An Anatomical Comparison of the Roots, Stems and Leaves of Big Bluestem (Andropogon Gerardi), Side-Oat Grama (Bouteloua Curtipendula) and Blue Grama (Bouteloua Gracilis) at a Three-Week Stage of Development

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AN ANATOMICAL COMPARISON OF THE ROOTS, STEMS AND LEAVES
OF BIG BLUESTEM (Andropogon gerardi), SIDE-OATS GRAMA
(Bouteloua curtipendula) AND BLUE GRAMA (Bouteloua gracilis) AT
A THREE WEEK STAGE OF DEVELOPMENT

being

A Thesis presented to the Graduate Faculty
of the Fort Hays Kansas State College in
partial fulfillment of the requirements for
the Degree of Master of Science

by

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Date Jan. 11, 1960 Approved

Approved

Major Professor

Chairman, Graduate Council
Big bluestem (Andropogon gerardi), side-oats grama (Bouteloua curtipendula) and blue grama (Bouteloua gracilis) represent a tall, mid and short grass type of vegetation respectively, and can typically be found growing in the mixed prairie. It was the purpose of this study to determine any variations in the internal anatomy of these plants and to compare these variations with respect to species.

Seeds of local source were planted in a greenhouse and, under ideal growing conditions, were allowed to germinate and develop for a period of three weeks. Samples of the roots, stems and leaves from each species were then collected and placed in F. A. A. solution. The samples were then sectioned, stained and mounted on slides. The diameter of the stem, the diameter of the root, the number and radial diameter of vascular bundles in the stem and root and the ratio of the diameter of the conductive tissue to diameter of total stem were the criteria used to measure the difference between the roots and stems of the three species. Criteria used to measure the difference in the anatomy of the leaf were the number, radial diameter and distance apart of the vascular bundles. Meristematic activity in the leaf was also used as a criterion for this study. Numerous measurements were made of each criterion, and photomicrographs were taken to aid in obtaining the resulting data.

The results revealed the stem and root of all species to consist of three tissue systems: (1) epidermal, (2) fundamental and (3) vascular. The three plants studied revealed only primary growth, but
differences in the manner of primary growth were noted. The vascular bundles in side-oats grama and blue grama were scattered throughout the transection, but in big bluestem the vascular bundles were formed more or less in two circles, the smaller bundles near the periphery, and the larger bundles deeper within the stem.

Since no vascular cylinder was formed in the stem, there was no distinction between the pith and cortex, but the central part of the root could be distinguished from the cortex by the presence of a pericycle. Big bluestem revealed the greatest stem diameter, root diameter, stem pith diameter and root conductive tissue diameter at this stage of growth. Blue grama showed the most vascular bundles and also the greatest radial diameter of the vascular bundles in the stem. The ratio of the radial diameter of conducting tissue to diameter of total stem was greatest in side-oats grama, with big bluestem and blue grama being approximately the same.

The leaf blades revealed many vascular bundles made up of two kinds of conducting tissues, the xylem and phloem. The leaves showed only primary growth, and large bulliform cells extending from one epidermis to the other were found between each vascular bundle. Side-oats grama contained the most bundles, but the diameter of the bundles was greatest in blue grama. Side-oats grama leaves had the greatest width, while big bluestem leaves had the greatest length. No meristemmatic activity was found at the base of the leaf blades.
ACKNOWLEDGEMENTS

The author wishes to express his gratitude to Dr. Gerald W. Tomanek for valuable suggestions in the choice of this problem, his wise counsel and helpful advice during the course of the problem and for reading and criticizing the manuscript. Thanks and appreciation are extended to Mr. Henry J. McFarland for allowing the author free access to the rotary microtome and for valuable assistance in photography and staining processes of specimens used. My sincere thanks are also extended to Dr. Edwin P. Martin for his helpful suggestions on the paraffin method of embedding plant specimens. These men, who gave helpful suggestions and constant advice throughout the investigation, were highly instrumental in making this study a success.
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INTRODUCTION

Of all the plants of the earth, the grasses are of the greatest use to the human race. The grasses furnish the principal breadstuffs of the world and a large part of the food of domestic animals. Grasses may be used for many purposes, some of which are as follows: (1) food grasses, (2) forage, pasture and range grasses, (3) hay grasses, (4) soil-holding grasses, (5) silage grasses and (6) ornamental grasses.

The purpose of this study was first, to compare the anatomy of the roots and stems of big bluestem (Andropogon gerardii), side-oats grama (Bouteloua curtipendula) and blue grama (Bouteloua gracilis), and secondly, to see if meristematic tissue can be found in the leaf blades of grasses. The fact that very few studies have been conducted on the meristems in leaves of grasses led the author to become interested in this phase of plant anatomy.

Big bluestem, side-oats grama and blue grama were used in this study to represent a tall, mid and short grass type of vegetation, and also because these grasses represent three of the most important species found in the mixed prairie.

Big bluestem is a tall, sod-forming, warm-season grass which occurs typically on lowland and hillside areas in the mixed prairie. Side-oats grama is a mid, sod-forming, warm-season grass which occurs typically on shallow hillside areas. Blue grama is a warm-season short grass, and under climax conditions is a dominant of deep upland soils.
The roots of grasses are fibrous with little modification. The primary root persists only a short time after germination, its place being taken by secondary roots produced from the nodes of the young culm. Besides the original root system at the base of the plant, secondary roots are often formed from nodes above the ground as in maize (prop roots), or from the nodes of creeping culms (rhizomes or stolons). Roots are never produced from the internodes of the culms.

The jointed stem of a grass, called a culm, is made up of a series of nodes and internodes. The internode may be hollow or solid. Perennial grasses may form a sod or mass of individuals by means of rhizomes or stolons, or they may form a crown or tuft by the continual formation of upright branches within the lower sheaths.

The leaves are borne on the culm in two ranks, one at each node. The leaf consists of a sheath and a blade. The sheath envelops the culm above the node to which it is attached, the margins overlapping or infrequently united into a cylinder for a part or a whole of the distance to the summit.

The following features of the plants selected were studied: (1) diameter of stem, (2) diameter of root, (3) number and radial diameter of vascular bundles in stem and root, (4) amount of pith, (5) ratio of diameter of conductive tissue to diameter of total stem, (6) number, radial diameter and distance apart of vascular bundles in the leaf and (7) meristematic activity, if any, in the leaf.
Throughout the contents of this paper, the symbol $\mu$ will represent microns.
RELATED STUDIES

Very little material has been published on the anatomy of grass seedlings. Almost all the texts and reference books deal with the mature structures without taking up their origin and development. The studies that have been made are mainly those of Zea, and only occasionally is some other grass used. The preparation and staining techniques for different plants are similar, but the anatomy of grass seedlings requires special techniques of its own. Though the vascular anatomy of the roots and aerial parts of grasses have many similarities, they are not all like corn and they are not all arranged as they are in corn. It is possible because of these variations among the grass tribes to identify many genera and even species by their vascular anatomy.

This was the object of a study conducted by Reynolds (1959). His work was done on the morphology and vascularization in selected members of the tribe Andropogoneae. In this study quantitative measurements were made to determine vein length per unit area, volumetric relationships of the various tissues, the ratio of the major blade unit width to its thickness and the ratio of the maximum major blade unit width to the width of a single wing.

As for corn, an excellent study was conducted by Zlab (1953) in partial fulfillment for the requirements of a Master of Science degree at the University of Nebraska.
One of the first studies on anatomical characters for certain Gramineae was made by Holm (1901). He later published "Studies in the Compositae" (1908).

A study on the vascular anatomy of sandreed grass (Calamovilfa longifolia) was done by Stover (1924). In this study, Stover investigated the development of the individual vascular bundle in the roots, stems and leaves from the embryo state until maturity.

Dr. Walter V. Brown, together with his students at the University of Texas, has contributed a number of papers dealing with grass phylogeny. Most of Brown's work was classifying grasses into groups based entirely on leaf anatomy. Brown and Emery (1957) wrote a fine paper on apomictic grasses of South Africa.

Brown and collaborators have investigated various disciplines which they relate to grass systematics, one of which is a paper written by Brown, Heimsch and Emery (1957) in which they report on the organization of the grass shoot apex.

In Texas, Dr. Frank W. Gould, curator of the Tracy Herbarium, Department of Range and Forestry, Texas A & M College, has been active in both cytological and taxonomic research pertaining to members of the genus Andropogon. Most of this work was done on chromosome size and number. He also constructed the "Tribal Triangle" (1955) to aid students in learning the subfamilies and tribes of grasses.

Irving W. Knobloch (1944), while employed by the Iowa State Agricultural Experiment Station, conducted a study of the gross morphology, anatomy, and development of smooth brome grass (Bromus
inennis). In this study, the anatomy of the roots, first internode, culm, leaves and rhizomes is described in detail.

The work of Lommasson (1957) on the leaf anatomy of Aristida provided some interesting results. He found that certain species of Aristida are unique in that the cells of the inner bundles sheath are large and contain a great concentration of chloroplasts which in most grasses are lacking in the inner sheath. The outer bundle sheath cells were found to be small. This work by Lommasson supported the placement of Aristida in a new tribe.

Cheadle (1942) reported on the role of anatomy in phylogenetic studies in the Monocots. The book on the Gramineae by Agnes Arber (1934) is a good reference work on grass morphology, and the texts of Esau (1953) and Eames (1947) on plant anatomy are excellent. Esau (1945) also wrote a good article on "Vascularization of the vegetative shoots of Helianthus and Sambucus." This article deals with the formation and vascularization of the first sieve tubes, first phloem cells and first xylem elements.

Cytogenetics and evolution of the grass family has been ably handled by an American worker, Stebbens (1956b), and he has also considered the taxonomy and evolution of grass genera (1956a).

Young (1938), a Chinese botanist, conducted a study on the developmental anatomy of the seedling of the rice plant which is native to his country. Lin (1949), while a student at Fort Hays Kansas State College, studied the stem anatomy of twelve forbs of the family Compositae growing near Hays, Kansas, in partial fulfillment of the
requirements for the degree of Master of Science. This study, as many others, was made on mature plants.
METHODS OF PROCEDURE

Collecting Materials

Seeds of local source were used in this study. They were planted in plots in a greenhouse and allowed to germinate and develop for a period of three weeks. Ideal growing conditions prevailed because the seeds were planted in a sifted loam soil and covered with a thin layer of fine sand. During this period of germination and development, the plots were watered once per day, and at the end of three weeks the grasses averaged about 3 1/2 inches in height above ground and about 4 inches in root length. Roots, stems and leaves were carefully removed from the plant, measured and cut into 1/2-inch-long sections. Each section was then marked to keep the species separate.

Killing and Preserving

The plant specimens were killed and preserved according to Johansen (1940). They were placed in bottles containing Formalin-Aceto-Alcohol. This fluid, more familiarly known as FAA, might almost be called the "standard preservative" of botanical microtechnique, since it is used more extensively than any other. The variations that have been proposed are almost endless, but the standard proportions are:

- 50% (or 70%) ethyl alcohol---90cc.
- Glacial Acetic Acid----------5cc.
- Formalin---------------------5cc.
This reagent may be used with almost any plant material intended for anatomical or morphological study. Material may be left in it almost indefinitely without appreciable damage, but the minimum time of fixation is 18 hours. The plant specimens were placed in this reagent to: (1) kill the plant tissue and preserve it as nearly life-like as possible, (2) soften the hard tissue and harden the soft tissue and (3) remove insoluble and soluble substances. The bottles were kept in a dark room.

Embedding, Sectioning and Mounting

The plant specimens were also embedded and sectioned according to Johansen (1940). The following steps for the process of embedding plant tissue in paraffin were used: (1) wash several times in 50% alcohol, (2) add 50% alcohol for 1 hour, (3) add 60% alcohol for 1 hour, (4) add 70%, 80%, and 95% alcohol for 12-24 hours each, (5) add 100% alcohol for 12-24 hours, changing once, (6) add 100% xylene for 1 hour changing once, (7) place in oven to melt paraffin and evaporate xylene for 12-24 hours and (8) pour in blocks and cool. These dehydration steps were employed to completely remove the water from the tissues. The transfer from one step to another in the dehydrating process must be very gradual, otherwise plasmolysis is certain to result. The process should never be rushed; but, on the other hand, tissues become brittle if left too long in one of the higher alcohol solutions. The paraffin blocks containing the plant specimens were then placed in the rotary microtome and sections were mounted on slides.
using Mayer's egg fixative. Hot water was then placed on the slides for the purpose of taking out wrinkles in the paraffin, and the excess water was allowed to evaporate for several days.

**Staining**

Safranin O and Fast Green FCF are the ideal stains for plant tissues, and thus were used in this study. Safranin O is 5.45% soluble in water and 3.41% soluble in alcohol. This is perhaps the most important stain known to botanists and is used extensively in both morphology and cytology. It stains lignified, cutinized and suberized structures as well as chromosomes, nucleoli and centrosomes. Fast Green FCF is 16.04% soluble in water and 0.33% soluble in alcohol. This dye is far superior to light green SF and fades very little. It stains more intensely in a shorter time. The plant specimens, already mounted on slides, were stained according to the following steps:

1. xylene 1 for 10 minutes, xylene 2 for 1 minute,
2. 60% alcohol for 5 minutes,
3. safranin for 8 hours,
4. 60% alcohol for 5 minutes,
5. 80% alcohol for 5 minutes,
6. 95% alcohol for 5 minutes,
7. fast green 4 to 6 dips,
8. 95% alcohol for 5 minutes,
9. absolute (100%) alcohol for 10 minutes,
10. xylene for 5 minutes and
11. mounted in piccolyte. The piccolyte was allowed to dry for a period of two weeks. Finished slides were available at the end of this two-week period.

**Collecting Data**

Data were obtained in two ways:

1. taking photomicrographs,
2. measuring under a microscope.
RESULTS

General Structure of the Stem

The vegetative body of the sporophyte of the vascular plant is customarily divided into three so-called organs: stem, leaf and root. The stem, as a part of the shoot, is organized during the development of the embryo.

A common feature of the stem in the primary state of development is its division into nodes and internodes. The plants used in this study, being only three weeks old, were not this far advanced, and thus did not have nodes and internodes.

Vascular systems composed of widely spaced strands and not restricted to one ring in cross sections are relatively uncommon in the dicotyledons, but in the monocotyledons similar and more complex systems are common.

In cross-sections of the grasses studied, the usual three tissue systems, the epidermal, the fundamental and the vascular, were present. The vascular bundles were distributed according to two basic plans. Either they were in two circles, one of the smaller bundles nearer the periphery, the other of larger bundles somewhat deeper within the stem, or they were scattered throughout the transection. In big bluestem, the bundles were formed more or less in two circles, while in side-oats grama and blue grama the bundles were scattered throughout the transection. The vascular bundles in all three species were collateral, each enclosed in a sheath of sclerenchyma. This means that the xylem
and phloem lie side by side on the same radius, the phloem being directed toward the outside of the stem and the xylem toward the center. Both the xylem and phloem are conductive in function, but the xylem also gives strength to the stem.

The stems at this early stage of development showed only primary tissue. The vascular tissues are in separate bundles, not arranged to form a hollow cylinder, but scattered irregularly to form a characteristic type of dictyostele. Between the scattered vascular bundles is undifferentiated parenchyma. Because no vascular cylinder is formed, there is no distinction between pith and cortex.

In big bluestem, in which the vascular bundles are arranged in two circles, there is a continuous cylinder of sclerenchyma close to the epidermis, with the outer smaller bundles imbedded in it. Inside the cylinder of sclerenchyma is the fundamental parenchyma in which the larger vascular bundles are imbedded (Figs. 1, 2). The central part of this parenchyma constitutes the pith. In side-oats grama and blue grama, where the vascular bundles occurred in a scattered arrangement, no sclerenchyma cylinder was formed, but the subepidermal parenchyma appeared strongly sclerified (Figs. 3, 4, 5, 6).

The average diameter of the stem of big bluestem at this stage of growth was 968.0u, while side-oats grama measured 768.0u and blue grama 771.8u (Table I). The radial diameter of the pith in big bluestem, side-oats grama and blue grama measured 573.0u, 392.0u and 498.0u, respectively. Big bluestem had the smallest
Figure 1. Cross section of big bluestem stem showing primary growth. The vascular tissues are in separate bundles, not arranged to form a hollow cylinder. X 150.
Figure 2. Cross section of big bluestem stem showing the fundamental parenchyma in which the larger vascular bundles are embedded. X 645.
Figure 3. Cross section of side-oats grama stem showing arrangement of the vascular bundles and the sclerified subepidermal parenchyma. X 150
Figure 4. Cross section of side-oats grama stem showing the absence of a sclerenchyma cylinder. X 645.
Figure 5. Cross section of blue grama stem showing the arrangement of the vascular bundles and the sclerified subepidermal parenchyma. X 150.
Figure 6. Cross section of blue grama stem showing the central portion of a vascular bundle. X 645.
Table I. Measurements in microns showing relationships between a tall, mid and short grass type at a three week stage of development

<table>
<thead>
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<th>Ratio of Diameter of Cond. Tissue to Diameter of Total Stem</th>
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<td>304.8</td>
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<tr>
<td>Side-oats grama</td>
<td>247.5</td>
<td>143.3</td>
<td>1:1.73</td>
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<tr>
<td>Blue grama</td>
<td>201.4</td>
<td>150.0</td>
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<th>Number of Bundles</th>
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<tr>
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<td>768.0</td>
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<td>Blue grama</td>
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<th>Diameter of Bundles</th>
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<td>1.02</td>
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<tr>
<td>Blue grama</td>
<td>1557.3</td>
<td>13</td>
<td>149.5</td>
<td>1.92</td>
</tr>
</tbody>
</table>
number of vascular bundles with 32, while side-oats grama was next with 35 and blue grama had the most with 39. The radial diameter of the vascular bundles was greatest in blue grama with an average measurement of 81.4μ. Side-oats grama again occurred midway between the other two grasses with an average radial diameter of 62.0μ, while the vascular bundles in big bluestem measured 56.0μ.

The stems of herbaceous monocotyledons (grasses) typically lack a cambium, therefore, no secondary tissue will be formed. Increase in diameter, which is slight, occurs entirely through the division and subsequent enlargement of the parenchymatous cells making up the ground tissue that lies between the vascular bundles.

General Structure of the Root

The root constitutes the underground part of the plant axis, specialized as an absorbing and anchoring organ. It occurs in the sporophytes of the vascular plants.

Roots display unlimited growth, elongation occurring only at their tips and growth in diameter, where present, taking place only in their older parts. Roots have practically the same kinds of permanent tissues as stems and leaves, but differ from these organs in the way in which their tissues are arranged.

Since stems and roots have many similarities in structure and show physical continuity, they are commonly treated as two parts of the same unit axis, and similar terms are applied to their tissue systems.
A parenchymatous region occurs in the center of the vascular cylinder of these grass roots, but according to Esau (1953) no complete agreement exists regarding the interpretation of this region. Commonly this region is referred to as pith or pith like, but there is some question whether it should not rather be regarded as potential vascular tissue that fails to differentiate as such. In view of the uncertain state of the interpretation of this area, the region will be referred to as pith in this study.

The central part of the root is occupied by the vascular cylinder composed of the vascular system and the associated parenchyma (Figs. 7, 8, 9). The vascular system of the root is more clearly distinguished from the cortex than that of the stem because of several distinctive anatomical features, one of which is the presence of a pericycle.

The roots of the grasses studied, like the stems, showed only primary growth. However, the root and stem showed some differences in the manner of primary growth. The root had a shorter elongation region of small cells and large cells. Thus, the root frequently develops no extensible types of protoxylem elements, whereas in the stem such elements are common.

The boundaries between the tissue systems appeared quite distinct in the root. The vascular cylinder is surrounded by a pericycle, while the evidence of an endodermis is somewhat distinguished. This differs from the stem in that the vascular tissues in the stem are not compactly arranged, and no distinct region meriting the name
Figure 7. Cross section of big bluestem root showing the vascular cylinder composed of the vascular system and the associated parenchyma. X 150.
Figure 8. Cross section of side-oats grama root showing the vascular cylinder composed of the vascular system and the associated parenchyma. X 150.
Figure 9. Cross section of blue grama root showing the vascular cylinder composed of the vascular system and the associated parenchyma. X 150.
of pericycle occurs between the cortex and the vascular tissues (Figs. 10, 11, 12).

The average diameter of the roots was as follows: big bluestem 304.8u, side-oats grama 247.5u and blue grama 201.4u (Table I). The average radial diameter of the conductive tissue of the roots of big bluestem was 225.0u, while side-oats grama measured 143.3u and blue grama 150.0u. This made the ratio of the radial diameter of the conducting tissue to diameter of total stem to be 1:1.35 in big bluestem, 1:1.73 in side-oats grama and 1:1.34 in blue grama.

General Structure of the Leaves

The leaf is the principal appendage or lateral organ borne by the stem. Like the stem, the leaf contains the same tissue systems—the dermal, the vascular and the fundamental. Also, as in the stem, the epidermis forms the outermost layer, and the vascular tissue is variously distributed in the ground tissue.

Although fundamentally alike in structure, the stem and the leaf differ from each other in details of growth and in relative arrangement of tissues. The leaf shows determinate apical growth as contrasted with the continued type of growth exhibited by the stem in its apical meristem. The differences in structure of the two organs are related to their functions. In the columnar shape of the stem, the vertical orientation of the vascular system and the abundance of mechanical elements and of storage parenchyma, suggest efficiency in longitudinal conduction of materials, support of the aerial body and storage of food.
Figure 10. Cross section of big bluestem root showing distinction between the cortex and the vascular tissues. X 645.
Figure 11. Cross section of side-oats grama root showing a distinction between the cortex and the vascular tissues. X 645.
Figure 12. Cross section of blue grama root showing a distinction between the cortex and the vascular tissues. X 645.
In the grass leaves studied the relatively large external surface, the extensive air space system, the abundance of chloroplasts in the ground tissue and the close spatial relation between the vascular and the ground tissues suggested a specialization related to photosynthesis. These characteristics favor exposure of the chloroplasts to the light and ready access of water and gases to the cells concerned with photosynthesis.

In the early stages of development of a grass leaf, such as was used in this study, no boundary is evident between the leaf blade and the leaf sheath. The writer believes that the initial part of the primordium represents only the leaf blade and that the leaf sheath is initiated somewhat later by intercalary growth near the foliar buttress. The blade itself must owe the major part of its growth to the activity of the intercalary meristem at its base. Although the writer found no intercalary meristem at this point, he still deems it necessary for the increase in length of the blade. Before the blade attains much length, a ligule (a thin projection from the top of the leaf sheath) develops from the adaxial protoderm and demarcates the sheath from the blade.

A cross section through a leaf blade reveals the presence of many veins (Figs. 13, 14, 15). This is due to the parallel venation found in grass leaves. Each vein occurs approximately equi-distance apart and consists of a single vascular bundle made up of two kinds of conducting tissues, xylem and phloem. As in the stem, these have a collateral arrangement, lying side by side, with the xylem above and
the phloem beneath. A bundle sheath composed of colorless cells was found surrounding each vein, its thickness being proportionate to the size of the vein (Figs. 16, 17, 18). Some mechanical tissue was also found in the vicinity of the larger veins, giving rigidity to the leaf. As in the stem and root, all the leaves studied showed only primary growth. Bulliform cells were present between each vascular bundle, and they extended from one epidermis to the other (Figs. 13, 14, 15).

At this three week stage of development, side-oats grama showed the most numerous bundles or veins with a total of 18. Blue grama numbered 13 and big bluestem had 11 (Table I). The diameter of the bundles was greatest in blue grama with an average diameter of 149.5μ. Side-oats grama averaged 100.6μ and big bluestem 88.8μ. The leaves of side-oats grama also showed the greatest width at this stage with an average of 2,025.0μ. The average width of blue grama leaves was 1,557.3μ, while big bluestem showed 1,399.0μ. Although blue grama leaves were wider than big bluestem, the leaves of big bluestem appeared much longer.
Figure 13. Cross section of big bluestem leaf showing the distribution of vascular bundles and arrangement of bulliform cells. X 150.
Figure 14. Cross section of side-oats grama leaf showing the distribution of vascular bundles and arrangement of bulliform cells. X 150.
Figure 15. Cross section of blue grama leaf showing the distribution of vascular bundles and arrangement of bulliform cells. X 150.
Figure 16. Cross section of big bluestem leaf showing the bulliform cells, inner bundle sheath, mesophyll, outer bundle sheath, phloem and xylem. X 645.
Figure 17. Cross section of side-oats grama leaf showing the bulliform cells, inner bundle sheath, mesophyll, outer bundle sheath, phloem and xylem. X 645.
Figure 18. Cross section of blue grama leaf showing the bulliform cells, inner bundle sheath, mesophyll, outer bundle sheath, phloem and xylem. X 645.
SUMMARY

A study of the anatomy of the roots, stems and leaves of big bluestem, side-oats grama and blue grama was undertaken to show the differences in their internal anatomy at a three week stage of development. Big bluestem, side-oats grama and blue grama were used in this study to represent a tall, mid and short grass type of vegetation, respectively.

The following features were studied: (1) diameter of stem, (2) diameter of root, (3) number and radial diameter of vascular bundles in stem and root, (4) amount of pith, (5) ratio of diameter of conductive tissue to diameter of total stem, (6) number, radial diameter and distance apart of vascular bundles in the leaf and (7) meristematic activity, if any, in the leaf.

The collecting of materials, killing and preserving, embedding, sectioning, mounting and staining were done according to Johansen (1940) with helpful hints from Sass (1951) and Gray (1958). Data were then obtained in two ways: (1) taking photomicrographs and (2) measuring under a microscope.

The results revealed the stem to consist of three tissue systems: (1) epidermal, (2) fundamental and (3) vascular. In big bluestem, the vascular bundles were formed more or less in two circles, the smaller bundles near the periphery, and the larger bundles deeper within the stem. In contrast to this type of arrangement, the vascular bundles in side-oats grama and blue grama were scattered throughout the
transection. The vascular bundles in all three species were collateral and enclosed in a sheath of sclerenchyma.

The stems at this early stage of development proved to be primary in origin, and because no vascular cylinder was formed, there was no distinction between the pith and cortex.

Big bluestem revealed the greatest stem diameter at this stage of growth, with blue grama next and side-oats grama the least in diameter. The radial diameter of the pith was greatest in big bluestem and least in side-oats grama. On the other hand, blue grama revealed the most vascular bundles, followed by side-oats grama and then big bluestem. Blue grama also showed the greatest radial diameter of the vascular bundles followed by side-oats grama and then big bluestem.

The central part of the root revealed a vascular cylinder, and could be distinguished from the cortex by the presence of a pericycle. The roots, like the stem, showed only primary growth, but differences in the manner of primary growth were noted.

As in the stem, big bluestem again revealed the greatest root diameter at this stage of growth. Side-oats grama was next in root diameter, with blue grama having the smallest. The radial diameter of conductive tissue was greatest in big bluestem, but this time blue grama was next with side-oats grama having the smallest diameter. This made the ratio of the radial diameter of conducting tissue to diameter of total stem to be greatest in side-oats grama, with big bluestem and blue grama being approximately the same.
In the grass leaves studied, no boundary was evident between the leaf blade and the leaf sheath. No meristematic activity was found at the base of the blade, but it is concluded that the blade itself must owe the major part of its growth to the activity of an intercalary meristem at this point.

Cross sections through the leaf blades revealed the presence of many veins, occurring approximately equal distances apart, and consisting of a single vascular bundle made up of two kinds of conducting tissues, the xylem and phloem. A bundle sheath was found surrounding each vein, and some mechanical tissue was also found in the vicinity of the larger veins. All the leaves showed only primary growth, and large bulliform cells extending from one epidermis to the other were found between each vascular bundle.

Side-oats grama contained the most bundles, followed by blue grama and then big bluestem. The diameter of the bundles was greatest in blue grama, side-oats grama was next and big bluestem had the smallest diameter. Side-oats grama leaves had the greatest width while big bluestem leaves had the greatest length. Although big bluestem leaves showed the greatest length, they also showed the narrowest width.
LITERATURE CITED


