THE EFFECTS OF POTASSIUM
FERTILIZATION AND NIGHT TEMPERATURES
ON THE ABSORPTION, TRANSLOCATION AND
ASSIMILATION OF IRON IN CENTIPEDEGRASS
(*EREMOCHLOA OPHIUROIDES*)

By
JON I. TENG

A DISSERTATION PRESENTED TO THE GRADUATE COUNCIL OF
THE UNIVERSITY OF FLORIDA
IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE
DEGREE OF DOCTOR OF PHILOSOPHY

UNIVERSITY OF FLORIDA

August, 1965

ACKNOWLEDGMENTS

The author wishes to express his sincere appreciation to Dr. W. L. Pritchett, Chairman of his Supervisory Committee, for his suggestions during these investigations, and for his guidance and constructive criticisms of the manuscript. Appreciation is also expressed to Dr. D. S. Anthony, Dr. G. C. Horn, Dr. F. B. Smith, Dr. D. O. Spinks and especially to Dr. T. L. Yuan for their constructive criticisms of the manuscript.

The author wishes to extend his appreciation to Dr. F. B. Smith, former Head of the Department of Soils, who awarded the assistantship, and to the American Potash Institute, whose research grant helped finance the study.

Thanks are also extended to Dr. H. H. Luke and Mr. Clarence Selin for their assistance in the laboratory phase.

The writer wishes to thank his wife for her patience and understanding throughout the study.

TABLE OF CONTENTS

	Page
ACKNOWLEDGMENTS	11
LIST OF TABLES	v
LIST OF FIGURES	xii
INTRODUCTION	1
REVIEW OF LITERATURE	4
Iron in the Soil	4
Absorption of Iron by Roots	11
Properties of roots	11
Factors affecting iron absorption	15
Translocation of Iron in the Plant	20
Mechanisms of translocation	20
Factors affecting the translocation of iron in plants	26
Assimilation of Iron in Plants	30
Concepts of iron assimilations in plants	30
Factors affecting assimilation of iron in plants	37
MATERIALS AND METHODS	42
Greenhouse-Cold Chamber Experiments	42
Experiment 1	42
Experiment 2	45

	Page
Experiments 3, 4 and 5	46
Laboratory Analyses	48
RESULTS AND DISCUSSION	55
Experiment 1	55
Experiment 2	59
Experiments 3, 4 and 5	67
Plant growth	67
Plant inorganic constituents	78
Some organic compounds in plants	93
Some properties of centipedegrass roots .	114
Soil mineral nutrients	117
General Discussion	124
SUMMARY AND CONCLUSIONS	136
APPENDIX	140
BIBLIOGRAPHY	188
BIOGRAPHICAL SKETCH	203

LIST OF TABLES

Table	Page
1. Some physical and chemical properties of Arredondo loamy fine sand.....	44
2. The composition of Hoagland nutrient solutions with and without potassium	46
3. Average maximum and minimum greenhouse temperatures.....	48
4. Analyses of variance of the effects of treatments on the yields and inorganic constituents in centipedegrass of experiments 4 and 5	74
5. Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the iron content in centipedegrass of experiments 4 and 5	81
6. Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the potassium content in centipedegrass of experiments 4 and 5	86
7. Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the calcium content in centipedegrass of experiments 4 and 5	89
8. Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the magnesium content in centipedegrass of experiments 4 and 5	91
9. Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the phosphorus content in centipedegrass of experiments 4 and 5	92

Table	Page
10. Effects of night temperatures and rates of potassium and iron applications on the nitrogen content in centipede-grass of experiments 4 and 5	94
11. Organic acids found in leaves, stolons and roots, in percentage of fresh weight of plant material	97
12. Analysis of variance of the effects of treatments on the organic acid content of the whole plant and leaves of centipede-grass of experiments 4 and 5	100
13a. Oxalic, citric, and malic acids in leaves, stolons, and roots of centipede-grass of experiments 4 and 5	102
13b. Oxalic, citric, malic, tartaric, aconitic, and succinic acids in leaves of centipede-grass of experiments 4 and 5	103
14. Effects of night temperatures, rates of potassium, and rates of iron on the total organic contents of the leaves, stolons, roots, and entire plants of centipede-grass	105
15. Organic acids contained in leaves of centipede-grass supplied with and without iron chelate at the lowest night temperature (-1.1° C.)	106
16. Analysis of variance of effects of treatments on the amino acid content of centipede-grass	110
17. Analysis of variance of the effects of treatments on the sugar content of centipede-grass	115
18. Analysis of variance of the effects of treatments on the extractable iron, potassium, magnesium, calcium and phosphorus contents of soils	119
19. Available nutrients in the soil	120

Table	Page
20. Effects of night temperatures and sources and rates of potassium on yield (gm. oven-dry wt.) of centipede grass of experiment 1	141
21. Effects of night temperatures and sources and rates of potassium on iron concentration (ppm) in centipede grass of experiment 1	142
22. Effects of night temperatures and sources and rates of potassium on percent potassium in centipede grass of experiment 1	143
23. Effects of night temperatures and rates of potassium on yield (gm. oven-dry wt.) of centipede grass of experiment 2 ..	144
24. Effects of night temperatures and rates of potassium on iron concentration (count per minute from Fe ⁵⁹) in centipede grass of experiment 2	145
25. Effects of night temperatures and rates of potassium on percent potassium in centipede grass of experiment 2	146
26. Analysis of variance of the effects of treatments on iron and potassium contents in centipede grass of experiment 2	147
27. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves, stolons, roots and entire plants yields (gm. oven-dry wt.) of centipede grass of experiments 3, 4 and 5	148
28. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves, stolons, roots and entire plants iron content (ppm) in centipede grass of experiments 4 and 5	151

29.	Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves, stolons, roots and entire plants percent potassium content in centipede grass of experiments 4 and 5	154
30.	Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaf, stolon, root and entire plant percent calcium content in centipede grass of experiments 4 and 5 ..	157
31.	Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaf, stolon, root and entire plant percent magnesium content in centipede grass of experiments 4 and 5	160
32.	Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves, stolons, roots and entire plants percent phosphorus content in centipede grass of experiments 4 and 5	163
33.	Effects of night temperatures, rates of potassium and iron applications on percent nitrogen content of leaves, stolons and roots of centipede grass of experiment 5	166
34.	Effects of night temperatures, rates of potassium and iron applications on percent oxalic acid content of leaves of centipede grass of experiments 4 and 5 ..	167
35.	Effects of night temperatures, rates of potassium and iron applications on percent oxalic acid content of leaves, stolons and roots of centipede grass of experiment 5	168

Table	Page
36. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent oxalic acid content in centipede-grass of experiments 4 and 5	169
37. Effects of night temperatures, rates of potassium and iron applications on leaves stolons and roots percent citric acid content in centipede-grass of experiment 5..	170
38. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent citric acid content in centipede-grass of experiments 4 and 5	171
39. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons, roots percent malic acid content in centipede-grass of experiment 5	172
40. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves malic acid content in centipede-grass of experiments 4 and 5	173
41. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent tartaric acid content in centipede-grass of experiments 4 and 5	174
42. Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent aconitic acid content in centipede-grass of experiments 4 and 5	175
43. Effects of night temperatures, rates of potassium and iron applications on leaves percent succinic acid content in centipede-grass of experiment 5	176

44. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons, and roots percent aspartic acid in centipedegrass of experiment 5	177
45. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent glutamic acid in centipedegrass of experiment 5	178
46. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent glycine in centipedegrass of experiment 5	179
47. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent histidine in centipedegrass of experiment 5	180
48. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent isolencine content in centipedegrass of experiment 5	181
49. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent reducing sugar content in centipedegrass of experiment 5	182
50. Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent cold water extractable carbohydrate in centipedegrass at experiment 5	183
51. Effects of night temperatures, rates of potassium and iron applications on the reductive capacity of root in centipedegrass of experiment 5	184

Table	Page
52. Effects of night temperatures, rates of potassium and iron applications on cation exchange capacity of roots in centipedegrass of experiment 5	185
53. Chemical analyses of Arredondo loamy fine sand at the conclusion of experiments 4 and 5	186
54. Soil nitrate (ppm) determined at the conclusion of experiment 5	187

LIST OF FIGURES

Figure	Page
1. The effect of night temperatures and potassium sources on the concentration and total absorption of Fe and K in centipedegrass.....	56
2. Iron and potassium in centipedegrass as affected by three rates and two sources of potassium.....	58
3. Effects of night temperatures and rates of potassium on yields on centipedegrass.....	61
4. Effects of night temperatures and rates of potassium on concentration and total absorption of iron in centipedegrass.....	63
5. Total iron (Fe ⁵⁹) and potassium contained in tops and roots of centipedegrass at three growth periods.....	65
6. Effects of night temperatures and rates of potassium in the presence of added iron on the appearance of centipedegrass.....	68
7. Effects of night temperatures and rates of potassium in the absence of added iron on the appearance of centipedegrass.....	69
8. Effect of night temperatures, rate of potassium and iron on leaves of centi- pedegrass.....	71
9. Effects of night temperatures, rates of potassium and iron on roots of centi- pedegrass.....	72
10. Effects of night temperatures and seasons on yields of centipedegrass.....	77
11. Effect of night temperature and rates of iron on the root yields of centipedegrass.....	79

12.	Effects of rates of potassium on yields of centipede grass.....	79
13.	Effects of night temperatures and rates of iron on the iron content of leaves, stolons, roots, and entire plants of centipede grass.....	84
14.	Effects of rates of potassium and night temperatures on the percent potassium in leaves of centipede grass.....	87
15.	Paper chromatogram of organic acids from leaves, stolons, and roots of centipede- grass.....	96
16.	Paper chromatogram of amino acids from leaves, stolons, and roots of centipede grass.....	108
17.	Effects of night temperatures on the percent of aspartic and glutamic acid in leaves, stolons and roots of centipede grass.....	111
18.	Effects of night temperatures on the percent of glycine and isoleucine in leaves, stolons and roots of centipede grass.....	112
19.	Effects of night temperatures on the percent of histidine in leaves, stolons and roots of centipede grass.....	113
20.	Effects of night temperatures on reductive capacities of roots of centipede grass.....	116

INTRODUCTION

In a modern, highly mechanized society, turf-grasses play an increasingly significant role by adding beauty to mundane environments and serving as foundations for play areas on which man can spend his leisure time. Grasses are the principal ingredient in landscaping for homes and public buildings and in golf courses, parks and playgrounds. Several varieties of turfgrasses are used for this purpose in Florida. Among these, centipedegrass has been one of the most popular since about 1920 due to the low maintenance requirements. However, there has been a gradual decline in the popularity of centipedegrass during the past decade due, in part, to the yellowish, chlorotic appearance often found in spring and early summer. Bryan reported in 1933 (32), that this unattractive discoloration could be ameliorated by intermittent spray of the chlorotic grass with an iron sulfate solution. Consequently, this yellowing of centipedegrass has been referred to as "iron chlorosis."

Centipedegrass grown in plots at the University of Florida Horticultural Unit, on Arredondo loamy fine sand, often exhibited a chlorotic appearance from early

spring to early summer when the night temperatures were relatively low. This chlorosis has been reported (72) to be closely related to applications of high rates of potassium fertilizers and to be more prevalent during periods of rapid growth, particularly where high levels of nitrogen were also applied (73). Among the soil factors often mentioned as being associated with soil iron availability are (a) a high content of phosphates, (b) a pH of 6.5 or above, (c) the presence of large amounts of bicarbonates, (d) an excess in heavy metals, (e) competition from soil microorganisms, and (f) a high oxidation-reduction potential of the soil. It is also believed that a chlorotic condition in plants may be induced by the physiological failure concerning the uptake of an adequate amount of iron, or the difficulty in the assimilation of iron by the plant. The chlorosis in centipedegrass associated with low night temperatures and/or high rates of potassium fertilizers may result from either a deficiency in available soil iron or with an inhibition in the absorption, translocation or assimilation of iron by the plant--or a combination of these factors.

This research was undertaken to study the effects of certain night temperatures and rates of potassium fertilizers on the absorption, translocation, and

assimilation of iron and on the organic and inorganic composition of centipedegrass, and to attempt to relate these properties of the plant to the prevalence of "iron chlorosis."

REVIEW OF LITERATURE

Iron in the Soil

Iron, the fourth most abundant element in the earth's crust, composes 6 to 7 percent of the soil mass and is found mostly as oxides (79). Iron occurs in primary minerals as ferromagnesian silicates. The iron-bearing secondary minerals are biotite micas, and the commonly occurring free iron groups are iron oxides and inorganic and organic iron salts (111). Iron oxides are the compounds that hold the greatest interest for agriculturists. Iron oxides include (a) hematite, Fe_2O_3 , red in color; (b) goethite, $\text{Fe}_2\text{O}_3 \cdot n\text{H}_2\text{O}$, brown or yellow; (c) matite, Fe_2O_3 , a dark iron compound; and (d) magnetite, Fe_3O_4 , a mixture of ferric and ferrous iron (111). Inorganic iron salts are compounds of hydroxides, carbonates and sulfates. Organic iron salts occur in soil organic colloids (71, 119).

Soil scientists are interested in free iron oxides in soil because they are involved in a great many soil problems. For example, poor drainage and other farming difficulties are often associated with the problems of iron concretions. Furthermore, the availability of phosphorus due to iron fixation at low soil

pH, or adsorption of phosphates on the surface of the iron oxides at high soil pH (111) is often a problem in soils high in iron. On the other hand, of significant importance is the nutritional value of iron to a green plant in the formation of chlorophylls (17, 38, 99).

Although the total amount of iron in soils is generally high, there often is a deficiency of available iron due to the stability of the ferric oxidation form, Fe^{3+} (49, 54, 79). Thus, to understand iron nutrition, it is necessary from the very beginning to study the transformation processes of soil iron under different conditions.

The majority of iron in agricultural soils is present in an insoluble form as Fe^{3+} , which is the stable oxidation state. The lower oxidation state of Fe^{2+} usually occurs in soils in quite small quantities. However, it is the ferrous iron, Fe^{2+} which has been reported (49, 119) as the form plant roots can most readily absorb, since ferrous salts have relatively higher solubility product values. The solubility product values of $\text{Fe}(\text{OH})_2$, FeCO_3 , and FeSO_4 are 1.6×10^{-14} at pH 8.0, 3.5×10^{-11} at pH 7.0, and 3.7×10^{-16} at pH 7.0, respectively (119). In contrast, $\text{Fe}(\text{OH})_3$ has a very low value, 6.0×10^{-38} , at pH 4.0 (148). Furthermore, ferrous iron fulfills a greater number of physio-

logical functions than ferric iron inside the plant (113, 116, 146, 149).

Both ferrous and ferric iron in soils are capable of movement in the form of organic complexes if sufficient amounts of low molecular weight organic acids are replenished by the decomposition of organic matter (49). Polyphenols, such as catchin, which can be released from fresh organic matter, are the other agents utilized in translocating iron compounds (111). Nevertheless, iron movement may be stopped if the soil undergoes alkaline reactions which result in low iron solubility, or if there is a sufficient concentration of transition elements present which tend to replace the iron from the organic complex (111).

The stability of an organic iron complex depends on the nature of its anion component (49). The order of these organic anions on the basis of their strength in forming organic complexes is:

citrate > oxalate > tartrate > acetate

The order for the inorganic anions is:

hydroxyl > arsenate > borate > thiocyanite >

sulfate > chloride > nitrate

Chemically, ferrous iron is generally predominant in soils with pH below 5.0, while above pH 6.0 ferric iron will be predominant (1). Whenever a high pH

condition is built up due to alkaline end products from soil microorganisms, or from other oxidation processes, ferric iron will be precipitated from the soil solution.

Oxidation-reduction potential plays an important role in determining the forms of iron in soils. There exist a great many redox systems. The oxidation potentials of these systems usually vary from the equivalent of hydrogen's redox system, 0.00 V, to that of oxygen's redox system, 1.23 V. Ponnampetuma (119) has discussed these systems in detail in his study of submerged soil. Several of them are reviewed below:

<u>Redox System</u>	<u>$E_o(V)$</u>	<u>$E(V)$ at pH 7.0</u>
$O_2 + 4H^+ + 4e^- \rightleftharpoons 2H_2O$	1.23	0.82
$Fe^{3+} + e^- \rightleftharpoons Fe^{2+}$	0.77	
$MnO_2 + 4H^+ + 2e^- \rightleftharpoons Mn^{2+} + 2H_2O$	1.23	0.43
$NO_3^- + H_2O + 2e^- \rightleftharpoons NO_2^- + 2OH^-$	0.00	0.41
$Fumarate + 2H^+ + 2e^- \rightleftharpoons Succinate$		-0.03
$Oxalate + 2H^+ + 2e^- \rightleftharpoons malate$		-0.10
$Fe(OH)_3 + 3H^+ + e^- \rightleftharpoons Fe^{2+} + 3H_2O$	0.77	-0.13 ¹
$Fe(OH)_3 + e^- \rightleftharpoons Fe(OH)_2 + OH^-$	-0.56	-0.14 ²

A general redox system is of the following nature:

¹At pH 6.5, calculated by the author.

²At pH 7.0, calculated by the author.

Then, the Nernst equation is used to calculate the oxidation potential:

$$E = E_0 + \frac{RT}{nf} \ln \frac{\text{Oxidant}}{\text{Reductant}}$$

In this equation, where E is the oxidation potential, R is the gas constant, T is the absolute temperature, n is the number of electrons lost in the change from reduced form to oxidized form, f is the value of Faraday and E_0 is the standard oxidation potential (20). When the concentration of oxidant and reductant has unit ratio, the oxidation potential E is equal to the standard oxidation potential E_0 (20, 38).

In the above list of redox systems any one system will be able to oxidize the one below it. Since the oxygen system has higher E_0 (1.23V+) in the soil, this system can oxidize the rest of the systems which appear in the list. A well-aerated soil, therefore, may have deficiencies of some nutrients due to this oxidation of soluble reduced forms of the nutrients to insoluble oxidized forms. The majority of soil pH values fall between 5.0 and 9.0 (1, 69), therefore a redox system occurring in soils should involve the concentration of hydrogen ions (119). A general equation involving the hydrogen ion is:

$$E_h = E_0 + \frac{RT}{nf} \ln \frac{\text{Oxidant} \cdot \text{H}^+{}^m}{\text{Reductant}}$$

E_h is the oxidation potential at a certain concentration of H^+ . Commonly, pH 7.0 is taken as the condition to calculate E_h . This equation clearly shows that a change in pH will influence a system's oxidation potential.

The standard oxidation potential of $Fe^{3+} \longrightarrow Fe^{2+}$ is 0.77 v, which indicates that Fe^{2+} is quite stable in an acid solution. Whenever a higher redox system, such as that of oxygen, exists with the $Fe^{3+} \longrightarrow Fe^{2+}$ system, a redistribution of the electrons will result in a spontaneous reaction favoring the reduction of oxygen to water (20, 119) and leaving the iron in Fe^{3+} form. The same thing happens when iron reacts with a manganese system. Ponnampeuma (119) believed that ferric iron could not be reduced until a considerable amount of manganese had been reduced, and that nitrate reduction could start only after nearly all of the manganese was in reduced form. When the soil pH gradually approaches neutrality the manganese and nitrate systems have about the same reduction potential, 0.43 v and 0.41 v, respectively, so that these systems are reduced simultaneously. However, the oxidation potential of the iron system drops drastically to -0.142v. Therefore, the reduction of iron will be very difficult because it is governed by all the systems above it. Evidently reduction of manga-

nese and nitrate plays an important role in controlling the iron system as soil pH increases. Manganese is one of the transition metals which is known to induce iron chlorosis in some tree species (29). Furthermore, soils high in nitrates and undergoing vigorous reduction processes have caused iron chlorosis in pineapple and other plants (1, 18). In a soil of pH 7.0, manganese and nitrate systems have retarded the reduction of iron and stabilized the potential at 0.2 V to 0.4 V (18). In addition, the formation of organic complexes have a marked effect on the oxidation potential of a redox system by reducing the oxidation potential and rendering metals to their reduced forms (46, 71). The reduced form of iron, Fe^{2+} , possibly functions as an exchangeable cation, since the amount of Fe^{2+} ion extracted by a KCl solution was much more than that extracted by an aqueous solution (74).

Iron transformation takes place in the presence of soil microorganisms, and the existing forms of iron are influenced by the type of microbes. A group of bacteria, called iron bacteria, oxidizes ferrous iron to ferric iron (1). Many heterotrophic species attack soluble organic iron salts causing the iron to precipitate out as ferric iron (1). Groups of bacteria can produce acid end-products that reduce ferric iron to ferrous iron

(71). Under anaerobic conditions, a large number of reducing agents, such as sulfides, reduce ferric to ferrous iron (1). Anaerobic microorganisms can also stabilize iron in the ferric state by utilizing the oxygen of nitrate in preference to oxygen of iron oxides (1, 49, 119).

Low soil temperature may affect the transformation of iron through its immediate influence on nitrate reduction (22). A decrease in nitrate reduction with decreased temperature results in more available iron. A low soil temperature and high moisture increased the solubility of iron due to delayed nitrate reduction in submerged soils (119).

Absorption of Iron by Roots

In grasses, there is a zone close behind the root tip bounded externally with piliferous roots. Grasses grown in sandy soils usually exhibit long and persistent root hairs (49). The root hair zone has several important properties which are responsible for the absorption of iron and other ions.

Properties of roots

The cation exchange capacity of the root has been extensively studied by a great many people (49, 50, 51). Jenny (84) produced a picture of mucilaginous substance

about roots believed to be pectic material on the root cell wall and thought to serve as cation exchange sites. Drake (49) reported that dicotyledons usually have higher root cation exchange capacity (CEC) than monocotyledons. Many workers (45, 51, 84) have shown that roots high in CEC absorbed more divalent cations than monovalent cations, while plants with low root CEC absorb a higher percentage of monovalent cations. An inverse relationship between potassium and CEC and a direct relationship between calcium and CEC has been recognized in legumes (49). However, the inverse potassium-CEC relationship can be found only when there exist a low concentration of potassium and a high concentration of calcium as explained by Wallace (45). It has been reported (49) that roots with a high CEC has greater uptake of iron than did roots with a low CEC. The root CEC of several grasses generally has been found to be below 30 m.e./100 gm. dry weight, which is lower than most dicotyledons but higher than most cereals (49).

Brown et al. (28) have shown that non-chlorotic Hawkeye soybeans have roots of higher reductive capacity and higher content of reduced cytochrome c than did chlorotic PI 54619-5-1 soybean. The reductive capacity was expressed as the capability of roots to reduce ferricyanide to ferrocyanide. This reductive capacity

in roots was also found (28, 49) to be influenced by the upper portion of the plant and the ability of the plant to synthesize various products.

(The presence of natural chelating agents has been observed in many plants roots (26, 46, 123, 151). For example, the chlorotic soybean PI 54619-5-1 was found by Brown (28) to have low root chelation capacity, while non-chlorotic Hawkeye soybean has high chelation capacity.) The most effective natural chelating agents in the roots are citric, tartaric, oxalic, and amino acids (49).

The specificity of a chelating agent is supposed to have great importance, according to Wallace (147). That is, certain chelating compounds are specific for absorption of sodium, magnesium, and iron; and others are involved in the accumulation of potassium, calcium, magnesium, and sodium in biological systems (148). This indicates that natural chelating agents are selective in regard to plant nutrients. In several exudates collected from plant species (123), there appeared to be both qualitative and quantitative differences in these natural chelating agents. It has been shown (123) that iron present in the plant exudate was in a chelated form. However, the organic component was not identified. There are also reports of the synthetic chelating agent entering into plant tissues with chelated metals (26).

They may or may not dissociate in the course of translocation, depending on the characteristics of the natural chelating agents and upon the pH of the plant sap (134). Plants have a great quantity of organic acids which is usually correlated with a highly active chelating nature. Citric, malic, and oxalic acids have been found in the roots of most plants (49). Ulrich (144) indicated that the organic acids in barley roots may be stimulated by a high absorption of K^+ in a solution of $KHCO_3$.

Generally, small amounts of reducing sugars can be found in root tissues. The reducing sugars D-glucose, D-fructose, D-galactose, and D-mannose are capable of reducing the higher oxidation state Fe^{3+} to the lower oxidation state Fe^{2+} (28). The cold water extractable carbohydrates which include α -malate, β -cellobiose, and β -lactose also have reducing powers (38). The high absorption capacity of a root is directly proportional to its reducing sugar content. It is reported that an increase in soluble reducing sugars can greatly enhance the uptake of cations (31). On the other hand, the lack of sufficient K^+ in the plant tissue may result in a high amount of reducing sugars in roots (39, 96, 134). This may facilitate iron absorption, but at the same time less iron would be used in metabolic activities due to a block in carbohydrate synthesis (93).

Growth of the root increases the number of binding sites and therefore, increases the total uptake of nutrients. One of the essential elements in increasing the growth of root is potassium (93, 105). Another important factor affecting plant root growth is temperature (37, 52, 64, 106, 107). In general, growth of roots decreased with increases in soil temperature, as observed in oats (22), buckwheat (62), and Kentucky bluegrass (23). Temperature also affects the morphological characteristics as reported by various researchers (120, 121). With a soil temperature at 10° C. roots were white, translucent, and relatively unbranched; at 15.6° C., roots were slender, profusely branched, and longer; and at 26.7° C. roots were very fine and fibrous but less extensive (121, 131). The total absorption of ions by roots was often related to the vigor of root growth which, in turn, was the function of soil temperature up to a limit of approximately 40° C. (37).

Factors affecting iron absorption

Iron is absorbed by roots through three mechanisms according to Glauser (60). The first is CO₂ pressure. The CO₂ evolved from cellular respiration of plants and soil microflora dissolves in the soil solution as carbonic acid which brings some ferric into the soil

solution to form ferrous iron. The second is synthesis of chelating agents. A third, surface contact, implies root contact exchange that is related to the CEC of the roots.

It is assumed that plant roots can not absorb Fe^{3+} directly. The ferric Fe^{3+} must first be converted to Fe^{2+} (88). However, this may not hold true for all plants. Bell et al. (8) found that one genotype of corn, could absorb Fe^{3+} where another failed to absorb Fe^{3+} when they were placed in the same ferric iron solution. The reason for a plant failing to absorb Fe^{3+} might be due to its accumulation in the root tip (89) or due to precipitation of Fe^{3+} on the roots' surface (46). The accumulations of ferric iron in the root tip were in the form of insoluble phosphates and organic iron complexes since tips of roots have high rates of metabolic activity but have no conducting tissue (89). Iron precipitated on the surface of roots primarily as ferric phosphates (11, 46).

The effect of potassium ion on the uptake of iron in plant roots has been observed to be an inverse relationship in many plants (63, 124). It was not clear as to which factor caused the deficiency of the other. Some workers (140, 146) in discussing high concentrations of potassium associated with low concentrations of iron

have designated iron as the cause and potassium as the result.

Shear's statement (124) and the cation-equivalent-constancy hypothesis in plants (146) indicate that at a given concentration of anions any increased accumulation of one or more cations must be accompanied by an equivalent decrease in one or more of the other cations. Therefore, whenever luxury consumption of potassium is favored, other cations suffer a depression. For example, the uptake of calcium, magnesium, and iron decrease in the presence of a larger quantity of potassium (108).

The absorption of potassium was reduced by treating the roots with CO₂ (49). High concentrations of CO₂ reduced nutrient uptake by the roots in the order of K > N > P > Ca > Mg (49). However, the uptake of iron was relatively increased as the CO₂ partial pressure increased in the soil (66). This suggests a possible cause for the inverse relationship between potassium content and iron content in a plant.

There have been many reports (10, 16, 41, 63, 65, 95, 118, 124) on the inverse relationship between potassium and iron in sugar cane, potato plants, pasture grasses, and tung trees. On the other hand, reports showed that potassium fertilization increased the

absorption of iron by increasing the rate at which the inorganic phosphates were converted into organic phosphates (3, 145). This activated the insoluble inorganic iron phosphates on the root surfaces and allowed them to move into the root cells (34). Ponnampuma (34, 119) reported that the addition of potassium caused some iron that had accumulated in the roots of sugar cane to pass upward in the plant.

Other cations which affect the absorption of iron appear to be zinc, calcium, manganese, and copper. Epstein (54) states that iron absorption in some plants was a function of the ratio of calcium to hydrogen in the soil. In soil solutions with the ratio of calcium concentration to that of hydrogen from 0.25 to 0.75, the plant roots could take up iron without trouble. Iron absorption decreased sharply when the ratio became 0.92. Manganese has been known to decrease iron content in some plants (54, 125, 130, 140). Copper, accumulated in Florida sandy soils, caused iron chlorosis in citrus (71). However, it has been reported (26) that a heavy application of copper increased ascorbic oxidase. To counteract the increasing ascorbic oxidase activity, the activity of the cytochrome oxidases increased, which resulted in greater iron absorption (26).

The anions which may possibly affect the absorption of iron are bicarbonates, phosphates, nitrates, and chlorides. Usually, the first two anions affect iron absorption to a greater extent than the latter two anions. Bicarbonates reportedly reduced the iron contents of some plant tissues (7, 25, 27, 29). As reported by Holmes et al. (71), irrigation caused chlorosis of peach trees, and the decomposition of organic matter intensified chlorosis of other fruit trees. The chlorosis was due to the high concentrations of bicarbonates produced in the processes. Bicarbonate effects iron absorption through increase in the soil pH and the solubility of phosphates (9), which decreases iron solubility in the soil solution (29). That phosphates will precipitate iron out of soil solution and deposit it on the surfaces of roots is rather well established (2, 48). Nitrate reduction, which takes place in root tips, will decrease the availability of iron (1, 71). The effect of chloride ions on iron absorption is less understood (71). The rate at which chloride is absorbed by the roots is about the same as the rate for nitrate ions (144).

Lundegardh (97) stated that the absorption of anion is a much more temperature-dependent process than the absorption of cations. He calculated the temperature

coefficient, Q_{10} , for the absorption of nitrate as 2.0 - 2.5 and Q_{10} for potassium as 1.4. Q_{10} is defined as: the ratio of reaction rates at two temperatures 10° C. apart (99). The equation given by Wallace and Sufi (147) is

$$Q_{10} = \frac{\text{uptake of } T_2}{\text{uptake of } T_1} \frac{1}{(T_2 - T_1)/10}$$

where T_1 and T_2 are temperatures in $^{\circ}$ C. A value of Q_{10} less than 1.5 is regarded as a physical phenomenon and larger than 2.0 as a chemical process (99).

A rising in soil temperature did increase the activity of bicarbonate ions (25, 71) which rendered iron deficiency. Soil iron availability had been decreased by lowering the pH of soil solution due to low temperature (83). Saric et al. (122) studied the relationship between soil temperature and iron absorption in wheat. They found that the iron content of leaves was greater at temperatures of from 4° to 30° C. while plants grown at temperatures on either side of this range had lower iron content.

Translocation of Iron in the Plant

Mechanisms of translocation

Ions in soil (5) taken up by roots either accumulate in the vacuoles or move in the cytoplasm.

Most probably the ions that enter the xylem to be translocated to the top tissues are the ones found in the cytoplasm. A scheme for translocation of ions in plants as described by Fried and Shapiro (58) is:

M represents the ions in soil solution, R is the carrier at the binding site in plant roots, k_2 is the rate of turnover of ion, and k_1 is the rate at which ion is translocated. Minerals or ions taken up by carriers on the protoplasmic membrane form complexes of MR. MR may deposit M in the vacuole or M can be released from the complex and enter into the xylem. The minerals or ions appearing in the xylem are carried up along the transpiration stream to the top of the plant. The rate of transpiration is slow during the night and rapid during the day, while the rate of root respiration is the opposite. Nevertheless, the energy for translocating minerals is produced by the mechanism of respiration (43, 91, 112, 138). The translocated minerals or ions may have four different destinations varying with plant tissues, individual minerals, and immediate metabolic conditions (11). The destinations are: (a) acceptance

by cells adjacent to xylem, (b) lateral movement via rays to the actively metabolizing cells, (c) depositions in leaves, and (d) to the apical primordia. Minerals accumulated in small vacuoles would possibly be transported, except for those in the xylem, through the plasmodesmata in the sieve tubes of the phloem. The vacuole translocation through plasmodesmata is usually conducted in darkness (134, 135).

Iron translocation in plants was shown by Brown et al. (21) through an application of Fe^{59} to a plant leaf. This iron isotope was translocated throughout the phloem to the newly formed leaves and to the tips of roots. This translocation of iron is equally effective when it is applied to roots (148).

The idea that the rate of nutrient translocation is directly proportional to the rate of transpiration probably does not hold, according to Sutcliffe's review (134), since an increase in the osmotic pressure of the external solution decreased transpiration but did not affect mineral absorption. Therefore, the translocation of minerals probably is in chelated form with organic compounds. There were a great number of researchers (12, 15, 56, 57, 82, 101, 136, 137) who had called attention to the consideration of the translocation of these organic compounds in the plants.

All organic compounds, such as the photosynthate, the metabolic intermediates, and other assimilates are translocated through the phloem to all non-synthesizing, but synthates requiring sites in the entire plants (6, 12, 15, 66). An interesting observation on the translocation of the organic compounds in the plant (89) is that when an organic compound is translocated from a green portion of a leaf to a colorless part of the same leaf, it first flows down the phloem to the roots and then rises again through the xylem to the colorless portion.

The destination for organic compounds from a green leaf depends on the leaf position (11). The lower leaves send organic compounds to roots; the uppermost leaves send them to the apex; while the intermediate leaves send them in both directions, all via the phloem. The rate of flow of organic compounds toward the roots is greater during the vegetative growth period because the roots need these compounds to maintain a high activity of absorption (141).

Sucrose is believed to be the first synthate sent into roots (136). It was found that glucose and fructose compose only 3-10 percent of the total sugars sent to roots of sugar beets (144). But reducing sugar was the primary form found in the phloem of the guayule tree (39, 40). Temperature has great influence on sugar

transportation. The Q_{10} for sugar translocation in tomato (153), in Bilox bean (89), and in sugar cane (152) was usually less than one. This led Went (152) to hypothesize that translocation of sugar from leaves to the other parts of the plant is faster at low temperatures. Went and Hall (153) found that in some plants localized sugar movement could be brought about to a stop at temperatures of 2° to 5° C., but to stop the plant's entire sugar movement the temperature should be 0° to 2°C.

Most of the organic acids seem to accumulate in surrounding cells of the phloem rather than entering into the flow path and therefore, are only weakly exported from assimilating sites of the leaves (89). However, citric and malic acids are exceptional in that they are transported in a considerable amount (89, 100, 144). Nevertheless, sugar in the phloem can be converted into very small amounts of organic acids. Ulrich (144) showed that temperature was negatively correlated with the amount of total organic acids in the excised barley roots. In another experiment Ulrich (144) and others (59, 61) showed that a positive correlation existed between total organic acids and total cations absorption. Kursanov (89), reported that amino acids comprise 0.2 to 12 percent and in some cases up to 50 percent of the total translocated assimilates. The rate of movement of amino acids

was estimated to be between 370-1,390 cm./hr. in 20-24 day-old soybeans. Nelson et al. (106) observed that glutamic acid and glycine moved slowly in the soybeans and that alanine, serine, and aspartic acid moved rapidly. Amino acids, like sugar, can be translocated from leaves to roots via phloem and also from roots to leaves via the xylem. Low temperature delays the transportation of amino acids from leaves to roots (137). Brown (22) reported that when the shoots and roots of Bermuda grass were held at 10° C. and those of orchard grass, Kentucky bluegrass, and Canada bluegrass held at 4.4° C., translocation of nitrogen assimilates was retarded. He attributed this retardation to the slow synthesis of nitrogen containing assimilates. Furthermore, temperature affected the types of amino acids found in the Robinia pseudocacia (89). During summer primarily glutamic acid, leucine, and valine were found in the phloem; during autumn, proline replaced glutamic acid and the others remained the same. The concentration of amino acids is usually higher in autumn than in summer (22, 89).

The effect of alternate day and night temperatures on the translocation of organic compounds in the conducting tissues has received considerable study. Nelson et al. (106) and Thrower (141) realized that the

process of translocation in a plant was much easier in the night than in the daytime. Kursanov (89) reviewed that organic compounds in the pumpkin plant were transported almost exclusively at night. The lower night temperatures appeared to affect the translocation of organic compounds from leaves to root of sugar beets much more than higher night temperature, as pointed out by Ulrich (144). He found that beet root had sugar concentrations of 11.9 percent at a night temperature of 2° C. and only 7.2 percent at a night temperature of 30° C.

The sap of conducting tissue is rich in K-containing compounds, approximately 2 percent of potassium was found in the phloem sap of Salix viminalis, according to Kursanov (89). However, he reported only low percentages of calcium and sodium in the sap. He explained the high concentration of potassium in phloem sap as due to potassium containing enzymes, such as phosphofructokinase, pyruvatekinase, and aminopherase.

Factors affecting the translocation of iron in plants

During the translocation of iron from roots to stem a change in the supply of oxygen partial pressure to the root will cause a change of speed in translocation. DeKock (46, 47) recorded that plants grown in an atmosphere of 1 percent oxygen contained much more iron in the stem than

those with 20 percent supply of oxygen. The author did not explain whether the low iron in the stems of the latter was a result of an oxidation state change from Fe^{2+} to Fe^{3+} .

Temperature may either accelerate or slow down the translocation of nutrients (98, 128). The temperature coefficient, Q_{10} , is a means to determine whether a given ion is transported or metabolically accumulated. Cool temperatures inactivated iron although it did not prevent iron absorption by plant roots (140). Branton and Jacobson (19) reported that the movement of iron in peas depended on metabolic energy. The metabolic energy is derived from cellular respiration and respiration is influenced by temperature (43, 44, 128).

The translocation of potassium in plants is a temperature-dependent phenomenon (107, 108, 117). Hoagland and Broyer (68) reported that the Q_{10} for potassium translocation was 3.0 at 0.5 to 2.0° C. Hartt (63) reported that iron was found accumulated in the root and stem in iron deficient sugar cane. The addition of potassium to the soil caused some of the accumulated iron to pass into the leaf blade. Ponnampeluma (119) reviewed that the accumulation of the iron in the nodes of corn plants was cleared up when heavy applications of potassium were made. A close association has

been observed between iron toxicity and potassium deficiency in rice, corn, sugar cane, and other members of the Gramineae (63, 87, 137). Ponnampcuma (119) commented on the relationship of potassium to iron translocation: "The ameliorative effect of potassium to which numerous investigators have drawn attention may be ascribed to the peculiar ability of potassium to mobilize the deposit of iron in the tissue."

Biddulph (11), who studied Fe^{55} distribution, showed that the translocation of iron was controlled by the prevailing status of pH and phosphorus. He indicated, first, that iron rapidly entered into and was distributed equally throughout the vein-mesophyll at a pH of 4.0 with medium phosphorus. Secondly, iron rapidly entered into the vein but with little or no distribution in the mesophyll at pH 7.0 with medium phosphorus. Thirdly, iron failed to enter the xylem and precipitated at the root surface at pH 7.0 with high phosphorus. This clearly shows the influence of the hydrogen ion and phosphorus concentrations in the substrate on the distribution and translocation of the iron. An experiment with Fe^{59} conducted on bush bean by Wallace (146) showed that the plant at pH 8.0, had a decrease in translocation of the iron from the vein to the mesophyll than at the lower pH. This is in agreement with Biddulph's finding

(11). In another experiment, iron was found to have precipitated in the conducting tissue at high pH, clogging conducting tissue (70).

Iron is a very important element in synthesizing several metallo-enzymes essential for biological systems. Nevertheless, the element may be inactivated by forming various organic salts. A great number of organic compounds may compete for iron; those organic compounds may be classified in four groups (121, 129):

(a) Hydroxymonocarboxylic acids, such as lactic acid ($\text{CH}_3\text{CHOHCOOH}$) and gluconic acid ($\text{CH}_2\text{OH}(\text{CHOH})_4\text{COOH}$),

(b) Dicarboxylic acids, such as oxalic (HOCCOOH), and malonic acid ($\text{HOOCCH}_2\text{COOH}$),

(c) Hydro^xdicarboxylic acid, such as tartaric acid ($\text{HOOC}(\text{CHOH})_2\text{COOH}$), and citric acid ($\text{HOOCCH}_2\text{COHCOOHCH}_2\text{COOH}$),

(d) Amino acids, which are also hydroxy or dicarboxylic acids, such as glutamic acid ($\text{HOOC}(\text{CH}_2)_2\text{CH}(\text{NH}_2)\text{COOH}$), Aspartic acid ($\text{HOOCCH}_2\text{CH}(\text{NH}_2)\text{COOH}$), and cysteine ($\text{HSCH}_2\text{CH}(\text{NH}_2)\text{COOH}$).

These hydroxyl and carboxyl groups react with iron to produce associated organic iron compounds. These inactivated iron compounds most probably would be found at the organic compound synthesizing sites of the leaves (42, 94, 129).

Due to the many difficulties that iron may encounter during its translocation and in the metabolically active sites, the active iron which actually participates in metabolic work may possibly be only a small portion of the total iron content of the plant.

Assimilation of Iron in Plants

Concepts of iron assimilations in plants

Iron problems are usually associated with chlorophyll formation in the plant. However, iron is not involved in the chlorophyll molecule, but it does function as a prosthetic group to a large number of enzymes in catalyzing chlorophyll synthesis (36, 38, 55). Furthermore, numerous physiological functions are performed by iron through its enzymatic activities.

Enzymes having heme iron (iron porphyrin complexes) as prosthetic groups are cytochromes, catalase, and peroxidase. The cytochromes act as mediations in cellular respiration (17, 38). In the electron transfer system, cytochromes carry oxygen to react with the hydrogen ion to form water and gives rise to ATP by oxidative phosphorylation. The cytochrome b_6 and cytochrome f are two enzymatic prosthetic groups which reside in the plastids of leaves and are primarily involved in photosynthesis. They do not act in respiratory processes as

do other groups of cytochromes (55, 126). Enzymes having non-heme iron are succinic dehydrogenase and aconitase (38, 130).

With ATP, the phosphorylation of amino acids can then proceed in making the protein portion of chloroplasts, enzymes, and other structural proteins (103). In this way iron is involved in protein synthesis. Pesur et al. (115) stressed that iron is also essential for initiating synthesis of chloroplastic protein.

The components of the porphyrin structure, the skeleton of iron-porphyrin complex, and the magnesium-porphyrin complex, are four pyrrole rings. The carbon and nitrogen constituents are from glycine and succinate (38). In the presence of sufficient iron, the coproporphyrinogen will be metabolized into protoporphyrinogen which can convert the coproporphyrin to a porphyrin structure. Carell and Price (36) used δ -amino levulinic acid to substitute for glycine and succinate as raw material and did synthesize the δ -amino levalinic acid to porphyrin in the presence of iron. The synthesis of a porphyrin molecule needs high energy, such as found in high energy phosphate bonds (17, 36, 99).

/ Iron oxidation states have a very significant influence on some physiological differentiation. Brown et al. (26) pointed out that during cell division of the

root tip, iron can either promote or prohibit root growth depending on the assimilated iron oxidation form involved. He indicated again that if Fe^{2+} had oxidized to Fe^{3+} , or simply by removing the less stable Fe^{2+} by means of an external chelating agent, the cell division will be hampered.

The relationship between iron and chlorophyll synthesis is not fully understood yet (24, 104, 139), but, is none-the-less a very interesting field. In studying the green color of plant leaves, it was learned that chlorophyll pigments occur as lipoprotein complexes with chloroplast. In fact, 30 to 40 percent of the total nitrogen of an oat plant is found in the chloroplasts (155). Chlorophyll-a is common to all plants and it contains about 10 percent of the total plant magnesium. The angiosperm seedling contains very small amounts of protochlorophyll in the darkness. However, when placed under illumination there is a conversion of chlorophyll-a from protochlorophyll in a few minutes (38, 155). This reaction is a type of photochemical reaction which can be completed even under relatively low light intensity. The difference between protochlorophyll and chlorophyll is that the former lacks two hydrogen atoms in one of its pyrrole rings (14). The gymnosperm seedling has its protochlorophyll converted to chlorophyll-a through enzymatic reaction

and therefore this reaction can be completed with no light (155). Bogorad (14) observed that the conversion of protochlorophyll to chlorophyll is not a chemical process either in light or darkness, since his value for Q_{10} in excised barley seedlings showed that in the dark at temperatures of 11.0°C. to 23.5°C. , Q_{10} was 1.69; and above 23.5°C. , Q_{10} was much smaller. In wheat, Q_{10} was less than 1.0 when temperature was raising from 25.0° to 35.0°C. (14). At higher temperatures, chlorophyll production usually declined. It was suggested that a higher temperature resulted in the destruction of the protochlorophyll or converted the protochlorophyll to new colorless compounds (14, 155).

Some direct correlations between total iron content and chlorophyll content in the plants have been found (80). Price and Carell (116) indicated that in Euglena the chlorophyll is linearly dependent on the iron content of the cells. No correlation was reported between total iron content and that of chlorophyll in the sunflower plants (80, 81) and others (35, 60, 80, 113, 126). Jacobson (80) reviewed more or less the same amounts of iron in chlorotic leaves as in green leaves. However, scientists have devised a workable term, active iron, which is based on the availability of iron rather than the total amount of iron in a plant (80, 113).

Active iron is that iron related to the concentration of chlorophyll (80). Oserkowsky (113) defined active iron as the iron extracted by 1.0 N HNO_3 . Active iron is present in relatively small amounts. For example, 0.3 to 0.5 mg. iron in 1.0 gm. fresh tissue was enough to keep pineapple plants greenish, because the iron was localized in the chloroplasts (126). The chloroplast also contains acid soluble iron, which is not active iron (127).

The non-chloroplast iron and chloroplast-acid-soluble iron of the cells are both inactive iron. They remain approximately constant as long as iron is a limiting factor. When iron is supplied liberally it accumulates more rapidly in the non-chloroplast fraction of the cell than in the chloroplasts (80, 81). Active iron can be transformed into residual iron, from which state it can not be utilized for chlorophyll formation (26). Pesur et al. (115) and others (77, 80) suggested that active iron was in organic combinations in the chloroplasts, and that it did affect the chloroplastic protein synthesis. Active iron, in the Fe^{2+} form (26, 140), may be oxidized into Fe^{3+} and then inactivated in protein combinations, followed by a disturbance in the protective protein-chlorophyll combination (140).

The failure of the assimilation of iron in plants leads to a number of disorders, such as, inactivation of iron porphyrin enzymes (36, 154) and iron chlorosis (90, 116, 154) on the one hand; iron toxicity (70, 87, 119) on the other.

Among the iron porphyrin enzymes, cytochromes c, b₆, and f are related to the chlorophyll synthesis, that is, cytochrome c acts in the electron transport system of respiratory cycle which provides energy for the synthesis of protein-chlorophyll combination (55, 155). Cytochromes b₆ and f are vital in the electron transport system of photophosphorylation which is also related to chlorophyll formation as proposed by Evan (55). He discussed cytochrome b₆ and f in using the following photophosphorylation scheme:

He suggested that the photolysis of water produce bound oxygen and bound hydrogen atoms. Two hydrogen will go through the electron transport system, F.H.N., Vit. K, ascorbate, and cytochromes to meet the bound oxygen and form a molecule of water. If a cytochrome, such as cytochrome f, is insufficient, this process would not be completed and the bound oxygen atoms would remain with the chloroplast. The chloroplasts, and a limited amount of cytochrome f, would be oxidized and finally destroyed, if there were insufficient available iron. Evan (55) pointed out that it seems highly probable that cytochrome f is the major constituent influenced by iron deficiency, because it has the highest concentration, 10^{-5} M, among the cytochromes group in normal plants.

Ponnampeuma (119) has reviewed a great many cases of iron toxicity in plants. Rice in culture solution with iron concentrations greater than 50 ppm had the growth and number of tillers reduced. The Hungary browning disease of rice is associated with an excess of reduced iron. The nodal discoloration in young corn plants was due to clogging of conducting tissue with ferrous iron. The disease "kalimati" in sugar cane is believed to be due to ferrous iron toxicity. These and other physiological diseases

induced by iron toxicity can be cured by an appropriate addition of potassium fertilizer. They may just as well be called potassium deficiency diseases.

Factors affecting assimilation of iron in plants

Potassium occurs mainly in ionic form in the cell vacuoles and, generally, it is the most abundant cation in the cell (105). Therefore, numerous phenomenon performed in the cell are associated with the potassium ion through enzymatic reactions (150). Lawton and Cook (93) pointed out that a number of enzymatic effects involving the transfer of the phosphate group of the phosphate pyruvate to the adenylic system involves potassium. This adenylic system involves high energy phosphate bond formations which act on the synthesis of proteins and porphyrins. Lawton and Cook (93) stated that potassium affects photosynthesis and other physiological processes indirectly through its influence on chlorophyll components. Often the reduction of nitrate takes place anywhere iron exists and the iron will most probably suffer a decrease in solubility following the reduction of nitrate (119). Potassium favors nitrate reduction (132). Therefore, a high concentration of nitrates accompanying a high concentration of potassium in the plant system means that the

assimilation of iron becomes a problem (132). Bollard (15) stated that an accumulation of ferric iron would result from a reduction of nitrates in plants. Lawton and Cook (93) stated that potassium aided in nitrate reduction for the synthesis of protein particularly in meristemic tissues. There have been observations that ammonia-nitrogen resulted in the formation of greater quantities of chlorophyll than nitrate-nitrogen (124, 126). Jones (86) found traces of chlorosis in potted gardenia at soil temperatures of 22° to 20° C., distinctive chlorosis at 18° C., and increasingly severe chlorosis at still lower temperatures. He found no deficiency in any nutrient elements, and even greater concentrations of some elements in the chlorotic leaves. Davidson (45) cured foliar chlorosis of gardenia induced by low soil temperatures of 15° C. or 20° C., by increasing light intensity, length of day, as well as, air temperature. During cold nights, nitrate reduction is at a minimum level of activity, while in the daytime, conditions will favor highest levels of activity of the nitrate reductase (109). This may result in the inactivation of iron assimilation (15).

There seems to be some uncertainty as to whether there is a greater total organic acid content in chlorotic leaves of plants or in non-chlorotic leaves.

Elliott (53) reported greater concentration of aliphatic acids in non-chlorotic barley shoots than chlorotic shoots. But Iljin (76) and others (131, 133) found a large amount of organic acid in chlorotic leaves of grapes and other plants. A greater concentration of total organic acids in non-chlorotic leaves than in chlorotic leaves would indicate both good translocation and good assimilation of iron, while a greater concentration of organic acids in chlorotic leaves could indicate the reverse.

Shear et al. (124) found that excess oxalic acid in the leaves of tung tree influenced iron utilization. This oxalic acid production was highly related to nitrate-nitrogen fertilization (59, 100) and could be counteracted by the application of calcium (59). Citric acid is usually higher in concentration in chlorotic plants than non-chlorotic plants. Iljin found this true in grape trees (76, 77), Brown et al. (28) in soybeans, and McGeorge (100) in other plants. The presence of citric acid affects iron assimilation in some ways which are not yet known. The research on soybean and other plants (80, 133, 134) indicated that chlorotic leaves contained higher amounts of citric acid than that of the non-chlorotic. The production of citric acid in the fruit was highly related to potassium

fertilization (39, 82). In the guayule plant, high citric acid was associated with high potassium fertilization (39, 40). Malic acid is usually in higher concentration in chlorotic plants than in non-chlorotic. Elliott (53) reported that of the acids found in chlorotic barley shoots, malic acid was 370 mg./gm. of fresh weight, while citric acid and succinic acids were 35 mg./gm. and 5 mg./gm., respectively.

Burström (34) reviewed the function of malic acid in the metabolism of cations, and reported that it acts as a malate ion:

If an excess of cations is added to such a system, the reaction goes to the right, and then an amount of malate equivalent to the cation excess is withdrawn from the respiration cycle. Potassium malate probably is the common potassium organic salt in cell. Iron malate can also be formed in the cell and the iron can not be freed from the salt by substitution of zinc and manganese (95). The amount of malic acid increases with increases in bicarbonate ions and simultaneously the amount of iron decreases.

Brown (28) found that chlorotic soybean (PI 54619-5-1) was higher in aspartic acid and asparagine than other varieties. Noggle (110) noticed that chlor-

otic plants were higher in glutamine, lysine, aspartic acid, and asparagine, but non-chlorotic plants were higher in alanine, glutamic acid, and glycine. Another report (28) stated that chlorotic bean plants were high in aspartic acid and glutamic acid. It is quite certain that the dicarboxylic amino acids and their derivatives affect iron assimilation to a noticeable extent.

The concentration of reducing sugar is usually high in photosynthetic sites of the leaves (28, 30, 40). The amount of reducing sugar in leaves was somewhat proportional to iron assimilation as reported by Brown (28). He found that the chlorotic soybean contained slightly less reducing sugar than non-chlorotic plants. An experiment (127) on grass carried out in culture solution with high iron content resulted in greater amount of sugar in plants than did a non-iron culture solution. Both suggested that iron and reducing sugar were directly related.

MATERIALS AND METHODS

In attempts to determine the effects of night temperature and potassium fertilization on the absorption, translocation, and assimilation of iron in centipedegrass, a series of five greenhouse-cold chamber experiments were conducted from 1963 to 1967. Descriptions of the experiments and of the laboratory analyses of soils and plant materials therefrom are given in the following sections.

Greenhouse-Cold Chamber Experiments

Experiment 1.--Cores of slightly chlorotic centipedegrass growing on Arredondo loamy fine sand were transferred from turf plots at the University of Florida Horticultural Unit to 11 cm. diameter plastic pots in the greenhouse. After a three-week period of acclimation, the grass was clipped to a uniform height of 1 inch, and the 800 gm. of soil in each pot was brought to a uniform moisture content of approximately 80 percent of field capacity. Some physical and chemical properties of the Arredondo soil used in this experiment are given in Table 1.

The treatments consisted of three rates each of potassium sulfate and potassium chloride and three

night temperatures. All treatments were replicated three times. The potassium rates were equivalent to 2, 5, and 8 pounds of potassium per 1,000 square feet. Other nutrients were uniformly applied in amounts equivalent to 8 pounds of nitrogen as calcium nitrate, 2 pounds of phosphorus as monocalcium phosphate, and 0.1 pound of iron as monosodium ferric diethylene-triamine-pentaacetate (NaFeDTPA) per 1,000 square feet. The grass was grown in the greenhouse during daylight hours at daily maximum temperatures ranging from 26.5 to 36.5° C. At night, one-third of the pots was placed in a cold chamber maintained at 1.6° C., another third was transferred to a chamber maintained at 7.2° C., and the remaining pots were left in the greenhouse proper, where the night temperature ranged from 15.5 to 21.0° C. Grasses grown at the two low night temperatures were placed in chambers at 8:00 P.M. and removed at 7:00 A.M. each day. In an attempt to maintain uniform humidity conditions for all treatments, pots were encased in plastic bags during the night.

Grasses were watered as needed and grown for eight weeks. At this time, the above-ground portion of the plant was harvested. The vegetative materials were dried for 48 hours at 70° C., weighed, and analyzed for iron, potassium, calcium, magnesium, and phosphorus,

Table 1.--Some physical and chemical properties of
Arredondo loamy fine sand.

Bulk density gm/cc	Moisture equivalent %	1 N NH_4OAc (pH 4.8) extractable nutrients				pH O.M. C.E.C.	
		Ca ppm	N ppm	P ppm	% me/100 gm.		
1.55	8.8	463	54	31	19	5.7	1.53 4.22

using methods described in Laboratory Analyses.

Experiment 2.--This factorial experiment, consisting of two levels of potassium (0 and $23\frac{1}{4}$ ppm) and two night temperatures (5° and $20-25^{\circ}$ C.), with six replications, was conducted on centipedegrass in acid-washed sand. Ten plugs of grass were planted in 1,000 ml. beakers containing 800 gm. of dry sand, after the soil had been removed by washing. The beakers were wrapped with aluminum foil, 300 ml. of Hoagland solution (67) added, and the grass allowed to acclimate in the greenhouse for 3 weeks. The nutrient solutions, containing the two rates of potassium, were added to the beakers at 50 mls. per day so as to sustain growth of grass. After the grass was established, 50 ml. aliquots of iron solution containing $2.575 \mu\text{c}$ of Fe^{59} in FeCl_3 were applied to the beakers. Minor elements were added uniformly to both potassium nutrient solutions, at rates suggested by Hoagland (67). Iron was added as the chelate, NaFeDTPA .

The nutrient contents of the two solutions--with and without potassium--are given in Table 2.

To obtain the night temperature treatments, one-half of the beakers were transferred to a cold chamber, maintained at 5° C., at 8:00 P.M. and removed at 7:00 A.M. the next day. The remaining beakers were left in

the greenhouse at night, where the temperature varied from 20 to 25° C. All pots were kept in the greenhouse during daylight hours where daily maximum temperatures varied between 35.0 and 42.0° C.

Table 2.--The composition of Hoagland nutrient solutions with and without potassium.

Component	Solution	Solution
	with Potassium ml./l.	without Potassium ml./l.
<u>M</u> MgSO_4	2	2
<u>M</u> $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$	5	5
0.05 <u>M</u> $\text{Ca}(\text{H}_2\text{PO}_4)_2 \cdot \text{H}_2\text{O}$	--	10
<u>M</u> KNO_3	5	--
<u>M</u> KH_2PO_4	1	--

The plants were harvested two replicates at a time on July 6, July 18, and July 30, 1964, after 12, 24, and 36 days of treatment, respectively. At harvest, the entire plant plus sand was dumped onto a screen-covered pail and the Fe^{59} -contaminated sand washed from the roots with a fine stream of distilled water.

Experiments 3, 4, and 5.--Three uniform experiments similar to Experiment 1 were conducted in the greenhouse and cold chambers; one in the spring

of 1964, one in the summer of 1964, and the third in the winter of 1964. The treatments used in each of the three experiments were the same, except that the daytime temperatures and the night temperatures in the greenhouse varied from season to season.

In these experiments, as in Experiment 1, centipede grass growing in Arredondo loamy fine sand at the Horticultural Unit (Table 1) was transplanted to 11 cm. plastic pots. Three weeks after transplanting, the grass was treated with the equivalent of 2, 5, and 8 pounds of potassium per 1,000 square feet, from potassium chloride. At each level of potassium, two rates of iron equivalent to 0 and 0.1 pound per 1,000 square feet were added as the chelate NaFeDTPA.

The day and nighttime temperatures prevailing inside the greenhouse during the period of the experiments are given in Table 3.

The temperatures inside the two cold chambers were maintained at -1.1°C . or 7.2°C .

After two months of growth, whole plants were removed from pots, soil washed from the roots and the plants separated into roots, stems, and leaves. The plant materials were dried, weighed, and analyzed for inorganic constituents; fresh plant

materials were used for organic constituents, both as outlined below.

Table 3.
Average Maximum and Minimum greenhouse temperatures

Season	Ave. night temperature		Ave. day temperature	
	max. °C.	min. °C.	max. °C.	min. °C.
Spring, 1964	21.0	18.0	39.7	21.0
Summer, 1964	26.5	22.0	48.0	26.5
Winter, 1964	17.7	16.0	28.6	17.7

Laboratory Analyses

Soil used in experiments 1, 3, 4, and 5 was Arredondo loamy fine sand, a phosphatic soil. Some of the chemical properties of this soil before treatment were given in Table 1. Soil samples were taken from individual pots at the termination of the experiments and used for determination of moisture content and for chemical analyses. Soil pH was measured with a glass electrode-potentiometer at a soil-water ratio of 1:2. Available nutrients were extracted by shaking 5 gm. of soil for 30 minutes in 25 ml. of N ammonium acetate solution buffered at pH 4.8. Calcium and potassium in the extract were determined by flame at 622 and 670 m μ , respectively, using a Beckman model B flame spectro-photometer with an oxygen-acetylene burner

assembly. Phosphorus was determined by a modified ammonium molybdate-stannous chloride procedure (143).

Exchangeable ferrous iron was extracted by a modification of the procedure described by Jackson (78). In this modified procedure, a 25 gm. sample of fresh soil was placed in a 500 ml. flask, shaken for 2 minutes, with 200 ml. of N ammonium acetate buffered at pH 6.0, and filtered into a 400 ml. beaker. A second extraction was made with 50 ml. of the solution. The filtrates were combined and evaporated to dryness. An aliquot of 20 ml. of aqua regia was added and the solution was again brought to dryness. The residue was placed in a muffle furnace and ignited at 500° C. for 15 minutes. After cooling, 3 ml. of concentrated hydrochloric acid were added, the contents were heated until colorless and then brought up to 100 ml. with 0.1 N hydrochloric acid. A 5 ml. aliquot was employed for the iron determination by the o-phenanthroline method (85).

The combined exchangeable ferrous and ferric iron and dilute acid soluble iron in soil were extracted by N ammonium acetate at pH 3.0 as described by Jackson (78). Iron was determined by the o-phenanthroline method (85).

Nitrates were determined by the nitrophenoldisulfonic acid colorimetric method (78) and read on the

Bausch and Lomb Spectronic 20 colorimeter at 410 $m\mu$ wavelength.

Plant samples analyzed for inorganic constituents were dried and ground in a Wiley mill to pass a 20-mesh sieve. One gram of dry plant material was ashed in a muffle furnace at 500° C. The ashed material was digested in 20 ml. of 40 percent hydrochloric acid. After evaporating to dryness on a hot plate, organic matter was removed by heating further with 2 ml. of concentrated nitric acid. The ash was taken up by 0.1 N hydrochloric acid to 100 ml. Aliquots were used for determinations of inorganic elements. Calcium was determined by a turbidimetric method described by Peach and English (114). Magnesium was determined by a colorimetric method provided by Mehlich (102). Potassium and phosphorus were determined on the spectrophotometer in the same manner as used for soil samples. Iron determinations were made by the o-phenanthroline method (85). Separate samples were used for total nitrogen determinations by the Kjeldahl-Gunning procedure (4).

The determination of Fe⁵⁹ in the plant was made by dry-ashing 1 gm. oven-dry plant material. The ashed material was taken up in 10 ml. of 0.1 N hydrochloric acid. Duplicated 0.5 ml. aliquots of this solution were pipetted into aluminum planchets, and dried under an

infrared light, counted on a Geiger-Müller counter and recorded as counts per minute per gm. of dry material.

Other amino acid and organic acid determinations were made on fresh plant materials. The fresh tissues were washed clean with distilled water and excess moisture blotted off. Tissues were separated into leaves, stolons, and roots and for the extraction of organic acids 10 gm. of leaves, 6 gm. of stolons, and 12 gm. of root tissue were macerated for 7 minutes in a Waring blender with 200, 150, and 100 ml., respectively, of boiling 85 percent ethyl alcohol. The material was filtered through double cheese cloth into a 400 ml. beaker and the residues remacerated twice with 50 ml. of hot 85 percent ethyl alcohol and added to the first filtrate. The combined filtrate was evaporated under the hood to about 25 ml. and then centrifuged at a speed of 12,000 rpm for 10 minutes. The pellets were centrifuged with 20 ml. of distilled water. The centrifugate was again reduced to about 25 ml. The reduced centrifugate was poured into cation exchange columns, 12 in. long by 0.5 in. in diameter and containing 5 gm. of Dowex 50-X8 resin with a small pad of glass wool at the bottom. The column was washed with 100 ml. of distilled water. The eluate containing organic acids minus amino acids was collected and evaporated to about 25 ml. Hereinafter, amino acids

will be reported and discussed separately from other organic acids. The amino acids which adsorbed on the cation exchange resin were leached out of the column with 100 ml. of 2N ammonium hydroxide and the eluate evaporated to dryness. The dried amino acids sample was taken up by 1 ml. of 10 percent isopropanol.

The organic acid eluate after being reduced to 25 ml. volume was poured into an anion exchange column containing 3 gm. Dowex 1-X8 resin which had been converted to the formate form. The column was then washed with 100 ml. of distilled water. All washing solutions were discarded. The organic acids absorbed on the anion exchange resin were leached out with 50 ml. of 50 percent formic acid. The eluate was dried and taken up with 1 ml. of 10 percent isopropanol.

Amino acids were spotted on Whatman No. 1 filter paper. The chromatogram was developed according to Block et al. (13) and Laurence et al. (92). The procedure was slightly modified so that 5, 5, and 10 microliters of leaves, stolons, and roots amino acid extracts, respectively, were spotted on paper strips. The strips were run through 75 percent phenol for 15 hours, then dried under a hood for at least 36 hours before dipping into 0.1 percent ninhydrin isopropanol solution. The over-spotting technique (13) was employed

to identify individual amino acid. The spots were cut out and dissolved in 3 ml. of 50 percent ethyl alcohol. After 30 minutes the color of the solution was measured with the Spectronic 20 colorimeter at $560\text{ m}\mu$. A similar-size area from a region of the paper which has been dipped in ninhydrin but which had not been traversed by amino acids was cut out to serve as blank.

Organic acid extracts were chromatographed in about the same manner as the amino acid. However, there were differences in sample size for spots and in the solvent and color developer. The spotting sizes for leaves, stolons, and roots were 15, 15, and 30 microliters, respectively. The solvent was a mixture of butanol, formic acid, and water in a ratio of 12:2:15 (13). The color developer was prepared from 1 gm. xylose, 1 ml. aniline and 3 ml. water diluted to 100 ml. with isopropanol (13). The colored spots were dissolved with 3 ml. of 50 percent ethyl alcohol and the color intensity was measured at $395\text{ m}\mu$ on the Spectronic 20.

Free **sugars** were extracted from 1 gm. of dry and finely ground plant material, according to the method described by Traub and Slattery (142). The reducing sugar content of monosaccharides of the extract was determined by Brown's (30) modification of the Smogyi's

semi-micro copper method (30). The determination of cold water soluble carbohydrates was also made using the hydrolyzate from the previous extraction and using the same procedure as that used for reducing sugars.

The ferric cyanide reduction method described by Brown et al. (28) was used to determine the reductive capacity of centipede grass roots. In this procedure, 75 ml. of aerated 10^{-4} M $K_3Fe(CN)_6$ and 5 ml. of 10^{-6} M $FeCl_3$ were placed in a 125 ml. flask. Ten pieces of grass roots were inserted in the solution. The degree of reduction of the mixtures was determined on the colorimeter at $400\text{ m}\mu$ every 12 hours. Data were recorded as optical density change per 12 hours per gm. of dry material.

Root cation exchange capacity was determined by the method described by Drake. et al. (50) in which fresh grass roots were dialyzed for 72 hours. They were thoroughly rinsed with distilled water and then centrifuged at 2,000 rpm for 5 minutes to provide the roots with approximately equal moisture content. A 3 gm. sample of the centrifuged roots was mixed with 150 ml. of KCl in a beaker for 30 minutes. The roots-KCl system was titrated with 0.05 M KOH to pH 7.0, as measured by a glass electrode. The weights of roots were determined after drying for 48 hours at 90°C . in the oven.

RESULTS AND DISCUSSION

Experiment 1

Neither sources nor rates of potassium fertilizer had significant effect on the growth of centipede-grasses, as reflected by yields shown in Table 23. The yield of grass grown at the prevailing night temperatures in the greenhouse ($15.5 - 21.0^{\circ}$ C.) averaged from 25 to 40 percent greater than grass kept in the cold chamber at night at the two lower temperatures. However, there was no significant difference between yield of grass maintained at 1.6° C. and that of grass maintained at 7.2° C. during night hours. It should be pointed out that no chlorosis was observed regardless of treatment probably because iron chelate was added uniformly to all pots. However, leaves of grass grown at the low night temperatures were short along the mid-rib and exhibited a purpling of the margins.

The concentration of iron (Table 21) in grass grown at two low night temperatures (1.6° C. and 7.2° C.) was greater than in grass kept in the greenhouse continuously, as shown in Figure 1. Increasing rates of potassium sulfate did not affect iron absorption, except at the lowest night temperature, 1.6° C., but

Figure 1. The Effects of Night Temperatures and Potassium Sources on the Concentration and Total Absorption of Fe and K in Centipedegrass.

increasing rates of potassium chloride generally reduced iron absorption at all three night temperatures (Table 21). It therefore appears that the anion components of the potassium salts played a more important role in iron absorption than the potassium itself, and that the chloride ion influenced iron uptake more than did the sulfate ion, at the three levels of potassium used (Figure 2).

Total amount of iron absorbed by the grass was not affected by the three night temperatures (Figure 1) as much as the concentration of iron in the plant. However, total iron content had a tendency to decrease with increases in potassium levels (Figure 2). There was a significantly greater uptake of iron where the chloride was used than sulfate (Figure 2).

Percentage potassium in the grass was increased with increase in the rate of potassium fertilizer, as shown in Figure 2. The response to potassium sulfate appeared to be better than that to potassium chloride, at all three levels. Nevertheless, increases in night temperature (Figure 1) increased the concentration of potassium in the plant, regardless of source. The potassium chloride source resulted in an almost linear increase, while the potassium sulfate source resulted in a decrease at the intermediate night temperature and

Figure 2. Iron and Potassium in Centipedegrass as Affected by Three Rates and Two Sources of Potassium.

then an increase at the highest temperature (15.5 - 21.0° C.).

The total potassium uptake by the plant generally increased with added potassium, but the effect is greater with potassium chloride than where potassium sulfate was used (Figure 2). Although potassium chloride was taken up fairly well in the entire temperature range from 1.6° C. to 15.5 - 21.0° C., potassium sulfate absorption was decreased between 1.6° C. and 7.2° C., but then absorption increased rapidly with increases in temperatures from 7.2° C. to 15.5 - 21.0° C.

An inverse relationship was observed between the absorption of potassium and the absorption of iron. Particularly noteworthy was the effect of potassium on iron absorption when applied as potassium chloride. This doubtlessly resulted from the fact that potassium chloride was absorbed more readily by centipedegrass than potassium sulfate, when these two potassium sources were compared at the higher rates of application.

Experiment 2

In this sand culture experiment using Fe⁵⁹, the yields of centipedegrass grown at the high night temperature (15.5 - 22.5° C.) were significantly

greater than yields at a night temperature of 4.5° C. (Table 23). This was particularly true of the yield of above-ground portions, or tops, as shown in Figure 3. The potassium-containing nutrient solution produced greater growth of both roots and tops than the solution without potassium. The greatest yield of tops from this nutrient was obtained at the higher night temperature (Figure 3). The general appearance, or color, of the grass changed from a slight yellow-green at the beginning of the experiment, to a very slight yellow-green at the first and second harvests, to a full green color at the end of 36 days.

The concentration of iron, as expressed by Fe^{59} in counts per minute per gm. of oven dry material, is shown in Table 24. The statistical analysis showed that there were highly significant differences in the concentrations of iron between that in the roots and that in the tops of plants, and that differences in iron concentrations were related to rates of potassium (Table 26). That is, the potassium in the nutrient solution appeared to decrease the percent of iron in the plant. The magnitude of the decrease is shown in Figure 4.

The concentration of iron in the roots, which contained three to four times more Fe^{59} than

Figure 3. Effects of Night Temperatures and Rates of Potassium on Yields of Centipede grass.

the tops, was greatly affected by the use of potassium in the nutrient solution. The concentration of iron in the grass was generally less at the higher night temperatures, and the effect of potassium on iron content was greater at these temperatures than at the lower temperature. On the other hand, there was little difference in iron content of tops of plants grown at the two night temperatures. The potassium addition had no effect on the content of iron at the $15.5 - 22.5^{\circ}$ C. night temperature, and only slightly reduced the iron content at the low night temperature. It seemed therefore, that potassium decreased the iron uptake in the roots through some mechanisms of competition for absorbing sites of the roots. Once the iron, or iron chelate molecule, entered the root it was apparently no longer affected by the presence of potassium at the higher night temperatures, and it was only slightly affected at the low night temperature (4.5° C.).

Total iron content, a product of concentration and yield, in the tops of the plants was decreased with the application of potassium at 4.5° C., but increased at high night temperatures ($15.5 - 22.5^{\circ}$ C.). However, the reverse was true for the roots (Figure 4). This suggests that at the 4.5° C. the potassium application slightly increased the growth of roots (Figure 3) and

CONCENTRATION IN PLANT

TOTAL ABSORPTION

Figure 4. Effects of Night Temperatures and Rates of Potassium on Concentration and Total Absorption of Iron in Centipede grass.

increased the total uptake of iron by the roots; but this iron was not readily translocated to the top of the plant. At high night temperatures roots continuously took up iron, due to increased root activity, and the iron was rapidly translocated to the tops, as a result by the greater growth of that portion of plant. Furthermore, as indicated by the three consecutive harvests, the total iron content of tops of the plants progressively increased at both temperatures, with or without potassium (Figure 5). This increase in iron may be at the expense of the iron content of the roots, as indicated in Figure 5.

The percentage of potassium in the grass was highly significantly influenced by the application of potassium to the solution. Moreover, the concentration of potassium in the tops was greater than that in the roots (Table 25). Total potassium uptake in the tops rapidly increased during the three consecutive growth periods, just as total iron content increased in that portion of the plant.

The following facts may be gathered from the results of this experiment. (a) High night temperatures (15.5 - 22.5° C.), or the application of potassium, increased the yield of tops and roots of centipedegrass, but affected the tops of the plants more than the roots.

Figure 5. Total Iron (Fe^{59}) and Potassium Contained in Tops and Roots of Centipedegrass at Three Growth Periods.

(b) The potassium application decreased the concentration of iron in the roots, particularly at high night temperatures (15.5 - 22.5° C.). (c) The potassium application decreased the total iron content of the tops of the plants at low night temperature (4.5° C.) but increased at higher night temperature. The total iron content in the roots was decreased at the higher night temperatures and increased at low night temperature. (d) Total iron content and total potassium content in the tops of the plants increased as a grass matured, while the total iron and total potassium of the roots decreased with the progressive growths.

Experiments 3, 4, and 5

Experiments 3, 4, and 5 were similar in all respects, except that they were conducted in three different seasons, and for this reason their results are presented and discussed together. The temperatures for the three seasons, expressed as the averaged maximum temperature for daylight hours, are shown in Table 3. While the night temperature in the greenhouse varied with seasons, the two cold-chamber night temperatures were maintained at -1.1° C. and 7.2° C. for all three seasons.

Plant Growth

Plant appearances developed during the progress of these experiments were significantly related to treatments. In general, plants that received iron chelate, NaFeDTPA, varied from very slightly yellowish-green at night temperature of -1.1° C. to dark green for those at 7.2° C. and $18.0 - 22.5^{\circ}$ C., as shown in Figure 6. Figure 7 shows representative pots of the series of treatments in which no iron chelate was applied to the soils. The grass appeared green in the high temperatures, but the three potassium treatments which were kept at -1.1° C. during the night had a definite yellowish-green appearance. The application

Fig. 6.--Effects of Night Temperatures and Rates of Potassium in the presence of Added Iron on the Appearance of Centipedegrass.

Note: Night temperature: $T_1 = -1.1^{\circ} \text{ C.}$, $T_2 = 7.2^{\circ} \text{ C.}$,
 $T_3 = 18.0 - 25.0^{\circ} \text{ C.}$
 Potassium rates: $K_1 = 2 \text{ lbs.}$, $K_2 = 5 \text{ lbs.}$,
 $K_3 = 8 \text{ lbs.}$, per 1000 sq. ft.
 Iron rates: $\text{Fe}_1 = 0.1 \text{ lb.}$ of iron, as a
 chelate, per 1000 sq. ft.

Fig. 7.--Effects of Night Temperatures and Rates of Potassium in the absence of Added Iron on the Appearance of Centipedegrass.

Note: For legend see Fig. 6.

of potassium fertilizer slightly intensified the development of the yellow color.

The leaves of centipedegrass which received the different treatments varied in size, as shown in Figure 8. Leaves in this photograph were taken from the third leaf position back from the apex of a random sample from each treatment. At a night temperature of -1.1° C. the leaves were all smaller than those at the higher night temperatures. Leaves that received an application of iron (Fe_1) were generally larger than leaves without additional iron (Fe_0).

An examination into the morphology of the roots reveals some differences due to treatments. That is, roots grown at low temperatures were generally white and soft, while roots grown at higher temperatures were darker in color, dense and very profused, as shown in Figure 9. This picture further illustrates that when comparing each pair in each row, the left one which received no additional iron (Fe_0), grew larger and more twisted than the mate that received the iron treatment (Fe_1). On the other hand, potassium applications did not appear to affect the morphology of the roots.

Plant yield data in the three experiments are shown in Table 27, for the leaves, stolons, roots, and entire plant, respectively. All yields were highly

Fig. 8.--Effects of Night Temperatures, Rates of Potassium and Iron on leaves of Centipede grass.

Note: For legend see Fig. 6.

Fig. 9.--Effects of Night Temperatures,
Rates of Potassium and Iron on Roots of Centipede-
grass.

Note: For legend see Fig. 6.

significantly influenced by temperatures, except for the leaves which were significantly affected (Table 4). The effect of night temperatures on yields are graphically presented in Figure 10. Total yields of the plant increased almost linearly as night temperatures increased. When the grasses were divided into component parts, the roots and stolons were found to exhibit trends in response to night temperature similar to that of the entire plant. However, leaf growth did not increase at temperatures above 7.2°C . One might speculate, therefore, that plant growth in response to a warm spell, when night temperatures are between 7.2°C . to $18.0\text{--}25.0^{\circ}\text{C}$., is composed of two responses, that of the stolons and the roots, while from -1.1°C . to 7.2°C ., plant growth is a combination of responses of leaves, stolons, and roots. These findings are somewhat different from the findings in the sand culture experiment in which leaf growth responded to warm night temperatures more than the roots. The yields of stolons as well as the yields of the entire plant were significantly different among the seasons as shown graphically in Figure 10. In general, the yields associated with the summer, or highest average daylight temperatures, and those associated with the winter, or the lowest temperatures, were about 20 to 30 percent less than

Table 4.--Analyses of variance of the effects of treatments on the yields and inorganic constituents in centipedegrass of experiments 4 and 5.

Source of variation	df.	Yield				df.	Iron			
		L		R			L		S	
		E		E			X102		R	
Night temp.	A 2	6.42*	23.51**	24.38**	139.99**	2	113.35**	79.74*	62.38**	288.25**
Sea. day										
max. temp.	B 2	2.55	1.26*	.46	10.46*	1	585.63**	396.67**	484.09**	1035.76**
AB	4	2.81	.61	.23	6.48	2	8.77	10.36	7.30	30.11
Potassium	C 2	2.47	.08	.75	1.31	2	21.93*	12.25	15.14	58.72*
AC	4	2.23	.82	.27	5.17	4	13.85	10.39	11.07	81.82*
BC	4	.28	.12	.83	1.24	2	6.04	4.14	16.97	8.28
Iron	D 1	.08	.28	.93	.73	1	224.00**	88.67*	22.40	1059.05**
AD	2	.79	.57	2.26**	1.26	2	132.59**	76.19*	10.99	614.47**
BD	2	1.27	.97	.21	1.93	1	.93	16.40	31.36	37.00
CD	2	1.40	.66	.53	2.61	2	2.05	3.23	.26	21.85
ABC	8	.77	.29	.28	2.04	4	4.59	19.05	12.13	3.45
ABD	4	1.28	.17	.22	1.62	2	11.13	27.00	8.80	27.38*
ACD	4	1.66	.34	.77	5.02	4	7.76	16.33	2.21	45.34**
BCD	4	1.95	.51	.32	3.83	2	.28	1.15	3.24	.56
ABCD	8	1.34	.43	.21	2.20	4	3.26	9.36	10.14	2.30
Error	28	1.30	.35	.33	2.70	16	5.33	14.70	7.63	16.29

Note: Values listed are mean squares;

*represents significance at 5% level; and

**represents significance at 1% level.

L = leaves, S = stems, R = roots, and E = entire plant.

Table 4.--Continued.

Source of variation	df.	Potassium			Calcium		
		L	S	R	L	S	R
							X10-2
Night temp. A	2	10.16*	4.07	8.93**	10.65*	2.56**	8.55**
Sea. day max.							
temp. B	1	78.02**	17.20	26.00**	.01	76.27**	53.55**
AB	2	16.04**	49.70**	.86	.71	.29	.69
Potassium C	2	160.71**	54.30**	16.46**	4.83	.01	.22
AC	4	11.93**	5.91	.20	4.27	.29	1.18**
BC	2	29.61**	10.10	1.26	2.08	.49	.01
Iron D	1	1.13	.26	4.84**	2.20	1.06*	.90*
AD	2	.49	1.51	1.40	3.06	.35	.63
BD	1	1.52	1.56	.59	7.02*	.07	.03
CD	2	.04	8.87	.13	.36	.17	.11
ABC	4	4.12	9.28	.06	1.48	.15	.06
ABD	2	.14	2.73	.28	.23	.12	.01
ACD	4	1.34	1.87	.85	2.53	.11	.63
BCD	2	.15	1.08	.48	.33	.08	.01
ABCD	4	2.08	3.98	.39	1.48	.33	.03
Error	16	1.92	4.25	.42	1.45	.17	.18

Note: Values listed are mean squares;

*represents significance at 5% level; and

**represents significance at 1% level.

L = leaves, S = stems, R = roots, and E = entire plant.

Table 4.--Continued.

Source of variation	df.	Magnesium			Phosphorus			Nitrogen		
		L	S	R	L	S	R	L	E	
		X10-4			X10-3			X10-3		
Night temp. A	2	4.11	61.86	5.30*	2.97	7.01*	23.63**	3.2**	8.98**	12.75**
Sea. day max. temp. B	1	.25	153.34	7.83*	.42	5.13	84.09**	.04	6.77**	1.14**
AB	2	22.33**	86.08	.79	.97	8.50**	3.42	.15	1.78**	460.00**
Potassium C	2	67.02**	64.28	.03	2.52	2.70	3.32	.26	.04	.63
AC	4	11.27*	97.10	2.25	3.78	2.85	3.89	.15	.28	29.82
BC	2	.83	84.20	.45	.17	3.83	1.87	.02	.94*	29.23
Iron D	1	4.69	115.56	.13	11.90*	.80	1.34	.27	.15	225.60*
AD	2	1.44	63.36	.47	.92	.72	3.11	.10	.08	37.40
BD	1	3.36	60.06	.28	.47	.13	1.34	.09	.01	15.60
CD	2	1.02	61.39	.23	.13	.53	2.93	.03	.58	6.29
ABC	4	7.91**	42.12	2.13*	.43	1.94	1.39	.02	.37	40.75
ABD	2	5.44**	46.26	.28	3.91	1.53	.71	.02	.01	1.15
ACD	4	1.77**	60.33	1.31	2.67	1.22	2.54*	.25*	.13	34.90
BCD	2	2.19**	68.81	.68	1.78	.10	.08	.01	.03	8.13
ABCD	4	.03	53.50	.22	1.76	.90	.34	.03	.31	30.00
Error	16	3.38	53.30	1.03	1.91	1.22	1.16	.08	.21	27.00

Note: Values listed are mean squares;

*represents significance at 5% level; and

**represents significance at 1% level.

L = leaves, S = stems, R = roots, and E = entire plant.

Figure 10. Effects of Night Temperatures and Seasons on Yields of Centipedegrass.

yields produced at the intermediate daylight temperatures prevalent during the spring period.

A highly significant interaction was observed between temperatures and applied iron on the growth of roots (Figure 11). At both the high night temperatures (18.0 - 25.0° C.) and the low night temperatures (-1.1° C.) root growth was less for grass that received an iron application as compared to grass that received no iron treatment.

Potassium had a tendency to increase plant growth, as shown in Figure 12. However, this increased growth was mostly a result of root growth, since the root was the only component which increased in growth with increases in potassium.

Plant Inorganic Constituents

Iron contents of grass in the three experiments are tabulated in Table 28. The analyses of variance (Table 4) showed that night temperatures significantly affected the iron concentrations in all parts of the grass. The iron concentration in plant parts was also markedly affected by night temperatures, seasons, rates of potassium, and rates of iron, as shown in Table 5. There was a definite tendency for the iron to decrease with increases in night temperatures. However, total

Figure 11. Effects of Night Temperature and Rates of Iron on the Root Yields of Centipedegrass.

Figure 12. Effects of Rates of Potassium on Yields of Centipedegrass.

iron in the entire plant (Table 5) increased with advanced night temperatures. The distribution of iron within the three major parts of the grass was employed as an indication of iron translocation. An examination of the data shows that there was no change in distribution under the three different night temperature regimes. Therefore, translocation seems to have operated independently of night temperatures. The averaged distribution of plant total iron among leaves, stolons, and roots for centipedegrass was 26.6, 22.5, and 50.9 percent. This indicates that one-half of the absorbed iron was stored in the roots and only one-quarter was transferred to leaves.

Iron absorption and distribution were affected tremendously in all three parts of the plant by season in which the experiment was conducted. In the summer of 1964, when the average daily maximum temperature was 48.0° C., the iron concentration was 145, 115, and 287 ppm for leaves, stolons, and roots, respectively. However, in the winter, with an average daily maximum temperature of 28.6° C. the iron contents were much higher; 212, 188, and 394 ppm, for the respective parts. Considering the entire plant (Table 5), the concentration of iron was 182 ppm for plants grown in the summer, while it was 263 ppm for plants grown in the winter.

Table 5.--Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the iron content in centipedegrass of experiments 4 and 5.

	<u>Leaves</u>		<u>Stolons</u>		<u>Roots</u>		<u>Total plants</u>	
	dist.%	ppm	dist.%	ppm	dist.%	ppm	ppm	Fe in mgm/pot
<u>Night temp. in °C.</u>								
-1.1	26.8	206	22.9	176	50.3	386	254	16.2
7.2	25.9	172	22.0	146	52.1	345	221	19.7
18.0-25.0	27.1	157	22.7	132	50.2	289	193	23.1
	<u>Night temperatures</u>							
<u>Sea. day max. temp. in ° C.</u>								
48.0 (Summer)	26.6	145	21.0	115	52.4	287	182	16.5
28.6 (Winter)	26.1	212	23.8	188	50.1	394	263	23.9
	<u>Seasons</u>							
<u>Lbs. per 1,000 sq. ft.</u>								
2	26.5	190	22.3	159	51.2	365	238	20.9
5	26.5	173	24.1	156	59.4	322	217	19.6
8	26.6	172	21.5	138	51.9	334	215	19.9
	<u>Potassium rates</u>							
<u>Lbs. per 1,000 sq. ft.</u>								
0.0	27.5	163	24.3	144	48.2	286	198	18.1
0.1	26.0	194	21.3	159	52.7	394	248	22.2
	<u>Iron rates</u>							

Total iron absorption by plants grown at the higher temperature (48.0° C.) was also less than by plants grown during the winter (28.6° C.). There was no change in distribution percentages of the iron due to changes in seasonal temperatures. These data indicate that neither night temperatures nor seasons affected iron translocation, but both affected iron absorption.

The rate of potassium application had a significant effect on iron concentration in the whole plant (Table 4). Plants that received the equivalent of 2 lb. of potassium per 1,000 square feet contained 238 ppm of iron, but grass that received 5 and 8 lbs. of potassium per 1,000 square feet contained only 217 and 215 ppm of iron, respectively (Table 5). However, total iron uptake was not affected by the rates of potassium.

Pots that received the iron chelate treatments exhibit an increase in the iron concentrations of the leaves, roots, and entire plants, as compared to pots to which no iron chelate was applied (Table 5). Calculations of distribution percentages of the iron from the chelate show that a considerable portion of added iron may have been retained in the roots rather than being translocated to tops of the grass. Nevertheless, the absolute amount of iron in the tops of the grass to

which iron was supplied was still significantly higher than in plant tops to which no iron was applied, as shown in Table 5. Several significant interactions of applied iron and night temperatures on the iron concentrations of centipedegrass are shown in Figure 13. For example, chelated iron was more effective at the lowest night temperature than at the higher temperatures, especially for the roots of centipedegrass.

Potassium contents in the tissue of centipedegrass are shown in Table 29 and the analyses of variance are listed in Table 4.

Highly significant differences were found among night temperatures (Table 4) and their effects on percent potassium of roots and leaves, and for the whole plants. The percent potassium in leaves, roots, and the entire plant grown at the medium night temperature (7.2°C.) was usually higher than when grown at the highest night temperatures ($18.0 - 25.0^{\circ}\text{C.}$), or the lowest night temperature (-1.1°C.). However, total potassium content of grass grown in the greenhouse at night temperatures of $18.0 - 25.0^{\circ}\text{C.}$ (Table 6) was greater than in grass grown at -1.1°C. or 7.2°C. The percentage distribution of potassium in the various parts of the grass may be an indication of the potassium translocation in that plant. This percentage distribu-

Figure 13. Effects of Night Temperatures and Rates of Iron on the Iron Content of Leaves, Roots, and Entire Plants of Centipedegrass.

tion in the grass roots and stolons was changed with changes in night temperatures. Relatively speaking, at night temperature of -1.1° C., the translocation of potassium from roots to stolons was much faster than at warmer night temperatures.

The relatively mild winter season with an average daily maximum temperature of 28.6° C. resulted in higher concentrations and greater total potassium absorption than did the hot summer season (48.0° C.), except for stolons, as indicated by the analysis of variance in Table 4. Translocation of potassium was also influenced by the season in which the experiment was conducted. A larger percentage of potassium was deposited in the stolons in the summer (30.3 percent) than in the winter (22.7 percent), as shown in Table 6. Furthermore, there was a higher percentage potassium in the leaves during the winter than during the summer.

Increases in amount of applied potassium highly significantly increased the amounts of absorbed potassium in grass, leaves, and roots. The increases in potassium rates increased the percent potassium content of the leaves of the grass grown at -1.1° C. much more than plants grown at warm night temperatures (Figure 14).

An application of iron decreased plant total potassium uptake; this decrease was mainly exhibited in

Table 6.--Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the potassium content in centipede grass of experiments 4 and 5.

	<u>Leaves</u>		<u>Stolons</u>		<u>Roots</u>		<u>Total plants</u>	
	dist. %	%	dist. %	%	dist. %	%	K in	% mgm/pot
<u>Night temp.</u>								
<u>in °C.</u>								
-1.1	53.6	1.570	28.8	.843	17.6	.514	.975	62.2
7.2	54.0	1.693	24.2	.756	21.8	.683	1.044	93.3
18.0-25.0	53.7	1.513	26.0	.732	20.3	.569	.938	112.3
<u>Night temperatures</u>								
<u>Ave. max. temp.</u>								
<u>in °C.</u>								
<u>Seasons</u>								
48.0 (Summer)	51.7	1.455	30.3	.846	18.0	.503	.931	84.4
28.6 (Winter)	55.7	1.739	22.7	.708	21.6	.673	1.040	94.5
<u>Potassium rates</u>								
<u>Lbs. per 1,000</u>								
<u>sq. ft.</u>								
2	57.2	1.321	23.1	.533	19.7	.454	.742	65.3
5	49.6	1.569	29.2	.923	21.2	.666	1.052	94.5
8	56.4	1.969	25.1	.875	18.5	.645	1.163	107.9
<u>Iron rates</u>								
<u>Lbs. per 1,000</u>								
<u>sq. ft.</u>								
0.0	53.6	1.630	25.8	.786	20.6	.625	.995	91.4
0.1	54.9	1.610	26.3	.768	18.8	.552	.976	87.7

Figure 14. Effect of Rates of Potassium and Night Temperatures on the Percent Potassium in Leaves of Centipedegrass.

in grass roots (Table 6). Plants that received 0.1 lb. of iron per 1,000 square feet had only 0.552 percent potassium in the roots, while the roots of the plants that had received no iron had 0.625 percent potassium.

Calcium contents are listed in Table 30 for leaves, stolons, roots, and the whole plants, respectively. The analyses of variance (Table 4) indicated a significant difference in calcium concentration in leaves, and highly significant differences in stolons, roots, and the entire plant due to night temperatures. Table 7 shows that percent calcium increased as night temperatures decreased in all parts of the plant, but total uptake of calcium in the entire plants was increased with increases in temperature.

In the summer season, calcium contents of roots and stolons were 60 percent and 170 percent more, respectively, than the corresponding parts in the winter season, because of less translocation of calcium to the leaves during the summer. Potassium applications had no effect on calcium content of the grass.

The addition of iron had a significant effect on the accumulation of calcium in the roots, reflecting a decrease in calcium translocation to the leaves.

Magnesium contents of centipedegrass are

Table 7.--Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the calcium content in Centipede grass of experiments 4 and 5.

	<u>Leaves</u>		<u>Stolons</u>		<u>Roots</u>		<u>Total plants</u>	
	dist. %	%	dist. %	%	dist. %	%	Ca in	% mgm/pot
<u>Night temp.</u>								
<u>in o C.</u>								
-1.1	53.5	1.136	19.6	.414	26.9	.569	.219	14.0
7.2	56.3	1.078	18.5	.354	25.2	.480	.199	17.8
18.0-25.0	57.0	.952	18.9	.315	24.1	.468	.165	19.8
<u>Night temperatures</u>								
<u>Seasons</u>								
Ave. max. temp.								
<u>in o C.</u>								
48.0 (Summer)	48.7	1.057	23.5	.509	27.8	.606	.208	18.9
28.6 (Winter)	67.4	1.065	9.7	.218	22.9	.362	.180	16.5
<u>Potassium rates</u>								
Lbs. per 1,000								
<u>sq. ft.</u>								
2	56.6	1.108	18.4	.362	25.0	.488	.193	16.9
5	53.4	.985	19.8	.365	26.8	.494	.197	18.0
8	53.3	.952	20.4	.365	26.3	.468	.195	18.0
<u>Iron rates</u>								
Lbs. per 1,000								
<u>sq. ft.</u>								
0.0	56.0	1.080	19.8	.381	24.2	.467	.197	18.0
0.1	54.9	1.031	18.5	.347	26.6	.499	.193	17.3

presented in Table 31. The analysis of variance in Table 4, shows that only the percent magnesium in the plant roots was significantly affected by night temperatures and seasons of the year. The lower the night temperatures the higher the percent magnesium content in the roots (Table 8). Furthermore, the total content of magnesium was increased by increases in night and daylight temperatures.

The percent magnesium in the leaves was highly affected by the rates of potassium. That is, increasing rates of potassium decreased percent magnesium in the grass. The addition of iron lowered the percent plant magnesium from .077 to .065 percent, and at the same time decreased the total magnesium content in the plant.

Phosphorus content of grass was statistically affected by night temperatures and seasons of the year in which the experiment was conducted (Table 32). Most parts of the grass contained lower percentages of phosphorus as temperatures increased during the night, but increased their total phosphorus content as night temperature increased (Table 9). However, stolons had a higher percent phosphorus content at the lowest night temperature than that at higher temperatures. Neither potassium nor iron application affected percentages of phosphorus in the grass.

Table 8.--Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the magnesium content in centipede grass of experiments 4 and 5.

	Leaves		Stolons		Roots		Total plants	
	dist. %	%	dist. %	%	dist. %	%	Mg in	%
<u>Night temperatures</u>								
Night temp. in ° C.								
-1.1	55.2	.128	23.4	.054	21.4	.050	.073	4.63
7.2	62.0	.133	20.0	.043	18.0	.039	.075	6.67
18.0-25.0	61.7	.121	19.2	.037	19.1	.037	.065	7.82
<u>Seasons</u>								
Ave. max. temp. in ° C.								
48.0 (Summer)	56.1	.126	23.2	.052	20.7	.047	.072	6.52
28.6 (Winter)	63.2	.127	18.4	.037	18.4	.037	.067	6.36
<u>Potassium rates</u>								
Lbs. per 1,000 sq. ft.								
2	64.1	.154	18.6	.045	17.3	.042	.075	6.65
5	56.2	.112	22.5	.045	21.3	.042	.066	6.09
8	57.3	.115	22.0	.044	20.7	.042	.070	6.55
<u>Iron rates</u>								
Lbs. per 1,000 sq. ft.								
0.0	58.7	.131	22.2	.049	19.1	.043	.077	7.04
0.1	60.3	.123	19.5	.040	20.2	.041	.065	5.84

Table 9.--Effects of night temperatures, seasons of the year, and rates of potassium and iron applications on the phosphorus content in Centipede-grass of experiments 4 and 5.

	Leaves		Stolons		Roots		Total plants	
	dist. %	%	dist. %	%	dist. %	%	P in % mgm/pot	
<u>Night temperatures</u>								
Night temp. in ° C.								
-1.1	40.9	.270	42.3	.278	16.8	.111	.219	14.0
7.2	45.4	.275	37.4	.226	17.2	.104	.199	17.8
18.0-25.0	46.4	.233	37.8	.190	15.8	.080	.165	19.8
<u>Seasons</u>								
Ave. max temp. in ° C.								
48.0 (Summer)	39.5	.246	44.9	.280	15.6	.090	.208	18.9
28.6 (Winter)	49.1	.272	33.1	.183	17.8	.079	.181	16.5
<u>Potassium rates</u>								
Lbs. per 1,000 sq. ft.								
2	46.5	.273	37.6	.221	15.9	.093	.193	16.9
5	40.7	.243	42.1	.251	17.2	.103	.197	18.0
8	40.9	.261	38.3	.223	16.8	.098	.195	18.0
<u>Iron rates</u>								
Lbs. per 1,000 sq. ft.								
0.0	43.4	.225	40.5	.238	16.1	.095	.197	18.0
0.1	44.7	.263	38.4	.226	16.9	.100	.193	17.3

Nitrogen content of the plants (Table 33) was highly significantly affected by night temperatures and iron applications, as shown by the analysis of variance in Table 4. In general, nitrogen percentages for the whole plant were 2.15, 1.74, and 1.18 for low, medium and high night temperatures (Table 10), respectively. However, the plants grown at intermediate night temperature contained 20 percent more total nitrogen than those grown at either the lowest temperature or the highest temperature. Plants that received the iron chelate contained approximately 15 percent more nitrogen, either expressed as percent nitrogen or as total nitrogen, than plants where no iron was added (Table 10).

The percent nitrogen in the leaves (Table 34) was markedly affected by night temperatures and season of the year. Generally, plants grown in low temperatures, either night temperature, or cool season day temperatures, accumulated higher percentages of nitrogen than those at higher temperatures.

Some Organic Compounds in Plants

Organic acids in centipedegrass were identified and quantitatively determined by paper chromatography and are listed in Tables from 33 to 43. It was found that organic acids detected in one part of the plant

Table 10.--Effects of night temperatures and rates of potassium and iron applications on the nitrogen content in centipedegrass of experiments 4 and 5.

	<u>Leaves</u>			<u>Stolons</u>			<u>Roots</u>			<u>Total plants</u>	
	dist. %	%		dist. %	%		dist. %	%		%	N in mgm/pot
<u>Night temp.</u>											
<u>in ° C.</u>											
-1.1	34.4	2.22		41.5	2.68		24.1	1.55		2.15	141.0
7.2	32.6	1.70		40.9	2.13		26.5	1.38		1.74	170.5
18.0-25.0	34.4	1.22		40.3	1.43		25.3	.90		1.18	143.0
<u>Night temperatures</u>											
<u>Lbs. per 1,000</u>											
<u>sq. ft.</u>											
<u>Potassium rates</u>											
2	33.8	1.67		40.7	2.01		25.5	1.26		1.65	170.6
5	33.7	1.75		41.6	2.16		24.7	1.28		1.73	186.8
8	33.7	1.71		41.0	2.08		25.3	1.28		1.69	184.5
<u>Lbs. per 1,000</u>											
<u>sq. ft.</u>											
<u>Iron rates</u>											
0.0	37.1	1.66		3.46	1.55		28.3	1.27		1.49	161.0
0.1	33.8	1.77		41.5	2.17		24.7	1.29		1.74	185.7

were not necessarily detectable in others, as shown in Figure 15. For example, 6 organic acids, oxalic, tartaric, citric, malic, succinic, and aconitic acids were found in the leaves, but succinic acid did not appear in stolons and roots, and tartaric and aconitic acids were missing in the roots. A summary of organic acids in the parts of centipedegrass under various treatments are shown in Table 11.

Oxalic, citric, and malic acids were the only three acids which were distributed throughout all major parts of the grass. The distribution percentages for the leaves, stolons, and roots, as shown in Table 13a, were 46.7, 39.4, and 13.9 for oxalic acid; 36.4, 27.4, and 36.2 for citric acid; 47.1, 35.6, and 17.3 for malic acid. There was a considerable decrease in the distribution of oxalic and malic acids from leaves to stolons to roots, while citric acid was of rather even distribution among these three plant parts. Under night temperature of -1.1°C ., 7.2°C ., and $18.0-25.0^{\circ}\text{C}$., citric acid was found to be 0.226, 0.246, and 0.138 percent, respectively, and malic acid was found to be 0.175, 0.241, and 0.256 percent. Apparently the medium night temperature (7.2°C .) favored the formation of citric acid while malic acid formation was favored at the high night temperatures ($18.0 - 25.0^{\circ}\text{C}$.).

Fig. 15.--Paper Chromatogram of Organic Acids from Leaves, Stolons, and Roots of the Centipede grass.

Table 11.--Organic acids found in leaves, stolons and roots, in percentage of fresh weight of plant material.

	In leaves				
	Oxalic	Tartaric	Citric	Malic	Succinic Aconitic
T ₁ K ₁ P ₆₀	.342	.195	.200	.335	.094 .424
T ₁ K ₁ P ₆₁	.390	.165	.190	.222	.112 .330
T ₁ K ₂ P ₆₀	.280	.140	.222	.337	.078 .458
T ₁ K ₂ P ₆₁	.252	.132	.190	.369	.152 .397
T ₁ K ₃ P ₆₀	.446	.125	.220	.406	.060 .331
T ₁ K ₃ P ₆₁	.218	.094	.172	.291	.060 .479
T ₂ K ₁ P ₆₀	.402	.139	.276	.342	.072 .384
T ₂ K ₁ P ₆₁	.385	.150	.372	.479	.208 .501
T ₂ K ₂ P ₆₀	.242	.155	.320	.463	.170 .510
T ₂ K ₂ P ₆₁	.200	.140	.250	.370	.160 .527
T ₂ K ₃ P ₆₀	.448	.160	.268	.300	.172 .450
T ₂ K ₃ P ₆₁	.334	.201	.294	.484	.186 .459
T ₃ K ₁ P ₆₀	.190	.121	.192	.312	.150 .364
T ₃ K ₁ P ₆₁	.326	.171	.158	.244	.160 .551
T ₃ K ₂ P ₆₂	.166	.087	.144	.245	.200 .609
T ₃ K ₂ P ₆₁	.432	.184	.296	.298	.250 .637
T ₃ K ₃ P ₆₀	.418	.176	.150	.251	.200 .689
T ₃ K ₃ P ₆₁	.352	.165	.092	.318	.310 .646
Average	.3235	.1500	.2231	.3373	.1552 .4858

Note: T₁ = -1.1° C., T₂ = 7.2° C., T₃ = 18.0° C.; K₁ = 2 lbs.,
 K₂ = 5 lbs.; P₆₀ = 8 lbs. of potassium per 1,000 sq. ft.; Fe₀ =
 no iron added; P₆₁ = 0.1 lbs. of iron per 1,000 sq. ft.

Table 11.--Continued.

	In stolons			In roots		
	Oxalic	Tartaric	Malic	Aconitic	Oxalic	Malic
T ₁ K ₁ FeO	.190	.123	.109	.095	.115	.147
T ₁ K ₁ Fe1	.190	.171	.134	.073	.135	.174
T ₁ K ₂ FeO	.280	.150	.107	.074	.113	.127
T ₁ K ₂ Fe1	.347	.222	.083	.084	.148	.098
T ₁ K ₃ FeO	.398	.250	.144	.106	.132	.108
T ₁ K ₃ Fe1	.463	.210	.138	.094	.100	.088
T ₂ K ₁ FeO	.165	.080	.076	.058	.123	.650
T ₂ K ₁ Fe1	.352	.117	.167	.089	.103	.346
T ₂ K ₂ FeO	.217	.022	.190	.040	.053	.203
T ₂ K ₂ Fe1	.237	.057	.248	.116	.120	.350
T ₂ K ₃ FeO	.117	.065	.170	.145	.055	.162
T ₂ K ₃ Fe1	.270	.192	.070	.039	.048	.026
T ₃ K ₁ FeO	.260	.145	.188	.184	.068	.343
T ₃ K ₁ Fe1	.272	.260	.208	.226	.058	.237
T ₃ K ₂ Fe2	.352	.192	.272	.210	.078	.332
T ₃ K ₂ Fe1	.108	.180	.239	.254	.012	.196
T ₃ K ₃ FeO	.307	.059	.288	.655	.105	.127
T ₃ K ₃ Fe1	.317	.090	.169	.121	.085	.272
Average	.269	.1436	.1666	.1479	.0917	.221
						.1167

Note: T₁ = -1.1° C., T₂ = 7.2° C., T₃ = 18.0 - 25.0° C.; K₁ = 2 lbs.,
 K₂ = 5 lbs., K₃ = 8 lbs. of potassium per 1,000 sq. ft.; FeO =
 no iron added, Fe1 = 0.1 lbs. of iron per 1,000 sq. ft.

Of the three acids, only citric acid was affected by potassium applications (Table 12). In the application of 2, 5, and 8 pounds of potassium in 1,000 square feet, citric acid content was detected as 0.231, 0.214, and 0.164 percent, respectively. None of these acids were affected by the addition of iron (Table 12).

Oxalic and citric acids in leaves were significantly affected by seasons, as seen in Table 13b. During the summer, oxalic acid content was 0.153 percent and citric acid content was 0.148 percent, while in the winter, oxalic acid increased to 0.313 percent and citric acid to 0.225 percent.

Aconitic acid in leaves was highly significantly affected by both night temperature and season of year (Table 12). Leaves contained 0.403, 0.479, and 0.591 percent of this acid when grown at night temperatures of -1.1° C., 7.2° C., and $18.6 - 25.0^{\circ}$ C., (Table 13b) respectively. Furthermore, they contained 0.435 and 0.546 percent when grown in the summer and winter, respectively.

Table 12.--Analysis of variance of the effects of treatments on the organic acid content of the whole plant and leaves of centipede grass of experiments 4 and 5.

Source of variation	d.f.	Organic acids found in whole plant		
		Oxalic10-3	Citric10-3	Malic10-3
Night temp. A	2	2.52	59.45**	33.05**
Sea.day.max.temp. B	1	239.48**	18.37	184.33**
AB	2	9.45	14.95	14.97*
Potassium C	2	11.34	22.08*	2.25
AC	4	8.52	7.86	5.48
BC	2	5.07	17.55*	4.30
Iron D	1	1.77	4.23	.09
AD	2	.67	11.76	.25
BD	1	1.33	2.82	2.95
CD	2	3.67	1.74	1.31
ABC	4	6.81	7.71	6.95
ABD	2	13.47	2.06	2.47
ACD	4	.47	3.15	3.25
BCD	2	9.11	6.14	1.80
ABCD	4	3.70	9.05	4.83
Error	16	6.30	6.41	4.44

Note: Values listed are means squares; *represents significance at 1% level and **represents significance at 5% level.

Table 12.---Continued.

Source of variation	d.f.	Organic acids found in leaves				
		Oxalic10-3	Citric10-3	Malic10-3	Tartaric10-3	Aconitic10-3
Night temp. A	2	0.01	4.53	49.60	0.75	107.18**
Sea, day max. temp. B	1	229.70**	49.80**	16.50	2.69	111.33**
AB	2	5.83	32.67**	78.88*	1.84	134.71**
Potassium C	2	0.49	1.57	2.06	1.00	29.75
AC	4	7.85*	6.30	3.14	3.47*	7.86
BC	2	8.20	2.35	0.28	1.61	4.16
Iron D	1	2.81	0.38	0.83	1.19	6.77
AD	2	17.10**	1.49	14.45	3.51*	2.01
BD	1	3.21	0.14	1.08	0.10	1.79
CD	2	5.51	2.32	3.04	0.47	2.71
ABC	4	4.30	3.51	25.71	0.14	22.35
ABD	2	1.48	7.53	11.39	0.02	5.99
ACD	4	4.16	5.67	16.57	1.71	13.46
BCD	2	0.15	0.12	15.15	0.68	2.96
ABCD	4	0.76	0.77	8.74	0.93	4.81
Error	16	2.51	3.44	16.08	0.75	11.27

Note: Values listed are means squares; *represents significance at 5% level and **represents significance at 1% level.

Table 13a.--Oxalic, citric, and malic acids in leaves, stolons, and roots of centipede grass of experiments 4 and 5.

Leaves dis.% acid%	Stolons dis.% acid%	Roots dis.% acid%	Night temp. in °C.		K in lbs.per 1000 sq.ft.		Fe in lbs. per 1000 sq.ft.						
			-1.1	7.2	18.0-	2	5	8	0.0	0.1			
25.0													
<u>Oxalic acid %</u>													
46.0	.323	39.4	.274	13.9	.097	.263	.215	.217	.225	.202	.267	.228	.235
<u>Citric acid %</u>													
36.4	.223	27.4	.167	36.2	.221	.226	.246	.138	.231	.214	.164	.212	.195
<u>Malic acid %</u>													
42.1	.316	35.6	.239	17.3	.116	.175	.241	.256	.229	.231	.210	.225	.222

Table 13b.--Oxalic, citric, malic, tartaric, aconitic, and succinic acids in leaves of centipede grass of experiments 4 and 5.

Night temp. in °C.		Season temp. in °C.		K in lbs.per 1000 sq.ft.		Fe in lbs.per 1000 sq.ft.			
-1.1	7.2	18.0-25.0	48.0	28.6	2	5	8	0.0	0.1
<u>Oxalic acid %</u>									
.234	.233	.232	.153	.313	.236	.226	.238	.242	.224
<u>Citric acid %</u>									
.164	.199	.193	.148	.223	.181	.198	.176	.182	.188
<u>Malic acid %</u>									
.328	.406	.278	.359	.316	.320	.348	.341	.359	.316
<u>Tartaric acid %</u>									
.142	.158	.151	.141	.158	.157	.139	.153	.144	.156
<u>Aconitic acid %</u>									
.403	.479	.591	.436	.547	.434	.523	.517	.478	.505
<u>Succinic acid %</u>									
.093	.161	.211	--	--	.132	.168	.164	.133	.177

The average percentages of total organic acids found in leaves, stolons and roots as affected by night temperatures, rates of potassium and iron applications are shown in Table 14. Total organic acid content of leaves and stolons usually increased with increases in night temperatures (with the exception of stolons at night temperature of 7.2° C.), increases in rates of potassium, and increases in the supply of iron. However, the average total organic acid percentage in the roots had a completely different picture. Roots grown at the medium night temperature (7.2° C.) contained a higher percentage of total organic acids than at other temperatures. Plants supplied with the highest rates of potassium, or supplied with iron had a lower average total organic acids percentages than when the low rates of these nutrients were used, warm nights and high rates of potassium and the addition of iron stimulated the production of total organic acids in the plant as a whole.

Table 15 lists the various organic acids percentages in the leaves of centipedegrass, with and without iron at the lowest night temperature. Occasionally leaves with iron were slightly chlorotic; and leaves without iron were mostly chlorotic. Organic acid contents, except for succinic acid, were higher in leaves

Table 14.--Effects of night temperatures, rates of potassium, and rates of iron on the total organic acids contents of the leaves, stolons, roots, and entire plants of centipedegrass.

Parts of plant	Night temp. in °C.			Rates of K in lbs. per 1000 sq.ft.		Rates of Fe in lbs. per 1000 sq.ft.	
	-1.1 %	7.2 %	18.0-25.0 %	2 %	5 %	8 %	0.1 %
Leaves	.810	1.626	1.644	1.446	1.596	1.584	1.494
Stolons	.930	.738	1.218	.883	.966	1.038	.954
Roots	.294	.552	.438	.572	.412	.304	.468
Entire plants	.678	.972	1.067	.969	.991	.975	.972
							.390
							.984

Table 15.--Organic acids contained in leaves of centipede grass supplied with and without iron chelate at the lowest night temperature (-1.1°C.).

	Fe-applied, lbs. per 1000 sq. ft.					
	0.0			0.1		
	K-applied, per 1000 sq.ft.			K-applied, per 1000 sq.ft.		
	2	5	8	2	5	8
	%	%	%	%	%	%
Oxalic	.510	.540	.564	.434	.444	.321
Citric	.346	.319	.349	.240	.486	.218
Malic	.671	.674	.812	.443	.758	.582
Aconitic	.849	.915	.662	.660	.793	.959
Tartaric	.390	.280	.250	.330	.264	.188
Succinic	.094	.078	.060	.112	.152	.060

of plants that received no iron. The high rate of potassium at low night temperatures without added iron increased the percentages of oxalic, citric and malic acid, but decreased tartaric and succinic acids in leaves. In leaves of plants supplied with iron, oxalic, citric, malic, and succinic acid contents were usually at their highest at the medium night temperature.

Free amino acids detected on paper chromatograms were aspartic acid (Table 44), glutamic acid (Table 45), glycine (Table 46), histidine (Table 47), and isoleucine (Table 48). These amino acids, identified by the over spotting method, were found in all three parts of the grass, as shown in Figure 16.

The amino acids significantly affected by night temperature treatments were glutamic acid and isoleucine, as shown by analyses of variance in Table 16. Grass grown at a night temperature of 7.2° C. had a higher glutamic acid content than grass grown either at -1.1° C. or $18.0 - 25.0^{\circ}$ C. Isoleucine increased with increases in night temperature.

The distribution percentages show that there were highly significant differences in the amounts of amino acids, except for glycine, in the leaves, stolons, and the roots of the plant. The amounts of aspartic acid and histidine were 1.5 to 1.9 times more concen-

Fig. 16.--Paper Chromatogram of Amino Acids from Leaves, Stolons, and Roots of Centipedegrass.

trated in the roots than in the leaves. Glutamic acid was the only amino acid for which the highest concentration was found in the leaves. Isoleucine was found in highest percentage in the stolons.

The relationships between night temperatures and the amino acid contents in various parts of the plant were highly significant, as indicated in Table 16. The interactions may be seen more clearly through the use of graphs, as presented in Figures 17, 18, and 19. For example, in Figure 17, roots of grass grown at a night temperature of -1.1° C. had smaller amounts of aspartic acid than those at higher night temperatures. Centipedegrass grown at the lowest night temperature had the lowest amount of glutamic acid in the leaves. Roots contained only a small quantity of glycine and isoleucine at night temperature of -1.1° C. Histidine was found in the highest amount in the leaves and the lowest amount in the root.

Since chlorotic leaves usually occurred at low night temperature, as previously discussed, and since low night temperature was found to be associated with lowest amounts of four of the amino acids, it appears that chlorosis of grass may be indirectly related to the low content of one or more of the four free amino acids (aspartic, glycine, isoleucine, and histidine)

Table 16.--Analysis of variance of the effects of treatments on the amino acid content of centipede grass.

Source of variation	d.f.	Aspartic acid				Glutamic acid				Glycine				Histidine				Isoleucine			
		10 ⁻⁴				10 ⁻⁴				10 ⁻⁴				10 ⁻⁴				10 ⁻⁴			
Night temp. (A)	2	.76	7.86*			.63	3.18			.63	3.18			.63	3.18			.63	3.18		
Parts of plant (B)	2	77.46**	92.80**			.15	201.85**			.15	201.85**			.15	201.85**			.15	201.85**		
AB	4	25.63**	7.16**			1.86**	28.66**			1.86**	28.66**			1.86**	28.66**			1.86**	28.66**		
Potassium (C)	2	2.29	4.55			.03	2.02			.03	2.02			.03	2.02			.03	2.02		
AC	4	.88	2.29			.53	5.82			.53	5.82			.53	5.82			.53	5.82		
BC	4	.72	3.54			.55	3.60			.55	3.60			.55	3.60			.55	3.60		
Iron (D)	1	.12	6.89			.02	.01			.02	.01			.02	.01			.02	.01		
AD	2	2.51	3.41			1.09	3.54			1.09	3.54			1.09	3.54			1.09	3.54		
BD	2	1.21	1.10			.06	2.16			.06	2.16			.06	2.16			.06	2.16		
CD	2	.04	3.23			.01	6.00			.01	6.00			.01	6.00			.01	6.00		
AEC	8	2.60	1.39			.40	5.29			.40	5.29			.40	5.29			.40	5.29		
ABD	4	.36	2.18			.25	.84			.25	.84			.25	.84			.25	.84		
ACD	4	2.21	1.51			.09	1.03			.09	1.03			.09	1.03			.09	1.03		
BCD	4	1.25	3.47			.13	3.67			.13	3.67			.13	3.67			.13	3.67		
ABCD	8	1.85	2.45			.32	6.92			.32	6.92			.32	6.92			.32	6.92		
Error	28	1.82	2.12			.27	4.28			.27	4.28			.27	4.28			.27	4.28		

Note: * represents significance at 5% level, ** represents significance at 1% level.

Figure 17. Effect of Night Temperatures on the Percent of Aspartic and Glutamic Acid in Leaves, Stolons and Roots of Centipedegrass.

Figure 18. Effects of Night Temperatures on the Percent of Glycine and Isoleucine in Leaves, Stolons and Roots of Centipede grass.

Figure 19. Effects of Night Temperatures on the Percent of Histidine in Leaves, Stolons and Roots of the Centipedegrass.

in the roots.

The total free amino acid percentages were 0.644, 0.775, and 0.782 for grasses grown at -1.1° C., 7.2° C., and $18.0 - 25.0^{\circ}$ C., respectively. They were 0.735, 0.792, and 0.695 percent for grasses which received 2, 5, and 8 pounds of potassium per 1,000 square feet, respectively. Grass that received iron was slightly lower in total free amino acids than grass that received no iron.

Free sugar content of the centipedegrass in these experiments, including the reducing sugars of monosaccharides and the cold water extractable carbohydrates are presented in Table 49 and 50. The analyses of variance of the effect of treatments on these sugars are shown in Table 17. The distribution percentages of sugars among the parts show that the stolons contained higher amounts of both reducing sugar and the potentially reducing power cold water extractable carbohydrate than roots and leaves.

Some Properties of Centipedegrass Roots

Root reductive capacities of centipedegrass as influenced by three night temperatures are presented in Table 51. A summary of the data presented in Figure 20 shows the reductive capacity of roots increased

Table 17.--Analysis of variance of the effects of treatments on the sugar content of centipedegrass.

Source of variation	d.f.	(1)	(2)	Sum of (1) and (2) 10^{-2}
		Reducing sugar 10^{-2}	Cold water extract. Carbo. 10^{-2}	
Night temp. A	2	1.87	38.72	25.79
Parts of plant B	2	42.53**	503.04**	747.22**
AB	4	7.48*	138.44**	93.41*
Potassium C	2	2.67	12.86	13.04
AC	4	9.26*	61.49*	60.67
BC	4	4.88	35.38	25.65
Iron D	1	5.54	4.74	13.80
AD	2	10.04*	80.08*	28.40
BD	2	9.04*	12.71	34.76
CD	2	1.68	23.40	39.65
ABC	8	3.70	17.05	16.05
ABD	4	3.39	24.62	41.26
ACD	4	3.00	17.86	9.57
BCD	4	49.35	28.83	27.07
ABCD	8	1.83	23.84	29.10
Error	28	2.56	21.87	24.03

Note: * represents significance at 5% level,
 ** represents significance at 1% level.

Figure 20. Effects of Night Temperatures on Reductive Capacities of Roots of Centipedegrass.

with increase in night temperature. Moreover, the reductive capacity of roots from plants grown at the intermediate night temperature steadily increased through a period of 36 hours.

Cation exchange capacities of centipedegrass roots are given in Table 52. The average exchange capacities of roots under night temperatures of -1.1° C., 7.2° C., and $18.0 - 25.0^{\circ}$ C. were 7.38, 6.43, and 6.13 me. per 100 gm. dry weight of plant roots, respectively. Roots of plants that received 2, 5 and 8 pounds of potassium per 1,000 square feet had cation exchange capacities of 7.08, 6.55, and 6.31 me. per 1,000 gm. dry weight of plant roots.

Soil Mineral Nutrients

Some properties of untreated Arredondo loamy fine sand were given in Table 2. The results of analyses of soil presented below were made after the centipedegrass had been harvested at the completion of the experiments.

Iron was extracted from soil by N Ammonium Acetate. Exchangeable ferrous iron was extracted with the solution buffered at pH 6.0, while the same solution buffered at pH 3.6 was used for extracting the combined exchangeable ferrous and ferric and acid soluble ferric

iron. The results are given in Table 53. The analysis of variance (Table 18) indicates that night temperatures, rates of potassium, and rates of iron significantly affected the amount of extractable iron. It further appears that there was highly significant differences in the amounts of the various forms of the iron extracted from the Arredondo loamy fine sand by the different buffered solutions. The average amounts of extractable iron under different treatments are found in Table 19. The amount of iron found in the plants are given in the table along with that extracted from the soil from each treatment series.

Since the amount of iron found in plants was much smaller than that found in the soil, the "available iron" (used herin to refer to the sum of the two sources) was of about the same magnitude as the soil component. Available iron decreased with increases in night temperatures. The extracting solution buffered at 3.0 extracted three times as much iron as that extracted by the pH 6.0 solution. The additions of 2 and 5 pounds of potassium per 1,000 square feet of soil did not affect the amounts of iron extracted, but when 8 pounds of potassium was added the "available iron" decreased almost 25 percent. An addition of iron chelate to the soils increased available soil iron

Table 18.--Analysis of variance of the effects of treatments on the extractable iron, potassium, magnesium, calcium and phosphorus contents of soils.

Source of variation	d.f.	Iron	Potassium	Magnesium	Calcium	Phosphorus
Night temp. A	2	3.744*	21,870**	5,357**	504,856**	16.33
Sea. day max. temp. B	1	195.207** ¹	17,556**	4,853**	8,306,884**	289.00**
AD	2	.106	4,980*	69	386,081*	.99
Potassium C	2	7.504**	91,683**	1,828*	66,695	8.33
AC	4	1.376	3,155*	54	62,684	39.41*
BC	2	1.071	3,980*	1,062	19,611	44.33*
Iron D	1	15.067**	971	11	30,218	2.77
AD	2	3.172*	379	980	66,507	17.44
BD	1	1.341	12	336	19,646	1.77
CD	2	1.185	4,101*	129	62,129	.11
ABC	4	.301	1,561	547	55,109	29.58
ABD	2	.982	21	703	71,608	4.78
ACD	4	.959	428	354	97,745	7.02
BCD	2	.112	1,463	352	27,034	5.44
ABCD	4	.954	841	168	100,884	5.19
Error	16	.690	893	399	75,763	11.72

¹Extracting solutions pH 6.0 and pH 3.0 of $\text{N NH}_4\text{OAc}$.

Note: * represents significance at 5% level,

** represents significance at 1% level.

Table 19.---Available¹ Nutrients in the soil.

	Night temp. in °C.	Sea. day max. temp. in °C.	K-applied lbs. per 1000 sq.ft.		Fe-applied lbs. per 1000 sq.ft.	
			2	5	8	0.1
	-1.1	7.2	18.0-25.0	48.0	28.6	
<u>Fe², mgm/800 gm. soil</u>						
Fe in plants	1.62	1.92	2.30	--	2.09	1.97
Fe in soil	8.94	8.01	7.16	4.32	11.76	8.62
Available Fe					8.95	6.60
in soil	10.56	9.93	9.46	4.32	11.76	10.71
					10.92	8.59
						8.81
						11.30
<u>K, mgm/800 gm. soil</u>						
K in plants	62.6	93.3	112.3	84.4	94.9	65.3
K in soil	119.9	90.8	51.2	118.2	69.6	24.6
Available K					74.4	94.5
in soil	182.5	184.1	163.5	202.6	164.5	89.9
					168.9	270.3
						174.4
						179.1
<u>Ca, mgm/800 gm. soil</u>						
Ca in plants	44.9	56.2	66.9	65.6	49.7	57.3
Ca in soil	1216.0	1100.0	892.8	1454.4	685.6	1136.0
Available Ca					1020.0	1053.6
in soil	1260.9	1156.0	959.7	1520.0	735.3	1193.3
					1076.4	1111.7
						1104.9
						1148.9
<u>Mg, mgm/800 gm. soil</u>						
Mg in plants	4.6	6.7	7.8	6.5	6.4	6.6
Mg in soil	86.0	83.8	60.0	104.4	132.1	89.8
Available Mg					70.8	75.8
in soil	90.6	90.5	67.8	110.9	138.5	95.4
					76.9	82.3
						111.5
						137.9

Table 19.---Continued.

	Night temp. in ° C.	Sea. day max. temp. in ° C.	K-applied lbs. per 1000 sq.ft.		Fe-applied lbs per.1000 sq.ft.					
	-1.1	7.2 18.0-25.0	48.0	28.6	2	5	8	0.0	0.1	
<u>P, mgm/800 gm. soil.</u>										
P in plants	1.40	1.78	1.98	1.89	1.65	1.69	1.80	1.80	1.73	
P in soil	15.60	15.80	14.30	12.90	17.50	15.20	15.80	14.50	15.40	
Available P in soil	17.00	17.58	16.18	14.79	19.15	16.89	17.60	16.30	16.70	17.13
<u>NO₃ - N, mgm/800 gm. soil</u>										
NO ₃ in soil	12.60	2.06	1.40	---	---	6.84	5.47	3.72	9.62	6.41

¹"Available" soil nutrients is expressed as the sum of that absorbed by plants plus that extractable from the soil after harvest.

²Fe extracted by N NH₄OAc, at pH 6.0 for 48.0° C. treatment and by N NH₄OAc at pH 3.0 for the 28.6° C. treatment.

about 28 percent at the medium night temperature (7.2° C.).

Other inorganic nutrients extracted from soil samples by N Ammonium acetate (pH 4.8) were potassium, calcium, magnesium, and phosphorus (Table 49). The seasonal effects on the availability of these four nutrients were highly significant, as were night temperatures. Soils kept in chamber at -1.1° C. in the night had the highest levels of available potassium, while those kept at the warmest temperature ($18.0 - 25.0^{\circ}$ C.) had the least amount of available potassium. Soil samples taken at the completions of the summer experiment (48.0° C.) contained a significantly greater amount of available potassium than soil taken at the conclusions of the winter experiment (28.6° C.). Calcium availability was similar to potassium in responses to treatments. Phosphorus, as well as magnesium, increased as the seasonal maximum temperature decreased. However, the two nutrients responded differently to night temperatures.

Nitrates found in the soil after plants were removed were greater at the night temperature of -1.1° C. than at 7.2 and $18.0 - 25.0^{\circ}$ C. (Table 54). The values for three night temperatures were 12.6, 2.0, and 1.4 ppm. of nitrate nitrogen, respectively. Soils to which 2, 5, 8, pounds of potassium per 1,000 square

feet were applied contained 6.8, 5.4, and 3.7 ppm, of nitrate nitrogen, respectively. Soils to which 0.1 pound iron per 1,000 square feet was added contained less nitrate than soils that received no iron.

GENERAL DISCUSSION

The data obtained from five related experiments, in which centipedegrass was fertilized at varying rates of potassium and subjected to different night temperatures, were not consistent in all respects, as might be expected. However, the following results were in general agreement throughout the experiments: (a) Lowering the night temperature increased iron concentration in centipedegrass. (b) The concentration of potassium was inversely related to that of iron in the grass. (c) Night temperature of -1.1° C. induced chlorotic condition in centipedegrass if no iron chelate was added.

Chlorotic plants grown at the low night temperature contained substantially smaller amounts of iron than did green plants which had been treated with iron chelate and grown at the same temperature. Yet, quite often, the same chlorotic plant contained as much, or more, iron than the healthy plant grown at higher temperatures. The effect of night temperature on iron contents (ppm) of leaves, stolons, and roots is clearly shown in the following table:

Night temp. in ° C.	Fe-applied, lbs. per 1,000 sq. ft.					
	0.0	0.1	0.0	0.1	0.0	0.1
	Leaves		Stolons		Roots	
	ppm	ppm	ppm	ppm	ppm	ppm
18.0-25.0	150	162	122	141	274	304
7.2	175	168	149	143	334	356
-1.1	163*	251	160*	195	252*	522

*From chlorotic plants

Chlorosis became quite serious when low night temperatures were associated with a high rate of potassium fertilizer. However, the chlorosis observed was not necessarily due to an increasing deficiency of iron in the plant, as shown in the following table:

K applied, lbs./ 1,000 sq. ft.	Fe concentration in ppm		
	Leaves	Stolons	Roots
	ppm	ppm	ppm
2	173	166	288
5	141	191	167
8	174	124	295

The data were obtained from chlorotic plants grown at low night temperature and without the addition of iron chelate. The severity of the chlorosis increased as the rate of potassium increased.

This enigma of high iron content in the chlorotic grass at low night temperatures posed a problem which demanded a thorough investigation. The results of the research on this problem, involving the availability of

iron in the soil and its absorption, translocation and assimilation in centipedegrass, occupies a major part of this discussion.

Iron absorption by roots of the grass depends on both the supply of available iron in the soil and the properties of those roots. Chemical analyses of soils have shown that extractable iron (and other nutrients) increased with decreases in temperature during the night. The near anaerobic conditions in soil transferred to the cold chamber (-1.1° C.) at night, resulting from moisture condensed around soil particles, rendered soil iron more soluble. The soil pH and the soil oxidation potential were apparently lowered (49, 119, 134) which reduced the iron to a more readily available form. On the other hand, potassium applications generally decreased the amount of available iron (and other cations). This may be an indirect effect on the soil oxidation potential, as a result of enhancing the reduction of soil nitrates (119). Data from the soil nitrate determinations supported this point (Table 54). While the greatest amounts of nitrate accumulated in the soil at the lowest night temperature (-1.1° C.), the application of potassium significantly reduced nitrate accumulation.

The roots of grass grown at the lowest night

temperature were abundantly supplied with nutrients, judging from the analysis of available soil nutrients (Table 18). It is possible several factors governed the ability of roots to absorb nutrients, such as the morphological and some physiochemical characteristics. Certainly, differences were found in the morphology of the roots grown at the different temperatures. For example, roots at -1.1° C. were white, soft and twisted (Figure 9). These are characteristics considered to be associated with high root activity for the absorption of plant nutrients (121, 131). Certain physiochemical characteristics of plant roots, such as reductive capacity, content of organic substances, and cation exchange capacity, are concerned with the absorption of iron and other nutrients (49).

The reductive capacity of roots grown at the low night temperature, -1.1° C., was lower than that of roots which were grown at higher night temperatures. Therefore, it appears that the reductive capacity of the roots was not responsible for the greater iron absorption at the low temperature in these experiments. While the content of reducing sugar in the roots may appear to be related to their reductive capacity, roots grown at -1.1° C. had neither larger amounts of reducing sugar nor higher contents of cold water-extractable

carbohydrates than at higher temperatures.

Cation exchange capacity of roots suggests a possible clue to understanding the peculiar characteristic of the grasses grown at -1.1° C., because cation exchange capacity of the roots increased with decreases in night temperature (Table 52). A further understanding of the absorption property of the roots may be found in the study of chelating agents in roots. The organic compounds detected in the roots which may serve this purpose were oxalic, citric and malic acids. Among them, citric acid content was the highest. However, the concentration of these three organic acids and the concentration of iron in the same root tissue exhibited an inverse relationship at the all night temperatures. That is, the organic acids, viewed as either concentration or total content, increased with increases in night temperature, the reverse was for iron absorption.

Aspartic acid and histidine were abundant in the roots. However, their concentrations increased with temperature increases, while iron decreased with increases in temperature. Therefore, these amino acids did not appear to be responsible for the greater uptake of iron by the plants grown at night

temperature of -1.1° C. Since the highest percentage of total nitrogen was observed in plants at the lowest night temperature, this may suggest that nitrogen-containing compounds were in some way connected with the absorption of iron. If so, they were probably proteins and peptide, but not free amino acids. While all the organic compounds mentioned above may have had some influence on the absorption of iron, none of them individually showed a marked correlation with high concentrations of iron at the low night temperature.

There were greater concentrations of iron in plants treated with iron chelate than in plants without iron chelate at the -1.1° C. night temperature whereas, the iron treatment made little difference in iron content of grass at the higher temperatures. This suggests that at low night temperature high concentrations of iron in the roots may have resulted from increased available ferrous iron from reduced soil conditions and from the chelated iron (NaFeDTPA). This large concentration of available iron may have resulted in a mass action effect on the roots. This mass flow then enabled the roots to take up iron with less energy expenditure at -1.1° C. than at either

7.2° C. or at 18.0 - 25.0° C. The Q_{10} was found to be less than 1.0, when calculated on the basis of iron concentrations of the entire plant. This suggests iron absorption of centipedegrass as possibly not being an energy dependent process.

Translocation of iron in the grass was found to be independent of night temperatures and the season of the year, as shown by the percentage distribution in different parts of the plant (Table 5). Furthermore, potassium applications did not appear to affect translocation, a fact contrary to the opinions of Hartt (63) and Ponnampetuma (119). Although the application of iron chelate increased absorption of iron, it did not increase translocation to any considerable degree. Most of the iron added was accumulated in roots. Calcium delayed the translocation of iron from the roots to stolons (Table 7), but apparently not from the stolons to the leaves. Nitrogen compounds (proteins, amides and peptides) increased the movement of iron from roots to stolons but slowed down the movement from stolons to leaves (Table 10). There was no obvious relationship of phosphorus or magnesium contents to iron translocation.

If one accepts the hypothesis that iron is

translocated as a chelate in the xylem vascular bundles and the phloem (33), the organic compounds which would become chelating agents for this purpose are probably oxalic, citric and malic acids. They were rapidly transported from leaves to all parts of the plant and have considerable strength in chelating metals (49). Over all, the concentration of organic acids in centipedegrass underwent a tremendous change at different night temperatures. At a temperature of -1.1° C., oxalic acid was the predominant acid. At a temperature of 7.2° C. citric and malic acids were in greater concentrations; while at the greenhouse temperatures of $18.0 - 25.0^{\circ}$ C., malic acid was dominant over the others (Table 13-a). This suggests that iron (and perhaps other nutrients) was chelated in different forms at the various night temperatures. For example, oxalic acid may have chelated a large proportion of the iron at lowest night temperature. This would result in a short supply of active iron (113, 125), because the association between iron and oxalic acid is very strong (49). In this case, added chelated iron (NaFeDTPA), entering the plant root as a molecule, avoided the effect of oxalic acid and organic compounds during the translocation flow. Therefore, grass that

received NaFeDTPA did not suffer from chlorosis, as did the grass that received no added iron chelate at the temperature of -1.1° C.

Organic acids occurring largely in the stolons and leaves were tartaric and aconitic acids. Their chance to act as chelating agents in transporting iron was much less than acids which were found in all three parts of the grass.

Iron Assimilation in a biological system is a process of incorporating iron into the vital position of an organoid, and it often proceeds with some difficulties, sometimes resulting in chlorosis. The causes of the chlorosis are mainly lack of sufficient (a) active iron in the tissue, (b) and porphyrin synthesis.

Iron translocation in the chlorotic tissue was possibly associated with large amounts of oxalic and citric acids. As soon as the iron reached the leaves it encountered an even higher concentration of oxalic acid, which in some cases was as high as 0.308 percent. Therefore, iron may have been inactivated as a tenaciously chelated organic complex. In some cases, calcium alleviates the pressure exerted on the iron from oxalic acid (124). However, under

the particular conditions which caused chlorosis of the centipedegrass in these experiments, calcium accumulated in the roots and probably had little influence on the oxalates and citrates in the other tissues.

In regard to porphyrin synthesis, it was reported (38) that succinic acid and glycine were the organic materials which provided carbon and nitrogen to form a molecule of porphyrin. Data (Table 43) in those experiments showed that there were only small amounts of succinic acid but large amounts of aconitic acid found in the leaves (Table 42). Although inferences as to metabolic pathways based on absolute amounts of any component (ignoring equilibria, steady state concentrations, turnover rates, etc.) are often in error, it is perhaps useful as a basis for further work to make such conjectures. The data on succinic and aconitic acid concentrations suggests the possibility that in the respiration system of centipedegrass leaves, the glyoxalate cycle, a cycle converting fatty acid to carbohydrates carried out only in higher plants (38), was operating to a noticeable extent. If the glyoxalate cycle is active, one might expect to find large quantities of citric, malic and oxaloacetic acids in the leaves. This was the case

in the present series of experiments, except that oxalic acid was found instead of oxaloacetic acid. This probably is due to the oxaloacetic acid being enzymatically converted into oxalic acid. It appeared that the glyoxalate cycle became less active little by little as temperatures increased. In the course of advancing temperature, the apparent importance of Krebs Cycle became noticeably larger. This supposition is based on the fact that when night temperatures were -1.1°C. , 7.2°C. and $18.0 - 25.0^{\circ}\text{C.}$, the amounts of succinic acid were 0.093, 0.161 and 0.211 percent, respectively (Table 43). One would assume from these data that in centipede-grass the glyoxalate cycle operated more actively at low temperatures than at high temperatures. Therefore, this species usually produced small amounts of succinic acid, a raw material for porphyrin synthesis, at the low temperatures. Glycine is the other raw material in the formation of porphyrin. This amino acid was present in very small amounts among the group of amino acids found in centipede-grass (Table 46).

It has been suggested (36) that porphyrin can not be formed without active iron in the same tissues. Therefore a deficiency of succinic acid, glycine, and

active iron resulted in a shortage of porphyrin compound. This deficiency, in turn, resulted in a small amount of magnesium porphyrin, a precursor of chlorophyll, and heme-porphyrin, which is the vital component for some essential biological processes (38). All of these are directly or indirectly involved in chlorophyll synthesis. Since all three of the original prerequisites for porphyrin synthesis were present in lesser amount at low night temperatures than at higher temperatures, a reasonable explanation of the observed chlorosis in centipede-grass at low night temperatures is provided.

SUMMARY AND CONCLUSIONS

Centipedegrass, one of the most popular turf-grasses in Florida, is often chlorotic during the spring and early summer when night temperatures are low. The chlorosis, apparently aggravated by heavy potassium fertilization, is reported (32) to be caused by a deficiency of iron.

A series of five factorial experiments were conducted in 1963 - 1964 to study the effects of night temperatures and potassium fertilization on the absorption, translocation and assimilation of iron. Although treatments varied among the pot experiments, they all involved a comparison of grass kept in cold chambers at night at temperatures of -1.1° C. and 7.2° C., with those maintained in the greenhouse at uncontrolled temperature, during various seasons of the year. Various levels of potassium were used in all tests and levels of iron and sources of potassium were used as additional treatments in some. The results from the experiments are summarized below:

1. Growth of centipedegrass increased with increases in night temperatures and to potassium additions, but not to applications of iron. Greater yield

increases were obtained in the spring than in the summer or winter.

2. Lowering night temperatures resulted in increases in iron concentration in the plant, but decreases in total uptake. However, the translocation of iron, was not affected.

3. Potassium applications decreased the iron concentration in grass. The chloride salt had a greater effect than sulfate. The potassium effect was more pronounced at cold night than at warm night temperatures. Potassium had no effect on iron translocation.

4. Extractable soil iron, potassium, calcium and magnesium but not phosphorus, increased with decreases in temperatures. Potassium fertilization decreased the availability of cations, except potassium. Nitrates accumulated in the soil at low night temperatures.

5. Chlorotic plants grown at low night temperatures often contained higher concentrations of iron than healthy plants grown at higher temperatures. Potassium applications increased the severity of chlorosis at low night temperatures.

6. Oxalic, citric and malic acids were distributed throughout roots, stolons and leaves; tartaric and aconitic acids in stolons and leaves; but succinic

acid was found in leaves only. Total organic acids in centipedegrass grown at -1.1° C., 7.2° C. and $18.0 - 25.0^{\circ}$ C. night temperatures were 2.03, 2.92, and 3.00 percent, respectively. Potassium had little influence on total organic acids content.

7. Of the amino acids, aspartic acid, glutamic acid, glycine, histidine and isoleucine were found in all three parts of the plant. Total amino acid contents of grass grown at night temperatures of -1.1° C., 7.2° C. and $18.0 - 25.0^{\circ}$ C. were 0.64, 0.77 and 0.78 percent respectively. Total amino acid content associated with application of 2, 5 and 8 pounds of potassium per 1000 square feet were 0.74, 0.79 and 0.70 percent, respectively.

8. Reducing sugar content of grass grown at night temperatures of -1.1° C., 7.2° C. and $18.0 - 25.0^{\circ}$ C. were 0.492, 0.500, 0.551 percent, respectively. Increase in potassium application decreased the concentration of reducing sugar in grasses but had no effect on cold water-extractable carbohydrate content.

9. These results lead to the following conclusion regarding the relation of night temperatures and potassium fertilization to chlorosis of centipedegrass:

a. Low temperatures probably resulted in a

decrease in soil oxidation potential, thus increasing available iron in the soil solution.

- b. High amounts of oxalic and citric acids present in plant tissue at low temperature probably chelated the iron to an inactive form.
- c. Low temperature probably affected plant's respiration system so that the activity of the glyoxalate cycle increased. This is inferred from the observed high amounts of oxalic, citric and malic acids, but little succinic acid in the grass.
- d. Glycine in plant tissue was very low at low temperatures.
- e. Therefore, a shortage of active iron and insufficient amounts of succinic acid and glycine appeared to have resulted in a deficiency of chlorophyll in the centipede grass at low temperature, even when the total content of iron in the plant was quite high.

APPENDIX

Table 20.--Effects of night temperatures and sources and rates of potassium on yield (gm. oven-dry wt.) of centipedegrass of experiment 1.

K-applied lbs. per 1,000 sq. ft.	K ₂ SO ₄			KCl		
	I	II	III	I	II	III
<u>Night temp. 15.5 - 21.0° C.</u>						
2	2.21	2.51	2.54	2.37	2.12	2.03
5	2.25	2.12	2.48	2.65	1.74	2.80
8	2.20	1.88	1.86	1.42	1.72	2.61
<u>Night temp. 7.2° C.</u>						
2	1.07	1.90	1.63	1.80	2.03	1.91
5	1.76	1.68	1.42	2.00	1.65	1.72
8	1.32	1.66	1.46	2.52	1.49	1.84
<u>Night temp. 1.6° C.</u>						
2	1.80	2.20	1.20	1.50	1.94	1.15
5	1.25	1.85	1.27	1.45	1.67	1.88
8	1.19	1.54	1.62	2.04	1.80	1.59

Table 21.--Effects of night temperatures and sources and rates of potassium on iron concentration (ppm) in centipodegrass of experiment 1.

K-applied lbs. per 1,000 sq. ft.	<u>K₂SO₄</u>			<u>KCl</u>		
	I	II	III	I	II	III
<u>Night temp. 15.5 - 21.0° C.</u>						
2	160	160	170	260	220	230
5	200	200	160	140	200	230
8	140	140	200	200	150	150
<u>Night temp. 7.2° C.</u>						
2	275	220	260	300	300	240
5	230	200	300	260	300	220
8	250	220	250	200	150	150
<u>Night temp. 1.6° C.</u>						
2	280	250	250	250	210	300
5	260	200	220	300	300	220
8	260	250	200	200	230	230

Table 22.--Effects of night temperatures and sources and rates of potassium on percent potassium in centipede grass of experiment 1.

K-applied lbs. per 1,000 sq. ft.	K ₂ SO ₄			Mol		
	I	II	III	I	II	III
<u>Night temp. 15.5 - 21.0° C.</u>						
2	2.65	2.55	2.10	2.30	2.85	2.55
5	2.75	2.50	2.50	2.75	2.77	3.00
8	2.75	3.00	3.25	3.30	2.60	2.75
<u>Night temp. 7.2° C.</u>						
2	3.20	2.00	1.80	2.25	1.90	2.10
5	2.05	2.30	2.15	2.65	2.50	2.20
8	2.50	2.40	2.40	2.65	2.45	2.70
<u>Night temp. 1.6° C.</u>						
2	2.10	2.35	2.20	2.60	1.40	2.20
5	2.70	2.55	2.60	2.25	2.15	1.37
8	3.00	2.75	2.75	2.15	2.40	2.20

Table 23.--Effects of night temperatures and rates of potassium on yield (gm. oven-dry wt.) of centipede-grass of experiment 2.

K in nutrient solution in ppm	Night temp. 4.5° C.				Night temp. 15.5-22.5° C.			
	I		II		I		II	
	Tops	Roots	Tops	Roots	Tops	Roots	Tops	Roots
<u>12 - day growth</u>								
0.0	2.36	1.12	2.31	1.90	2.78	1.52	2.21	1.38
234.0	2.26	1.88	2.60	1.52	2.80	2.48	2.11	2.14
<u>24 - day growth</u>								
0.0	2.51	.55	3.00	.83	4.02	1.01	4.29	1.03
234.0	5.34	2.33	.95	1.19	6.03	.98	3.26	1.11
<u>36 - day growth</u>								
0.0	5.13	.88	3.72	1.19	5.61	1.21	4.50	1.00
234.0	6.66	1.46	2.85	.83	9.06	1.51	5.91	1.06

Table 24.--Effects of night temperatures and rates of potassium on iron concentration (counts per minute from Fe⁵⁹) in centipodegrass of experiment 2.

K in nutri- ent solution ppm	Night temp. 4.5°C				Night temp. 15.5-22.5°C			
	I		II		I		II	
	Tops	Roots	Tops	Roots	Tops	Roots	Tops	Roots
<u>12 - day growth</u>								
0.0	930	3815	788	4559	1786	4159	1134	4150
234.0	644	2478	668	2730	836	3090	963	2242
<u>24 - day growth</u>								
0.0	2117	5191	1151	4972	1355	4128	704	4072
234.0	526	4502	896	3523	1149	2271	970	2756
<u>36 - day growth</u>								
0.0	1699	3225	723	2082	869	3290	999	3830
234.0	818	3618	1167	4001	957	2948	799	2440

Table 25.--Effects of night temperatures and rates of potassium on percent potassium in centipedegrass of experiment 2.

K in nu- trient so- lution ppm	Night temp. 4.5° C				Night temp. 15.5-22.5°C			
	I		II		I		II	
	Tops	Roots	Tops	Roots	Tops	Roots	Tops	Roots
<u>12 - days growth</u>								
0.0	.40	.10	1.00	.50	.60	.10	1.40	.40
234.0	3.35	2.10	2.70	1.55	4.00	1.80	3.10	1.90
<u>24 - days growth</u>								
0.0	.62	.19	.80	.30	.36	.08	.62	.10
234.0	3.94	1.30	2.28	1.41	2.91	2.60	2.46	1.55
<u>36 - days growth</u>								
0.0	.44	.17	.94	.27	.37	.05	.46	.17
234.0	3.67	1.37	2.58	1.06	3.34	1.64	3.26	2.45

Table 26.--Analysis of the effects of treatments on iron and potassium contents in centipede grass of experiment 2.

Source of variation	a.f.	Iron Fe59		Potassium	
		concentration	total	concentration	total
Hight temp. (A)	1	721,043	2,248,570	1,496	146,082
Rate of K (B)	1	3,932,510**	574,440	477,603**	5,744,984**
Harvest 2 (C)	2	928,416	2,152,365	2,109	674,170
Parts (D)	1	76,151,897**	3,158,540	104,907**	5,125,844**
AB	1	118,895	1,155,377	4,641	184,512
AC	2	2,472,525	6,625	540	96,683
AD	1	466,291	2,113,864	1,262	64,829
BC	2	976,748	1,534,620	197	567,523
BD	1	1,345,017	1,373,590	26,508**	2,691,377
CD	2	394,100	32,363,960**	495	1,400,556
Error	34	814,371	2,212,713	1,760	15,154,316

Note: Values listed are means squares; *represents significance at 5% level and **represents significance at 1% level.

Table 27.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves, stolons, roots and entire plants yields (gm. oven-dry wt.) of centipedegrass of experiments 3, 4 and 5.

K-applied lbs. per 1000 sq. ft.	<u>Spring 1964</u>		<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied</u>		<u>Fe-applied</u>		<u>Fe-applied</u>	
	lbs. per		lbs. per		lbs. per	
	1000 sq.ft.		1000 sq.ft.		1000 sq.ft.	
	0.0	0.1	0.0	0.1	0.0	0.1

Night temp. 18.0 - 25.0° C.

Leaf

2	5.28	3.33	4.45	3.80	4.85	4.15
5	3.43	6.15	4.77	3.30	5.50	3.53
8	4.55	4.88	4.45	4.30	5.25	5.68

Stolon

2	3.97	3.43	2.52	3.52	2.22	3.97
5	3.03	5.01	3.84	2.91	3.96	4.31
8	3.87	3.88	3.41	3.29	4.06	5.48

Root

2	4.70	3.80	4.30	3.20	4.90	3.80
5	4.95	2.60	4.50	3.60	4.20	2.00
8	3.30	4.15	5.05	4.10	2.75	4.00

Entire plant

2	13.95	10.56	11.27	10.57	11.97	11.92
5	11.41	13.76	13.11	9.84	13.66	9.84
8	11.72	12.90	12.91	11.67	12.06	15.16

Night temp. 7.2° C.

Leaf

2	5.65	6.15	4.40	2.60	2.10	7.15
5	5.17	7.10	4.96	4.48	5.20	4.83
8	5.93	4.75	4.80	3.15	4.13	4.08

Table 27.--Continued.

K-applied lbs. per 1000 sq.ft.	Spring 1964		Summer 1964		Winter 1964	
	Fe-applied		Fe-applied		Fe-applied	
	lbs. per		lbs. per		lbs. per	
	1000 sq.ft.		1000 sq.ft.		1000 sq.ft.	
	0.0	0.1	0.0	0.1	0.0	0.1

Night temp. 7.2° C.Stolon

2	3.96	2.90	2.43	2.02	1.42	3.48
5	2.84	2.61	2.34	1.14	2.28	2.22
8	2.63	2.15	1.70	1.84	1.91	1.92

Root

2	3.05	3.45	2.00	3.65	3.25	3.95
5	3.85	3.75	1.80	3.15	2.35	2.55
8	2.35	2.65	2.85	3.00	1.85	2.00

Entire plant

2	12.66	12.50	8.83	8.27	5.77	13.07
5	11.86	13.36	9.10	8.77	9.83	9.66
8	10.91	9.55	8.55	7.99	7.89	7.99

Night temp. -1.1° C.Leaf

2	2.50	3.28	2.45	3.12	2.30	3.13
5	3.28	2.85	3.69	4.00	3.20	3.43
8	5.75	4.95	6.50	4.70	3.85	1.78

Stolon

2	.95	1.72	.91	1.47	.84	1.68
5	1.40	1.37	1.16	1.93	1.58	1.75
8	2.17	1.61	2.66	.91	1.12	.60

Root

2	1.80	1.35	1.90	1.30	1.90	1.65
5	1.70	1.45	1.45	1.45	2.40	.90
8	1.95	1.65	1.40	1.55	1.80	.50

Table 27.--Continued.

K-applied lbs. per 1000 sq. ft.	<u>Spring 1964</u>		<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied</u>		<u>Fe-applied</u>		<u>Fe-applied</u>	
	<u>lbs. per</u>		<u>lbs. per</u>		<u>lbs. per</u>	
	<u>1000 sq.ft.</u>		<u>1000 sq.ft.</u>		<u>1000 sq.ft.</u>	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. -1.1° C.</u>						
2	5.25	6.35	5.26	5.89	5.04	6.46
5	6.38	5.67	6.30	7.38	6.78	6.08
8	9.87	8.21	10.56	7.16	6.77	2.88

Table 28.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaf, stolon, root and entire plant iron content (ppm) in centipedegrass of experiments 4 and 5.

K-applied lbs.per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied per 1000 sq. ft.		Fe-applied per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. 18.0 - 25.0° C.

Leaf

2	133	165	153	186
5	111	105	157	217
8	150	110	197	193

Stolon

2	75	110	195	175
5	88	100	137	170
8	88	113	152	182

Root

2	220	294	300	360
5	260	266	330	349
8	229	241	304	317

Entire plant

2	143	189	216	240
5	153	157	208	245
8	156	155	218	231

Night temp. 7.2° C.

Leaf

2	152	157	228	175
5	135	135	265	198
8	127	156	145	190

Table 28.--Continued.

K-applied lbs. per 1000 sq. ft.	Summer 1964		Winter 1964	
	Fe-applied per 1000 sq. ft.		Fe-applied per 1000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. 7.2° C.</u>				
<u>Stolon</u>				
2	100	125	200	188
5	113	140	183	152
8	88	135	207	122
<u>Root</u>				
2	330	260	450	390
5	265	356	330	474
8	255	280	375	375
<u>Entire plant</u>				
2	194	181	293	251
5	171	210	260	275
8	157	190	242	229
<u>Night temp. -1.1° C.</u>				
<u>Leaf</u>				
2	145	182	202	402
5	123	200	158	275
8	150	177	197	270
<u>Stolon</u>				
2	88	175	244	239
5	113	150	268	265
8	125	140	248	330
<u>Root</u>				
2	230	481	345	721
5	135	375	200	525
8	260	425	330	615

Table 28.--Continued.

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied per 1000 sq. ft.		Fe-applied per 1000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. -1.1° C.</u>				
<u>Entire plant</u>				
2	154	279	264	454
5	124	242	209	322
8	178	247	217	358

Table 29.--Effects of night temperatures, seasons of the years, rates of potassium and iron applications on leaf, stolon, roots and entire plant percent potassium content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied, per 1000 sq. ft.		Fe-applied, per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. 18.0 - 25.0° C.

Leaf

2	.87	.73	1.35	1.35
5	1.36	1.55	1.75	1.75
8	1.50	1.65	2.25	2.05

Stolon

2	.46	.50	.45	.41
5	1.05	1.00	.55	.49
8	1.30	1.10	.85	.65

Root

2	.43	.29	.60	.44
5	.55	.47	.80	.71
8	.69	.43	.80	.62

Entire plant

2	.59	.51	.80	.73
5	.99	1.01	1.03	.98
8	1.16	1.06	1.30	1.11

Night temp. 7.2° C.

Leaf

2	1.00	1.07	1.45	1.60
5	1.62	1.88	1.82	1.82
8	1.73	1.73	2.30	2.30

Table 29.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied, per 1000 sq. ft.</u>		<u>Fe-applied, per 1000 sq. ft.</u>	
	0.0	0.1	0.0	0.1

Night temp. 7.2° C.Stolon

2	.48	.42	.47	.55
5	1.05	1.78	.65	.50
8	1.05	.98	.68	.47

Root

2	.45	.45	.60	.62
5	.73	.58	1.00	.85
8	.60	.77	.85	.70

Entire plant

2	.64	.65	.84	.92
5	1.13	1.41	1.16	1.06
8	1.13	1.16	1.28	1.16

Night temp. -1.1° C.Leaf

2	1.20	1.44	1.45	1.35
5	1.69	1.47	1.06	1.06
8	1.69	1.83	2.25	2.35

Stolon

2	.60	.55	.90	.61
5	.70	.85	1.03	1.45
8	.82	.55	1.06	1.00

Root

2	.35	.28	.52	.42
5	.50	.45	.70	.66
8	.43	.60	.64	.60

Table 29.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied, per 1000 sq. ft.		Fe-applied, per 1000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. -1.1° C.</u>				
	<u>Entire plant</u>			
2	.72	.76	.96	.79
5	.96	.92	.93	1.06
8	.99	.99	1.32	1.31

Table 30.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaf, stolon, root and entire plant percent calcium content in centipede grass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied lbs.		Fe-applied lbs.	
	per 1000 sq. ft.		per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. 18.0 - 25.0° C.

Leaf

2	1.03	.94	1.16	.84
5	.95	1.01	.99	.75
8	.91	1.05	.84	.95

Stolon

2	.50	.47	.19	.16
5	.47	.32	.22	.15
8	.49	.46	.27	.18

Root

2	.48	.49	.29	.29
5	.48	.58	.28	.35
8	.47	.51	.28	.31

Entire plant

2	.67	.63	.55	.43
5	.63	.64	.50	.42
8	.62	.67	.46	.48

Night temp. 7.2° C.

Leaf

2	1.03	1.01	1.20	.95
5	1.12	1.04	1.30	.90
8	1.10	1.10	1.16	1.03

Table 30.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied lbs.</u>		<u>Fe-applied lbs.</u>	
	<u>per 1000 sq. ft.</u>		<u>per 1000 sq. ft.</u>	
	0.0	0.1	0.0	0.1

Night temp. 7.2° C.Stolon

2	.52	.50	.20	.19
5	.54	.50	.27	.18
8	.46	.50	.25	.14

Root

2	.56	.53	.34	.32
5	.66	.67	.40	.40
8	.61	.57	.37	.34

Entire plant

2	.70	.68	.58	.49
5	.77	.74	.66	.49
8	.72	.72	.59	.50

Night temp. -1.1° C.Leaf

2	1.20	1.29	1.20	1.45
5	.99	1.10	.80	.87
8	1.01	1.15	1.45	1.12

Stolon

2	.58	.58	.20	.25
5	.50	.60	.34	.29
8	.64	.54	.22	.23

Root

2	.69	.91	.41	.55
5	.69	.64	.40	.38
8	.64	.72	.37	.43

Table 30.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied lbs.		Fe-applied lbs.	
	per 1000 sq. ft.		per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. -1.1° C.

Entire plant

2	.82	.93	.60	.75
5	.73	.78	.51	.51
8	.76	.80	.68	.59

Table 31.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaf, stolon, root and entire plant percent magnesium content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	Summer 1964		Winter 1964	
	Fe-applied lbs. per 1000 sq. ft.		Fe-applied lbs. per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. 18.0 - 25.0° C.

Leaf

2	.15	.16	.19	.15
5	.11	.10	.12	.09
8	.08	.10	.12	.08

Stolon

2	.065	.060	.010	.020
5	.045	.037	.020	.020
8	.046	.045	.040	.040

Root

2	.040	.045	.023	.035
5	.036	.042	.030	.025
8	.049	.038	.048	.041

Entire plant

2	.085	.088	.074	.068
5	.064	.060	.057	.045
8	.058	.061	.069	.054

Night temp. 7.2° C.

Leaf

2	.13	.13	.17	.17
5	.11	.11	.14	.16
8	.13	.11	.13	.10

Table 31.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied lbs.</u>		<u>Fe-applied lbs.</u>	
	<u>per 1000 sq. ft.</u>		<u>per 1000 sq. ft.</u>	
	0.0	0.1	0.0	0.1

Night temp. 7.2° C.

Stolon

2	.053	.065	.060	.040
5	.058	.045	.040	.020
8	.049	.050	.020	.010

Root

2	.050	.044	.055	.031
5	.045	.049	.032	.025
8	.043	.044	.022	.020

Entire plant

2	.078	.080	.095	.080
5	.071	.068	.071	.068
8	.074	.068	.101	.043

Night temp. -1.1° C.

Leaf

2	.17	.18	.12	.13
5	.13	.11	.08	.08
8	.13	.13	.14	.13

Stolon

2	.065	.030	.040	.030
5	.056	.047	.100	.050
8	.070	.050	.050	.060

Table 31.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied lbs. per 1000 sq. ft.		Fe-applied lbs. per 1000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. -1.1° C.</u>				
<u>Root</u>				
2	.055	.055	.034	.030
5	.050	.050	.070	.055
8	.048	.054	.034	.059
<u>Entire plant</u>				
2	.097	.034	.065	.063
5	.079	.069	.083	.062
8	.083	.078	.075	.083

Table 32.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaf, stolon, root and entire plant percent phosphorus content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied, lbs. per 1000 sq. ft.		Fe-applied, lbs. per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. 18.0 - 25.0° C.

Leaf

2	.25	.20	.28	.29
5	.17	.17	.25	.28
8	.17	.21	.25	.25

Stolon

2	.23	.20	.14	.15
5	.22	.20	.15	.15
8	.24	.25	.19	.16

Root

2	.065	.084	.080	.084
5	.077	.078	.085	.098
8	.075	.068	.088	.072

Entire plant

2	.183	.160	.167	.140
5	.157	.150	.163	.177
8	.163	.177	.187	.160

Night temp. 7.2° C.

Leaf

2	.28	.28	.28	.29
5	.23	.33	.26	.32
8	.24	.19	.28	.32

Table 32.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied lbs. per 1000 sq. ft.		Fe-applied lbs. per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. 7.2° C.Stolon

2	.30	.31	.16	.17
5	.32	.30	.18	.17
8	.30	.23	.17	.11

Root

2	.103	.082	.098	.088
5	.110	.126	.100	.120
8	.105	.108	.100	.104

Entire plant

2	.227	.223	.180	.183
5	.220	.253	.180	.170
8	.217	.177	.183	.177

Night temp. -1.1° C.Leaf

2	.29	.34	.27	.23
5	.27	.27	.21	.16
8	.26	.29	.33	.32

Stolon

2	.37	.28	.18	.16
5	.30	.38	.27	.37
8	.35	.26	.21	.21

Root

2	.110	.112	.104	.108
5	.098	.110	.114	.115
8	.100	.130	.100	.125

Table 32.--Continued

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied lbs. per 1000 sq. ft.		Fe-applied lbs. per 1000 sq. ft.	
	0.0	0.1	0.0	0.1

Night temp. -1.1° C.

Entire plant

2	.257	.243	.183	.167
5	.227	.253	.197	.213
8	.237	.227	.213	.217

Table 33.--Effects of night temperatures, rates of potassium and iron applications on percent nitrogen content of leaves, stolons and roots of centipedegrass of experiment 5.

K-applied lbs. per 1000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied, lbs. per 1000 sq.ft. 0.0	Fe-applied, lbs. per 1000 sq.ft. 0.1	Fe-applied, lbs. per 1000 sq.ft. 0.0	Fe-applied, lbs. per 1000 sq.ft. 0.1	Fe-applied lbs. per 1000 sq.ft. 0.0	Fe-applied lbs. per 1000 sq.ft. 0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	1.28	1.23	1.39	1.52	.87	.97
5	1.14	1.29	1.36	1.61	.91	.90
8	1.15	1.24	1.21	1.49	.81	.95
<u>Night temp. 7.2° C.</u>						
2	1.54	1.61	1.85	2.06	1.21	1.28
5	1.84	1.95	2.39	2.30	1.50	1.39
8	1.65	1.61	2.17	2.04	1.57	1.33
<u>Night temp. -1.1° C.</u>						
2	1.95	2.43	2.40	2.86	1.73	1.54
5	2.22	2.11	2.58	2.72	1.39	1.61
8	2.14	2.49	2.64	2.90	1.40	1.65

Table 34.--Effects of night temperatures, rates of potassium and iron applications on percent oxalic acid content of leaves of centipedegrass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied,</u>		<u>Fe-applied,</u>	
	<u>lbs.per 1000</u>		<u>lbs.per 1000</u>	
	<u>sq. ft.</u>		<u>sq. ft.</u>	
	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>				
2	1.28	1.23	2.89	2.67
5	1.14	1.29	2.22	2.60
8	1.15	1.24	2.60	2.75
<u>Night temp. 7.2° C.</u>				
2	1.54	1.61	2.85	3.15
5	1.84	1.95	3.02	2.85
8	1.65	1.61	2.88	3.24
<u>Night temp. -1.1° C.</u>				
2	1.95	2.43	2.74	3.05
5	2.22	2.11	2.82	3.17
8	2.14	2.49	2.64	2.98

Table 35.--Effects of night temperatures, rates of potassium and iron applications on percent oxalic acid content of leaves, stolons and roots of centipedegrass of experiment 5.

K-applied lbs.per 1000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied lbs.		Fe-applied lbs.		Fe-applied lbs.	
	per 1000 sq.ft.	per 1000 sq.ft.	per 1000 sq.ft.	per 1000 sq.ft.	per 1000 sq.ft.	per 1000 sq.ft.
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.190	.326	.260	.272	.068	.058
5	.166	.432	.352	.108	.078	.012
8	.418	.352	.307	.317	.105	.085
<u>Night temp. 7.2° C.</u>						
2	.402	.385	.165	.352	.123	.103
5	.242	.200	.217	.237	.053	.120
8	.448	.334	.117	.270	.055	.048
<u>Night temp. -1.1° C.</u>						
2	.342	.390	.190	.190	.115	.132
5	.280	.252	.280	.347	.113	.148
8	.446	.218	.498	.463	.132	.199

Table 36.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent oxalic acid content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1,000 sq. ft.	Summer 1964		Winter 1964	
	Fe-applied, per 1,000 sq. ft.		Fe-applied, per 1,000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>				
2	.071	.130	.258	.312
5	.082	.212	.172	.406
8	.222	.164	.382	.377
<u>Night temp. 7.2° C.</u>				
2	.219	.181	.396	.319
5	.206	.125	.289	.238
8	.050	.028	.399	.351
<u>Night temp. -1.1° C.</u>				
2	.247	.146	.263	.268
5	.217	.155	.323	.289
8	.232	.076	.332	.245

Table 37.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent citric acid content in centipede-grass of experiment 5.

K-applied lbs. per 1000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1000 sq. ft.		per 1000 sq. ft.		per 1000 sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.192	.158	.109	.134	.147	.174
5	.144	.296	.107	.083	.127	.098
8	.150	.092	.144	.138	.108	.088
<u>Night temp. 7.2° C.</u>						
2	.276	.372	.076	.167	.650	.346
5	.320	.250	.190	.248	.203	.350
8	.268	.294	.170	.070	.162	.026
<u>Night temp. -1.1° C.</u>						
2	.200	.190	.188	.208	.343	.237
5	.222	.190	.272	.239	.332	.196
8	.220	.172	.288	.169	.127	.272

Table 38.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent citric acid content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1,000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	Fe-applied, per 1,000 sq. ft.		Fe-applied, per 1,000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>				
2	.063	.208	.200	.190
5	.112	.220	.222	.190
8	.251	.276	.220	.172
<u>Night temp. 7.2° C.</u>				
2	.116	.162	.276	.372
5	.168	.090	.320	.250
8	.048	.027	.268	.294
<u>Night temp. -1.1° C.</u>				
2	.154	.082	.192	.158
5	.175	.190	.144	.296
8	.199	.126	.150	.092

Table 39.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons, roots percent malic acid content in centipedegrass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000 sq. ft.		per 1,000 sq. ft.		per 1,000 sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.230	.344	.307	.292	.039	.154
5	.369	.376	.352	.289	.240	.118
8	.379	.404	.297	.237	.081	.091
<u>Night temp. 7.2° C.</u>						
2	.250	.384	.147	.212	.420	.205
5	.384	.308	.162	.260	.056	.138
8	.384	.378	.207	.170	.145	.120
<u>Night temp. -1.1° C.</u>						
2	.306	.275	.200	.245	.089	.030
5	.254	.305	.244	.207	.065	.028
8	.199	.166	.226	.246	.042	.022

Table 40.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves malic acid content in centipedegrass of experiments 4 and 5.

K-applied, lbs. per 1000 sq. ft.	<u>Summer 1964</u>		<u>Winter 1964</u>	
	<u>Fe-applied</u>		<u>Fe-applied</u>	
	lbs.per 1000	sq. ft.	lbs. per 1000	sq. ft.
	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>				
2	.406	.143	.230	.344
5	.121	.220	.369	.376
8	.123	.231	.379	.404
<u>Night temp. 7.2° C.</u>				
2	.433	.575	.250	.384
5	.542	.432	.384	.308
8	.217	.590	.384	.378
<u>Night temp. -1.1° C.</u>				
2	.365	.168	.306	.275
5	.420	.453	.254	.305
8	.613	.416	.199	.166

Table 41.--Effects of night temperatures, seasons of the year, rates of potassium and iron applications on leaves percent tartaric acid content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	Summer 1964		Winter 1964	
	Fe-applied, lbs.per 1000 sq. ft.		Fe-applied, lbs.per 1000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>				
2	.110	.152	.132	.190
5	.096	.178	.078	.190
8	.140	.166	.212	.164
<u>Night temp. 7.2° C.</u>				
2	.087	.130	.192	.170
5	.146	.138	.164	.142
8	.150	.174	.170	.228
<u>Night temp. -1.1° C.</u>				
2	.170	.174	.220	.156
5	.166	.134	.114	.130
8	.134	.100	.116	.088

Table 42.--Effects of night temperatures, seasons of the year, rates of potassium applications on leaves percent aconitic acid content in centipedegrass of experiments 4 and 5.

K-applied lbs. per 1000 sq. ft.	Summer 1964		Winter 1964	
	Fe-applied,		Fe-applied,	
	lbs.per 1000 sq. ft.		lbs.per 1000 sq. ft.	
	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>				
2	.278	.483	.551	.619
5	.600	.653	.619	.621
8	.600	.645	.779	.646
<u>Night temp. 7.2° C.</u>				
2	.351	.382	.416	.620
5	.316	.395	.704	.658
8	.253	.193	.707	.767
<u>Night temp. -1.1° C.</u>				
2	.457	.318	.392	.342
5	.516	.436	.399	.357
8	.365	.605	.297	.354

Table 43.--Effects of night temperatures, rates of potassium and iron applications on leaves percent succinic acid content in centipedegrass of experiment 5.

K-applied lbs. per 1000 sq. ft.	Night temp. 18.0-25.0°C.		Night temp. 7.2° C.		Night temp. -1.1° C.	
	Fe-applied, lbs.per 1000 sq. ft.		Fe-applied, lbs.per 1000 sq. ft.		Fe-applied, lbs.per 1000 sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
2	.090	.110	.070	.255	.130	.168
	.098	.114	.074	.191	.170	.153
5	.085	.144	.198	.155	.232	.230
	.059	.160	.142	.165	.170	.270
8	.080	.060	.175	.187	.214	.308
	.040	.061	.170	.186	.187	.312

Table 44.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons, and roots percent aspartic acid in centipedegrass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000		per 1,000		per 1,000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.026	.026	.029	.040	.024	.028
5	.028	.037	.048	.015	.031	.047
8	.024	.028	.071	.082	.059	.098
<u>Night temp. 7.2° C.</u>						
2	.020	.018	.069	.066	.077	.080
5	.064	.026	.045	.045	.070	.080
8	.027	.023	.043	.024	.090	.057
<u>Night temp. -1.1° C.</u>						
2	.029	.037	.080	.076	.037	.045
5	.019	.020	.096	.093	.043	.068
8	.021	.018	.076	.066	.047	.049

Table 45.--Effects of night temperatures, rates of potassium and iron application on leaves, stolons and roots percent glutamic acid in centipedegrass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000		per 1,000		per 1,000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.056	.042	.019	.021	.014	.003
5	.072	.060	.021	.006	.011	.005
8	.042	.058	.007	.019	.011	.009
<u>Night temp. 7.2° C.</u>						
2	.067	.060	.019	.019	.023	.017
5	.162	.050	.019	.021	.022	.019
8	.050	.050	.026	.008	.022	.008
<u>Night temp. -1.1° C.</u>						
2	.040	.051	.035	.027	.017	.011
5	.039	.035	.019	.020	.017	.014
8	.028	.032	.022	.018	.011	.011

Table 46.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent glycine in centipede grass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000		per 1,000		per 1,000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.021	.014	.011	.019	.016	.026
5	.018	.019	.010	.012	.020	.023
8	.016	.019	.027	.030	.015	.028
<u>Night temp. 7.2° C.</u>						
2	.021	.018	.018	.019	.032	.020
5	.037	.016	.020	.015	.015	.020
8	.016	.016	.014	.008	.026	.016
<u>Night temp. -1.1° C.</u>						
2	.021	.022	.018	.020	.011	.006
5	.013	.025	.024	.024	.008	.007
8	.011	.010	.025	.024	.009	.009

Table 47.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent histidine in centipede grass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000		per 1,000		per 1,000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.022	.018	.101	.175	.096	.093
5	.030	.039	.092	.082	.105	.119
8	.022	.035	.135	.115	.093	.108
<u>Night temp. 7.2° C.</u>						
2	.020	.024	.091	.074	.096	.113
5	.081	.023	.080	.146	.119	.122
8	.023	.022	.127	.083	.128	.088
<u>Night temp. -1.1° C.</u>						
2	.080	.097	.050	.075	.096	.083
5	.055	.055	.075	.075	.104	.093
8	.049	.038	.074	.074	.093	.074

Table 48.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent isolencine content in centipede-grass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000		per 1,000		per 1,000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.032	.016	.034	.060	.027	.041
5	.042	.027	.065	.053	.061	.053
8	.015	.026	.052	.048	.061	.040
<u>Night temp. 7.2° C.</u>						
2	.028	.019	.023	.017	.033	.037
5	.049	.015	.013	.020	.029	.025
8	.018	.028	.051	.035	.025	.021
<u>Night temp. -1.1° C.</u>						
2	.028	.013	.017	.023	.012	.013
5	.006	.028	.033	.033	.008	.016
8	.007	.011	.030	.030	.013	.008

Table 49.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots on percent reducing sugar content in centipede grass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000 sq. ft.		per 1,000 sq. ft.		per 1,000 sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.34	.48	.76	1.35	.83	.74
5	.50	.20	.54	.68	.70	.38
8	.10	.20	.52	.78	.50	.33
<u>Night temp. 7.2° C.</u>						
2	.28	.40	.46	.42	.47	.90
5	.30	.70	.44	.66	.32	.42
8	.20	.30	.58	1.08	.44	.63
<u>Night temp. -1.1° C.</u>						
2	.54	.30	.34	.26	.69	.45
5	.56	.20	.46	.66	.74	.68
8	.30	.18	.42	.68	.70	.70

Table 50.--Effects of night temperatures, rates of potassium and iron applications on leaves, stolons and roots percent cold water extractable carbohydrate in centipedegrass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000 sq. ft.		per 1,000 sq. ft.		per 1,000 sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.38	.12	.24	1.05	1.28	.22
5	.20	.22	.16	1.04	.56	.87
8	.42	.32	.48	1.14	1.54	2.02
<u>Night temp. 7.2° C.</u>						
2	.72	.52	3.10	2.56	.71	.82
5	.60	.14	2.50	.42	.51	.59
8	.08	.04	.94	1.36	1.44	.43
<u>Night temp. -1.1° C.</u>						
2	.18	.42	1.22	2.18	.06	.33
5	.11	.80	1.92	1.53	.82	.36
8	.66	.67	1.00	2.54	.02	.74

Table 51.--Effects of night temperatures, rates of potassium and iron applications on the reductive capacity of root in centipedegrass of experiment 5.

K-applied lbs. per 1,000 sq. ft.	Leaves		Stolons		Roots	
	Fe-applied		Fe-applied		Fe-applied	
	per 1,000 sq. ft.		per 1,000 sq. ft.		per 1,000 sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
<u>Night temp. 18.0 - 25.0° C.</u>						
2	.08	.07	.09	.04	.13	.06
5	.10	.07	.11	.04	.15	.04
8	.09	.08	.07	.09	.08	.08
<u>Night temp. 7.2° C.</u>						
2	.07	.08	.13	.09	.20	.13
5	.05	.09	.13	.05	.13	.05
8	.08	.09	.05	.15	.05	.20
<u>Night temp. -1.1° C.</u>						
2	.08	.08	.10	.07	.18	.14
5	.16	.08	.26	.07	.36	.06
8	.07	.10	.05	.07	.09	.06

Table 52.--Effect of night temperatures, rates of potassium and iron applications on cation exchange capacity of roots in centipedegrass of experiment 5.

K-applied, lbs. per 1000 sq. ft.	Night temp. 18.0-25.0°C.		Night temp. 7.2°C.		Night temp. -1.1°C.	
	Fe-applied,		Fe-applied,		Fe-applied,	
	lbs.per 1000		lbs.per 1000		lbs.per 1000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
2	11.0	7.0	4.5	5.5	6.0	7.0
	9.8	7.0	6.6	5.9	7.0	7.5
Average	10.4	7.0	5.5	5.8	6.5	7.3
5	7.2	6.8	7.8	4.8	4.4	6.6
	7.9	6.9	7.2	7.0	6.2	6.2
Average	7.5	6.9	7.3	5.9	5.3	6.4
8	5.0	7.4	6.6	6.9	5.1	6.0
	6.8	5.8	6.8	7.9	5.0	6.3
Average	5.9	6.6	6.7	7.4	5.1	6.2

Table 53.---Chemical analyses of Arredondo loamy fine sand at the conclusion of experiments 4 and 5.

K-applied lbs. per 1000sq.ft.	Summer 1964										Winter 1964									
	Extractable nutrients* ppm										Extractable nutrients ppm									
	Soil					pH (6) (3)					Soil					pH (6) (3)				
	Fe	Fe	K	Ca	Mg P	Fe	Fe	K	Ca	Mg P	Fe	Fe	K	Ca	Mg	P				
2	0.0	6.3	5.1	18.9	48	2091	92	13	6.2	5.92	14.31	36	856	133	22					
5	0.1	6.2	6.9	15.1	62	2623	131	22	6.3	7.26	9.00	48	939	154	25					
	0.0	5.9	7.6	18.3	223	2183	75	14	6.3	6.94	10.71	89	908	138	19					
	0.1	6.2	8.7	18.0	143	1557	94	17	6.2	8.20	14.97	114	826	120	22					
8	0.0	6.5	4.8	11.0	277	1659	80	17	6.4	7.42	11.67	164	885	120	22					
	0.1	6.0	4.6	14.9	410	2856	137	18	6.0	10.12	15.18	185	868	116	23					
Night temp. 18.0-25.0° C.																				
Night temp. 7.2° C.																				
2	0.0	6.5	6.0	12.8	20	1639	82	11	6.4	7.20	14.31	23	865	144	24					
	0.1	6.1	9.1	19.9	32	1843	86	9	6.5	10.12	20.64	17	960	156	25					
5	0.0	6.2	3.3	11.0	145	1757	106	28	6.3	3.54	10.41	88	910	108	23					
	0.1	6.4	7.3	19.9	83	2151	77	27	6.3	4.62	17.33	62	874	82	21					
8	0.0	6.3	2.9	10.4	237	2014	92	16	6.0	3.06	11.64	167	770	116	20					
	0.1	6.0	4.1	13.2	271	1935	127	12	6.3	3.94	15.33	206	793	82	22					
Night temp. -1.1° C.																				
2	0.0	6.2	5.4	11.6	28	1927	99	19	6.3	7.12	18.30	21	890	114	23					
	0.1	6.5	4.2	14.2	17	1463	52	12	6.4	4.20	20.64	18	946	105	23					
5	0.0	6.3	2.3	12.1	49	1367	61	13	6.5	4.42	11.64	31	818	69	21					
	0.1	6.3	5.9	18.9	50	1243	63	15	6.3	5.62	16.71	39	708	69	18					
8	0.0	6.8	2.8	10.9	112	1254	73	14	6.6	4.76	12.24	112	763	82	18					
	0.1	6.1	5.7	13.3	154	1162	38	14	6.4	4.90	11.31	146	852	75	22					

*Extracted with N ammonium acetate, buffered at pH 4.8, except Fe(6) was buffered at pH 6.0, and Fe(3) was buffered at pH 3.0.

Table 54.--Soil nitrate (ppm) determined at the conclusion of experiment 5.

K-applied lbs. per 1000 sq. ft.	Night temp. 18.0-25.0 C.		Night temp. 7.2° C.		Night temp. -1.1° C.	
	Fe-applied		Fe-applied		Fe-applied	
	lbs.per 1000		lbs.per 1000		lbs.per 1000	
	sq. ft.		sq. ft.		sq. ft.	
	0.0	0.1	0.0	0.1	0.0	0.1
2	1.5	1.3	2.5	2.0	21.0	16.5
	1.5	1.3	1.5	2.5	20.0	11.5
	1.3	1.3	1.5	1.5	19.5	15.0
5	1.0	1.3	1.3	3.5	20.0	13.5
	1.5	1.3	1.3	3.5	13.5	3.0
	1.3	2.0	2.0	3.5	19.5	5.5
8	1.3	1.3	1.5	2.0	5.5	3.5
	1.5	1.3	1.5	1.5	16.5	5.0
	1.3	1.3	2.0	2.0	10.5	7.5

BIBLIOGRAPHY

1. Alexander, M. Introduction to Soil Microbiology. John Wiley and Sons, Inc., New York, pp. 387-400. 1961.
2. Apple, S.B., Jr., and Butts, J.S. Soil temperature studies: I. The effect of soil temperature and phosphorus on growth and phosphorus uptake by pole beans. Amer. Soc. Hort. Sci. Proc. 61: 325-332. 1953.
3. Arnon, D.I. Phosphorus and the biochemistry of photosynthesis. Agrochimica 3: 108-139. 1959.
4. Association of Official Agricultural Chemists, Official and Tentative Methods of Analysis. Washington, D.C., ed. 6. 1945.
5. Barber, S.A. A diffusion and mass flow concept of soil nutrient availability. Soil Sci. 93: 39-49. 1962.
6. Barker, J., and Mapson, L.W. Studies in the respiration and carbohydrate metabolism of plant tissues. J. Expt. Bot. 15: 272-283. 1964.
7. Baxter, P., and Belcher, R. The role of the bicarbonate ion in the lime-induced chlorosis. J. Austral. Agr. Sci. 21: 32-34. 1955.
8. Bell, W.O., Bogarad, L., and McGrath, W.J. Response of yellow stripe maize mutant (ys.) to ferrous and ferric iron. Bot. Gaz. 120: 36-39. 1958.
9. Benson, A.A., and Calvin, M. The path of carbon in photosynthesis: VII. Respiration and Photosynthesis. J. Expt. Bot. 1: 63-68. 1950.

10. Berger, K.C., Potterton, P.E., and Hobson, E.L. Yield, quality, and phosphorus uptake of potatoes as influenced by placement and composition of potassium fertilizer. *Amer. Potato J.* 38: 272-285. 1961.
11. Biddulph, O. Translocation of minerals in plants. In *Mineral Nutrition of Plants*. Truog, ed., Univ. of Wisc. Press, Madison, Wisc., pp. 261-275. 1955.
12. _____, and Cory, R. Translocation of C¹⁴ metabolites in the phloem of bean plant. *Plant Physiol.* 40: 119-129. 1965.
13. Block, R.J., Durrum, E.L., and Zweig, G. *A Manual of Paper Chromatography and Paper Electrophoresis*. Ed. 2, John Wiley and Sons, Inc., New York, pp. 110-154, 215-229. 1958.
14. Bogorad, L. Factors associated with the synthesis of chlorophyll in the dark in seedlings of pinus. *Bot. Gaz.* 111: 221-241. 1949.
15. Bollard, E.G. Translocation of organic nitrogen in the xylem. *Austral. J. Biol. Sci.* 10: 292-301. 1957.
16. Bolle-Jones, E.W. The interrelations of iron and potassium in the potato plant. *Plant and Soil* 6: 129-173. 1955.
17. Bonner, J., and Galston, A.W. *Principle of Plant Physiology*. John Wiley and Sons, Inc., New York, pp. 11-44. 1952.
18. Bradfield, R., Batjer, L.P., and Oskamp, J. Soil in relation to fruit growing in New York: Part IV. The significance of the oxidation-reduction potential in evaluating soils for orchard purposes. *Cornell Univ. Agr. Expt. Sta., Bull.* 592. 1934.
19. Branton, D., and Jacobson, L. Iron localization in pea plants. *Plant Physiol.* 37: 546-551. 1962.
20. Brey, W.S., Jr. *Principles of Physical Chemistry*. Appleton-Century-Crofts, Inc., New York. 1958.

21. Brown, A.L., Yamaguchi, S., and Loal-liaz, Jaime. Evidence for translocation of iron in plants. *Plant Physiol.* 40: 35-38. 1965.
22. Brown, E.M. Some effect of temperature on the growth and chemical composition of certain pasture grasses. *Mo. Agr. Expt. Sta., Res. Bull.* 299. 1939.
23. _____. Seasonal variation in the growth and chemical composition of Kentucky bluegrass. *Mo. Agr. Expt. Sta., Res. Bull.* 360. 1943.
24. Brown, J.C. Iron Chlorosis. *Ann. Rev. Plant Physiol.* 7: 171-190. 1956.
25. _____. An evaluation of bicarbonate induced iron chlorosis. *Soil Sci.* 89: 246-247. 1960.
26. _____, Holmes, R.S., Shapiro, R.E., and Specht, A.W. Effect of phosphorus and copper salts on iron chlorosis of rice in flooded and non-flooded soil and the associated enzymatic activity. *Soil Sci.* 79: 363-371. 1955.
27. _____, Lunt, O.R., Holmes, R.S., and Tiffin, L.O. The bicarbonate ion as an indirect cause of iron chlorosis. *Soil Sci.* 88: 260-266. 1959.
28. _____, Holmes, R.S., and Tiffin, L.O. Iron chlorosis in soybeans as related to the genotype of rootstalk. *Soil Sci.* 86: 75-82. 1958.
29. _____, Holmes, R.S., and Tiffin, L.O. Hypotheses concerning iron chlorosis. *Soil Sci. Soc. Amer. Proc.* 23: 231-234. 1959.
30. Browne, C.A. *Physical and Chemical Method of Sugar Analysis.* Ed. 3, John Wiley and Sons, Inc., New York. 1941.
31. Broyer, T.C. The nature of the process of inorganic solute accumulation in roots. In *Mineral Nutrition of Plant.* Troug, ed., Univ. of Wisc. Press, Madison, Wisc., pp. 187-245. 1951.

32. Bryan, O.C. Yellowing of centipedegrass and its control. Fla. Agr. Expt. Sta., Press Bull. 450. 1933.
33. Burström, H. Growth regulation by metals and chelates. In Advan. in Bot. Res. 1: 73-98. 1963.
34. _____. The mechanism of ion absorption. In Mineral Nutrition of Plant. Broyer, ed., Cornell Univ. Press, Ithaca, New York. 1951.
35. Burtch, L.M., Thorne, D.W., and Wann, F.B. The effect of light, soil temperature, and soil moisture on high-lime chlorosis. Soil Sci. Soc. Amer. Proc. 13: 394-398. 1948.
36. Carell, E.F., and Price, C.A. Porphyrin and the iron requirement for chlorophyll formation in Euglena. Plant Physiol. 40: 1-7. 1965.
37. Carroll, J.C. Effect of drought, temperature, and nitrogen on turf grasses. Plant Physiol. 18: 19-36. 1943.
38. Conn, E.E., and Stumpt, P.K. Outline of Biochemistry. John Wiley and Sons, Inc., New York, pp. 24-48, 245. 1963.
39. Cooil, B.J. Potassium deficiency and excess in guayule: II. Cation-anion balance in the leaves. Plant Physiol. 23: 403-424. 1948.
40. _____, and Slattery, M.C. Effect of potassium deficiency and excess upon certain carbohydrates and nitrogenous constituents in guayule. Plant Physiol. 23: 425-439. 1948.
41. Cooper, H.P., Mitchell, J.W., and Page, H.H. The relation of the energy properties of soil nutrients to the chemical composition of crop plants. Soil Sci. Soc. Amer. Proc. 12: 364-369. 1947.
42. Cram, D.J., and Hammond, G.S. Organic Chemistry. McGraw-Hill Book Company, Inc., New York. 1959.

43. Curtis, C.F. Studies on solute translocation in plants. Experiments indicating that translocation is dependent on the activity of living cells. *Amer. J. Bot.* 16: 154-168. 1929.
44. _____, and Herty, S.D. The effect of temperature on translocation from leaves. *Amer. J. Bot.* 23: 528-532. 1936.
45. Davidson, O.W. Effect of temperature and growth and flower production of gardenia. *Amer. Soc. Hort. Sci. Proc.* 39: 387-390. 1941.
46. DeKock, P.C. Iron nutrition of plants at high pH. *Soil Sci.* 79: 167-175. 1955.
47. _____, and Morrison, R.I. The mechanism of chlorotic leaves: 1. Amino acids; 2. Organic acids. *Biochem. J.* 70: 266-277. 1958.
48. Doney, R.C., Smith, R.L., and Wiebe, H.H. Effects of various levels of bicarbonate, phosphorus, and pH on the translocation of foliar-applied iron in plants. *Soil Sci.* 89: 269-275. 1960.
49. Drake, M. Soil chemistry and plant nutrition. In *Chemistry of Soils*. Ed. 2, Bear, ed., Reinhold Pub. Corp., New York, pp. 395-444. 1964.
50. _____, Jones, V., and Colby, W.G. Cation exchange capacity of plant roots. *Soil Sci.* 72: 139-149. 1951.
51. Dunham, C.W., Hamner, C.L., and Asen, S. Cation-exchange properties of the roots of some ornamental plant species. *Amer. Soc. Hort. Sci. Proc.* 68: 556-563. 1956.
52. Ehrler, W., and Bernstein, L. Effect of root temperature, mineral nutrition, and salinity on the growth and composition of rice. *Bot. Gaz.* 120: 67-74. 1958.
53. Elliott, D.C. Detection of glycolic acid in etiolated barley shoots. *J. Expt. Bot.* 5: 353-356. 1954.

54. Epstein, E., and Stout, P.R. The micronutrient cations: iron, manganese, zinc, and copper; their uptake by plants from the adsorbed state. *Soil Sci.* 72: 47-65. 1951.
55. Evans, H.L. The biochemical role of iron in plant metabolism in mineral nutrition of trees. In *Mineral Nutrition of Trees*. Duke Univ., School of Forestry Bull. 15. 1959.
56. Franzen, H., and Helwert, F. The chemical constituents of green plants: XXII. The presence of succinic acid and of oxalic acid in the Currant (*Ribes Rubrum*). *Physiol. Chem.* 124: 65-74. 1922. Abstract in (London) *Chem. Soc. Jour.* 124(1): 77. 1923.
57. _____, and Keyssner, E. The chemical constituents of green plants: XXIII. Malic acid in plants. *Biochem. Ztschr.* 135: 183-216. 1923. Abstracts in (London) *Chem. Soc. Jour.* 124(1): 427. 1923.
58. Fried, M., and Shapiro, R.E. Soil-plant relationships in ion uptake. *Ann. Rev. Plant Physiol.* 12: 91-112. 1961.
59. Gibert, S.G., Shear, C.B., and Cropp, C.A. The effect of the form of nitrogen and the amount of base supply on the organic acid of tung leaves. *Plant Physiol.* 26: 750-756. 1951.
60. Clauser, R., and Jenny, H. Two-phase studies on availability of iron in calcareous soils. *Agrochimica* IV, no. 4, Univ. of Cal., Berkely, Cal. Sept., 1960.
61. Goodall, D.W., and Gregory, F.G. Chemical composition of plants as an index to their nutrient status. *Imp. Bur. Hort. and Plant Crops, Tech. Comm.* no. 17. 1947.
62. Hall, W.C. Growth and development of buckwheat under differential temperature gradients. *Bot. Gaz.* 111: 331-342. 1949.

63. Hartt, C.E. Some effects of potassium upon the growth of sugar cane and upon the absorption and migration of ash constituents. *Plant Physiol.* 9: 399. 1934.
64. Hellmers, H. Effects of soil and air temperatures on growth of Redwood seedling. *Bot. Gaz.* 124: 172-177. 1962.
65. Hewitt, E.J., and Belle-Jones, E.W. Iron deficiency in crops: II. The interrelationships of iron and potassium in the potato plant. *J. Hort. Sci.* 28: 185-195. 1953.
66. Hewitt, S.P., and Curtis, O.F. The effect of temperature on loss of dry matter and carbohydrate from leaves by respiration and translocation. *Amer. J. Bot.* 35: 746-765. 1948.
67. Hoagland, D.R., and Arnon, D.I. The water culture method for growing plants without soil. *Agr. Expt. Sta., Calif. Circ.* 347. 1938.
68. _____, and Broyer, T.C. General nature of the process of salt accumulation by roots description of experimental methods. *Plant Physiol.* 11: 471-507. 1936.
69. Hodgson, J.F. Chemistry of the micronutrient elements in soils. *Advan. Agron.* 15: 119-159. 1963.
70. Hoffer, G.W., and Carr, R.H. Accumulations of aluminum and iron compounds in corn plants and its probable relation to root rots. *J. Agr. Res.* 23: 801-823. 1923.
71. Holmes, R.S., and Brown, J.C. Iron and soil fertility. In U.S.D.A. Yearbook, pp. 111-115. 1957.
72. Horn, G.C. Yellowing of turfgrass. *Florida Turfgrass Association* 10: 1-5. 1963.
73. _____, and Pritchett, W.L. The relationship of fertilization to various turf disorders. *Soil and Crop Sci. Soc. Fla. Proc.* 22:107-115. 1962.

74. Ignatieff, V. Determination and behavior of ferrous iron in soils. *Soil Sci.* 51: 249-265. 1941.
75. Iljin, V.A. Salts and organic acids in relation to calciophiles *Flora* 37: 265-299. 1944.
76. _____. Metabolism of plants affected with lime-induced chlorosis (calciöse). *Plant and Soil* 3: 339-351. 1951.
77. _____. Metabolism of plants affected with lime-induced chlorosis: III. Mineral elements. *Plant and Soil* 4: 11-28. 1952.
78. Jackson, M.L. Soil chemical analysis. Prentice-Hall, Inc., Englewood Cliffs, N.J., pp. 197-199, 391-394. 1958.
79. _____. Chemical composition of soils. In *Chemistry of the Soil*, ed. 2, Bear, ed. Reinhold Pub. Corp., New York, pp. 71-141. 1964.
80. Jacobson, L. Iron in the leaves and chloroplasts of some plants in relation to their chlorophyll content. *Plant Physiol.* 20: 233-245. 1945.
81. _____, and Certli, J.J. The relation between iron and chlorophyll contents in chlorotic sunflower leaves. *Plant Physiol.* 31: 199-204. 1956.
82. _____, and Ordin, L. Organic acid metabolism and ion absorption in roots. *Plant Physiol.* 29: 70-75. 1954.
83. _____, Overstreet, R., Carlson, R.M., and Chastain, J.A. The effect of pH and temperature on the absorption of potassium and bromide by barley roots. *Plant Physiol.* 32: 658-662. 1957.
84. Jenny, H., and Overstreet, R. *Soil Sci.* 47: 257-272. 1939.
85. Johnson, C.M., and Ulrich, A. Analytical methods. *Cal. Agr. Expt. Sta. Bull.* 766: 66-67. 1959.

86. Jones, L.H. Relation of soil temperature to chlorosis of Gardenia. J. Agr. Res. 57: 611-621. 1938.
87. Kapp, L.C. Study of relation of growth to nutrition of the rice plant. Ark. Agr. Expt. Sta., Bull. 335. 1936.
88. Kliman, S. The importance of ferrous iron in plants and soils. Soil Sci. Soc. Amer. Proc. 2: 385-392. 1937.
89. Kursanov, A.L. Metabolism and the transport of organic substances in the phloem. Advan. in Bot. Res. I, pp. 209-274. 1963.
90. Kurtz, E.B., and Maier, R.H. Acid chlorosis and iron uptake by soybean grown in solution culture. Agron. J. 52: 486-487. 1960.
91. Laties, G.G. Active transport of salt into plant tissue. Ann. Rev. Plant Physiol. 10: 87-112. 1959.
92. Laurence, J.M., Miller, J.U., and Rasmussen, L.W. Estimation of amino acids in plant tissue by paper chromatograph. Wash. Agr. Expt. Sta., Technical Bull. 32. 1960.
93. Lawton, K., and Cook, R.L. Potassium in plant nutrition. Advan. Agron. VI, pp. 253-298. 1954.
94. Lindner, R.C., and Harley, C.P. Nutrient interrelations in lime-induced chlorosis. Plant Physiol. 19: 420-429. 1944.
95. Lingle, J.C., Tiffin, L.O., and Brown, J.C. Iron uptake-transport of soybeans as influenced by other cations. Plant Physiol. 38: 71-76. 1963.
96. Loustalot, A.J., Gilbert, S.G., and Drosdoff, H. The effect of nitrogen and potassium levels in tung seedlings on growth, apparent photosynthesis and carbohydrate composition. Plant Physiol. 25: 394-412. 1950.

97. Lundegardh, H. Anion respiration, the experimental basis of theory of absorption, transport, and exudation of electrolytes by living cells and tissues. Symp. Soc. Exp. Biol. 8: 262-296. 1954.
98. _____. Mechanisms of absorption, transportation, accumulation and secretion of ions. Ann. Rev. Plant Physiol. 6: 1-24. 1955.
99. Machlis, L., and Torrey, J.G. Plants in Action. W.H. Freeman and Company, San Francisco, Cal. 1956.
100. McGeorge, W.T. Lime-induced chlorosis: The relation between active iron and citric and oxalic acids. Soil Sci. 68: 381-390. 1949.
101. _____, and Breazeale, E.L. Application of Neubauer technique and applied potential to the study of immobilization of iron in plants. Soil Sci. 82: 329-336. 1956.
102. Mehlich, A. Improvement in the colorimetric magnesium and ammonium methods with sodium polysacrylate. Assoc. Off. Agr. Chem. Jour. 39: 518-523. 1956.
103. Miller, C.D., Ross, W., and Louis, L. Hawaiian-grown vegetables proximate composition: calcium; phosphorus; total iron; available iron; and oxalate content. Hawaii Agr. Expt. Sta., Tech. Bull. 5: 45-50. 1947.
104. Motomura, S. The relationship between ferrous iron formation and nitrogen metabolism in soil. Soil Sci. and Plant Nutr. 8: 177-185. 1962.
105. Nakanigawa, K., and Okuda, A. The role of potassium in plant physiology. J. Sci. Soil (Tokyo) 31: 279-280. 1960.
106. Nelson, C., and Gorham, P. Uptake and translocation of C^{14} labelled sugars applied to primary leaves of soybean seedling. Canad. J. Res. (Botany) 35: 339-347. 1957.

107. _____, and Gorham, P. Translocation of C¹⁴ labelled amino acids and amides in the stems of young soybean plants. *Canad. J. Res. (Botany)* 37: 431-438. 1959.
108. Nielson, K.F., Halstead, R.L., Maclean, A.J., Holmes, R.M., and Bourget, S.J. The influence of soil temperature on the growth and mineral composition of oats. *Canad. J. Soil Sci.* 40: 255-263. 1960.
109. Nitrate accumulation and toxicity. *Agron. Mimeo* No. 64-66, Dept. Agron., Cornell Univ., Ithaca, New York. April, 1963.
110. Noggle, G.R. Nitrogen transformation and utilization in plants. *Soil and Crop Sci. Soc. Fla. Proc.* 21: 255-261. 1961.
111. Oades, J.W. The nature and distribution of iron compounds in soils. *Soils and Fert.* XXVI, pp. 69-78. 1963.
112. Ordin, L., and Jacobson, L. Inhibition of ion absorption and respiration in barley roots. *Plant Physiol.* 30: 21-27. 1955.
113. Oserkowsky, J. Quantitative relation between chlorophyll and iron in green and chlorotic pear leaves. *Plant Physiol.* 8: 449-467. 1933.
114. Peach, M., and English, L. Rapid microchemical soil test. *Soil Sci.* 57: 167-195. 1944.
115. Pesur, N.G., Smith, R.L., and Wiebe, H.H. Effect of iron chlorosis on protein fraction of corn leave tissue. *Plant Physiol.* 36: 736-739. 1961.
116. Price, C.A., and Carell, E.F. Control by iron of chlorophyll formation and growth in Euglena gracilis. *Plant Physiol.* 39: 862-868. 1964.
117. Priestley, C.H.B. Heat conduction and temperature profiles in air and soil. *Austral. Inst. Agr. Sci. J.* 25: 94-107. 1959.

118. Pritchett, W.L., and Horn, G.C. Turf fertilization research. Turf Management Conference X, pp. 209-215. 1962.
119. Ponnampeuma, F.N. The chemistry of submerged soils in relation to the growth and yield of rice. Ph.D. Thesis, Cornell Univ., Ithaca, New York, Univ. Microfilm 11916, Ann Arbor, Mich. 1955.
120. Richards, S.J., Hagan, R.H., and McCalla, T.M. Soil temperature and plant growth. In Soil Physical Conditions and Plant Growth. Shaw, ed., Academic Press, Inc., New York, Agron. 2: 303-462. 1952.
121. Roberts, A.N., and Kenworthy, A.L. Growth and composition of the strawberry plant in relation to root temperature and intensity of nutrition. Amer. Soc. Hort. Sci. Proc. 68: 157-168. 1956.
122. Saric, M., and Curic, R. The uptake of P. Ca. S. and Fe. by the plants in dependence of temperature. Agrochimica 7: 173-184. 1963.
123. Schmid, W.E., and Gerloff, G.C. A naturally occurring chelate of iron in xylem exudate. Plant Physiol. 36: 226-231. 1961.
124. Shear, C.E., Crane, H.L., and Specht, A.W. Paper presented before Amer. Soc. Plant Physiol., Columbus, Ohio. Sept., 1950.
125. Sherman, G.D. Manganese and soil fertility. U.S.D.A. Yearbook, pp. 137-139. 1957.
126. Sideris, C.P., and Young, H.Y. Effects of iron on chlorophyllous pigments, ascorbic acid, acidity, and carbohydrates of Ananas comosus (L.) Merr., supplied with nitrate or ammonium salts. Plant Physiol. 19: 52-75. 1944.
127. _____, and Young, H.Y. Effect of potassium on chlorophyll, acidity, ascorbic acid, and carbohydrates of Ananas comosus (L.) Merr. Plant Physiol. 20: 649-670. 1945.

128. Smith, R.L., and Wallace, A. Influence of cation ratio, temperature, and time on adsorption absorption of calcium and potassium by citrus and other plant species. *Soil Sci.* 82: 9-19. 1956.
129. Smythe, C.V., and Schmidt, C.L.A. Studies on the mode of combination of iron with certain proteins, amino acids, and related compounds. *J. Biol. Chem.* 88: 241-268. 1930.
130. Somers, L.I., and Shive, J.W. The iron manganese relation in plant metabolism. *Plant Physiol.* 17: 582-602. 1942.
131. Starkey, R.L. Relations of micronutrients to development of microorganisms. *Soil Sci.* 79: 1-13. 1955.
132. Steinberg, R.A. Correlations between protein-carbohydrate metabolism and mineral deficiencies in plants. In *Mineral Nutrition of Plants*, Trought, ed., Univ. of Wisc. Press, Madison, Wisc. 1951.
133. Su, L.Y., and Miller, G.W. Chlorosis in higher plants as related to organic acid content. *Plant Physiol.* 36: 415-420. 1961.
134. Sutcliffe, J.F. Factors affecting salt absorption. In *Mineral Salt Absorption in Plants*. Pergamon Press, Oxford, New York, pp. 47-71. 1962.
135. _____. Structural aspects of salt absorption in cells. In *Mineral Salt Absorption in Plants*. Pergamon Press, Oxford, New York, pp. 95-110. 1962.
136. Swanson, C., and El-shishing, E. Translocation of sugars in the concord grape. *Plant Physiol.* 33: 33-37. 1959.
137. Swanson, C.A. Translocation of organic solutes. In *Plant Physiology*. Steward, ed. 2: 481-551. 1959.
138. Thimann, K.V. The chemistry and physiology of growth. Parpart, ed., Princeton Univ. Press, Princeton, N.J., pp. 61-71. 1949.

139. Thomas, B., Eseritt, J.R., and Trinder, N. The minor elements of common heather Calluna vulgaris. Emp. J. Expt. Agr. 13: 93. 1945.
140. Thorne, D.W., Wann, F.B., and Robinson, W. Hypotheses concerning lime-induced chlorosis. Soil Sci. Soc. Amer. Proc. 15: 254-258. 1951.
141. Thrower, S.L. Translocation of labelled assimilates in the soybean: II. The pattern of translocation in intact and defoliated plants. Austral. J. Biol. Sci. 15: 629-649. 1962.
142. Traub, H.P., and Slattery, M.C. Analysis of levulins, inulin, and monosaccharides in guayule, Parthenium argentatum A. Gray. Bot. Gaz. 108: 295-299. 1946.
143. Truog, E. The determination of the readily available phosphorus of soils. J. Amer. Soc. Agron. 22: 874-882. 1930.
144. Ulrich, A. Metabolism of non-volatile organic acids in excised barley root as related to cation-anion balance during salt accumulation. Amer. J. Bot. 28: 526-537. 1941.
145. Wallace, A. Possible effects of root cation-exchange capacity on the relative amounts of Ca and K in plants. In Solute Uptake by Intact Plants. Edwards Bros., Inc., Ann Arbor, Mich., pp. 127-129. 1963.
146. _____. Effect of nutrient deficiencies on K, Na, and Fe uptake by Bush bean plants. In Solute Uptake by Intact Plants. Edwards Bros., Inc., Ann Arbor, Mich., pp. 191-104. 1963.
147. _____, and Sufi, S.M. Some investigations of factors which influence transfer of solutes from roots to shoots. In Solute Uptake by Intact Plants. Edwards Bros., Inc., Ann Arbor, Mich., pp. 33-39. 1963.

148. _____, Carnack, C., and Wallace, G.A. Ethylenediamine Di(o-Hydroxyphenyl Acetate) influence on Fe uptake and translocation in Bush bean. In Solute Uptake by Intact Plants. Edwards Bros., Inc., Ann Arbor, Mich., pp. 147-155. 1963.
149. _____, and Lunt, O.R. Iron chlorosis in horticultural plants. Am. Soc. Hort. Sci. Proc. 75: 819-841. 1935.
150. Ward, G.H. Potassium in plant metabolism: II. Effect of potassium upon the carbohydrates and mineral composition of potato plants. Canad. J. Plant Sci. 39: 243-252. 1959.
151. Weinstein, L.H., Robbins, W.R., and Perkins, H.F. Chelating agents and plant nutrition. Science 120: 41-42. 1954.
152. Went, F.W. Temperature. In Encyclopedia of Plant Physiology. Ruhland, ed., Springer-Verlag XVI, pp. 1-22. 1961.
153. _____, and Hull, H.A. The effect of temperature upon translocation of carbohydrate in the tomato plant. Plant Physiol. 24: 505-526. 1949.
154. Wilkie, G.W., and Miller, G.W. Iron nutrition of Nicotiana tabacum L. in relation to riboflavin-5-phosphate, and flavinadenine dinucleotide content. Plant Physiol. 35: 516-520. 1960.
155. Withrow, R.B. Modifying influences of various environmental factors upon mineral nutrition. In Mineral Nutrition of Plant. Truog, ed., Univ. of Wisc. Press, Madison, Wisc., pp. 384-410. 1955.

BIOGRAPHICAL SKETCH

Jon I. Teng was born October 19, 1930, at Kienow, Fukien, China. In August, 1954, he received the degree of Bachelor of Science in Agriculture from National Taiwan University. From 1955 to 1957 Mr. Teng taught chemistry and soil science at the Yilan Agriculture School, Taiwan. From 1957 until 1960 he was employed as a manager of one of the sugar cane research stations of Taiwan Sugar Corporation. In 1961 he enrolled in the Graduate School of the South Dakota State University. He worked as a graduate assistant in the Department of Agronomy until December, 1962, when he received the degree of Master of Science in Agronomy. In January, 1963, he enrolled in the Graduate School of the University of Florida. Mr. Teng worked as a graduate assistant in the Department of Soils from January, 1963, to the present time and has pursued his work toward the degree of Doctor of Philosophy.

Jon I. Teng is married to the former Chim-Miao Tsai and is the father of one child. He is a member of the American Society of Agronomy and Alpha Zeta Society.

This dissertation was prepared under the direction of the chairman of the candidate's supervisory committee and has been approved by all members of that committee. It was submitted to the Dean of the College of Agriculture and to the Graduate Council, and was approved as partial fulfillment of the requirements for the degree of Doctor of Philosophy.

August 14, 1965

Dean, College of Agriculture

Dean, Graduate School

Supervisory Committee:

Chairman

463.34
T292e

AGRI-
CULTURAL
LIBRARY

UNIVERSITY OF FLORIDA

3 1262 08553 7917