the results indicate that drought tolerance of blue grama seedlings increases with increasing age. However, both the mechanism by which drought tolerance increases and the stage of seedling development at which drought tolerance reaches a maximum remain unknown and warrant further investigation.

Literature Cited


