SEASONAL FLUCTUATION OF ETHANOL-EXTRACTABLE WATER-SOLUBLE CARBOHYDRATES IN THREE NATIVE RANGE GRASSES

By

VAL KENNETH KERR

Bachelor of Science

Southwestern State College

Weatherford, Oklahoma

1960

Submitted to the Faculty of the Graduate School
of the Oklahoma State University
in partial fulfillment of the requirements
for the degree of
MASTER OF SCIENCE
July, 1966

SEASONAL FLUCTUATION OF ETHANOL-EXTRACTABLE WATER-SOLUBLE CARBOHYDRATES IN THREE NATIVE RANGE GRASSES

Thesis Approved:

J. F. Crockett
Thesis Adviser

Gene Guinn

W. W. Hawes

J. W. Bozzo
Dean of the Graduate School

ACKNOWLEDGMENTS

I am indebted to all who aided in the completion of this study, particularly the members of my committee, Drs. Hansen, Crockett and Guinn for suggesting and supervising the study; to Drs. Basler, M. Brooks, Crockett, Guinn and Todd for use of their equipment; and to James Mac Stewart for technical assistance rendered. I am grateful to my wife, Elva, for her patience and moral support. Without her assistance, this study would have been much more difficult.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

CHAPTER I

INTRODUCTION

Among native forage grasses the level of carbohydrate reserves stored in the roots is an index to general plant vigor. The quantity of these reserves may be correlated directly with such outward manifestations of plant vigor as regeneration of top growth, size and number of plants in a given area, forage and seed production, initiation of spring growth, and deterioration of root and shoot systems (Kinsinger 1953, McCarty 1938, Biswell and Weaver 1933, Albertson, et al. 1953, Cook, et al. 1955, Buckley and Weaver 1939, Aldous 1930, Weaver and Darland 1947, and Garber, et al. 1927).

Carbohydrate level also fluctuates during the different seasons of the year. An inverse correlation exists between this seasonal variation and growth rate. The advent of the growing season is marked by a rapid depletion of root reserves associated with increased physiological activity and rapid expansion growth. An accumulation of reserves is associated with a low or declining growth rate that terminates in dormancy (Hischke 1961, Hyder and Sneva 1959, Kinsinger 1953, Dodd and Hopkins 1957, and Crockett 1960).

The seasonal variation in carbohydrates should be an important consideration in determination of stocking rates on native pastures, for when root reserves are at a low level, forage plants are more subject to damage by grazing or mowing (Hischke 1961, McCarty and Price

1942, Cook, et al. 1958, and Stoddart and Smith 1955).

Continued excessive removal of herbage results in deterioration and death to range plants since root reserves must be utilized for regeneration of photosynthetic tissue. A perennial plant must have sufficient reserves to maintain physiological activity during dormancy and to initiate the new season's growth (Kinsinger 1953, Stoddart and Smith 1955, and Sampson and McCarty 1930).

Investigations by McCarty and Price (1942) on six mountain forage species indicated that approximately 75 per cent of the carbohydrate root reserves are exhausted in producing 10 per cent of the herbage growth during initial spring growth before the plant becomes self-sustaining.

Much of the research data gathered concerning carbohydrate fluctuation in the roots as affected by season, growth cycle, and removal of herbage has involved comparisons of protected and clipped herbage plots. An attempt has been made in this experimentation to measure carbohydrate variation in three native forage grasses, Andropogon gerardi Vitman (big bluestem), Andropogon scoparius Michx. (little bluestem), and Sorghastrum nutans (L.) Nash. (Indiangrass), subjected to continuous grazing conditions. The scope of this particular research was limited to measurement of ethanol-extracted water-soluble sugars. Investigations were conducted from September, 1962, through August, 1963. The common names of these grasses will be used throughout the paper and will follow Anderson (1961).

CHAPTER II

DESCRIPTION OF STUDY AREA

The study site was a one hundred acre fenced pasture one mile north and one mile west of Stillwater (SE $\frac{1}{4}$ of Section 5, 19N, 2E), Payne County, Oklahoma. The pasture is owned by the city of Stillwater and is leased to individuals for grazing purposes.

Topography

The elevation of the area is about 880 feet above sea level. The pasture can be described briefly as a tall grass loamy prairie range site with a predominately Zaneis loamy soil type but with no sizeable hills. The topography is rolling (with small clay pan sites randomly interspersed in the pasture).

Composition

At the beginning of the study, the range site was in excellent condition (Table I) with 85.9% of the grasses being decreasers (Sims 1962). However, in the late fall of 1962 and during the 1963 grazing season, the pasture was severely overgrazed. A herd of thirty mature cattle, along with the subsequent calf crop, and a herd of horses varying in number from two to six utilized the range. These grazing animals were removed during the winter months but were returned early in March of 1963. Precipitation for the months of May and June of 1963 was

TABLE I

VEGETATIVE COMPOSITION CLASSIFICATION OF RANGE SITE*

Species	Relative Composition	
Andropogon scoparius Michx.	63.0	
Andropogon gerardi Vitman	18.8	Decreasers
Sorghastrum nutans L.	3.5	
Panicum virgatum L.	0.6	
Eragrostis spectabilis (Pursh) Steud.	2.9	
Sporobolus asper Michx. Kunth	5.9	Increasers
Bouteloua curtipendula Michx.Torr.	0.6	and
Eragrostis curtipedicellata Buckl.	1.2	Invaders
Chloris verticillata Nutt.	0.6	
Carex sp.	2.9	
	100.0	

*The study area was a loamy prairie range site of a Zaneis loam soil type with a slope of 3-5%. Range condition was excellent with 85.9% of the grasses being decreasers. This is well within the range of 75-100% decreasers which constitutes excellent condition.

3.30 inches below normal (Figure 1).

This dry period, coupled with the excess stocking rate, accounted for a visible deterioration of the range site. Although composition did not change significantly in this period, deterioration was marked by disappearance of mulch, increased trampling, excessive grazing of decreasers, and an apparent visual decrease in range vigor.

Climate

Records from the United States Weather Bureau for Stillwater, Oklahoma, for the years 1893-1960 show a mean annual precipitation of 30.83 inches (Figure 1). About 81% of this moisture (24.95 in.) falls during the months of March through October, which approximates the grazing season. The mean average temperature for the sixty-seven year period was 60.9° F. (Figure 2). The average frost-free period is approximately 207 days, generally extending from mid-April through late October.

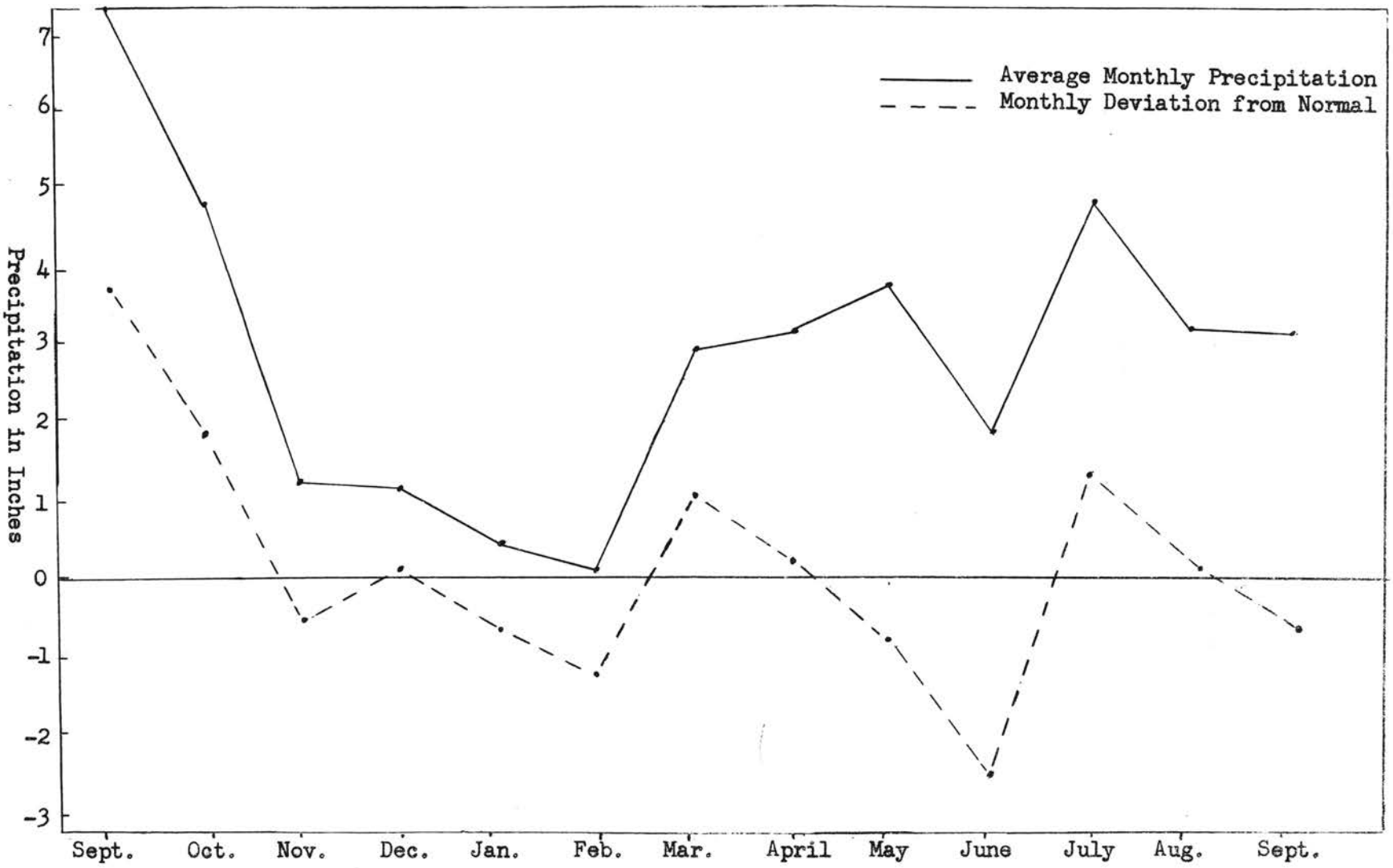

Figure 1. Average Monthly Precipitation and Deviation From Normal.

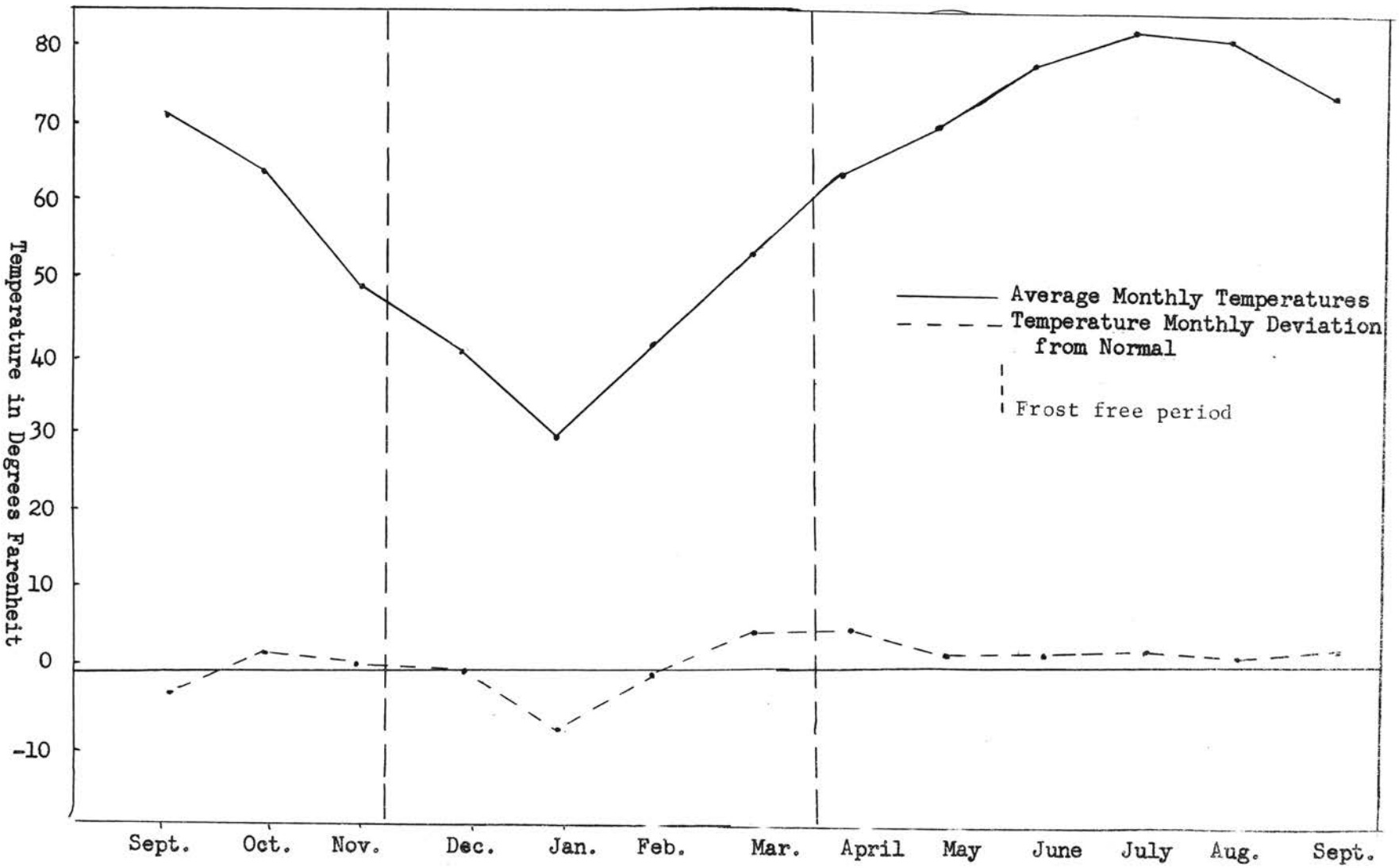

Figure 2. Average Monthly Temperatures and Deviation From Normal.

For the year 1962 the frost-free period extended from April 2 to November 4, a total of 216 days. Precipitation for the year was 32.43 inches, 1.6 inches above the yearly mean. The mean average temperature for 1962 was 60.6° F. Precipitation during the eight-month period from March through October was 29.04 inches.

The frost-free period for 1963 was initiated on March 23, 1963. During the eight-month period extending from March through October, 24.76 inches of moisture fell. This amount approaches the average; however, rainfall during May was .84 inch below normal and during June fell 2.46 inches below normal.

CHAPTER III

METHODS

Collection and Preparation Procedures

Collections of big bluestem and little bluestem were made at approximately one-month intervals for a period extending from September 28, 1962, to September 5, 1963, while collections of Indiangrass were made from September 28, 1962, to October 7, 1963.

Three samples of each species were chosen for analysis each month. Samples were selected on the basis of being free of contamination by other species. Difficulty was encountered in separating the entangled network of roots in clumps composed of several species. No statistically oriented sampling technique was used but since grazing was uniform throughout the pasture, it was felt that sampling was not significantly biased.

Collections were made some time between the hours of 9:00 a.m. and 11:00 a.m. on the collection dates. Mulch cover was removed from the samples but top growth was left intact. A four-inch cube of sod was removed from the center of the clump.

The soil was removed from the roots by directing a warm water spray, regulated by a nozzle, at the roots. Forbs, annuals, and perennial grass contaminants were then removed from the clumps after measurement. (No height measurements were taken during dormancy.) The shoot growth was cut to crown level using a small pair of pruning

shears. The larger woody rhizomatous masses and the densely compacted aggregates of roots in the tillering regions were also cut and separated at this time.

The roots were thoroughly washed a second time under a hand faucet to insure the removal of all soil. The samples were then put in labeled paper sacks and placed in a drying oven. Enzymatic activity of the roots was halted by heating at 100° C. for two hours. It was essential that the washing operation be completed as quickly as possible since enzymatic breakdown of carbohydrates continues in excised roots (Hischke 1961 and Kinsinger 1953). The washing operation was normally completed within an hour. Drying was completed by heating at 80°C. from 24 to 36 hours. Because of the woody nature of the roots, each sample was pulverized in a large cast iron mortar and pestle before grinding in a Wiley mill to pass a forty-mesh screen. A vacuum cleaner was used to clean the mill after each grinding operation. The ground samples were collected in small bottles, redried at 80° C. from 12 to 24 hours and then tightly sealed.

Extraction

Extraction was accomplished using a procedure suggested by Guinn (1962)[1].

Twenty milliliters of 80% ethanol were added to a 100 milligram sample in a plastic centrifuge tube. After the sample was thoroughly agitated in an automatic shaker for one hour, the ethanol extract was separated from the bulk root tissue by centrifuging at 9,750 X g for

---

[1]Guinn, Gene, 1962. Unpublished data. Oklahoma State University, Stillwater, Oklahoma.

ten minutes and filtering the digest through a Buchner funnel under suction into test tubes.

The ethanol was eliminated from the samples by evaporating in a boiling water bath. Ten milliliters of each ethanol solution were pipetted into 25-milliliter graduated test tubes. Small carborundum stones were placed in each graduated tube to insure even boiling. After approximately seven milliliters of the ethanol were eliminated by boiling, five milliliters of distilled and deionized water were added. The other three milliliters of the ethanol were then eliminated. This operation dispersed the carbohydrates in a water medium.

The carbohydrates were quantitatively transferred to ten-milliliter graduated tubes. To insure complete transfer of the carbohydrates, three separate one-milliliter rinsings were made and these rinsings added to the ten-milliliter graduated tubes. The volume was then made to ten milliliters.

Five milliliters of the water-carbohydrate solution were transferred to a glass centrifuge tube and 0.2 milliliters of saturated lead acetate solution were added. The solution was thoroughly stirred and allowed to set for at least five minutes. Lead acetate will coagulate any protein present in the tube. Excess lead acetate was precipitated by adding 0.6 milliliters of saturated sodium diphosphate. (Excess sodium phosphate does not interfere appreciably with the analysis.)

The contents of the tube were centrifuged for ten minutes at 9,750 X g and the clear supernatent was removed for analysis. All of the ethanol-extractable, water-soluble carbohydrates were present in the supernatent.

Analysis

The samples were analyzed by a colorimetric method developed by DuBois, et al. (1951 and 1956) as modified by Guinn[2] (1962).

One milliliter of the sugar sample was added to the test tube. Two milliliters of 5% phenol were added to the tube and the solution was mixed with the Vortex mixer. Five milliliters of concentrated sulfuric acid were pipetted directly into the solution with a fast delivery pipette within ten to twenty seconds. The tube was allowed to cool to room temperature before absorbance was measured with a Klett Summerson colorimeter equipped with a green No. 54 filter.

The basis for the reaction is that simple sugars, oligosaccharides, polysaccharides, or their derivitives that contain a free or partially free reducing group will give a stable orange-yellow color when treated with phenol and concentrated sulfuric acid. Although the color is not strictly proportional, it can be used for quantitative microcolorimetric determination of reducing sugar when compared to a standard curve.

The mechanism of the reaction is not known, but concentrated sulfuric acid does split polysaccharides, freeing potentially active aldose and ketose reducing groups. A high maximum temperature is desired for the reaction because it increases the sensitivity of the test. Therefore, the sulfuric acid must be directed rapidly against the liquid surface to insure a rapid reaction with the resultant liberation of a great quantity of heat.

A series of blanks had to be prepared for each group of samples to eliminate optical density error. Concentrated sulfuric acid gives

---

[2]See footnote, p. 9

some color with the samples in the absence of phenol. The optical density of the sulfuric acid and phenol will also introduce error. To enable a net reading indicating true optical density, three types of blanks were prepared.

(1) Water blank - three ml. of H_2O and five ml. of concentrated H_2SO_4.

(2) Sample blank - one ml. of sample, two ml. of H_2O and five ml. of concentrated H_2SO_4.

(3) Reagent blank - one ml. of H_2O, two ml. of 5% phenol and five ml. of concentrated H_2SO_4.

To eliminate error, the following steps were carried out:

(1) The colorimeter was adjusted to zero using the water blank and the optical density of the sample blanks were determined.

(2) The colorimeter was readjusted to zero using the reagent blank and the optical density of the samples were determined.

(3) The sample blank readings were subtracted from the sample reading to give the net colorimetric readings which are proportional to the reducing sugars present.

All of the samples collected during one month were analyzed at one time. Only one water blank and one reagent blank were required, but a sample blank was required for each sample. Variation in sugar content of the different samples caused fluctuation in the sample blank density error.

Two series of tests were run on each month's sample. If a difference greater than six colorimetric units was present when the two values of the samples were compared, the sample was rerun. The restriction of no greater variation than six colorimetric units was

arbitrarily chosen after several test runs.

Standard Curve

The net colorimetric readings were compared against a standard curve prepared from a series of known glucose concentrations. A stock solution of 200 micrograms per milliliter was prepared by mixing 200 milligrams of glucose with one liter of water. The dilution series ranged from ten micrograms per milliliter to 200 micrograms per milliliter. The preparational procedure used in making the dilutions is illustrated in Table II.

TABLE II

DILUTION SERIES FOR PREPARATION OF STANDARD CURVE

Dilution mg./liter	ml. stock	Ml. H_2O
200	undiluted solution	0
150	6 " "	2
100	25 " "	25
80	8 (100 mg. dilution)	2
60	6 " " "	4
40	4 " " "	6
20	2 " " "	8
10	1 " " "	9

To assure an accurate comparison between the standard curve and the samples, identical steps were used in preparing and analyzing the standard dilutions as were used in sample preparations. The glucose blank was determined against the water blank and the standard glucose sample was determined against the reagent blank. Lead acetate and saturated disodium phosphate were also added to each sample.

Three trials were made for each dilution and a mean value determined. These values are given in Table III.

TABLE III

NET COLORIMETRIC READINGS OF STANDARD

Conc. mg/l	10	20	40	60	80	100	150	200
Trial 1	12.7	19.9	38.6	54.0	71.5	84.8	115.8	149.1
" 2	10.2	21.6	43.5	57.2	74.0	87.6	118.5	150.3
" 3	13.2	18.2	40.2	56.3	73.9	86.5	120.7	153.3
$\bar{X}$	12.0	20.0	40.8	55.8	73.1	86.3	118.3	150.9

The average Klett readings were then plotted against the known dilutions to obtain the standard curve (Figure 3).

Conversion of Standard Units to Root Sample Content

The comparison of standard units was determined mathematically. Initially the carbohydrate extract from 100 milligrams of sample was diluted in twenty milliliters of ethanol. The ethanol was evaporated from only ten milliliters of the digest and dilution was made back to ten milliliters with distilled water. Since lead acetate and sodium diphosphate were added in both the sample determinations and the standard determinations, the resulting dilutions can be neglected. One milliliter of the ten milliliter solution was analyzed for sugar content. The concentration of sugar in the sample was obtained by comparing each reading with the standard curve and then multiplying this value by 0.2 to convert to milligrams per gram dry weight of sample.

The conversion factor of 0.2 was obtained as follows:
[ug/ml (from standard curve) X 1 mg/1000 ug X 20 ml/100 mg X 1000 mg/g = 0.2 mg/g (milligrams of sugar per gram dry weight or sample)].

A summary of the colorimetric readings, standard glucose units, conversion units, and height and condition of the samples is given in Appendix A.

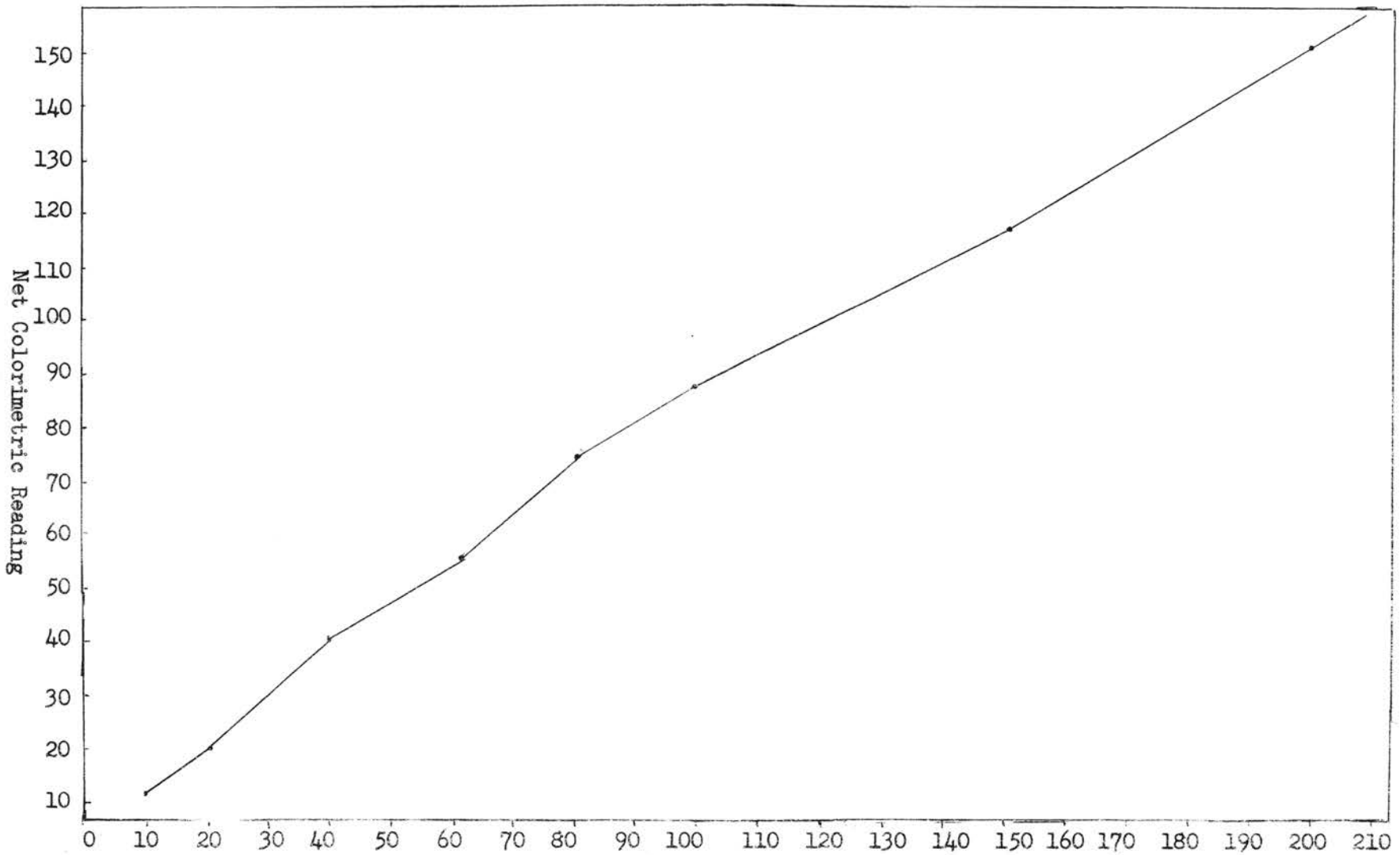

Figure 3. Standard Curve (Micrograms/milliliter of Glucose).

Statistical Analysis

Since carbohydrate level is known to vary during the different seasons of the year, the samples taken each month were considered as separate treatments. Statistical analyses were run to test for significant differences between the treatments. Groupings were made of the months that had no significant differences.

Because the different treatments represented the criterion for classification of the data, a one-way classification utilizing subsampling units was used (Steele and Torre 1960).

Duncan's new multiple range test was used to test for significant differences. Duncan's test allows comparison of each treatment mean with all of the treatment means (Steele and Torre 1960). Analysis of variance tables were set up and standard errors computed. Significant differences were tested at the 5% level.

Ranges for the number of means involved in the comparison were obtained from tables (Steele and Torre 1960) and were multiplied by the standard error to give the least significant ranges.

The treatment means were then ranked in increasing order. If the least significant difference for the number of means involved in a particular comparison was less than the subtracted difference of the treatment values, the difference in treatments was declared non-significant.

The statistical design with the determined experimental data, analysis of variance tables, and the Duncan's new multiple range tests for each species are listed in Appendix B.

The various mathematical operations and required formulae are given in Appendix C.

CHAPTER IV

RESULTS AND DISCUSSION

Phenologically, all three grasses were showing the first signs of spring rejuvenation by April 4. Greening of older stalks and the appearance of some new stalks were observed on that date. Rapid expansion growth occurred in May with blade length averaging 22.5 inches at the collection time. Only slight additional blade expansion occurred during June, July, and August.

Culm elongation was generally completed by September. Seed head formation and anthesis were observed in the bluestem grasses, but flowering seemed to lag in Indiangrass. Many of its inflorescences were in the boot stage. Seed formation and dispersal were evident in all three species during October.

A wide diversity in flowering dates was evident among the clones of each species. Some clones formed no stalks while many heavily grazed clones had only one or two seed stalks. Lack of seed stalk formation might have been partly caused by overgrazing, but Hutchinson (1960) observed that many clones of the bluestem grasses and Indiangrass failed to flower in a relict area. Rice (1950) suggests that a shoot may not initiate flowering if it hasn't acquired a certain number of expanded blades by a certain prerequisite date. The number of blades and the date varies with the species.

The first frost was November 4, 1962. Both of the bluestem grasses appeared dormant at the November collection date, but scattered shoots of Indiangrass still appeared green. A search of the literature revealed no other information to substantiate this, but Hazell (1964) observed that Indiangrass was a later maturing grass than the two bluestem grasses. All three species appeared dormant by the end of the month. As continuous heavy grazing occurred, uncontaminated clones of big bluestem became increasingly difficult to locate.

There was overlapping in the significant ranges of each of the species (Table IV). This might be expected because of the wide variation among individuals within a species. In spite of the overlapping of the significant ranges, some specific trends are indicated.

The largest drop in carbohydrates occurred in May in all three grasses during the period of rapid growth (Figure 4). However, this drop in Indiangrass and little bluestem was not significantly different from a second general plateau which included the months of June, July, August, and September. Since dormancy is broken in April, one would expect an increase in enzymatic activity along with a corresponding decrease in the carbohydrate level. After this period of rapid expenditure of the stored carbohydrates in May and to a lesser degree in June, the carbohydrate level would tend to increase. The carbohydrate level can only maintain itself during this period because some growth is occurring. The months of July and August are generally dry and hot. The harsher environmental conditions discourage growth. Culm growth, flowering, and seed production might also tend to cause some expenditure of root reserves.

TABLE IV

SIGNIFICATE RANGE FOR SEASONAL FLUCTUATION

Big Bluestem

12 May 62	3 Aug. 63	11 Jun. 63	5 Apr. 62	28 Sept. 62	5 Sept. 63	11 Jul. 63	7 Mar. 63	10 Nov. 62	28 Nov. 62	11 Jan. 63	7 Feb. 63
3.2	9.1	12.6	13.1	13.1	13.3	17.0	20.6	22.7	24.0	25.7	29.9

Little Bluestem

12 May 63	5 Apr. 63	11 Jun. 63	28 Sept. 62	5 Sept. 63	3 Aug. 63	11 Jul. 63	10 Nov. 62	29 Nov. 62	7 Mar. 63	11 Jan. 62	7 Feb. 63
8.3	10.6	12.5	13.4	13.4	14.9	15.5	17.5	18.7	20.1	21.0	23.9

Table IV (Continued)

Indiangrass

12 May 63	11 Jun. 63	5 Sept. 63	28 Sept. 62	7 Feb. 63	7 Mar. 63	11 Jun. 63	5 Apr. 63	3 Aug. 63	10 Nov. 62	11 Jan. 63	7 Oct. 63	29 Nov. 62
6.2	13.7	15.5	20.9	22.3	22.8	23.5	25.2	26.2	26.7	28.4	29.9	34.7

The treatment means are ranked in ascending order. If the difference of the treatment means in a comparison is numerically greater than the least significant range, the difference was declared significant. Any two means not underscored by the same line are significantly different. Any means underscored by the same line are not significantly different.

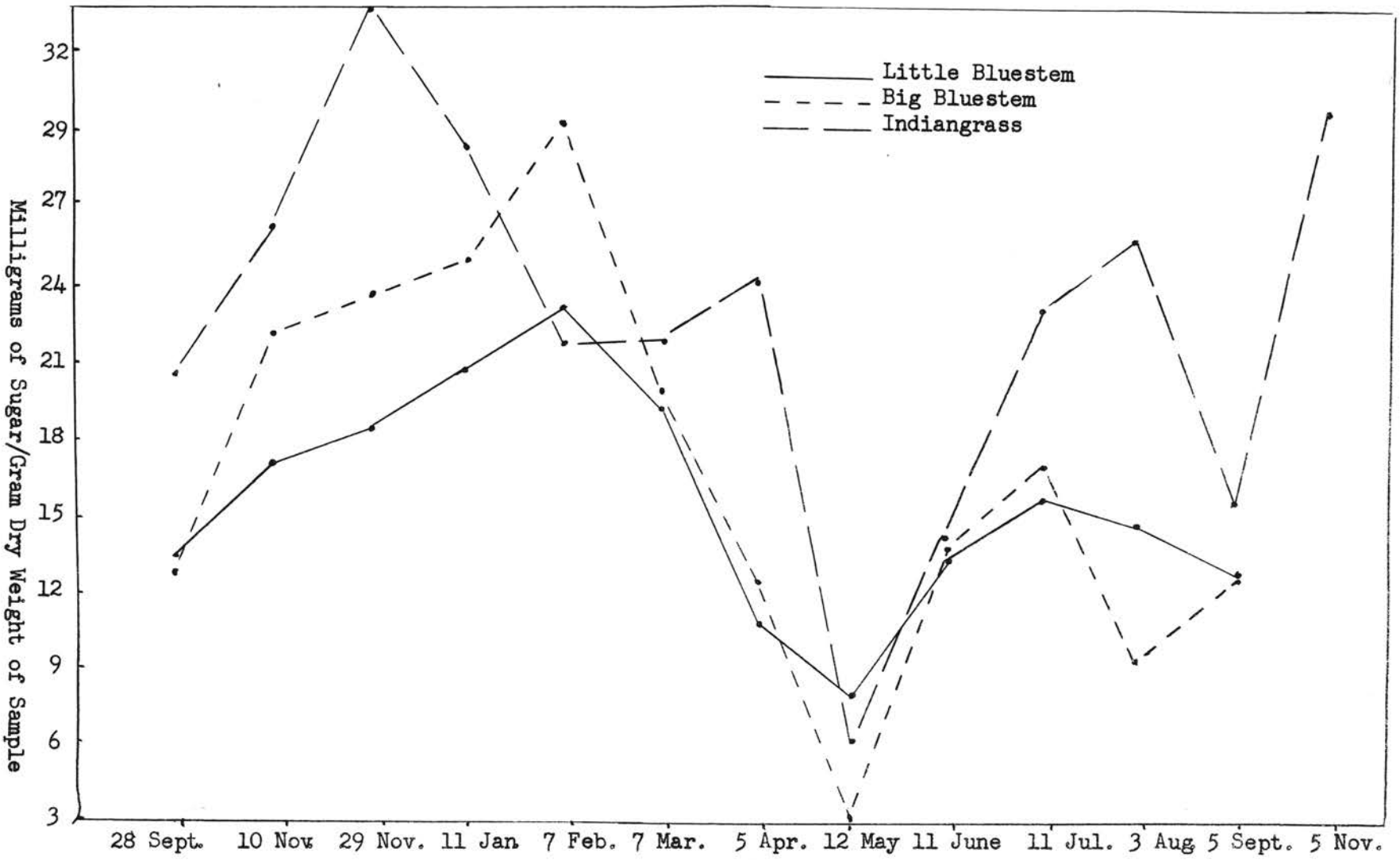

Figure 4. Milligrams of Sugar / Gram Dry Weight of Sample.

The months of October, November, December, January, February, and March constitute a third plateau of ranges. At dormancy little enzymatic activity occurs and the carbohydrate content is maintained at a high level.

The average carbohydrate level of Indiangrass was higher in April than the values for the bluestem grasses. At the collection date on September 5, 1963, the average value was also lower than that of the bluestem grasses. For this reason samples of Indiangrass were collected and analyzed for the month of October. The mean carbohydrate level showed an increase at this date.

It is difficult to draw assumptions since only three samples were collected each month and variations were introduced because of extremes in grazing and moisture conditions. But these data indicate that Indiangrass is slower in breaking dormancy in the spring as well as accumulating reserves for winter. At the date of the September collection, Indiangrasses lagged behind the bluestem grasses in flowering. This would indicate a crop of carbohydrates in early flowering. Hischke (1961) measured no drop in carbohydrates during flowering; McCarty (1938) did find a decrease during this period.

All three grasses are decreasers on the tall grass prairie (Weaver and Hansen 1941, Weaver and Tomanek 1951). Apparent loss of vigor was more evident in big bluestem than the other grasses. It is a highly nutritive grass preferred by livestock and is the first of the three climax grasses to decrease under unfavorable grazing conditions (Weaver 1954). The extremely low value for big bluestem registered in August, 1963, may be due to the factors of extended overgrazing and lack of moisture.

The carbohydrate level of little bluestem was comparatively lower than the other two species, although it is the last of the three species to decrease in the disturbed tall grass prairie. It is a bunch grass with the younger, more succulent blades appearing at the base of the clumps. The older central portion of the clumps are coarse and less palatable. The grass is harder to graze and is disturbed to a lesser degree by livestock when more palatable grasses are available. It is also more predominant on upland areas characterized by more shallow soil while Indiangrass and big bluestem inhabit the lowland areas characterized by deeper soils. These lowland areas are probably more accessible to grazing because of the terrain. Little bluestem, however, does not withstand grazing as well as big bluestem or Indiangrass (Weaver 1954), lingering longer on rougher, lesser grazed areas.

The mean carbohydrate level of Indiangrass was somewhat higher than big bluestem. In mixed clumps of the two grasses, Indiangrass seemed to increase as the vigor of big bluestem decreased. Weaver (1954) states that Indiangrass increases in big bluestem communities when occasional flooding or repeated rains occur. Young seedlings of Indiangrass are also more drought resistant. Hazell (1964) also noted that Indiangrass was more abundant in a cemetery mowed late in the growing season as compared to a native meadow in excellent grazing condition. He suggests that the later maturation date of Indiangrass may be a factor, late mowing being less detrimental.

It might be noted, however, that in ranges severely disturbed by grazing over a period of years, occasional fruiting culms of big bluestem can be observed while Indiangrass and little bluestem are absent. These culms are generally short in height, low in vigor,

and occur singularly.

Since the experimentation measured only ethanol extracted water-soluble sugars; carbohydrates such as starch and hemicellulose were eliminated. McCarty and Price (1942) noted that as dormancy approaches, high values of soluble sucrose are associated with lower values of nonsoluble starch. These factors may be associated with winter hardening. A fluctuation of hemicellulose was also noted, but McCarty and Price feel that hemicellulose is mainly important as a structural component of the cell wall rather than a storage material.

McCarty (1938) and Kinsinger (1953) found that hemicellulose in big bluestem decreased during dormancy as starch increased. Hischke (1961) noted a decrease in sucrose and reducing sugars and an increase in starch formation during the winter months. He observed a total increase in carbohydrates and felt that structural carbohydrate components may be converted to carbohydrates available for physiological activity. At spring rejuvenation Hischke observed a drop in sucrose and reducing sugars but an increase in starch.

McCarty (1938), however, found that the individual carbohydrates generally showed similar reactions to the combined results. This agreed more with the results of this experimentation. Monthly variation was observed within the groupings, but these differences were not generally significant. Fluctuation of carbohydrate values was observed among the samples collected each month. This variation was due in part to the different levels of carbohydrates in the roots.

CHAPTER V

SUMMARY

Variations in the root reserves of three native prairie grasses, big bluestem, little bluestem, and Indiangrass, were studied. The study was conducted on a range previously subjected to continuous moderate grazing. The range, however, was subjected to a period of severe grazing during the study which undoubtedly altered results.

The little bluestem and big bluestem were collected at approximate monthly intervals over a twelve-month period extending from September 25, 1962, to September 5, 1963, while Indiangrass was collected over a thirteen-month period extending from September 25, 1962, to October 7, 1963.

The ethanol-extractable water-soluble carbohydrates were measured. An inverse correlation between growth and reserves was found. During periods of rapid growth in the spring, carbohydrate values were low. As winter dormancy approached, growth declined and carbohydrate levels increased.

The carbohydrate values could be grouped into three general plateaus. Dormancy was broken in April and rapid expansion growth occurred in May. The carbohydrate level dropped in April and decreased greatly in May.

The months of June, July, August, and early September constituted a second general plateau. Carbohydrate values increased during these months.

There was little expansion of blade growth but culm elongation and flowering occurred during this period. Data were inconclusive as to the importance of flowering in depleting reserves, but there was indication that accumulation did not occur during the flowering and culm elongation period.

The months of October, November, December, January, February, and March constituted a third plateau. Carbohydrate level began to increase in September and a high accumulation was evident in October. A fluctuation in carbohydrate level occurred during the winter months, but these differences were not statistically significant.

The level of ethanol-extracted water-soluble carbohydrates was less in little bluestem than in Indiangrass and big bluestem. Indiangrass maintained a higher level than big bluestem. Indiangrass also seemingly lagged behind both of the bluestem grasses in initiation of spring growth, flowering, accumulation of carbohydrates, and entering dormancy.

There was an apparent drop in vigor of big bluestem and an increase in vigor of Indiangrass. Little bluestem remained relatively unchanged. These observations were made by visual inspection only.

The higher level of carbohydrates gives Indiangrass a temporary advantage. Little bluestem is less palatable and is less severely grazed. Little bluestem, however, does not withstand as severe grazing as the other two grasses (Weaver 1954) and remains longest on grazed areas. The comparatively lower level of carbohydrates in little bluestem supports these statements. Although there is a temporary increase in vigor in Indiangrass under grazing conditions, personal observations in other study areas indicate that big bluestem also has a higher

vigor than Indiangrass. In pastures subjected to extremely heavy grazing over a period of years, occasional struggling stalks of big bluestem can still be found, while little bluestem and Indiangrass are absent.

The greatest drop in the measured carbohydrates was during the month of May in all three species. This was the period of most rapid growth. The greatest accumulation levels were during late fall as dormancy approached and during the winter period when enzymatic activity normally decreases.

LITERATURE CITED

Anderson, K. L., 1961. Common names of a selected list of plants. Kan. Agr. Exp. Station Tech. Bull. 117.

Aldous, A. E., 1930. Relation of food reserves to growth of some pasture plants. Jour. of Amer. Soc. Agron. 22: 385-392.

Biswell, H. H. and J. E. Weaver, 1933. Effect of clipping on the development of roots and tops of grasses in prairie sods. Ecology. 14(4): 368-390.

Buckey, I. S. and S. N. Weaver, 1939. Effects of frequent clippings on the underground food reserves of certain prairie grasses. Ecology 20: 246-252.

Cook, C. W., L. A. Stoddart and Floyd E. Kinsinger, 1958. Responses of crested wheat grass to various clipping treatments. Ecol. Monog. 28: 231-272.

Crockett, Jerry J., 1960. Effects of intensity of clipping on three range grasses from grazed and ungrazed areas in West Central Kansas. Unpublished Master's Thesis. Fort Hays Kansas State College, Fort Hays, Kansas.

Dodd, Jimmie D. and Harold H. Hopkins, 1958. Transactions of the Kansas Academy of Science 61(3): 280-287.

Dubois, M., K. Gilles, J. K. Hamilton, P. A. Rebers and F. Smith, 1951. A colorimetric method for determination of sugars. Nature 168: 167.

_____Michel, K. A. Gilles, J. K. Hamilton, P. A. Rebers and Fred Smith, 1956. Colorimetric determination of sugars and related substances. Anal. Chem. 28: 350-356.

Graber, L. A., 1931. Food reserves in relation to other factors limiting the growth of grass. Plant Physiol. 6: 42-72.

_____, N. T. Nelson, W. A. Luckol and W. B. Albert, 1927. Organic food reserves in relation to the growth of alfalfa and other perennial herbaceous plants. Univ. Wis. Agr. Exp. Res. Bull. 80.

Hazell, Don B., 1964. Forage production, vegetative composition and plant vigor in relation to range condition. Unpublished doctor's thesis. Oklahoma State University, Stillwater, Oklahoma.

Hischke, Herbert H., Jr., 1961. Carbohydrate reserves of three native prairie grasses. Unpublished master's thesis. Fort Hays Kansas State College, Fort Hays, Kansas.

Hutchinson, H. L., 1963. Developmental growth of four species of range grasses in North Central Oklahoma. Unpublished master's thesis. Oklahoma State University, Stillwater, Oklahoma.

Hyder, D. W. and F. A. Sneva, 1959. Growth and carbohydrate trends in crested wheat grass. Jour. Range Mgt. 12: 273-276.

Kinsinger, Floyd E., 1953. Forage yield and carbohydrate content of underground parts of grasses affected by clipping. Unpublished master's thesis. Fort Hays Kansas State College, Fort Hays, Kansas.

McCarty, B. C. and R. Price, 1942. Growth and carbohydrate content of important mountain forage plants in Central Utah as affected by clipping and grazing. U. S. Dept. Agr. Tech. Bull. 818.

_____, Edward, 1938. The relationship of growth to the varying carbohydrate content in mountain brome. U. S. Dept. Agr. Bull. 598.

Parker, K. W. and A. W. Sampson, 1931. Growth and yield of certain Gramineae as influenced by production of photosynthetic tissue. Hilgardia, 5(10): 361-381.

Rice, E. L., 1950. Growth and floral development of five species of range grasses in Oklahoma. Bot. Gaz. 11: 361-367.

Sampson, Arthur W. and Edward C. McCarty, 1930. The carbohydrate metabolism of Stipa pulchra. Hilgardia, 5: 61-100.

Sims, Phillip E., 1964. The pattern of retrogression of native vegetation in North Central Oklahoma. Unpublished master's thesis. Oklahoma State University, Stillwater, Oklahoma.

Steele, Robert G. D. and James H. Torrie, 1960. Principles and Procedures of Statistics. McGraw-Hill Book Company, Inc., New York, 99-133.

Stoddart, Laurence A. and Arthur D. Smith, 1955. Range Management, 2nd ed. McGraw-Hill Book Company, Inc., New York, 98-113.

United States Weather Bureau, 1963. Climatological Data for Oklahoma. Washington, D. C.

Weaver, J. E., 1954. North American Prairie, Johnson Publishing Company, Lincoln, Nebraska. 23-53 and 274-276.

_____ and G. W. Tomanek, 1951. Ecological studies in a midwestern range. The vegetation and effects of cattle on its composition and distribution. Univ. Nebraska Cons. and Surv. Div. 31.

Weaver, J. E. and R. W. Darland, 1946. Changes in vegetation and production of forage resulting from grazing in a lowland prairie. Ecology 29: 1-29.

_____ and W. W. Hansen, 1941. Native midwestern pastures, their origin, composition, and degeneration. Univ. Nebr. Cons. and Surv. Div. Bull. 22.

APPENDIX A

CONVERSION OF NET ADJUSTED COLORIMETRIC READINGS TO STANDARD GLUCOSE UNITS (MICROGRAMS PER MILLILITER SAMPLE) AND CONCENTRATION OF ETHANOL-EXTRACTED WATER-SOLUBLE SUGARS PER GRAM DRY WEIGHT OF ROOT (MILLIGRAMS OF SUGAR PER GRAM DRY WEIGHT)

Big Bluestem

Date of Collection	Sample	Trial	Net Colorimetric Readings	Standard Units Ug/ml	Height	Gm.dry wt.
	1	1	53.7	57	35"	11.4
		2	53.9	57		11.4
28 Sept. 62	2	1	60.9	65	42"	13.0
		2	64.4	69		13.8
	3	1	66.6	72	29"	14.4
		2	66.7	72		14.4
	1	1	102.0	125	Dormant	25.0
		2	98.1	116		23.2
10 Nov. 62	2	1	95.8	116		23.2
		2	98.7	120		24.0
	3	1	85.0	99		19.8
		2	89.1	105		21.0
	1	1	103.1	127		25.4
		2	106.3	132		26.4
28 Nov. 62	2	1	96.5	117		23.4
		2	102.9	127		25.4
	3	1	90.2	108		21.6
		2	92.4	111		22.2
	1	1	126.9	164		32.8
		2	127.6	165		33.0
11 Jan. 63	2	1	94.5	114		22.8
		2	97.3	118		23.6
	3	1	87.4	102		20.4
		2	90.6	108		21.6
	1	1	138.8	167		33.4
		2	143.7	190		38.0
7 Feb. 63	2	1	119.6	153		30.6
		2	112.2	141		28.2
	3	1	97.8	119		23.8
		2	102.7	126		25.2

Appendix A (Continued)

Date of Collection	Sample	Trial	Net Colori-metric Readings	Standard Units Ug/Ml	Height	Mg. sugar/ Gm.dry wt.	
	1	1	18.9	90		18.0	
		2	79.7	91		18.2	
7 Mar. 63	2	1	101.4	124		24.8	
		2	96.6	116		23.2	
	3	1	83.7	98		19.6	
		2	81.7	94		18.8	
	1	1	57.2	61	Greening	12.2	
		2	58.0	64	of older	12.8	
5 Apr. 63	2	1	78.9	90	stalks	18.0	
		2	76.4	86	observed;	17.2	
	3	1	41.9	44	some new	8.8	
		2	46.0	48	growth	9.6	
	1	1	15.6	14	23"	2.5	Blade growth
		2	15.9	15		3.0	
12 May 63	2	1	15.3	14	27"	2.8	
		2	12.6	11		2.2	
	3	1	20.1	119	28"	3.8	
		2	22.2	22		4.4	
	1	1	56.0	60	23"	12.0	
		2	52.4	56		11.2	
11 Jun. 63	2	1	52.9	56	22"	11.2	
		2	55.6	60		12.0	
	3	1	70.5	77	33"	15.4	
		2	64.7	70		14.0	
	1	1	64.2	69	18"	13.8	
		2	66.7	72		14.4	
11 Jul. 63	2	1	85.2	100	11"	20.0	
		2	89.1	106		21.2	
	3	1	73.4	81	11"	16.2	
		2	74.9	82		16.3	
	1	1	46.3	49	23"	9.8	
		2	42.1	44		8.8	
3 Aug. 63	2	1	43.1	45	18"	9.0	
		2	48.5	52		10.4	
	3	1	37.6	39	20"	7.8	
		2	40.7	43		8.6	
	1	1	66.1	71	18"	14.2	
		2	65.4	69		13.8	
5 Sept. 63	2	1	60.7	65	29"	13.0	flowering observed
		2	61.6	66		13.2	
	3	1	59.8	64	23"	12.8	
		2	58.4	62		12.4	

Appendix A (Continued)

Little Bluestem

Date of Collection	Sample	Trial	Net Colorimetric Readings	Standard Units Ug/ml	Height	Mg. Sugar/ Gm.dry wt.
	1	1	58.2	63	27"	12.6
		2	62.5	67		13.4
28 Sept. 62	2	1	64.2	69	26"	13.8
		2	66.5	70		14.0
	3	1	61.8	66	23"	13.2
		2	62.6	67		13.4
	1	1	75.8	85	Dormant	17.0
		2	78.8	90		18.0
10 Nov. 62	2	1	65.1	70		14.0
		2	64.5	69		13.8
	3	1	86.5	102		20.4
		2	91.7	109		21.8
	1	1	72.0	79		15.8
		2	69.2	75		15.0
29 Nov. 62	2	1	95.0	115		23.0
		2	97.0	118		23.6
	3	1	77.4	87		17.4
		2	74.2	82		16.4
	1	1	83.7	97		19.4
		2	88.7	105		21.0
11 Jan. 63	2	1	91.0	109		21.8
		2	95.2	115		23.0
	3	1	87.0	102		20.4
		2	86.4	101		20.2
	1	1	110.1	138		27.6
		2	115.0	145		29.0
7 Feb. 63	2	1	85.2	101		20.2
		2	88.1	104		20.8
	3	1	95.2	115		23.0
		2	95.1	115		23.0
	1	1	95.4	115		23.0
		2	93.5	112		22.4
7 Mar. 63	2	1	93.5	112		22.4
		2	89.4	106		21.2
	3	1	75.0	83		16.6
		2	76.6	86		17.2
	1	1	58.1	62	Greening	12.4
		2	59.5	64	of older	12.8
5 Apr. 63	2	1	38.7	40	stalks	8.0
		2	35.6	37	observed	7.4
	3	1	54.1	58		11.6
		2	54.4	58		11.6

Appendix A (Continued)

Date of Collection	Sample	Trial	Net Colori-metric Readings	Standard Units Ug/ml	Height	Mg. sugar/ Gm.dry wt.
	1	1	39.2	41	18"	8.2
		2	36.0	37		7.4
12 May 63	2	1	28.7	24	17"	5.8
		2	31.9	31		6.2
	3	1	49.1	52	17"	10.4 Blade growth
		2	55.0	58		11.6
	1	1	55.7	60	25"	12.0
		2	60.6	65		13.0
11 June 63	2	1	67.1	72	25"	14.4
		2	64.2	69		13.8
	3	1	51.0	54	28"	10.8
		2	52.3	56		11.2
	1	1	71.2	78	18"	15.6
		2	60.6	65		15.2
11 July 63	2	1	82.0	95	17"	19.0
		2	76.4	86		17.2
	3	1	77.1	87	19"	17.4
		2	81.4	94		18.8
	1	1	67.1	73	20"	14.6
		2	71.0	78		15.6
3 Aug. 63	2	1	62.4	67	16"	13.4
		2	60.0	65		13.0
	3	1	71.4	78	19"	15.6
		2	76.5	86		17.2
	1	1	63.0	68	36"	13.6
		2	62.2	62		13.4
5 Sept. 63	2	1	69.8	76	25"	15.2 flowering observed
		2	65.0	70		14.0
	3	1	54.3	58	34"	11.6
		2	57.3	61		12.2

Appendix A (Continued)

Indiangrass

Date of Collection	Sample	Trial	Net Colorimetric Readings	Standard Units Ug/ml	Height	Mg.Sugar/ Gm.dry wt.
	1	1	79.0	90	35"	18.0
		2	77.3	87		17.4
28 Sept. 62	2	1	126.0	161	30"	32.2
		2	129.2	167		33.4
	3	1	73.7	82	40"	16.4
		2	79.5	91		18.1
	1	1	83.0	91	Dormant	19.2
		2	81.2	93		18.6
10 Nov. 62	2	1	92.2	110		22.0
		2	93.6	113		22.6
	3	1	144.5	191		38.2
		2	149.7	199		39.8
	1	1	144.3	190		38.0
		2	143.8	189		37.8
29 Nov. 62	2	1	143.5	190		38.0
		2	146.1	193		28.6
	3	1	107.7	135		27.0
		2	113.9	144		28.8
	1	1	105.8	131		26.2
		2	107.3	134		16.8
11 Jan. 63	2	1	127.8	165		33.0
		2	129.1	167		33.4
	3	1	102.6	126		25.2
		2	104.0	130		26.0
	1	1	89.8	107		21.4
		2	92.8	111		22.2
7 Feb. 63	2	1	54.8	59		11.8
		2	56.6	61		12.2
	3	1	126.2	163		32.6
		2	129.4	168		33.6
	1	1	117.2	149		29.8
		2	115.0	146		29.2
7 Mar. 63	2	1	83.2	97		19.4
		2	86.5	102		20.4
	3	1	82.3	95		19.0
		2	83.1	96		19.2

Appendix A (Continued)

Date of Collection	Sample	Trial	Net Colorimetric Readings	Standard Units Ug/ml	Height	Mg. sugar/ Gm.dry wt.	
	1	1	73.1	81	Greening	16.2	
		2	71.7	79	of older	15.8	
5 Apr. 63	2	1	109.1	137	stalks	27.4	
		2	101.7	131	observed;	26.2	
	3	1	125.9	162	some new	32.4	
		2	128.1	166	growth	33.2	
	1	1	39.2	41	23"	8.2	
		2	40.7	43		8.6	
12 May 63	2	1	37.0	38	18"	7.6	
		2	33.3	39		7.8	blade growth
	3	1	13.3	12	17"	2.4	
		2	14.8	13		2.6	
	1	1	58.5	63	24"	12.6	
		2	59.0	64		12.8	
11 June 63	2	1	63.7	69	29"	13.8	
		2	66.3	71		14.2	
	3	1	60.6	69	31"	13.0	
		2	64.0	69		13.8	
	1	1	99.7	122	18"	14.4	
		2	97.6	119		23.8	
11 July 63	2	1	102.0	125	17"	25.0	
		2	106.2	132		26.4	
	3	1	90.8	108	19"	21.6	
		2	85.2	101		20.2	
	1	1	115.5	146	13"	29.2	
		2	115.9	147		29.2	
3 Aug. 63	2	1	106.8	133	5"	26.6	
		2	105.4	131		26.2	
	3	1	15.5	116	9"	11.6	
		2	94.3	114		22.8	
	1	1	82.2	95	24"	19.0	
		2	85.1	106		20.2	
5 Sept. 63	2	1	52.9	156	27"	11.2	flowering observed
		2	55.0	59		11.8	
	3	1	72.8	80	20"	16.0	
		2	68.9	74		14.8	
	1	1	121.0	15.5	36"	31.0	
		2	115.7	148		29.6	
7 Oct. 63	2	1	132.2	172	19"	34.4	
		2	135.1	171		34.2	
	3	1	117.1	149	24"	29.8	
		2	119.0	152		30.4	

APPENDIX B

MILLIGRAMS OF SUGAR PER GRAM DRY WEIGHT OF ROOT
BIG BLUESTEM

	28 Sept. 1962			10 Nov. 1962			28 Nov. 1962			11 Jan. 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	11.4	13.0	14.4	25.0	23.0	19.8	25.4	23.4	21.6	33.8	22.8	20.4
Trial 2	11.4	13.8	14.4	23.2	24.0	21.0	26.4	25.4	22.2	33.0	23.6	21.6
Σ	22.8	26.8	28.8	48.2	47.2	40.8	51.8	48.8	43.8	66.8	46.4	42.0
Σ all trials		78.4			136.2			144.4			154.2	
$\bar{X}$		13.1			22.7			24.0			25.7	

	7 Feb. 1963			7 Mar. 1963			5 Apr. 1963			12 May 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	35.4	30.6	23.8	18.0	24.8	19.6	12.2	18.0	8.8	2.8	2.8	3.8
Trial 2	38.0	28.2	25.2	18.2	23.2	18.8	12.8	17.2	9.6	3.0	2.2	4.4
Σ	71.4	58.8	49.0	36.2	48.0	38.4	25.0	35.2	18.4	5.8	5.0	8.2
Σ all trials		179.2			123.6			78.2			19.0	
$\bar{X}$		29.9			20.6			13.0			3.2	

APPENDIX B - Bluestem (Continued)

	11 June 1963			11 July 1963			3 Aug. 1963			5 Sept. 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	12.0	11.2	15.4	13.8	20.0	16.2	9.8	9.0	7.8	14.2	13.0	12.8
Trial 2	11.2	12.0	14.0	14.4	21.2	16.3	8.8	10.4	8.6	13.8	13.2	12.4
Σ	23.2	23.2	29.4	28.2	41.2	32.5	18.6	19.4	16.4	28.0	26.2	25.2
Σ all trials		75.8			102.2			54.4			79.8	
$\bar{X}$		12.6			17.0			9.1			13.3	

APPENDIX B (Continued)

MILLIGRAMS OF SUGAR PER GRAM DRY WEIGHT OF ROOT
LITTLE BLUESTEM

	28 Sept. 1962			10 Nov. 1962			28 Nov. 1962			11 Jan. 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	12.6	13.8	13.2	17.0	14.0	20.4	15.8	23.0	17.4	19.4	21.8	20.4
Trial 2	13.4	14.0	13.4	18.0	13.8	21.8	15.0	23.6	16.4	21.0	23.0	20.2
Σ	26.0	27.8	26.6	35.0	27.8	42.2	30.8	46.6	33.8	40.4	44.8	40.6
Σ all trials		80.4			105.0			111.2			125.8	
$\bar{X}$		13.4			17.5			18.7			21.0	

	7 Feb. 1963			7 Mar. 1963			5 Apr. 1963			12 May 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	27.6	20.2	23.0	23.0	22.4	16.6	12.4	8.0	11.6	8.2	5.8	10.4
Trial 2	29.0	20.8	23.0	22.4	21.2	17.2	12.8	7.4	11.6	7.4	6.2	11.6
Σ	56.6	41.0	46.0	45.4	43.6	33.8	25.2	15.4	23.2	15.6	12.0	22.0
Σ all trials		143.6			122.8			63.8			49.6	
$\bar{X}$		23.9			20.1			10.6			8.3	

APPENDIX B - Little Bluestem (Continued)

	11 June 1963			11 July 1963			3 Aug. 1963			5 Sept. 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	12.0	14.4	10.8	15.6	19.0	17.4	14.6	13.4	15.6	13.6	15.2	11.6
Trial 2	13.0	13.8	11.2	15.2	17.2	18.8	15.6	13.0	17.2	13.4	14.0	12.2
Σ	25.0	28.2	22.0	30.8	36.2	26.2	30.2	26.4	32.8	27.8	29.2	23.8
Σ all trials		75.2			93.2			89.4			80.2	
$\bar{X}$		12.5			15.5			14.9			13.4	

APPENDIX B (Continued)

MILLIGRAMS OF SUGAR PER GRAM DRY WEIGHT OF ROOT
INDIANGRASS

	28 Sept. 1962			10 Nov. 1962			29 Nov. 1962			11 Jan. 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	18.0	32.2	16.4	19.2	22.0	38.2	38.0	38.0	27.0	26.2	33.0	25.2
Trial 2	17.4	33.4	18.1	18.6	22.6	39.8	37.8	38.6	28.8	26.8	33.4	26.0
Σ	35.4	65.6	24.5	37.8	44.6	78.0	75.8	76.6	55.8	53.0	66.4	51.2
Σ all trials		125.5			160.4			208.2			170.6	
$\bar{X}$		20.9			26.7			34.7			28.4	

	7 Feb. 1963			7 Mar. 1963			5 Apr. 1963			12 May 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	21.4	11.8	32.6	29.8	19.4	19.0	16.2	27.4	32.4	8.2	7.6	2.4
Trial 2	22.2	12.2	33.6	29.2	20.4	19.2	15.8	26.2	33.2	8.6	7.8	2.6
Σ	43.6	24.0	66.2	59.0	39.8	38.2	32.0	53.6	65.6	26.8	25.4	5.0
Σ all trials		133.8			137.0			151.3			37.2	
$\bar{X}$		22.3			22.8			25.2			6.2	

APPENDIX B - Indiangrass (Continued)

	11 June 1963			11 July 1963			3 Aug. 1963			5 Sept. 1963		
Sample	1	2	3	1	2	3	1	2	3	1	2	3
Trial 1	12.6	13.8	13.0	24.4	25.0	21.6	29.2	26.6	23.0	19.0	11.2	16.0
Trial 2	12.8	14.2	13.8	23.8	26.4	20.0	29.4	26.2	22.8	20.2	11.8	14.8
Σ	25.4	28.0	26.8	48.2	51.4	41.6	58.6	52.8	45.8	39.2	23.0	30.8
Σ all trials		80.2			141.2			157.2			93.0	
$\bar{X}$		13.7			23.5			26.2			15.5	

	7 Oct. 1963		
Sample	1	2	3
Trial 1	31.0	34.4	29.8
Trial 2	29.6	34.2	30.4
Σ	50.6	68.6	60.2
Σ all trials		179.4	
$\bar{X}$		29.9	

APPENDIX B (Continued)

ANALYSIS OF VARIANCE TABLES

Big Bluestem

Source of variance	DF*	SS**	MS***
Among samples	35	4388.5	-----
Treatments	11	3867.2	351.6
Among samples within treatment (Experimental error)	24	521.3	21.3
Among trials within samples (Sample error)	36	161.5	4.5
Total	71	4550.0	-----

$S_{\bar{X}} = 1.9$

Little Bluestem

Source of variance	DF*	SS**	MS***
Among samples	35	1409.4	-----
Treatments	11	1078.2	97.4
Among samples within treatment (Experimental error)	24	331.2	13.8
Among trials within samples (Sample error)	36	326.8	9.1
Total	71	1736.2	-----

$S_{\bar{X}} = 1.5$

*DF = Degrees of freedom
**SS = Sum of squares
***MS = Mean squares

APPENDIX B - Analysis of Variance Tables (Continued)

Indiangrass

Source of variance	DF*	SS**	MS***
Among samples	38	5409.6	------
Treatments	12	3345.0	278.8
Among samples within treatment (Experimental error)	26	2064.6	79.4
Among trials within samples (Sample error)	39	853.3	21.9
Total	77	6262.9	------

$S_{\bar{X}} = 3.6$

*DF = Degrees of freedom
**SS = Sum of squares
***MS = Mean squares

The variance tables are determined from data in the statistical design. Completion of the tables yielded information necessary for computation of the standard deviation ($S_{\bar{X}}$) as well as a convenient check for mathematical accuracy of the tables (Steele and Torre 1960).

$$S_{\bar{X}} = \sqrt{\frac{\text{experimental error mean square}}{\text{number of trials in a treatment}}}$$

The mathematical computations are given in Apprendix B.

APPENDIX B (Continued)

DETERMINATION OF LEAST SIGNIFICANT RANGES
FOR DUNCAN'S NEW MULTIPLE RANGE TEST

Big Bluestem

*P	2	3	4	5	6	7	8	9	10	11	12
**SSR(24DF)	2.92	3.07	3.15	3.22	3.28	3.31	3.34	3.37	3.38	3.40	3.41
(SSR · $S_{\bar{X}}$)	5.55	5.83	5.98	6.12	6.19	6.29	6.35	6.40	6.42	6.45	6.48

Little Bluestem

P	2	3	4	5	6	7	8	9	10	11	12
SSR(24DF)	2.92	3.07	3.15	3.22	3.28	3.31	3.34	3.37	3.38	3.40	3.41
(SSR · $S_{\bar{X}}$)	4.38	4.60	4.72	4.83	4.92	4.97	5.01	5.05	5.07	5.10	5.12

Indiangrass

P	2	3	4	5	6	7	8	9	10	11	12	13
SSR(24DF)	2.91	3.06	3.14	3.21	3.27	3.30	3.84	3.36	3.38	3.40	3.41	3.42
(SSR · $S_{\bar{X}}$)	10.50	11.00	11.30	11.56	11.77	11.88	12.02	12.09	12.20	12.30	12.30	12.40

*P = Number of means for range being tested
**SSR = Studentized ranges

Duncan's test allows comparison of one treatment mean with other treatment means. Comparisons are then made between treatments (i.e. between two treatments, three treatments, four treatments, etc.). The studentized ranges were determined at the 5% level with the experimental error degree of freedom 24. The studentized ranges are multiplied by the standard deviation to give the least significant ranges (SSR·$S_{\bar{X}}$) (Steele and Torre 1960).

APPENDIX C

MATHEMATICAL COMPUTATIONS FOR STATISTICAL ANALYSES

Big Bluestem

CF	$(11.4+...+25.2)^2 / 72 = 20920.4$
Total SS	$(11.4)^2+...+(25.2)^2 - CF$ $25470.4 - 20920.4 = 4550.0$
Sample SS	$(22.8)^2+...+(25.2)^2 / 2 - CF$ $25308.9 - 20920.4 = 4388.5$
Among trials within treatment	$4550.0 - 4388.5 = 161.5$
Degrees of Freedom	
Total DF	$72 - 1 = 71$
Sample DF	$36 - 1 = 35$
Among trials within Sample DF	$71 - 35 = 36$
Among samples within treatment DF	$35 - 11 = 24$
Treatment DF	$12 - 1 = 11$

$$S_{\bar{X}} = \sqrt{\frac{\text{Experimental error}}{\text{No. trials in a treatment}}} \qquad \sqrt{\frac{21.7}{6}} = 1.9$$

APPENDIX C (Continued)

Little Bluestem

CF	$(12.6+...+12.2)^2 / 72 = 18368.0$
Total SS	$(12.6+...+12.2)^2 - CF$ $20104.2 - 18368.0 = 1736.2$
Sample SS	$(26.0)^2+...+(23.8)^2 / - CF$ $19777.4 - 18368.0 = 1409.4$
Among trials within Sample SS	$1746.2 - 1409.4 = 326.8$
Treatment SS	$(80.4)^2+...+(80.2)^2 / 6 - CF$ $19446.2 - 18368.0 = 1078.2$
Among samples	$1409.4 - 1078.2 = 331.2$
Degrees of Freedom	
Total DF	$72 - 1 = 71$
Sample DF	$36 - 1 = 35$
Among trials within Sample DF	$71 - 35 = 36$
Among samples within treatment DF	$35 - 11 = 24$
Treatment DF	$12 - 1 = 11$

$$S_{\bar{X}} = \sqrt{\frac{\text{Experimental error}}{\text{No. trials in a treatment}}} \quad \sqrt{\frac{13.8}{6}} = 1.5$$

APPENDIX C (Continued)

Indiangrass

CF	$(18.0+...+30.8)^2 / 78 = 41303.4$
Total SS	$(18.0)^2+...+(30.8)^2 - CF$ $47576.3 - 41303.4 = 6262.9$
Sample SS	$(35.4)^2+...+(60.2)^2 / 2 - CF$ $46713.0 - 41303.4 = 5409.6$
Among trials within Sample SS	$6262.9 - 5409.6 = 853.3$
Treatment SS	$(125.5)^2+...+(179.4)^2 / 6 - CF$ $44648.4 - 41303.4 = 3345.0$
Among Samples within treatment	$5409.6 - 3345.0 = 2064.6$
Degrees of Freedom	
Total DF	$78 - 1 = 77$
Sample DF	$39 - 1 = 38$
Among trials within Sample DF	$77 - 38 = 39$
Among samples within treatment DF	$38 - 12 = 26$
Treatment DF	$13 - 1 = 12$

$$S_{\bar{x}} = \sqrt{\frac{\text{Experimental error}}{\text{No. trials in a treatment}}} \quad \sqrt{\frac{79.4}{6}} = 3.6$$

VITA

VAL KENNETH KERR

Candidate for the degree

of

Master of Science

Thesis: SEASONAL FLUCTUATION OF ETHANOL-EXTRACTABLE WATER-SOLUBLE CARBOHYDRATES IN THREE NATIVE RANGE GRASSES

Major Field: Botany and Plant Pathology

Biographical:

Personal Data: Born August 27, 1937 at Perry, Oklahoma, the son of James C and Emma Kerr.

Education: Attended first grade at Perry Public Schools and received the remainder of the public school at Geary, Oklahoma graduating May 23, 1955. Attended Oklahoma Baptist University and Southwestern State College (Oklahoma). Graduated with a Bachelor of Science degree in Biology from Southwestern State College in July, 1960.

Experience: Served as a laboratory assistant in the Biology Department at Oklahoma Baptist University and Southwestern State College and as a graduate teaching assistant while completing the course requirements for the Master of Science degree at Oklahoma State University. Taught in the Public Schools two years and is a member of the Oklahoma National Guard.

Professional Organizations: Member of National Education Association, Oklahoma Education Association, Oklahoma Academy of Science, and Beta Beta Beta, honorary society for biological science majors.