THE EFFECT OF FERTILIZER TREATMENT ON THE CHEMICAL COMPOSITION OF WHEAT GRASS AND GRAIN IN SOUTHEAST KANSAS

by

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B. S., University of California,
College of Agriculture, 1937

A THESIS

submitted in partial fulfillment of the

requirements for the degree

MASTER OF SCIENCE

Department of Chemistry

KANSAS STATE COLLEGE
OF AGRICULTURE AND APPLIED SCIENCE

1948

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INTRODUCTION

Many investigators have studied the effect of fertilizers on the quality and chemical composition of the wheat grain, but only a few have dealt with its influence on the composition of the growing wheat plant. Since cattle in Kansas are often pastured on wheat grass, a knowledge of the mineral content and the factors affecting it are very important. Therefore, the primary purpose of this research was to study the effect of certain fertilizer treatments on the chemical composition of the wheat plant at different stages of growth. The influence of fertilization on the mineral content of the wheat grain was also investigated.

In 1937, Sotola (34) reported on his studies of the chemical composition of wheat and other cereal hays at different stages of maturity. He suggested that young cereals afforded a high protein pasture and, with the exception of phosphorus and calcium, are almost identical with legume pasture in chemical makeup. Wheat grass has also been suggested as a raw material in chicken feed. It is necessary to maintain the proper mineral content in an animal's ration in order to avoid the development of deficiency diseases. Since a knowledge of the mineral content of the feed is valuable, a study of the factors influencing the composition is an important problem.

Wheat is also an important source of food for human consumption. The baking quality of the flour depends on the composition of the ash. Bayfield (6), in his report on the investigation of the effect of fertilizers on the ash of wheat, stated that the per cent of ash in flour is an important criteria of its quality. He said that wheat of high ash frequently produces high ash flour of undesirable quality. Millers of quality flour try to maintain uniform ash content.

From the standpoint of nutrition it is important to know what part of man's mineral requirement comes from this cereal. When Greaves and Carter (19) reported their findings on the affect of irrigation water on the composition of wheat, they stated that individuals requiring greater quantities of minerals may turn to irrigated grains for a sufficient amount to prevent nutritional disorders. They continued:

> Individuals in need of more ash in the diet could turn to the irrigated grains in preference to the non-irrigated, although by doing so would be getting less protein, and conversely where it is desired to restrict the mineral intake of an individual the non-irrigated grains should be used. . . . .
>
> For the feeding of farm animals in which the production of bone is considered, the irrigated grain would be superior. . . . . . Considering the extent to which irrigation water has modified the mineral elements in these grains it is easy to see how a ration of irrigated cereals would carry sufficient calcium and phosphorus if fed to swine to produce strong normal bones, which is not the case with corn.

Since fertilization also affects the mineral composition of plants, these same conclusions could be made about fertilizer treated cereals. In fact, Beeson and LeClerc (10) stated in their review of the significance of the soil to human and animal nutrition:

> It has been shown that fertilization can affect very materially the composition of the plant, and when it is

> economicel, fertilization of tha plant is probably the baat meens of corracting e mineral deficiency in enimals . . . . Meny inveatigetors in the field of animel nutrition believe thet it ie not only e deficiency of minerala in the plent that leeds to nutritionel disordera in the enimal, but thet it ia often e problem of unbalanced mineral elementa in the plant.

It mey be poeaible to correct theae unbalanced conditiona, if the influence of the fectors effecting the composition were better understood. Kraybill (21), in hie review of the effect of climete end fertilizers on the composition of wheet, stated thet the uee of fartilizera offered e practical meens of influencing the quality of this cereal. However, e note of warning es voiced by Browne (11), ahould ba kept in mind. He werned egeinat exaggereted and sensationel claims for minerelization of humen and enimel foode.

Excellent surveye of the aubject of nutritionel diseasea in cattle have been givan by Browna (11), Beeson (9), end Beeaon end LeClerc (10). Auchter (5) has preeented e aurvey ahowing the interrelation of soil and enimal end human nutrition. Many eccounts were given showing that deficiency diaeases developed ee e result of deficient feed. A few of the striking ceaea will be cited here in order to show the importance of maintaining the proper minerel content end balence in the cettle feed.

In 1920 Cery (12) deacribed e nutritionel disease called "sweeny" or "creeping aickness," which he said was ectuelly osteomalecia. The soil was described ea sendy and deficient in lime. Becker, et el. (8) deecribed e condition in Floride known ae "atiffa" or "aweeny" in which animals chewed bones, wood, and such material and became stiff and lame. It was said to be caused by a shortage of phosphorus in forage, since subnormal inorganic phosphorus in the blood plasma was found. In Pennsylvania, Forbes and Johnson (16) found a phosphorus deficiency disease among cattle that were wintered largely or exclusively on less nutritive roughage such as cereal straw and corn stover without feeding supplements or concentrated feeds.

In 1926 Eckles, et al. (14) reported that the deficiency disease found in 32 counties in Minnesota was more severe in years of drought. The soil in these localities and the alfalfa and prairie hay were low in phosphorus. It was estimated that a cow receiving the phosphorus deficient prairie hay along with the usual ration of oats would receive only 68.6 per cent of this mineral necessary for maintenance and milk supply. It is known that the milk flow is reduced by a lack of phosphorus in the feed.

Hart, et al. (20) in Wisconsin investigated a dairy cattle disease characterized by weakness and distortion of the bones, emaciation, stiffness, unthrifty condition, and a loss of appetite. They found that a ration rich in phosphorus would result in rapid improvement of the cattle and suggested that the land and pastures should be treated with phosphates.

A report on bone chewing in Montana was made by Welch (38) in 1924. The disease was usually associated with feeding wild hay grown on low ground. The cattle fed on alfalfa, clover, or other legumes did not show these symptoms.

Becker, et al. (7), in 1931, stated that the disease "salt sick" in Florida was caused by cattle grazing on forage crops low in iron and copper. Archibald, et al. (4) reported on the nutritional anemia in cattle in southeastern Massachusetts. They stated that the disease "neck ail" was caused by insufficient amounts of iron in the native forage, which in turn was due to the low iron in the soil. Adding iron compounds to the soil restored this mineral in the grasses.

REVIEW OF THE LITERATURE

A review of the literature showed that there have been many studies on the effect of fertilizer treatments on the yield and quality of the wheat grain and on the relation between mineral composition and baking quality. There have been a considerable number of investigations on the effect of fertilization on the mineral content of different plants including wheat. The research on wheat was usually confined to the grain or the straw of this cereal. It seems that very few studies have been focused on the relation between fertilization and the growing wheat plant. Since this present research dealt with fertilizer applications and the wheat grass and grain, the review of the literature was confined mostly to this phase of the subject.

The most noted investigations on wheat are the classical Rothamsted experiments in England. Allison (1) gave the centenial report and said that these studies have been a tremendous force in the development of agricultural science in general and soil science in particular.

The remarks made by Browne (11) were certainly found to be true in attempting to evaluate the literature on the subject.

> The influence of fertilization upon the composition of the mineral matter of crops is exceedingly complex. For not only do the variable effects of variety of crop, climate, water supply and other environmental conditions apply to the fertilizers incorporated with soils, but there is also the additional complication of the influence which the presence of one element exercises upon the absorptive powers of the plant for other mineral nutrients of the soil or fertilizers. . . . . The mutual effects of all the various soil nutrients on the yield of crops and their assimilation of mineral matter under different environmental conditions are so complicated that the literature on the subject presents a chaos of contradictions.

Browne listed the following factors influencing the mineral composition of crops: type of soil; continuous cropping; variety of crop; successive cuttings in case of grass or hay crops; climatic conditions such as humidity, rainfall, sunshine, and altitude; water supply; and fertilization.

<u>Climate</u> <u>and</u> <u>Season</u>. Ames (2) in 1910 discussed some of these factors that influenced the composition of the wheat plant. He reported considerable variations due to favorable or unfavorable weather conditions such as temperature and moisture. Lawes and Gilbert (23), in studying the seasonal influences, reported lower percentages of potash, phosphorus, and nitrogen in the better mature grains of favorable seasons. They showed that the proportion of potassium and phosphorus in the grain varied considerably from season to season.

Russell and Watson (31) in 1940 presented an excellent review of the Rothamsted fertilizer experiments in which they showed the effect of season, rainfall, and manuring on the growth and composition of the wheat plant.

In order to show the influence of soil and climate on the composition of wheat, LeClerc and Yoder (24) made a three way exchange of the soil between Maryland, Kansas, and California. The Turkey variety of wheat was grown on each of these plots in the three states. The wheat grown in Kansas, whether on the Kansas, Maryland, or California soil, had more protein and potash in the ash than that grown on the same soils in the other two states. The protein content of wheat grown on the three soils in California was 13.0 per cent, in Maryland 11.0 per cent, and Kansas 18.0 per cent. The average protein content of that grown on California soil in the three states was 13.88 per cent, that grown on Maryland soil was 15.44 per cent, and that grown on Kansas soil was 13.94 per cent. These results showed the effect of climatic conditions on the protein content of wheat.

On the other hand, Gericke (17) stated that the low protein in the wheat in California was not due to climatic conditions but to an insufficient supply of available nitrogen at certain growth periods. He showed that nitrogen fertilizers increased the protein in the grain of the wheat grown on nitrogen deficient soil.

In their investigation of the composition of three varieties of wheat in Kansas, Schrenk and King (32) observed that areas receiving the most rainfall produced good yields with slightly below average ash and low protein content.

In 1923 Greaves and Carter (19) attempted to show the effect of water supply on the wheat plant by a series of irrigation experiments. With progressive additions of irrigation water, increases occurred in the percentages of ash, potassium, calcium, magnesium, and phosphoric acid in the grain of wheat, oats and barley.

Variety. Many reports indicated very little variety difference in chemical composition. Schrenk and King (32), in their study of the composition of three varieties of wheat grown in different localities in Kansas, observed that the mineral content was not greatly influenced by variety. Location was a more important factor. However, Beeson (9) stated that it is entirely possible that in some species one variety is a better feeder on certain nutrients than other varieties of the same species. The results reported by Greaves, Bracken, and Hirst (18) substantiate this. They showed that spring wheat had a greater percentage of ash, calcium, magnesium, potassium, iron, phosphorus, and sulfur than winter wheat grown on the same soil.

Sotola (34) also observed that certain varieties of wheat and barley showed a higher content of digestible nutrients than others as the plant progressed from the milk to the ripe stage.

Stage of Growth. Donaldson (13) in 1938 studied the mineral nutrition of Marquis wheat by analyzing plants at intervals during the growth period. He found that the total amount of dry matter, phosphorus, sulfur, and nitrogen increased progressively.

Potassium reached a maximum one month before harvest and then suffered subsequent loss.

Soil and Fertilization. It is generally accepted that the composition of a plant is greatly dependent upon the soil in which it grows. Schrenk and King (32) showed that localities in Kansas producing high mineral content wheat did so consistently over a three year period. This indicated that differences due to rainfall and other factors did not greatly affect the mineral content. Locality was a more important factor than others in causing variations.

Beeson (9) states that, in general, many instances of known deficiencies of some particular element can be remedied by adding that element to the soil. Indeed there have been many experimental results to substantiate this conclusion. Gericke (17) showed that the protein in wheat grown on low nitrogen soils in California was increased by nitrogen applications. Stubblefield and DeTurk (35) also showed that applications of certain chemicals to the soil caused an increase of those elements in the plant.

Protein and Nitrogen. There have been many experiments to show that the protein of wheat can be increased by fertilization. A few of these will be summarized here. Snyder (33) in his studies of the influence of fertilizers found that increasing the nitrogen in the soil slightly increased the nitrogen in the grain. The highest percentage of nitrogen was secured from wheat grown on plots receiving nitrogen alone or complete fertilizer treatment. Ames, Boltz, and Stenius (3) produced marked changes in the chemical composition of the wheat grain by changing the condition of the soil. By applications of nitrogen alone and by nitrogen and potash together the percentage of protein in wheat was increased, while applications of phosphorus decreased it.

Murphy (29) in 1930 studied the effect of fertilizer in Oklahoma, and found that the protein content of wheat rises as nitrogen fertilizer is added. Ames (2) showed that nitrogen fertilizers increased the nitrogen in the crop. Addition of phosphorus fertilizer without nitrogen decreased the nitrogen in the plant. In 1934 Swanson (36) studied the factors influencing the protein content of wheat. He found that by applications of urea and urine, the protein content was increased four per cent. The use of nitrates increased the protein in proportion to the need of the soil for nitrogen. Swanson concluded that the most important factor in determining the percentage of protein of wheat was the availability of nitrogen in the soil, particularly during the latter stages of growth.

Total Ash. In 1937 Bayfield (6) observed that the amount and composition of the ash in the grain is influenced by the supply of plant nutrients available in the soil. He said that the supply of the nutrients can be varied by fertilization, crop rotation, and tillage practices. Liming the soil increased the ash content by over 0.25 per cent. There was a tendency for the phosphate fertilizers to increase the ash, while nitrates had an opposite effect. However, Lawes and Gilbert (23) were able to show an increase in the ash constituants of the total crop due to fertilization, but the amount stored in the grain was not influenced to any great extent. They also showed that manure had little effect on the potassium and phosphorus content of the grain.

Schrenk and King (32) reported that the ash and mineral content of wheat grown in different parts of the state of Kansas varied appreciably. It was closely correlated to the available nutrients in the soil. Areas producing wheat high in mineral matter also produced those higher in protein.

King and Perkins (22) showed that limestone applied to soil decreased the percentage ash in plants grown on acid soil but increased the ash content of plants grown on basic soil.

The report on the Rothamsted experiments on wheat showing the sixteen year average results (1848-1863) was given by Russell (30). He reported that ammonium sulfate fertilizer decreased the total mineral matter and the percentage of phosphoric acid and of potash in the ash. This application also resulted in slight increased lime in the straw and in the grain and a marked increase in the magnesia of the straw. The experiments through an 18 year period (1856-1873) in which potassium salts were used as fertilizer resulted in increases in the total ash and potash but lower percentage of other constituents.

Minerals. Ames (2) investigated the influence of the soil and fertilizers on the composition of the wheat. He showed that adding fertilizers to depleted soils increased the relative amounts of the essential elements. He reported that the percentage of nitrogen and potassium in the crop were in accord with the amounts found in the soil.

It was further shown by Ames that if the soil were deficient in phosphorus and potassium, applications of these minerals on the soil caused their increase in the crop. Potassium increased the nitrogen, and phosphorus decreased the nitrogen in the grain. Phosphorus fertilizers increased the potassium in the grain. Additions of lime to the soil increased the phosphorus assimilated by the plant and subsequently decreased the nitrogen and increased the potassium in the grain. An increase in the potassium supply of the soil resulted in a greater potassium content of the straw.

Ames also investigated the phosphorus content at maturity, and he reported that 80 per cent of it was in the grain. With an increased supply of available phosphorus in the soil, there was a tendency for greater transference of phosphorus from the straw to the grain. Additions of nitrogen to the soil decreased the total phosphorus in the plant and in the grain. Of the phosphorus carriers, manure had the greatest effect on increasing the phosphorus in the plant.

When Schrenk and King (32) studied the composition of the wheat grown in different localities in Kansas, they observed that the general downward trend of phosphorus as the eastern border of the state was approached was reversed in the wheat grown on the eastern plots. This they ascribed to the fertilizer these plots received.

Studies of the application of phosphate fertilizers in China were made by Lee (25). In 1940 he reported increases in the phosphorus in the wheat grown on phosphate treated soils. The experiments were carried out on yellow earth, which was seriously low in this mineral. Early applications were more successful, since the phosphate is absorbed early by the plant.

Greaves, et al. (18) showed that the use of green manure increased the phosphorus content of wheat. Murphy (29) observed that the phosphorus in the grain increased 20 per cent when superphosphate was used, and that potash and nitrogen fertilizers did not increase this mineral very much.

King and Parkins (22) reported that large amounts of iron in the soil reduced the percentage of phosphorus in the wheat plant. Phosphorus applications increased the percentage of this mineral absorbed by the plant on iron treated soils.

McCalla and Woodford (27) studied the effect of the potassium supply on the composition of wheat. They observed that by limiting the supply of potassium to wheat plants a decreased nitrogen content and a markedly increased calcium and magnesium content of the dry matter resulted. There was a reduced amount of nutrients absorbed by the plant. The nitrogen and ash content of the grain was in rough agreement with the absorption by the whole plant.

Carotene. Carotene in wheat has not been studied to the extent that protein or the minerals have. Fifield, et al. (15) determined the carotene content of six varieties of wheat grown at five different locations in the Northwest. Variety differences and the effect of environment were noticed, but no relation between carotene and the protein of the grain were found.

Thomas and Moon (37) investigated the effect of fertilizer treatments on the carotene content of grass. They showed that applications of ammonium sulfate and of limestone increased the carotene content of the pasture. The carotene content did not vary greatly during the growth period. These investigators reported a relation between the carotene content and the percentage of crude protein present.

In a later report of the influence of fertilizers on the carotene content of poor pasture grass Moon (28) showed a 28 per cent increase by ammonium sulfate or sodium nitrate applications. Potassium sulfate treatment resulted in a 6.2 per cent increase. Seasonal variations were also noticed. Moon reported an increase in the ash content as a result of potassium sulfate, limestone, or superphosphate fertilization. Sodium nitrate, ammonium nitrate and superphosphate treatments increased the protein in the grass.

METHODS OF ANALYSIS

Several plots of wheat in the Columbus and Thayer areas under supervision of the Agronomy Department were given treatments with the following fertilizers: lime, superphosphate, potash, manure, and legumes. The samples were collected by the men on the field and then sent to the Chemistry Department for analysis.

Samples of the wheat grass were collected for two years, in the springs of 1947 and 1948; both sets of samples received the same fertilizer treatments. The grass was cut at three different physiological stages of growth, and as nearly to the same stage each year as possible. The first sample was taken when the wheat was about three inches tall, and the other samples were cut at about ten day intervals. These represent the stages of growth at which cattle are usually allowed to pasture on the wheat grass in the spring.

The grass was clipped immediately above the ground, packed in dry ice, and shipped to the laboratory. The samples represent the whole plant above the ground. They were then dried, ground, and analyzed for nitrogen, phosphorus, calcium, potassium, and magnesium. Data on the total ash and carotene content of the samples were furnished.

Analysis for Nitrogen

Analysis for nitrogen was made by the usual A.O.A.C. Kjeldahl procedure. One gram samples were digested for about three hours with concentrated sulfuric acid in the presence of a catalyst consisting of a mixture of mercuric oxide, cupric sulfate, and potassium sulfate. For distillation, a strong alkali solution with a catalyst was added. The nitrogen was distilled over as ammonia into N/5 sulfuric acid and then titrated with N/14 sodium hydroxide, with methyl red as the indicator.

Analysis for Phosphorus, Calcium, and Potassium

The photoelectric method developed by Wolfe (39) was used for the determination of the phosphorus, calcium, and potassium content. The L values of the galvanometer readings on the photometer were compared in each case with a series of standards carried through the same procedures in order to calculate the percentages.

<u>Digestion of Samples</u>. The dry samples were digested according to the method suggested by Wolfe. A 0.2 gram sample was used and digested with concentrated sulfuric acid by heating and the addition of hydrogen peroxide until they were clear. These clear solutions were diluted with an acetate buffer. This ash extract was then analyzed for the minerals: phosphorus, calcium, and potassium.

<u>Analysis for Phosphorus</u>. In the Wolfe method for the determination of phosphorus, ammonium molybdate solution and aminonaphtholsulfonic acid reagent were added to the extract. Readings were taken on an Evelyn photometer using a 420 $m\mu$ blue filter.

Wolfe stated that the blue color formed increased in intensity for a number of hours unless heat was used. This increase in intensity of color was studied in order to determine the best time for taking the readings on the photometer. The results of this study are given in Table 1. These results confirm the statement by Wolfe that reproducible results can be obtained in the cold after 15 minutes standing. The results indicated that there was very little change in intensity in the blue color in either the tubes containing the standard solution or the ash extract. It was decided to take the readings 15 to 20 minutes after the addition of the reducing agent, amino-naphtholsulfonic acid.

Table 1. Influence of time of reading on the phosphorus determination.*

Min.	Standard solution 4 ml		6 ml		Samples A	B	C	D
5.0	43.0	39.5	26.2	24.2	59.2	60.5	52.8	54.0
7.5	43.0	39.0	26.0	24.0	55.0	56.5	49.2	49.0
10.0	42.5	39.0	25.8	23.8	52.5	53.5	46.5	46.0
12.5	42.5	39.0	25.5	23.5	51.0	52.0	45.0	44.5
15.0	42.0	38.8	25.5	23.5	50.0	52.2	44.0	44.0
17.5	42.0	38.8	25.2	23.2	49.5	51.0	43.2	43.0
20.0	41.5	38.2	25.0	23.0	49.0	50.5	43.0	42.8
22.5	41.5	38.2	25.0	23.0	49.0	50.5	43.0	42.8
25.0	41.2	38.0	25.0	23.0	49.0	50.5	42.5	42.8
30.0	41.2	38.0	25.0	23.0	49.0	50.8	42.8	42.5

* Figures represent galvanometer readings.

Analysis for Calcium. The Wolfe method for calcium is a turbidity method in which ground ammonium oxalate is used as the precipitating agent. A small amount of gum arabic was used to stabilize the colloidal calcium oxalate formed. Readings were taken on an Evelyn photometer using a 420 $m\mu$ blue filter.

Analysis for Potassium. In the photometric method suggested by Wolfe for the determination of potassium, formaldehyde was added to eliminate the interference of ammonia. A small amount of gum arabic was added as a stabilizer, and the potassium was precipitated as the colored cobaltinitrite with a sodium cobaltinitrite reagent in the presence of iso-propyl alcohol. Readings were taken on an Evelyn photometer using a 660 $m\mu$ red filter.

The Wolfe method suggested that the test tubes be allowed to stand 15 minutes after adding the iso-propyl alcohol, the last reagent to be added, before readings are taken on the photometer. The influence of the time of reading on the results was studied. The results are given in Table 2. These indicated that the best results were taken ten to fifteen minutes after the addition of the alcohol.

Table 2. The influence of the reading time on the potassium determination.*

Min.	0.5 ml Samples								0.5 ml standard solution
	A		B		C		D		
1	90.5	93.0	----	----	90.2	92.0	86.2	88.5	95.5
5	88.2	89.5	86.8	88.2	86.0	87.0	84.2	86.5	94.0
10	87.2	88.0	87.5	88.2	85.5	86.8	84.2	86.2	93.8
15	87.2	88.5	86.5	88.0	85.8	87.0	84.0	86.5	93.8
20	88.5	90.0	87.5	89.0	86.5	88.0	86.0	88.0	95.0
25	89.5	90.0	88.5	89.8	87.5	88.5	87.0	89.2	96.2
30	90.0	91.0	89.2	90.8	88.5	89.5	88.5	90.5	97.2
35	91.0	91.8	90.8	91.5	90.0	91.2	90.5	92.0	98.2
40	91.5	92.2	92.2	93.2	91.5	92.5	91.0	92.5	88.8
45	93.5	94.0	93.8	94.8	93.0	94.0	92.2	94.0	99.5

* Figures represent galvanometer readings.

Analysis for Magnesium

The method by Wolfe was also attempted for the determination of magnesium, but the color did not develop adequately in the titan yellow reaction even with varying amounts of sodium hydroxide solution. Since satisfactory results could not be obtained by this procedure, it was discarded, and the method developed by Lindner (26) was used to digest the dry sample and to analyze the extract for magnesium.

In this method a small amount of hydroxylamine hydrochloride solution was first added and then the titan yellow as the colorimetric reagent. One ml of 40 per cent sodium hydroxide solution was then added as recommended by Lindner. Readings were taken on an Evelyn photometer using a 540 mμ green filter, the log values of which were compared to a series of standards carried through this same procedure.

This procedure was used first, and it was satisfactory providing the pH was kept constant. However it was found difficult to duplicate the results. Lindner, in his discussion of the method, stated, that, among other factors, the readings are influenced by pH and temperature. In order to decrease the influence on the pH due to errors in titration, 2.5 ml of 15 per cent sodium hydroxide were used instead of one ml of 40 per cent alkali. There was an improvement in the results since the probable error decreased from 0.0030 to 0.0022 as shown by comparing the data in Table 3 with that in Table 4.

Table 3. The standard deviation and probable error in magnesium determination using one ml of 40 per cent sodium hydroxide.

Sample	Standard deviation	Probable error
63	0.0076	0.0030
66	0.0115	0.0026
67	0.0146	0.0033
99	0.0156	0.0043
108	0.0052	0.0018
Average	0.0109	0.0030

Table 4. The standard deviation and probable error in the magnesium determination using 2.5 ml of 15 per cent sodium hydroxide.

Sample	Standard deviation	Probable error
63	0.0057	0.0019
75	0.0094	0.0026
99	0.0101	0.0028
100	0.0108	0.0026
101	0.0029	0.0010
103	0.0060	0.0014
107	0.0073	0.0016
108	0.0159	0.0048
151	0.0049	0.0016
157	0.0076	0.0018
Average	0.0081	0.0022

Lindner (26) suggested that readings should be taken immediately after the solution in the tube had reached the constant room temperature. The test tubes were placed in a water bath held at room temperature for five minutes, and then the readings were taken. The influence of the time of reading after the addition of the sodium hydroxide was studied in order to determine how the readings on the photometer changed with time. The results are given in Table 5. These results indicated that the color of the solution changed with time. This accounted for some of the results which could not be duplicated very well.

Table 5. The influence of reading time on the magnesium determination.*

Min.	Samples					1 ml standard solution
	A	B	C	D	E	
1	60.0	57.0	56.5	56.0	55.0	67.5
3	61.2	57.5	57.0	56.8	55.5	68.5
5	61.5	57.5	57.8	58.0	56.0	69.2
7	61.2	58.0	58.8	58.0	57.0	69.2
10	61.8	59.2	60.0	58.5	58.2	69.2
12	62.0	59.2	61.5	59.5	58.0	69.2
15	63.5	60.0	63.0	59.8	58.0	69.2
20	63.5	61.5	64.5	60.2	58.2	69.2

* Figures represent galvanometer readings.

The influence of the temperature of the solution on the photometric readings were also studied. The test tubes were allowed to remain in the water bath five minutes, and the average temperature of the bath was observed. The results are given in Table 6. These indicated that within the range of the temperatures of running tap water there is no variation in the readings.

Table 6. The influence of temperature on the magnesium determination.*

Av. temp.	0.75 ml standard solution		Sample A	
21.0°	75.0	76.5	65.0	65.5
16.0°	75.8	74.2	64.2	64.2
25.2°	75.0	75.0	65.5	66.0

* Figures represent galvanometer readings.

Analysis of the Wheat Grain

The methods for the determination of calcium and potassium suggested by Wolfe did not study the analysis of these two minerals in the wheat grain. Evidently the concentration of these two minerals in the grain was too small to be detected by the procedure followed in the analysis of the grass. Satisfactory results were obtained when a one gram sample of the ground grain was digested and used in the ash extract. The one gram sample was digested by using three ml of concentrated sulfuric acid, heat treatment, and subsequent additions of 30 per cent hydrogen peroxide until the solution cleared up, following the method for digestion suggested by Wolfe (39). The samples were then analyzed for calcium and potassium according to the method already described.

FERTILIZER TREATMENT

The fertilizer treatment of the wheat in the crop rotation series at Columbus, Kansas, were carried out by the men in the field under the supervision of the Agronomy Department. The fertilizers that were used are the ones that are usually used by the farmers in that area. The soil has been found to be deficient in nitrogen, phosphorus, and calcium. The same treatments each year were given to the same plots on which the wheat was grown. The control plot received no fertilization and no legume treatment.

The lime was applied before the legumes, alfalfa or sweet clover, were grown on the plots. Alfalfa and sweet clover were grown in the rotation on all plots except the control. Manure was applied before plowing in preparation for the wheat at the rate of eight tons per acre.

The 20 per cent superphosphate was applied at the time the wheat was seeded at the rate of 80 pounds per acre for the 1947 crop and at the rate of 150 pounds per acre for the 1948 crop. The 50 per cent muriate of potash was applied at the rate of 10-2/3 pounds per acre at the time the wheat was seeded for the 1947 crop and at the rate of 60 pounds per acre for the 1948 crop.

RESULTS

Wheat Grass

Table 7 presents data regarding the effect of various fertilizer treatments on the ash, carotene, and nitrogen contents of the wheat grass grown at Columbus in 1947. Table 8 shows the same data for 1948. The two-year averages as taken from Tables 7 and 8 are shown in Table 9. The data in Table 7 are the averages of the compositions of these constituents at the three different stages of growth as presented in Table 13. Table 8 presents the averages of the three stages of growth as taken from Table 14.

Table 10 shows the effect of the various fertilizer treatments on the percentage of the major minerals found in the wheat grass grown at Columbus. These figures are the averages of these constituents at the three different stages of growth in 1947 as taken from Table 13. Table 11 presents the same data for the year 1948 and is taken from Table 14. The averages of the 1947-1948 results are tabulated in Table 12. These results are presented in graphic form in Fig. 1 and 2.

<u>Carotene</u>. There was a noticeable decrease in the carotene content of the wheat grass especially when the superphosphate treatment was given. Lime alone had less effect than the other fertilizer combinations. The greatest decrease showed up in the grass receiving the "no legume" treatment, in which the average

carotene content decreased from 48.0 to 36.6 mg per 100 g of dry material. This was a decrease of over 23 per cent. The least decrease was observed in the case of manure treatments; it was less than six per cent. Approximately the same average decreases were observed each year. There also seemed to be a correlation between the carotene and nitrogen content. This confirms the results obtained by Thomas and Moon (37).

Nitrogen. There was a decrease in the nitrogen content of the wheat grass grown on the fertilizer treated soil, especially when superphosphate was added. Those samples receiving the superphosphate treatment showed an average decrease of over 18 per cent in the nitrogen content. These results confirmed the conclusions drawn by Ames (2). The greatest reduction was noticed in the wheat which received a "no legume" treatment. In this case there was an average 28 per cent decrease in its nitrogen content. The addition of manure seemed to reestablish the content of nitrogen, but it did not restore the amount to that found in the "no treatment" wheat grass.

Fairly consistent yearly effects of the fertilizer treatment on the nitrogen were noticed. The total average for 1948 was only slightly higher than that for 1947. There was less loss of nitrogen in the wheat grown on the "no legume" soil in 1948 than in 1947.

Total Ash. Definite decreases in the total ash were observed in the wheat grass that received phosphate fertilizer treatments. The greatest decreases resulted from the lime and superphosphate combination and from the lime, superphosphate and manure treatment. Lime showed very little effect on the total ash either year. The percentage of ash in the 1948 crop was higher than in the 1947 crop, except in plants from the "no legume" plots. In general there was a greater decrease in the ash content in 1948.

Very noticeable increased percentages of phosphorus and calcium were obtained as a result of fertilization; however, an appreciable decrease in the total ash was found. There was an average increase of 0.05 per cent for both the calcium and phosphorus contents in the grass from all fertilizer treated plots, but there was an average decrease in potassium content of 4.7 per cent as compared to the grass from the control plots. Since the decrease in potassium was greater than the increases in the other two minerals, a decrease in the total ash resulted.

Phosphorus. There was a substantial increase in the phosphorus content of the wheat grass as a result of the fertilizer treatments. The use of superphosphate especially resulted in significant results. The greatest increase occurred in the wheat grown on soil receiving the "no legume" treatment. In this case there was a 57 per cent increase in the phosphorus content. The lime, potash or the manure did not seem to have any significant influence on the percentage of phosphorus in the grass.

The greatest gain was consistently seen in the 1947 samples. In 1947 there was an average increase of 48 per cent in the phosphorus content of the grass from the fertilized plots, as compared with only a 16 per cent gain in the phosphorus content in 1948.

Calcium. Significant increases in the calcium content of the wheat grass were seen as a result of the fertilizer treatments which included lime. The percentage of calcium in the samples increased from 0.32 to 0.37 as a result of the lime treatment. Greatest increases were seen when superphosphate and superphosphate plus potash were used with the lime. In general the 1947 results indicated a more definite trend than the 1948 data.

Potassium. The percentage of potassium in the wheat grass was less when grown on fertilizer treated soil. Substantial reductions were caused by superphosphate treatments. The average potassium content for all of the samples from the superphosphate treated soils was 2.17 per cent. The control samples had 2.80 per cent. No significant effect was noticed as a result of the potash combination or the treatment consisting of only lime.

The results for the two years were fairly consistent. However the potassium content in 1947 was in general lower than in 1948. Greater reductions in the samples receiving fertilizer treatments also occurred in the 1947 samples. In 1947 there was an average decrease in the potassium content of almost 25 per cent, while in 1948 the average decrease was about 15 per cent.

Magnesium. No significant effect of the various fertilizer treatments on the magnesium content was noticed. The results for the two years were in general about the same. The wheat receiving the "no legume" treatment showed a slight decrease in magnesium.

Table 7. The effect of various fertilizer treatments on the average ash, carotene, and nitrogen contents of the wheat grass in 1947.*

Treatment**	Ash		Carotene		Nitrogen	
	per cent	% gain or loss	mg***	% gain or loss	per cent	% gain or loss
Control	10.6	-----	47.8	-----	4.03	-----
L.	10.5	- 0.9	48.3	+ 1.0	4.00	- 0.7
L.Sp.	8.8	-17.0	45.1	- 5.6	3.41	-15.4
L.Sp.	8.7	-17.9	41.3	-13.6	3.20	-20.6
L.Sp.P.	9.7	- 8.5	43.7	-15.7	3.24	-19.6
L.Sp.M.	7.9	-25.4	44.6	- 6.7	3.58	-11.1
L.Sp.NL.	10.9	+ 2.8	34.1	-28.6	2.63	-34.7
Average	9.6	-12.1	43.1	-11.5	3.44	-17.0

* Composition was analyzed on a dry weight basis.

** Control received no treatment; L=lime; Sp=superphosphate; P=potash; M=manure; and NL=no legume.

*** Mg per 100 g of dry weight.

Table 8. The effect of various fertilizer treatments on the average ash, carotene, and nitrogen contents of the wheat grass in 1948.*

Treatment**	Ash		Carotene		Nitrogen	
	per cent	% loss or gain	mg***	% loss or gain	per cent	% loss or gain
Control	12.1	-----	48.1	-----	4.07	-----
L.	11.9	- 1.6	43.4	- 9.8	3.67	- 9.8
L.Sp.	10.2	-15.7	42.0	-12.7	3.20	-21.4
L.Sp.	8.9	-26.4	44.1	- 8.3	3.69	- 9.3
L.Sp.P.	10.4	-14.0	39.7	-17.4	3.19	-21.6
L.Sp.M.	9.8	-19.0	45.9	- 4.6	3.63	-10.8
L.Sp.NL.	9.7	-19.8	39.1	-18.7	3.16	-22.3
Average	10.4	-16.1	43.2	-11.9	3.52	-15.9

* Composition was analyzed on a dry weight basis.

** Control received no treatment; L=lime; Sp=superphosphate; P=potash; M=manure; and NL=no legume.

*** Mg per 100 g of dry weight.

Table 9. The average two-year effect of various fertilizer treatments on the ash, carotene, and nitrogen content of the wheat grass.*

Treatment**	Ash per cent	Ash % loss or gain	Carotene mg***	Carotene % loss or gain	Nitrogen per cent	Nitrogen % loss or gain
Control	11.4	-----	48.0	-----	4.05	-----
L	11.2	- 1.8	45.8	- 4.6	3.84	- 5.2
L.Sp.	9.5	-16.6	43.6	- 9.2	3.30	-18.5
L.Sp.	8.8	-22.4	42.7	-11.0	3.44	-15.0
L.Sp.P.	10.0	-12.3	41.7	-13.1	3.22	-20.5
L.Sp.M.	8.8	-22.4	45.2	- 5.8	3.60	-11.1
L.Sp.NL.	10.3	- 9.6	36.6	-23.7	2.90	-28.4
Average	10.0	-14.1	43.2	-11.2	3.48	-16.4

* Composition was analyzed on a dry weight basis.

** Control plot received no fertilizer treatment; L=lime; Sp=superphosphate; P=potash; M=manure; and NL=no legume.

*** Mg per 100 g of dry weight.

Table 10. The effect of various fertilizer treatments on the mineral content of wheat grass in 1947.*

Treetment**	P		Ca		K		Mg	
	per cent	per cent gain or loss	per cent	per cent gain or loss	per cent	per cent gain or loss	per cent	per cent gain or loss
Control	0.199	-----	0.30	-----	2.71	-----	0.132	-----
L.	0.217	+ 9.5	0.40	+33.3	2.29	-15.5	0.144	+ 9.1
L.Sp.	0.289	+47.3	0.43	+43.4	1.78	-34.3	0.151	+14.4
L.Sp.	0.307	+54.3	0.42	+40.0	1.94	-28.4	0.153	+15.9
L.Sp.P.	0.298	+49.7	0.45	+50.0	1.99	-26.5	0.151	+14.4
L.Sp.M.	0.297	+49.2	0.35	+16.6	1.94	-28.4	0.150	+13.6
L.Sp.NL.	0.358	+79.9	0.32	+ 6.7	2.28	-15.8	0.118	-10.6
Average	0.281	+48.4	0.38	+31.7	2.13	-24.8	0.143	+ 9.5

* Composition was analyzed on a dry weight basis.

** Control plot received no fertilizer treatment; L = lime; Sp = superphosphate; P = potash; M = manure; and NL = no legume.

Table 11. The effect of various fertilizer treatment on the mineral composition of wheat grass in 1948.*

Treatment**	P		Ca		K		Mg	
	per cent	per cent gain or lose	per cent	per cent gain or lose	per cent	per cent gain or lose	per cent	per cent gain or loss
Control	0.169	-----	0.34	-----	2.89	-----	0.155	-----
L.	0.167	- 1.2	0.34	0.0	2.98	+ 3.1	0.141	- 9.3
L.Sp.	0.203	+20.1	0.32	- 5.9	2.66	- 8.0	0.134	-13.5
L.Sp.	0.206	+21.8	0.43	+26.4	1.80	-38.0	0.167	+ 7.7
L.Sp.P.	0.201	+18.9	0.36	+ 5.9	2.63	- 9.0	0.123	-20.6
L.Sp.M.	0.197	+16.6	0.34	0.0	2.46	-14.8	0.152	- 1.9
L.Sp.NL.	0.221	+30.8	0.41	+21.9	2.23	-22.8	0.134	-13.5
Average	0.195	+16.4	0.36	+ 8.0	2.52	-14.9	0.144	- 8.5

* Composition was analyzed on a dry weight basis.

** Control plot received no fertilizer treatment; L = lime; Sp = superphosphate; P = potash; M = manure; and NL = no legume.

Table 12. The average two-year effect of various fertilizer treatment on the mineral composition of wheat grass.*

Treatment**	P		Ca		K		Mg	
	per cent	per cent gain or loss	per cent	per cent gain or loss	per cent	per cent gain or loss	per cent	per cent gain or loss
Control	0.184	-----	0.32	-----	2.80	-----	0.144	-----
L.	0.192	+ 4.4	0.37	+15.6	2.64	- 5.7	0.142	- 1.4
L.Sp.	0.246	+33.7	0.38	+18.7	2.22	-20.7	0.142	- 1.4
L.Sp.	0.256	+39.1	0.42	+31.2	1.87	-33.2	0.160	+11.1
L.Sp.P.	0.250	+35.8	0.40	+25.0	2.31	-15.3	0.137	- 4.9
L.Sp.M.	0.247	+34.2	0.34	+ 6.2	2.20	-21.4	0.151	+ 6.2
L.Sp.NL.	0.290	+57.6	0.36	+12.5	2.26	-19.3	0.126	-12.5
Average	0.238	+36.1	0.37	+17.4	2.33	-19.3	0.143	- 0.5

* Composition was analyzed on a dry weight basis.

** Control plot received no fertilizer treatment; L = lime; Sp = superphosphate; P = potash; M = manure; and NL = no legume.

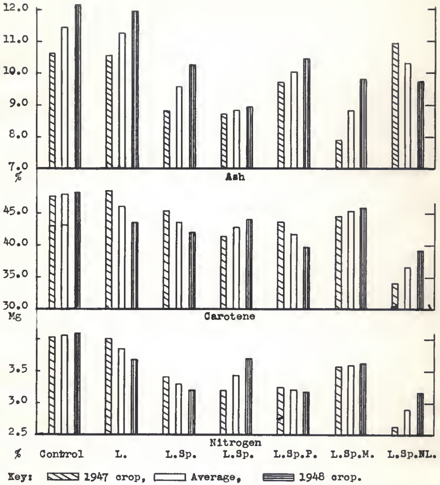

Fig. 1. The effect of fertilizer treatment on the composition of nitrogen, carotene and total ash of the wheat grass.

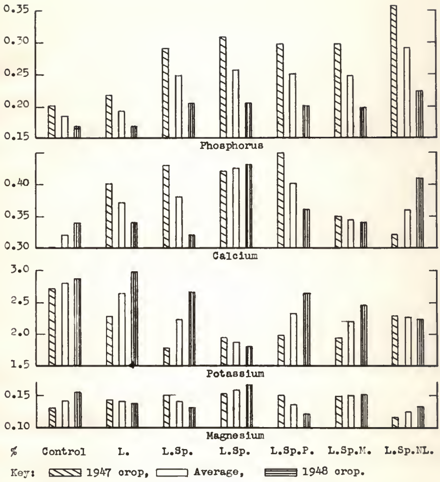

Fig. 2. The effect of fertilizer treatment on the mineral composition of wheat grass.

Stage of Growth

Table 13 gives the data showing the chemical composition of the wheat grass at the different stages of growth in 1947 for each of the various fertilizer treatments. Table 14 gives the same data for 1948. The averages of the percentages for these two years are given in Table 15.

Carotene. The carotene content of the wheat grass consistently decreased each year between the first and third stages of growth. Often an increase was observed in the second stage, but not consistently. This gain was in each case more than offset by the loss in the next period. The total decrease was greater for the fertilizer treated grass.

Nitrogen. In the wheat grass grown on the "no treatment" plots there was a decline in the percentage of nitrogen for the progressive stages of growth. This same decrease in the nitrogen content was found in all samples receiving the various fertilizer treatments. However, a greater decrease was noticed in the grass which received superphosphates than in those plants receiving only lime.

In 1947 there were greater and more consistent decreases in the nitrogen content of the wheat grass. This was true for samples from the "no treatment" plots as well as those from the treated plots. By comparing Tables 7 and 8 it was noticed that wheat grass in 1947 had slightly less nitrogen than in 1948.

Total Ash. The ash content of the wheat grass decreased with age between the first and third cuttings. No significant fertilizer effect was apparent. In 1948 the average total decrease in the percentage ash content was 1.1 per cent compared to a 2.3 per cent decrease for 1947.

Phosphorus. The results for phosphorus did not show a decrease with stages of growth in every case as was found in the nitrogen. However, in the wheat grasses from the plots receiving superphosphate treatment in which there were distinct increases in the phosphorus content according to Table 12, there was, in every case, a decline with age.

It was already pointed out that the wheat grass grown in 1947 on the plots which received the superphosphate fertilizer had a much greater phosphorus content than those grown on lime and "no treatment" soil. This was seen from Table 10. From Table 13 it was seen that in 1947 there was a decrease with the stage of growth in the phosphorus content in those cases in which the plants received the superphosphate fertilizer. Such consistent results did not occur in 1948.

According to Table 14, there was sometimes an increase and at other times a decrease with age in the percentage phosphorus of the plant. From Table 11 it can be seen that the phosphorus content was not as high in the plants receiving superphosphate in 1948 as in 1947. It seems that in those cases in which a fertilizer treatment has effected a substantial increase in the percentage phosphorus of the plants, there was a decrease in the phosphorus content with the age of the plant.

Calcium. No consistent changes with the stage of growth was noticed in the calcium content of the wheat grass. In general the grass from the plots receiving lime treatments seemed to be but slightly higher in calcium content in the second stage of growth.

According to Tables 13 and 14, the changes in calcium percentage with stage of growth for each year were neither consistent nor very pronounced.

Potassium. According to Table 15 potassium showed a decline with the stage of growth in each case. There were greater losses of this mineral in the progressive stages of growth for these plants grown on the fertilized plots. Usually the greatest loss was found in the period between the first two stages.

A much greater average decline with growth was seen in 1948 than in 1947. The average total decrease of the percentage content in the latter year was 0.73 compared to a 0.43 decrease in the percentage content for the former year.

Magnesium. In the case of magnesium Table 15 shows that there was no loss of magnesium between the first and second stages of growth, and there was usually a loss of magnesium between the second and third stages. This trend was seen in most cases in both the 1947 and 1948 crops.

No correlation seemed apparent between the loss or gain of the magnesium content at the different stages of growth with either the total amount of the minerals present or the kind of fertilizer used. Except, there was a slight gain in total magnesium content when manure was used and a slight loss in the samples of the "no legume" treatment. No marked yearly difference was noticed.

Table 13. The effect of various fertilizer treatments on the chemical composition of wheat grass at three different stages of growth in 1947.*

Treatment***	Growth stage	Ash %	Carotene**	N %	P %	Ca %	K %	Mg %
Control	1st	11.7	49.6	4.50	0.208	0.28	3.04	0.127
	2nd	10.7	49.7	4.24	0.210	0.32	2.28	0.143
	3rd	9.3	44.1	3.36	0.178	0.31	2.80	0.126
L.	1st	11.6	50.8	4.60	0.233	0.40	2.57	0.136
	2nd	10.6	53.3	4.02	0.218	0.37	2.00	0.153
	3rd	9.2	40.8	3.37	0.200	0.42	2.30	0.144
L.Sp.	1st	9.9	50.3	4.40	0.312	0.42	2.20	0.169
	2nd	8.8	54.6	3.47	0.292	0.39	1.42	0.176
	3rd	7.7	30.4	2.35	0.263	0.47	1.72	0.107
L.Sp.	1st	9.8	41.6	4.03	0.335	0.34	2.09	0.163
	2nd	8.5	49.4	3.25	0.315	0.50	2.06	0.194
	3rd	7.8	32.9	2.32	0.271	0.42	1.68	0.102
L.Sp.P.	1st	11.4	50.1	4.12	0.322	0.43	2.33	0.147
	2nd	9.6	55.5	3.28	0.302	0.45	1.85	0.159
	3rd	8.2	25.4	2.32	0.270	0.46	1.80	0.146
L.Sp.M.	1st	9.2	43.0	4.67	0.352	0.29	2.28	0.141
	2nd	7.7	60.5	3.64	0.288	0.40	1.94	0.149
	3rd	6.6	30.2	2.44	0.251	0.36	1.60	0.161
L.Sp.NL.	1st	12.0	38.0	3.34	0.372	0.31	2.53	0.118
	2nd	11.2	42.7	2.64	0.374	0.33	2.20	0.132
	3rd	9.5	21.6	1.90	0.328	0.32	2.12	0.103

* Composition analyzed on the basis of dry weight.

** In mg per 100 grams of dry weight.

*** Control plot received no fertilizer treatment; L = lime; Sp = superphosphate; P = potash; M = manure; NL = no legume.

Table 14. The effect of various fertilizer treatments on the chemical composition of wheat grass at three different stages of growth in 1948.*

Treat-ment***	Growth stage	Ash %	Caro-tene**	N %	P %	Ca %	K %	Mg %
Control	1st	13.0	43.9	4.24	0.158	0.30	3.07	0.132
	2nd	12.4	54.0	4.16	0.183	0.26	3.10	0.186
	3rd	10.8	46.5	3.80	0.166	0.46	2.50	0.147
L.	1st	12.0	45.1	4.06	0.156	0.30	3.31	0.132
	2nd	12.8	50.1	4.00	0.157	0.37	3.36	0.154
	3rd	10.8	35.1	2.94	0.189	0.34	2.27	0.137
L.Sp.	1st	10.4	46.3	3.74	0.219	0.34	2.93	0.131
	2nd	10.8	42.9	3.17	0.188	0.33	2.93	0.139
	3rd	9.5	36.8	2.68	0.203	0.30	2.12	0.133
L.Sp.	1st	8.8	42.0	3.98	0.206	0.48	2.02	0.154
	2nd	9.0	49.7	3.56	0.201	0.42	1.86	0.186
	3rd	8.8	40.5	3.52	0.212	0.38	1.53	0.161
L.Sp.P.	1st	10.3	43.5	3.82	0.207	0.36	2.88	0.132
	2nd	11.4	42.6	2.89	0.210	0.38	2.71	0.121
	3rd	9.4	33.1	2.86	0.186	0.34	2.30	0.115
L.Sp.M.	1st	10.5	47.7	4.00	0.210	0.35	3.04	0.142
	2nd	9.2	48.0	3.42	0.184	0.38	2.08	0.164
	3rd	9.6	42.0	3.46	0.197	0.29	2.25	0.151
L.Sp.NL.	1st	10.6	40.6	3.76	0.244	0.43	2.78	0.133
	2nd	9.2	40.7	3.12	0.216	0.47	1.98	0.156
	3rd	9.3	36.0	2.61	0.202	0.33	1.92	0.112

* Composition analyzed on the basis of dry weight.

** In mg per 100 grams of dry weight.

*** Control plots received no fertilizer treatments; L = lime; Sp = superphosphate; P = potash; M = manure; NL = no legume.

Table 15. The average two-year effect of various fertilizer treatments on the composition of wheat grass at three different stages of growth.*

Treat-ment***	Growth stage	Ash %	Carotene**	N %	P %	Ca %	K %	Mg %
Control	1st	12.4	46.8	4.37	0.183	0.29	3.06	0.130
	2nd	11.6	51.8	4.20	0.196	0.29	2.69	0.164
	3rd	10.0	45.3	3.58	0.172	0.38	2.65	0.136
L.	1st	11.8	48.0	4.33	0.194	0.35	2.94	0.134
	2nd	11.7	51.7	4.01	0.188	0.37	2.68	0.154
	3rd	10.0	38.0	3.16	0.194	0.38	2.28	0.140
L.Sp.	1st	10.2	48.0	4.07	0.266	0.38	2.56	0.150
	2nd	9.8	48.8	3.32	0.240	0.36	2.18	0.158
	3rd	8.6	33.6	2.52	0.233	0.38	1.92	0.120
L.Sp.	1st	9.3	41.8	4.00	0.270	0.41	2.06	0.158
	2nd	8.8	49.6	3.40	0.258	0.46	1.96	0.190
	3rd	8.3	36.7	2.92	0.242	0.40	1.60	0.132
L.Sp.P.	1st	10.8	46.8	3.97	0.264	0.40	2.60	0.140
	2nd	10.5	49.0	3.08	0.256	0.42	2.28	0.140
	3rd	8.8	29.2	2.59	0.228	0.40	2.05	0.130
L.Sp.M.	1st	9.9	45.4	4.34	0.281	0.32	2.66	0.142
	2nd	8.4	54.2	3.53	0.236	0.39	2.01	0.156
	3rd	8.1	36.1	2.95	0.224	0.32	1.92	0.156
L.Sp.NL.	1st	11.3	39.3	3.55	0.308	0.37	2.66	0.126
	2nd	10.2	41.7	2.88	0.295	0.40	2.09	0.144
	3rd	9.4	28.8	2.26	0.265	0.32	2.02	0.108

* Composition analyzed on the basis of dry weight.

** In mg per 100 grams of dry weight.

*** Control plots received no fertilizer treatments; L = lime; Sp = superphosphate; P = potash; M = manure; NL = no treatment.

Veriety Differences

Teble 16 gives the differences in the chemicel composition for the three varieties; Tenmarq, Turkey, and Bleckhull, which are commonly grown in the southeestern part of Kanses. These percentages ere the everages of the dete for the three steges of growth for each of the three varieties given in Tables 18 and 19. Teble 17 is the everage of the date for the Columbue eamples end the two-yeer's semplee from Thayer. The chemical composition et the three etages of growth for the three verieties are given in Tebles 18 and 19. These same date are averaged in Table 20.

Only elight variety differences in the minerel composition were noticed. The Bleckhull wheet hed a elightly higher cerotene content. It calculated to be 50.0 mg per 100 g of dry weight. There wes no greet difference in the percentage esh. The Turkey samples were elightly lower in nitrogen and the Blackhull e little higher, according to Table 17. No significent end coneistent difference in the phoephorus or the magnesium content was observed. The Bleckhull veriety wae only elightly higher in calcium end in poteesium content.

Very little variety difference wes obeerved in the decreeee of the different conetituents with the ege of the plent.

<u>Yearly Differences</u>. Some distinct yeerly differencee were observed. The Tenmarq end Bleckhull samplee for Theyer, 1947, were eomewhet lower in nitrogen. The Turkey veriety did not

show such a definite difference. All three varieties from Thayer were lower in phosphorus in 1948 than in 1947, but they were definitely higher in potassium and magnesium content. The average ash content for the Thayer, 1948 samples, was 10.4 per cent as compared to 8.6 per cent as the average of the previous year's growth. The carotene content was about 18 per cent lower in the Thayer, 1948 plants, compared with the samples from the same area the year before.

Area Difference. All three varieties grown in the Columbus area in 1947 had a much lower potassium content but a higher percentage of magnesium than the Thayer samples.

Table 16. Variety differences in the chemical composition of the wheat grass.*

Area and year	Variety	Ash per cent	Caro-tene mg**	N per cent	P per cent	Ca per cent	K per cent	Mg per cent
Columbus	Tenmarq	8.2	48.9	4.13	0.266	0.39	1.71	0.199
1947	Turkey	8.2	47.8	3.84	0.271	0.41	1.84	0.163
	Black-hull	8.4	50.0	4.12	0.265	0.47	1.86	0.181
	Average	8.2	48.9	4.03	0.267	0.42	1.80	0.181
Thayer	Tenmarq	8.2	51.6	3.89	0.268	0.42	2.20	0.135
1947	Turkey	8.6	53.2	3.93	0.248	0.33	2.26	0.126
	Black-hull	8.8	53.0	3.95	0.267	0.45	2.34	0.138
	Average	8.6	52.6	3.92	0.261	0.40	2.27	0.133
Thayer	Tenmarq	10.1	42.1	4.02	0.179	0.33	2.64	0.145
1948	Turkey	11.1	41.4	3.99	0.173	0.30	2.62	0.135
	Black-hull	10.1	46.2	4.28	0.178	0.32	2.68	0.151
	Average	10.4	43.2	4.09	0.177	0.32	2.65	0.144

* Composition analyzed on the basis of dry weight.

** In mg per 100 g of dry weight.

Table 17. The two-year average variety differences in the chemical composition of the wheat grass.*

Variety	Ash per cent	Carotene mg**	N per cent	P per cent	Ca per cent	K per cent	Mg per cent
Tenmarq	8.8	47.5	4.01	0.238	0.38	2.18	0.160
Turkey	9.3	47.6	3.92	0.231	0.35	2.24	0.141
Blackhull	9.1	49.7	4.12	0.237	0.41	2.29	0.157
Average	9.1	48.3	4.02	0.235	0.38	2.24	0.153

* Composition analyzed on the basis of dry weight.

** In mg per 100 g of dry weight.

Table 18. Variety differences in the total ash, carotene and nitrogen contents of the wheat grass at three different stages of growth.*

Area and year	Variety	Growth stage	Ash %	Carotene**	N %
Columbus, 1947	Tenmarq	1st	9.2	49.1	4.76
		2nd	8.2	56.7	4.18
		3rd	7.1	41.0	3.44
	Turkey	1st	9.8	48.4	4.70
		2nd	7.8	56.5	3.78
		3rd	7.1	38.6	3.04
	Blackhull	1st	10.0	51.9	4.78
		2nd	8.2	56.8	4.19
		3rd	7.0	41.4	3.40
Thayer, 1947	Tenmarq	1st	8.8	62.6	4.76
		2nd	8.4	46.1	3.52
		3rd	7.6	46.1	3.40
	Turkey	1st	9.0	59.6	4.67
		2nd	8.8	47.2	3.50
		3rd	8.0	52.7	3.63
	Blackhull	1st	8.8	64.2	4.89
		2nd	9.2	47.5	3.62
		3rd	8.4	47.2	3.35
Thayer, 1948	Tenmarq	1st	10.2	43.8	4.36
		2nd	10.9	38.0	4.21
		3rd	9.2	44.4	3.48
	Turkey	1st	11.7	43.6	4.22
		2nd	12.4	45.3	4.14
		3rd	9.2	36.3	3.61
	Blackhull	1st	10.0	44.1	4.58
		2nd	10.7	50.5	4.58
		3rd	9.5	44.0	3.69

* Composition analyzed on a dry weight basis.

** In mg per 100 g of dry weight.

Table 19. Variety differences in the mineral composition of the wheat grass at three different stages of growth.*

Area and year	Variety	Growth stage	P %	Ca %	K %	Mg %
Columbus, 1947	Tenmarq	1st	0.291	0.32	2.02	0.207
		2nd	0.270	0.43	1.42	0.204
		3rd	0.236	0.42	1.68	0.186
	Turkey	1st	0.315	0.43	2.16	0.159
		2nd	0.255	0.40	1.78	0.169
		3rd	0.244	0.41	1.58	0.161
	Blackhull	1st	0.276	0.38	2.37	0.156
		2nd	0.260	0.42	1.72	0.170
		3rd	0.260	0.60	1.48	0.217
Thayer, 1947	Tenmarq	1st	0.293	0.45	2.28	0.151
		2nd	0.250	0.42	2.20	0.130
		3rd	0.260	0.40	2.11	0.124
	Turkey	1st	0.286	0.36	2.31	0.152
		2nd	0.222	0.34	2.26	0.116
		3rd	0.236	0.30	2.20	0.111
	Blackhull	1st	0.300	0.51	2.26	0.168
		2nd	0.265	0.48	2.48	0.134
		3rd	0.235	0.36	2.28	0.112
Thayer, 1948	Tenmarq	1st	0.202	0.35	2.86	0.122
		2nd	0.176	0.32	2.60	0.162
		3rd	0.160	0.31	2.46	0.152
	Turkey	1st	0.202	0.33	2.88	0.121
		2nd	0.168	0.29	2.66	0.136
		3rd	0.148	0.28	2.32	0.148
	Blackhull	1st	0.203	0.38	2.69	0.132
		2nd	0.171	0.31	2.74	0.169
		3rd	0.160	0.27	2.60	0.153

* Composition analyzed on the basis of dry weight.

Table 20. The two-year average variety differences of the chemical composition of the wheat grass at three different stages of growth.*

Variety	Growth stage	Ash %	Carotene mg**	N %	P %	Ca %	K %	Mg %
Tenmarq	1st	9.4	51.8	4.63	0.262	0.37	2.39	0.160
	2nd	9.2	46.9	3.97	0.232	0.39	2.07	0.165
	3rd	8.0	43.8	3.44	0.219	0.38	2.08	0.154
Turkey	1st	10.2	50.5	4.53	0.268	0.37	2.45	0.144
	2nd	9.6	49.7	3.81	0.215	0.34	2.23	0.140
	3rd	8.1	42.5	3.43	0.209	0.33	2.03	0.140
Blackhull	1st	9.6	53.4	4.75	0.260	0.42	2.44	0.152
	2nd	9.4	51.6	4.13	0.232	0.40	2.31	0.158
	3rd	8.3	44.2	3.48	0.218	0.41	2.12	0.161

* Composition analyzed on the basis of dry weight.

** In mg per 100 g of dry weight.

Wheat Grain

Table 21 shows the effect of the various fertilizer treatments on the percentage of total ash and of nitrogen in the wheat grain. The percentages of minerals in the grain at the various fertilizer treatments is presented in Table 22. Unfortunately only grain samples for one year's crop were available. Consequently definite conclusions regarding the effect of fertilizer on the chemical composition of the wheat grain cannot be drawn. However, some definite trends were noticed.

Total Ash. The ash content was definitely higher in the grain samples from the fertilized plots. The greatest increase was observed in those samples from the plots which had received the superphosphate treatment. The grain from the "no legume" plot had 1.76 per cent ash, the highest value.

Nitrogen. The nitrogen content was less in those plants receiving fertilizer treatment. The greatest effect seemed to result from superphosphate treatment. As in the case of the wheat grass, the manure treatment effected a slight increase in this element.

Phosphorus. There was an increase in the percentage of phosphorus in the grain as a result of fertilizer treatment. The lime treatment effected a slight increase, but substantial gains were observed in those receiving superphosphate. The grains from the plots which had received manure and "no legume" treatment had at least 0.30 per cent phosphorus. These observatione were the same as the corresponding ones made on the wheat grass, and are in accord with the findings of Ames (2), Lee (25), Murphy (29) and others.

Calcium. The results for calcium did not show such definite affects, however, a distinct trend was observed. The fertilizer treatments seemed to increase the calcium in the grain, especially in that wheat grown on the plots receiving the lime treatment. When the "no legume" treatment was given there was less calcium found in the grain, a 36 per cent decrease as compared to the control samples.

Potassium. Definite increases were observed in the potassium content of the grain grown on the fertilizer treated plots. This was opposite of the effect of fertilizer treatment on the potassium content on the grass. Superphosphate seemed to have a greater effect than the lime treatment alone. The potash treatment did not seem to cause any substantial increase of this mineral. The largest amount of potassium was found in the grain receiving the "no legume" treatment; it had 0.310 per cent potassium content, an increase of over 16 per cent as compared with the control.

Magnesium. There were also increases in the magnesium content of the grain grown on fertilizer treated plots. The addition of superphosphate had a greater effect than the lime treatment alone.

Table 21. The effect of various fertilizer treatments on the ash and nitrogen contents of the wheat grain.*

Treatment**	Ash		Nitrogen	
	per cent	per cent gain or loss	per cent	per cent gain or loss
Control	1.37	-----	2.00	-----
L.	1.49	+ 8.8	1.87	- 6.5
L.Sp.	1.70	+24.1	1.65	-17.5
L.Sp.	1.66	+21.2	1.66	-17.0
L.Sp.P.	1.65	+20.4	1.64	-18.0
L.Sp.M.	1.58	+15.3	1.78	-11.0
L.Sp.NL.	1.76	+28.4	1.64	-18.0
Average	1.60	+19.7	1.75	-14.7

* Composition was analyzed on a dry weight basis.

** Control plot received no fertilizer treatment; L = lime; Sp = superphosphate; P = potash; M = manure; and NL = no legume.

Table 22. The effect of various fertilizer treatments on the mineral composition of the wheat grain.*

Treatment**	P		Ca		K		Mg	
	%	% gain or loss	%	% gain or loss	%	% gain or loss	%	% gain or loss
Control	0.250	-----	0.044	-----	0.266	-----	0.109	-----
L.	0.262	+ 4.8	0.072	+63.6	0.275	+ 3.4	0.113	+ 3.7
L.Sp.	0.285	+14.0	0.064	+45.4	0.302	+13.5	0.119	+ 9.2
L.Sp.	0.297	+18.8	0.037	-15.9	0.284	+ 6.8	0.138	+26.6
L.Sp.P.	0.293	+17.2	0.050	+13.6	0.298	+12.0	0.129	+18.3
L.Sp.M.	0.300	+20.0	0.048	+ 9.1	0.280	+ 5.3	0.108	- 0.4
L.Sp.NL.	0.306	+22.4	0.028	-36.4	0.310	+16.5	0.123	+12.8
Average	0.285	+16.2	0.049	+13.2	0.288	+ 9.6	0.120	+11.7

* Composition was analyzed on a dry weight basis.

** Control plot received no fertilizer treatment; L = lime; Sp = superphosphate; P = potash; M = manure; and NL = no legume.

CONCLUSIONS

Definite effects of the fertilization on the chemical composition of the wheat grass and grain were observed. A marked decrease in the carotene content and in the percentage of nitrogen were shown to exist in the grass from the fertilized plots, especially where superphosphate was used.

Significant decreases in the total ash were observed in the wheat plants which had received superphosphate treatment. The 1948 crop had a higher ash content than the one from the previous year. Significant increases were noticed in phosphorus and calcium contents of the samples which had received superphosphate. The greatest gains of phosphorus were found in the 1947 crop. The percentage of potassium in crops receiving superphosphate decreased.

The application of potash seemed to have no effect on the potassium content, while applications of lime and superphosphates caused increases in the calcium and phosphorus contents respectively. No significant effect on the magnesium content was observed.

In the study of three different stages of growth of the wheat plant decreases in the carotene, nitrogen, total ash, phosphorus, potassium contents with age were observed. Greatest decreases were noticed in the nitrogen and phosphorus contents in the samples from the superphosphate treated plots. In those plants in which fertilization caused marked increases in the phosphorus content, there was a decrease of this element with age.

Only slight variety differences were observed in the carotene, nitrogen, calcium, and potassium contents.

Some yearly differences in chemical composition were observed in the variety samples as well as in the fertility treated ones.

The effect of fertilization on the constituents in the grain was observed to be decreased nitrogen content and increased total ash, phosphorus, calcium, and potassium contents.

ACKNOWLEDGMENTS

Indebtedness is acknowledged to Dr. J. S. Hughes and Dr. W. G. Schrenk for their assistance in choosing this problem; to Dr. Schrenk for his patience, helpful suggestions and supervision during this investigation; and to Dr. H. H. Laude and the Department of Agronomy of Kansas State College and various men in the field who carried out the treatments and collected the samples. All data on the fertilizer treatments were very generously furnished by the Department of Agronomy. The patience, encouragements, and unselfish assistance given by my wife, Georgina Johnson, during the course of this research are also gratefully acknowledged.

LITERATURE CITED

(1) Allison, Robert V.
The Rothamsted centenial. The original field plot studies at Rothamsted. Soil Sci. Soc. Am., Proc. 8: 6-11. 1943.

(2) Ames, J. W.
The composition of wheat. Ohio Agr. Expt. Sta. Bul. 221, 37 p. 1910.

(3) Ames, J. W., G. E. Boltz and J. A. Stenius.
Effect of fertilizers on the physical and chemical properties of wheat. Ohio Agr. Expt. Sta. Bul. 243: 567-587. 1912.

(4) Archibald, J. G., K. J. Kucinski, R. O. Brooke and S. L. Freeman. Nutritional anemia in cattle in southeastern Massachusetts. Jour. Dairy Sci. 21: 59-68. 1938.

(5) Auchter, E. C.
The interrelation of soils and plant, animal and human nutrition. Sci. 89: 421-442. 1939.

(6) Bayfield, E. G.
Effect of fertilizer on the ash of wheat. Ohio Agr. Expt. Sta. Bul. 579: 31-32. 1937.

(7) Becker, R. B., W. M. Neal and A. L. Shealy.
I. Salt sick: Its cause and prevention. II. Mineral supplements for cattle. Fla. Agr. Expt. Sta. Bul. 231: 23 p. 1931.

(8) Becker, R. B., W. M. Neal, A. L. Shealy and Gus York. Stiffs or Sweeny (phosphorus deficiency) in cattle. Fla. Agr. Expt. Sta. Bul. 264: 27 p. 1933.

(9) Beeson, Kenneth C.
The mineral composition of crops with particular reference to the soils in which they were grown. U.S.D.A. Misc. Pub. 369: 164 p. 1941.

(10) Beeson, K. C. and J. A. LeClerc.
The significance of the soil to human and animal nutrition. Soil Sci. Soc. Amer. Proc. 2: 335-341. 1937.

(11) Browne, C. A.
Some relationships of soil to plant and animal nutrition. The major elements. Soils and Men. U. S. Dept. Agr. Yearbook, 1938: 777-806. 1938.

(12) Cery, C. A.
Deficiency diseeses. Amer. Vet. Med. Assoc. Jour. 56: 609-614. 1920.

(13) Doneldson, Frenk T.
A study of the minerel nutrition of wheet es influenced by fertilizer combinetions. Plent Phys. 13: 737-766. 1938.

(14) Eckles, C. H., R. B. Becker end L. S. Pelmer.
A minerel deficiency in the retions of cattle. Minn. Agr. Expt. Sta. Tech. Bul. 229, 49 p. 1926.

(15) Fifield, C. C., S. R. Snider, Herland Stevens and Ray Weaver. The carotene content of wheat verieties in the Pecific Northwest. Cer. Chem. 13: 463-469. 1936.

(16) Forbes, E. B. end S. R. Johnson.
Phosphorus deficiency among cattle in Pennsylvenia. Pe. Agr. Expt. Ste. Bul. 371: 25 p. 1939.

(17) Gericke, W. F.
Protein content of wheat. Sci. 52: 446-447. 1920.

(18) Greeves, J. E., A. F. Brecken and C. T. Hirst.
The influence of variety, season end green manure upon the composition of wheats. Jour. Nut. 19: 179-186. 1940.

(19) Greaves, J. E. and E. G. Carter.
The influence of irrigation weter on the composition of greins end the relstionship to nutrition. Jour. Biol. Chem. 58: 531-541. 1923.

(20) Hert, E. B., B. A. Beach, E. J. Delwiche and E. G. Beiley.
Phosphorus deficiency and e deiry cattle disease. Wis. Agr. Expt. Sta. Bul. 389: 10 p. 1927.

(21) Kreybill, Henry R.
Effect of plent nutrition on the composition of wheat. Cer. Chem. 9: 71-80. 1932.

(22) King, H. H. and A. T. Perkins.
Plant nutrition investigations. Kan. Agr. Expt. Ste. Rept. 1932-1934: 28. 1934.

(23) Lawes, J. B. and J. H. Gilbert.
On the composition of the ash of wheat grein, and wheat strew, grown et Rothemsted, in different seasons, and by different manures. (London) Roy. Soc. Phil. Trans. (B) 192: 139-210. 1900.

(24) LeClerc, J. A. and P. A. Yoder.
Tri-local soil exchange experiments with wheat.
8th Internatl. Cong. Appl. Chem. 26: 137-150. 1912.

(25) Lee, Ching-Kwei.
Variations in yield and composition of the wheat plant as affected by the time of applying phosphatic fertilizers. Jour. Amer. Soc. Agron. 32: 782-788. 1940.

(26) Lindner, R. C.
Rapid analytical methods for some of the more common inorganic constituents of plant tissues. Plant Phys. 19, 1: 76-89. 1944.

(27) McCalla, A. G. and E. K. Woodford.
The effect of potassium supply on the composition and quality of wheat. II. Can. Jour. Res. 13. e. No. 6: 339-354. 1935.

(28) Moon, F. E.
The influence of fertilizer treatment on the carotene content of poor pasture grass, and on the relation of this constituent of the ash and organic fractions. Jour. Agr. Aci. 29: 524-543. 1939.

(29) Murphy, H. F.
Effect of fertilizers on the yield and composition of wheat. Amer. Soc. Agron. Jour. 22: 765-770. 1930.

(30) Russel, Sir E. John.
Soil conditions and plant growth. Ed. 7: 655 p. London, N. Y. 1937.

(31) Russell, E. J. and D. J. Watson.
The Rothamsted field experiments on growth of wheat. Com. Bur. Soil Sci. Tech. Comm. 40: 1-163. 1940.

(32) Schrenk, W. G. and H. H. King.
Composition of three varieties of Kansas-grown wheat. Mineral analysis of wheat and soil. Cer. Chem. Vol. 25. No. 1: 61-71. 1948.

(33) Snyder, H.
Influence of fertilizer on the composition of wheat. Jour. Amer. Chem. Soc. 30: 604-608. 1908.

(34) Sotola, Jerry.
The chemical composition and nutritive value of certain cereal hays as affected by plant maturity. Jour. Agron. Res. 54: 399-415. 1937.

(35) Stubblefield, F. M. and E. E. DeTurk.
The composition of corn, oats, and wheat as influenced by soil, soil treatment, seasonal conditions and growth. Soil Sci. Soc. Amer. Proc. 5: 120-124. 1940.

(36) Swanson, C. O.
Factors influencing the protein content of wheat. Kans. Agr. Exp. Sta. Rept. 1932-1934: 52-53. 1934.

(37) Thomas, Brynmor and E. E. Moon.
A preliminary study of the effects of fertilizer treatments and of age on the carotene content of grass. Emp. Jour. Expt. Agr. 6: 235-245. 1938.

(38) Welch, Howard.
Bone chewing by cattle. Mont. Agr. Expt. Sta. Cir. 122: 8 p. 1924.

(39) Wolfe, B.
Rapid tests for major nutrients in soils and plants. (mimeographed manuscript.) Res. Div. Seabrook Farms, Bridgeton, N. J. 1946.