RESPONSE OF SORGHUM GENOTYPES TO IRON DEFICIENCY IN CALCAREOUS SOIL AND NUTRIENT SOLUTION

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CHAPTER I

ABSTRACT

Sorghum (Sorghum bicolor (L.) Moench) grown on calcareous soils is subject to a number of limiting factors, one of which is iron (Fe) deficiency. The American Great Plains, including Western Oklahoma, contain many calcareous soils where moderate to severe Fe-deficiency problems exist. Substantial sorghum yield losses in both grain weight and dry matter from Fe deficiency have been observed, but are not well documented and may occur even when symptoms are moderate. Nutrient solutions and calcareous soils have been used to screen sorghum cultivars for Fe efficiency however, across sorghum genotypes there have been varying degrees of response to these growth media. Research correlating leaf chlorophyll content with visual scores has established the usefulness of visual evaluation of Fe-deficiency chlorosis.

The objectives of this research were (1) to screen ten sorghum genotypes for iron (Fe) efficiency in the growth chamber using an Fe-deficient solution and in the greenhouse and field using a calcareous, Fe-deficient soil from Northwestern Oklahoma; and (2) to evaluate the effect of Fe deficiency on leaf chlorophyll content, plant dry matter, and grain weight of the ten genotypes grown under growth chamber, greenhouse, and field conditions, respectively.

Ten sorghum cultivars were grown in 1987 at the Southern Great

Plains Research Station, Woodward, Oklahoma, on two soils (a Woodward loam-Typic Ustochrepts, calcareous, and a Carey loam-Typic Argiustolls, non-calcareous) to determine their tolerance to iron-deficiency field by a visual evaluation method and the effect of Fe deficiency on grain weight. The same ten genotypes were grown simultaneously in a growth chamber, the same year, in two nutrient (+Fe and -Fe) solutions. In 1988, they were grown in a greenhouse in two soils one of which was collected from the Woodward trial site (Woodward loam) and the other from Perkins (a Teller loam-Udic Argiustolls). These growth media were used to evaluate the Fe-deficiency tolerance of those cultivars and the effect of Fe shortage on their plant biomass and leaf chlorophyll content. In 1988 the field experiment was repeated on Woodward loam only for the same objectives previously described except visual symptoms and leaf chlorophyll content were evaluated.

Narrow ranges of Fe-deficiency responses were obtained among genotypes grown in the three environments. However, some cultivars exhibited distinct chlorosis symptoms in the growth chamber and field (1987), indicating that -Fe solution and Woodward loam could be used to screen sorghum genotypes for Fe efficiency. The correlation of field chlorosis scores with those from the growth chamber under iron-deficient conditions was significant ($r = +0.64$) suggesting that field Fe-deficiency performances of new sorghum cultivars could be predicted using results found in the growth chamber. Significant linear relationships were found between greenhouse and field chlorosis scores, and leaf chlorophyll content of genotypes ($r = -0.56$ & -0.69), indicating that chlorophyll analysis could be substituted for or

supplement visual rating of chlorotic sorghum plants. Plant total dry matter was not significantly reduced by the iron stress in the -Fe solution or Fe-deficient soil, in the greenhouse. Nevertheless, reductions of at least 15% of leaf chlorophyll content were obtained for each cultivar grown on the Fe-deficient soil (greenhouse) or in the -Fe solution. Also a minimum reduction of 50% of plant grain weight was obtained for all cultivars grown in the field on the Fe-deficient soil.

CHAPTER II

LITERATURE REVIEW

Iron deficiency chlorosis, commonly known as lime-induced chlorosis or iron chlorosis, is a mineral element deficiency disorder often observed in plants grown on calcareous soils. Young leaves of a green plant become chlorotic if there is a shortage of iron (Fe) uptake while the older parts of the plant remain green due to the relative immobility of Fe in the plant. Iron-deficiency chlorosis appears on plant leaves because they do not contain sufficient active Fe for metabolism (Banin and Navrot, 1972; Dekok et al., 1979; Oserkowsky, 1933). Iron must be present for the chloroplast synthesis, but it is not a part of the chlorophyll molecule. It is used in nitrogen fixation, oxidative metobolism and electron transport in plants (Bates, 1982). Therefore, under Fe-deficiency conditions, plant growth is affected and roots are reduced in growth (Kannan, 1983).

Sorghum (Sorghum bicolor (L.) Moench) grown on carbonaceous alkaline or calcareous soils is subject to a number of limiting factors, one of which is the Fe deficiency. According to Chen and Barak (1982), the most widespread occurence of Fe deficiency in plants is found in calcareous soils which cover over 30 percent of the earth's land surface. Large acreages of soil that produce Fe-deficiency symptoms during the production of wheat (Triticum aestivum

L.), sorghum, soybean (Glycine max (L.) Merr.) and peanut (Arachis hypogea L.) have been identified in the American Great Plains and western third of Oklahoma (Clark, 1982a). Soil or foliar amendments are often the immediate cure for Fe deficiency in plants so that the crop will produce some yield. However, these corrective methods are not permanent solutions for correcting the disorder and usually not cost effective when considering the returns of the crop. Mortvedt and Giordano (1973) reported a doubling of forage sorghum yields and Fe uptake in sorghum plants from the band application of the combination of iron sulfate ($FeSO_4$) and ammonium polyphosphate. Datin and Westerman (1982) suggested the existence of a synergistic effect between phosphorus (P) and Fe in calcareous soils after getting an increase in grain sorghum yield when they jointly applied ammonium polyphosphate and $FeSO_4$. Regardless of the material used, Fe must be added frequently to maintain green plants because the residual effect of applied Fe amendments is low (Mortvedt and Giordano, 1971).

Sorghum genotypes have shown differences in their ability to remain green, grow and develop under Fe-deficient conditions (Brown and Jones, 1976; Esty et al., 1980). These genetic differences that influence the susceptibility of plants to Fe deficiency should hold a potential for developing plants with the ability to withstand low Fe environment. The successful identification of Fe-efficient sorghum genotypes in a controlled environment, greenhouse, or field conditions is necessary to solve the problem of Fe deficiency. Specific levels of tolerance have been difficult to identify and the level desired difficult to quantify. Visual scores assigned to Fe-chlorotic plants have been frequently used to assess the degree of Fe chlorosis. Clark

et al. (1982) and Williams et al. (1982) reported that visual scorings were essential as screening tools.

The effects of Fe deficiency on sorghum plants like reduction in yield, plant dry matter and leaf chlorophyll content, have not been intensively studied (Clark et al., 1982). Fe deficiency can vary over small areas due to environmental differences such as soil pH, partial pressure of carbon dioxide (pCO_2), bicarbonate (HCO_3), soil moisture, soil fertility, and others; thus complicating the evaluation of sorghum genotypes for Fe deficiency. It has also been observed (Clark et al., 1982) that with time from emergence to maturity most chlorotic sorghum plants may become green (regreening process) and eventually produce a harvestable product with no readily apparent visual deficiency problems in leaf color at maturity. Yields may be reduced, but the extent of the losses is not generally known. The severity of Fe deficiency has been reported to vary from season to season even on the same soils and on sites within fields (Clark et al., 1982). Numerous experiments in controlled environments have shown that nitrogen (N) nutrition can also influence the occurence of Fe chlorosis (Colgrove and Roberts, 1956; Cain, 1954). In a study to determine the effects of nitrogen source on Fe nutrition of sorghum, Bernado et al. (1984a and 1984b) produced a pH rise to near 7 using nitrate (NO_3) as the sole source of N and a decrease to near or below pH 4 after adding ammonium (NH_4) to a nutrient solution. Sorghum plants grown in NO_3 developed severe Fe chlorosis in the leaves whereas those in a mixture of NO_3 and NH_4 solution were normal and did not develop Fe chlorosis. They observed a higher dry matter yield for plants nourished with some NH_4 compared to those grown with NO_3 only

and explained the cause of Fe deficiency in plants as Fe not being translocated to leaves instead of lack of Fe uptake by roots. The alkalinization of the free space of plant tissues and neutralization of Fe^{++} by excess NO_3 (Mengel and Geurtzen, 1986) reduces the translocation of Fe to the leaves (Ulrich and Novacky, 1981).

McDaniel and Brown (1982) observed several distinct Fe-chlorosis levels among pure-line oat (Avena Sativa L.) genotypes using both field trials at Beeville, Maryland, and a controlled environment experiment. A Quinlan soil (Typic Ustochrepts) from Woodward, Oklahoma, and Tripp soil (Aridic Haplustolls) and Millville soil (Typic Haploxerolls) were used to induce Fe deficiency in plants in greenhouse and field experiments, respectively. Significant increases in dry matter yields resulted for two oat genotypes, 'Tam-O-312' and 'Coker', when either Fe or zinc (Zn) was added to Quinlan soil, and when Fe was added to Tripp soil.

Soybean genotypes are usually screened for Fe-chlorosis susceptibility in field nurseries with calcareous soil. However, results from fields may not always be consistent due to soil heterogeneity within the nursery and fluctuation of environmental conditions. In a study of Fe chlorosis in soybean, Al-Showk et al. (1986) found that an entry 'Essex' exhibited the least chlorotic symptoms of the cultivars tested in Quinlan soils. Also Essex had a higher chlorophyll concentration in leaves. Inskeep and Bloom (1986) pointed out that decreases in chlorophyll content were associated with increases in soil moisture, pCO_2 and solution bicarbonate when 'Anoka' soybean was grown on several calcareous soils.

Williams et al. (1982) found a fairly close relationship between

Fe-deficiency symptoms and dry matter when they used nutrient solution (modified Hoagland solution) to screen two groups of sorghum inbreds for Fe efficiency. A non-significant correlation coefficient below 0.3 was reported between dry weight and Fe-deficiency chlorosis scores. Based upon Fe-deficiency visual ratings as the criteria for evaluation of cultivars, 'Plainsman' was among the most tolerant to Fe-deficiency conditions and 'Redlan' among the most susceptible. 'Sc118-15E', Redlan, and 'Wheatland' developed Fe-deficiency symptoms more rapidly while 'Sc 33-9-8-E4', and 'BTX 623' remained green and tolerated Fe stress for a longer period of time. Clark and Gross (1986) also categorized Redlan and Sc118-15E as susceptible to Fe-deficiency.

Clark et al. (1982) used different potted calcareous soils maintained at field capacity and a calcium carbonate ($CaCO_3$) buffer solution to screen sorghum genotypes for Fe efficiency. They reported greater differences in dry matter yields among genotypes when they were grown in soils under greenhouse conditions than in nutrient solution culture. Many genotypes in Fe-deficient soils did not respond the same way they did in the nutrient solution. Nevertheless, they drew a conclusion that the differences among genotypes existed no matter what screening process was used and recommended the use of NO_3 as sole source of nitrogen in nutrient solutions to induce Fe-deficiency in plants. Pau (1987) reported that calcareous soil from Western Oklahoma could be used to screen sorghum cultivars for Fe efficiency in the growth chamber, and the use of ammonium polyphosphate contributed to an increase in dry matter and chlorophyll content of the fertilized sorghum plants grown in pots. This latter

result suggested a combined effect of phosphate and ammonium in making Fe more available to plants in calcareous soils.

McKenzie et al.(1984) visually evaluated sorghum genotypes for Fe-deficiency symptoms. They assigned a score to each plant on a daily basis ranking from 0 = no Fe-deficiency to 4 = severe Fe deficiency. The ranking was based on the plant overall appearance and development of chlorosis in the leaves. This was after plants were put into treatment solutions. They found a high correlation between the visual rating system and leaf chlorophyll content of sorghum plants grown under field and growth chamber conditions when evaluating sorghum cultivars for Fe chlorosis tolerance. They concluded that visual ratings could be substituted for chlorophyll analyses which are tedious and complex.

The objectives of this research were (1) to screen ten sorghum genotypes for Fe efficiency in the growth chamber, using an Fe-deficient solution, and in the greenhouse and field using a calcareous, Fe-deficient soil from Northwestern Oklahoma; and (2) to evaluate the effect of Fe deficiency on leaf chlorophyll content, plant dry matter, and grain weight of those cultivars grown in the above-mentioned environments.

CHAPTER III

MATERIALS AND METHODS

Field Experiments

Experiments were conducted from June to November, 1987, and June to September, 1988, at the Southern Great Plains Research Station, Woodward, Oklahoma, to evaluate ten grain sorghum genotypes ('OK 322', Wheatland, Redlan, 'ROKY 8', Plainsman, 'ROKY 40', 'ATX 622', '85-3026', '85-3062', '10H2416') in an Fe chlorosis-inducing field environment. The experiment of 1987 was carried out on two different soils, about 100 meters distant each from the other. The Fe-deficient area was a Woodward loam (Typic Ustochrepts), a calcareous soil known to induce Fe-deficiency in sorghum plants (Clark, 1982b; Nance and Gray, 1978). The Fe sufficient area, a Carey loam (Typic Argiustolls), had characteristics similar to the Woodward loam (Nance and Gray, 1978), but was sufficient in available Fe. In 1988, the same experiment was repeated on the Fe-deficient area only because the Fe sufficient area previously used was no longer available. The main plant available nutrients of those soils are listed in Table I. Both soils were treated similarly and amended with recommended amounts of nitrogen (N) based on yield goal of 408 Kg/ha (Johnson and Tucker, 1982) for sorghum cultivation, except iron. Supplemental water was added to sustain plant growth up to maturity.

An experimental unit consisted of a 3.6 m long by 1 m wide row in

the field and the treatments were the 10 sorghum genotypes. Each cultivar was planted at a rate of 14 seeds m^{-1}, then thinned

TABLE I

MAIN PLANT AVAILABLE NUTRIENTS AND PH OF SOILS USED IN EXPERIMENTS

	Teller loam (Typic Arguistolls)	Woodward loam (Typic Ustochrepts)	Carey loam (Typic Arguistolls)
pH	6.1	8.0	7.1
NO_3-N(kg ha^{-1})	90.7	19.0	80.7
P_2O_5+(kg ha^{-1})	128.8	32.5	50.4
K_2O+ (kg ha^{-1})	245.3	669.5	621.5
Fe# (mg kg^{-1})	23.7	0.0	5.1
Zn+ (mg kg^{-1})	0.6	0.6	0.8

\+ Melich III extractable
\# DPTA extractable

to 7 plants m^{-1} after emergence. The final density after thinning was 66,667 plants ha^{-1}. After one month of growth, five plants per row were randomly selected and evaluated for Fe chlorosis. The top three leaves of each selected plant were scored on alternate weeks with a modification of the visual evaluation system used by McKenzie et al. (1984) and Williams et al. (1982). A plant with no chlorotic leaves received a score of 1.0 and one expressing severe chlorosis was assigned a score of 5.0. Scores of 2.0, 3.0, or 4.0 were assigned to plants having one, two or three chlorotic leaves, respectively.

Grain weight (weight) of scored plants was determined at maturity for the 1987 experiment and leaf chlorophyll content (chlt) was evaluated three times before heading in the 1988 field experiment. Chlorosis scores, chlt and grain weight were subjected to statistical analyses based on the mean chlorosis score, leaf chlt and grain weight for five plants, respectively.

A split plot design and randomized complete block design (RCBD) consisting of four replications (Cochran and Cox, 1957) were used for the 1987 and 1988 field experiments, respectively. For the split plot, the main plots were the two locations (soils treated the same way as far as water and management were concerned) and the subplots were the 10 sorghum genotypes. Analysis of variance was performed on all genotypes for plant chlorosis score, grain weight and chlt. The replication x genotype, within treatment, and soil type x genotype interaction mean squares were used to test for genotype effect and effect of Fe deficiency, respectively.

Growth Chamber Experiment

Tray Preparation, Germination of Seeds and Growth Conditions

The same 10 sorghum genotypes used in the field were utilized in the growth chamber. Seeds were treated and germinated at 30°C between moist rolled paper towels in a germinator. Three day old-uniform sized seedlings of 10 genotypes were transferred to rectangular plastic trays having 90 holes each, consisting of ten rows and nine columns. The interval between two consecutive rows or columns was 2.5 cm. Trays used in the experiment were prepared to fit inside 10 L

plastic household dishpans containing nutrient solutions (Clark et al., 1982). All plants were grown for six days in 3.5 L of full strength nutrient solution (+Fe solution) containing 2.7 x 10^{-3} mM of Fe as $Fe(NO_3)_3.9H_2O$ (Table II).

TABLE II

COMPOSITION OF NUTRIENT SOLUTIONS

Solution Number	Salt	+Fe Solution Conc. (g/l)	+Fe Solution Stock + Solution (ml/l)	-Fe Solution Conc. (g/l)	-Fe Solution Stock Solution (ml/l)
1	$Ca(NO_3)_2 \cdot 4H_2O$	270.00	6.6	369.70	9.04
	NH_4NO_3	33.80			
2	KCl	18.60	7.2	18.60	9.2
	K_2SO_4	44.00		44.00	
	KNO_3	24.60		49.20	
3	$Mg(NO_3)_2 \cdot 6H_2O$	142.40	2.8	156.60	3.1
4	KH_2PO_4	17.60	0.5	17.60	0.5
5	$Fe(NO_3)_2 \cdot 9H_2O$	13.31	1.5	none	
	HEDTA	8.68		none	
6	$MnCl_2 \cdot 4H_2O$	2.34	1.5	2.34	1.5
	H_3BO_3	2.04		2.04	
	$ZnSO_4 \cdot 7H_2O$	0.88		0.88	
	$CuSO_4 \cdot 5H_2O$	0.20		0.20	
	$Na_2MoO_4 \cdot 2H_2O$	0.26		0.26	

+ The stock solution was made by mixing in one container all salts of each solution number
conc. = concentration

After six days of growth in the +Fe solution, seedlings from five trays were transferred to five others containing the modified full strength solution (-Fe solution). At this time, their roots were briefly rinsed with distilled water before being put into -Fe solutions. The modified full strength solution was made by not adding

Fe in solution and using (nitrate nitrogen) NO_3-N as the sole source of N, a common method used to induce Fe-deficiency in plants (Brown and Jones, 1962; Esty et al., 1980; Kannan 1980a, 1980b). The pH of nutrient solutions with NO_3-N increases or remains relatively high (near pH 7 or above) as plants grow in them. This high pH causes Fe to be more unavailable for plant use (Williams et al., 1982).

The other seedlings from the +Fe solution received an additional quantity of full strength nutrient solution and remained unchanged up to harvest. The growth conditions for seedlings were 16 hours of light (340 $\mu E\ m^{-2}\ sec^{-1}$ at plant height) at 29°C and 8-h darkness at 22°C. Lamps providing the light were 16-cool/white fluorescent 72 inch and 8-60 watt incandescent bulbs, in a SHERER Model CEL (Controlled Environment Laboratory) 37-14. Treatments were arranged in a split plot design with nutrient solutions as main plots and genotypes as subplots, replicated 5 times.

Plant Measurements

Iron deficiency symptoms were visually recorded for 5 plants selected at random, as in the field experiment, on a daily basis three days after plants were put into treatments. They were recorded to the nearest 0.5 score for intermediate ratings.

Four leaf discs of 0.24 cm^2 each were taken per plant per genotype from the rated leaves for chlorophyll determination. These discs were put into transparent tubes and diluted into 3.5 ml of pure N, N, Dimethylformamide (D.M.F) then placed upright on a horizontal tray and refrigerated for 48 hours. Absorbance (abs) was measured at 664.5 and 647-nm wavelengths with a Bausch and Lomb model

Spectronic 710 Spectrophotometer. Chlorophyll A, chlorophyll B, and total leaf chlorophyll content were calculated from the absorbance readings based on the following equations (Inskeep and Bloom, 1985):

$$\text{Chlorophyll A (ug cm}_{-2}) = 12.7 * \text{abs664.5} - 2.79 * \text{abs647}$$

$$\text{Chlorophyll B (ug cm}_{-2}) = 19.9 * \text{abs647} - 4.62 * \text{abs664.5}$$

$$\text{Total chlorophyll} = \text{chlorophyll A} + \text{chlorophyll B}$$

Total chlorophyll was equivalent to leaf chlt of five plants per genotype expressed in $_{\mu}g\ cm^{-2}$. To terminate the experiment, sixteen day old plants were harvested and each plant biomass was separately oven-dried in an envelope at 63°C for 48 h. The total dry matter (TDM) was determined and average weight of five plants per genotype was calculated.

Greenhouse Experiment

Two soils were utilized in the greenhouse, Woodward loam from the area where the field experiment was carried out and Teller loam (Typic Argiustolls), an Fe-sufficient soil from Perkins, Oklahoma (Table I). A supply of the Teller loam soil was sufficiently at hand and contained available Fe and was utilized instead of Carey loam. Each soil was sieved through a 3.2 mm screen and mixed thoroughly. The potted soils were maintained at field capacity (FC). The estimate of FC each soil was determined after drying 100 g of a water saturated soil sample at 49°C for 72 h and weighing it. The difference in soil weight before and after the drying was assumed to be the quantity of water necessary to increase the soil water content to the FC. The

quantity of water estimated to bring the potted soil to FC was approximately 500 ml. Soil was evently packed to a volume of 2.1 L and bulk density of 1.3 g cm^{-3} (3.25 kg / pot) in 2.4 L undrained-plastic pots (Pau, 1987). Plant available iron was determined by the DPTA extraction method (Lindsay and Norvel, 1978). The method outlined by McLean (1982) was used to determine soil pH (1:1 soil-H_2O ratio). Nitrogen was added at the recommended rates to grow grain sorghum based on the yield goal of 408/kg (Johnson and Tucker, 1982). The soil in each pot was kept at FC throughout the experiment by daily waterings.

The same ten sorghum genotypes used in the previous experiments were utilized. Eight seeds per genotype were directly planted 2 cm deep in a pot and seedlings were thinned to three per pot ten days after germination. Pots were randomly placed on a table considered as a block with a factorial arrangement of soil x genotype treatment combinations (2 X 10), replicated 4 times. Supplemental light with a photosynthetic photoflux density 40 μmol m^{-2} s^{-2} was supplied at night and during cloudy days. Fifteen day old plants were assigned visual iron-chlorosis scores as previously described based upon the level of yellowing of the three most expanded top leaves. Leaf discs were taken from the rated plants and plant shoots harvested at the soil surface level for leaf chlorophyll content and total dry matter evaluations, respectively.

Statistical Analysis

The data sets collected from the growth chamber, greenhouse and field experiments were analyzed using the ANOVA and the General Linear

Model procedures, to identify significant effects. The Statistical Analysis System (SAS) and Turbostat (Nofziger et al., 1986) were the statistical sofware packages utilized. All significant tests were at the 0.05 level of probability. Significantly different means were compared using LSD (least square difference) and OSL (observed significance level). For the growth chamber and 1987 field experiments, the split plot design considered had the two nutrient solutions and the two soil types as the main plots, respectively, and each genotype as subplot. Error a and error b were used to test the main plot and genotype x soil type (or solution) interaction main effects, respectively. In the 1988 experiment, the replication x genotype mean square was used to test the genotype main effect. The error was used to test all main effects of factors involved in the analysis for the 10 x2 factorial arrangement considered in the greenhouse (soil type, genotype, soil x genotype interaction).

CHAPTER IV

RESULTS AND DISCUSSION

Growth of Plants

Seedlings in nutrient solutions appeared healthy, and no deficiency was noted until they were transferred to treatments used to impose the Fe deficiency. Plants grew normally in the Fe-deficient soil in the greenhouse for about 14 days from planting and then Fe-deficiency symptoms appeared. Expression of Fe-deficiency symptoms was exhibited in the field on plants grown on calcareous soil after three weeks, but growth was not inhibited extensively until they became moderately Fe deficient.

Screening of Sorghum Cultivars in Nutrient Solution and Soil for Fe Efficiency

Fe chlorosis scores taken in Woodward, the greenhouse and the growth chamber were averaged over the dates of scoring because the genotype x date interaction was not significant. Plants in the Fe-deficient growth media rapidly developed low to moderate Fe-deficiency symptoms and the most distribution of differences in score among genotypes for Fe-deficiency ratings was noted in -Fe nutrient solution (Table III and Figure 1). Mean chlorosis score for Plainsman in figure 1 was the lowest and significantly different from others in -Fe solution. This means that the genotypes used exibited a wide range of

TABLE III

MEANS FOR CHLOROSIS SCORES OF CULTIVARS EVALUATED IN THE GROWTH CHAMBER, GREENHOUSE, AND FIELD USING CHLOROSIS-INDUCING SOIL AND -Fe SOLUTION

	Cultivar	-Fe Soln^ Growth chamber score	GH# score	Woodward loam Field score 1987	Field score 1988	Average score over environments	
1	Plainsman	1.91	2.33	1.90	1.54	1.99	T
2	BTX 622	1.94	2.20	2.64	2.21	2.19	Ms
3	1082H16	2.04	2.48	2.14	1.83	2.17	Ms
4	OK 632	2.05	2.35	2.74	1.92	2.24	MS
5	ROKY 8	2.10	2.05	2.68	2.13	2.18	MS
6	Redlan	2.10	2.50	3.00	2.46	2.44	S
7	85-3062	2.20	2.56	2.55	2.13	2.37	S
8	85-3026	2.23	2.38	2.73	2.08	2.34	S
9	ROKY 40	2.28	2.58	2.64	2.46	2.47	S
10	Wheatland	2.73	2.58	2.64	1.92	2.53	S
	LSD (0.05)	+	NS	0.48	NS	0.28	

+ Because of missing data, comparison of any two means within the column was made using the observed significance level (OSL) at the 0.05 probability level

S, MS susceptible and moderately susceptible, respectively

T, tolerant

NS, non significant at the 0.05 probability

^ sol, solution

GH, greenhouse

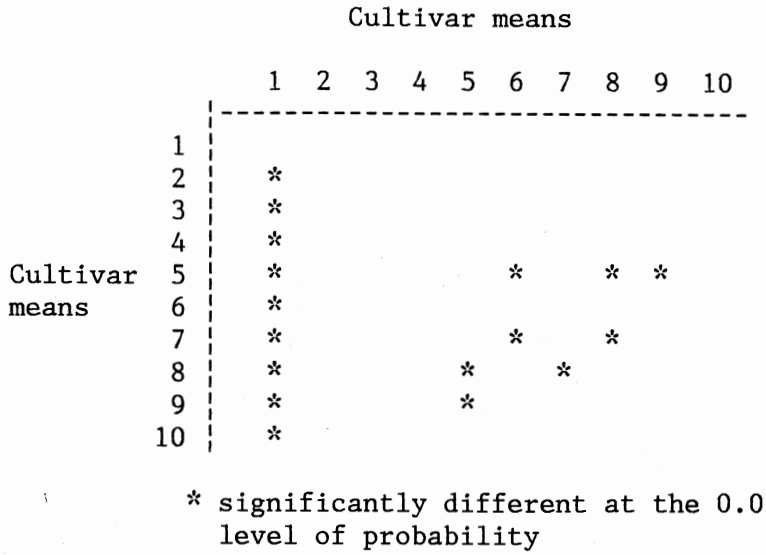


Figure 1. Comparison of any two score means from the growth chamber using the observed significance level

response to the Fe stress more in the nutrient solution than in the other environments. Competition for any available Fe among genotypes in solution may have contributed to the low of chlorosis symptoms observed. It has been reported that in case of Fe stress, root membranes of Fe-efficient plants exudate hydrogen ions (H^+) which acidify their rhizosphere, then enhance dissolution of any insoluble Fe form and its uptake (Ambler and Brown, 1972). Possibly some Fe-efficient cultivars had influenced some susceptible ones through the nutrient solution. Sorghum genotypes grown in the Fe-deficient soil under field conditions distinctly responded to the Fe deficiency only in 1987, but these responses were not necessarily the same as those noticed in 1988 and in the greenhouse. Differences in Fe-chlorosis scores among the 10 sorghum genotypes grown in both environments may be related to the varying tolerances of these cultivars to Fe chlorosis (Brown and Holmes, 1956; Brown and Jones, 1962; and Byron

and Lambert, 1983). Plants grown in the field in 1987 exhibited the most severe Fe-stress symptoms. The highest score was 3.0 (in 1987) for Redlan versus 2.08 and 2.50 for the same cultivar in the growth chamber and greenhouse, respectively, and the lowest was 1.54 for Plainsman on Woodward soil (1988 experiment).

Correlation between greenhouse scores from the Fe-deficient soil and those from the -Fe solution was significant ($r = +.55$) despite the lack of a significant genotype effect for chlorosis score in the greenhouse experiment (Table IV). Also, the field average scores over two years were significantly correlated to those of the -Fe solution ($r = +.64$). These relations mean that visually screening growth chamber-grown seedlings to predict greenhouse and field Fe-deficiency chlorosis performance could be effective. It is possible that other lower correlations among growth environments were in part due to the natural variation expressed in Fe chlorosis when the same genotypes are planted in different environments. Plants in Teller loam and +Fe solution showed no chlorosis, (Tables V, VI & VII) indicating that these growth media had enough available Fe to sustain plant growth. However, the failure of Woodward loam soil to induce distinct intensive chlorosis symptoms in most of the genotypes in the greenhouse and 1988 field experiments was unexpected and could not be explained. Light chlorosis symptoms were observed on some few plants grown on the Carey loam (Table III). This might be attributed in part to the non-homogeneity of soil to produce consistent Fe-chlorosis symptoms in case of the field situation.

TABLE IV

MEAN SQUARES FOR CHLOROSIS SCORES, CHLOROPHYLL CONTENT, PLANT BIOMASS AND PLANT GRAIN WEIGHT FROM THE GROWTH CHAMBER, GREENHOUSE AND FIELD EXPERIMENTS

		Mean Square			
Source	df	Score^	Chlt	TDM	Weight
		Growth chamber			
Nutrient	1	--	3167.0*	13519*	--
Error a	4	--	43.5	5008	--
Genotype	9	0.275**	108.0	66814*	--
Nutr.x Geno.	9	--	244.0*	2189	--
Error b#	34	0.047	--	--	--
	56	--	71.0	--	--
	67	--	--	2432	--
		Greenhouse			
Soil	1	39.223*	2549.0*	1911	--
Genotype	9	0.062	99.2	30152	--
Soil x Geno.	9	0.063	64.8	11102	--
Error	57	0.176	90.0	14394	--
		Field			
Soil	1	219.500*	--	--	22970.74*
Error a	3	0.076	--	--	246.76
Genotype+	9	1.420*	616.2	--	311.81
	27	--	1703.2	--	--
Soil x Geno.	9	0.766	--	--	178.00
Error b	54	0.060	--	--	198.82

* Significant at the 0.05 probability level
-- Not estimated
Df for Error b is 34, 56, 67 for score, Chlt and TDM respectively
+ Df for genotype is 27 for Chlt
^ Chlt chlorophyll content
Score chlorosis
TDM plant biomass
Weight grain weight

TABLE V

CHLOROSIS SCORE, CHLOROPHYLL CONTENT AND TOTAL DRY MATTER OF 10 SORGHUM CULTIVARS GROWN IN THE GROWTH CHAMBER IN NUTRIENT SOLUTIONS

	Cultivar	Score# +Fe soln^	Score# -Fe soln	Chlt +Fe soln	Chlt -Fe soln	TDM +Fe soln	TDM -Fe soln
				....ug cm^{-2}.		.....mg.....	
1	Plainsman	1.0	1.91	50.40	24.40	163.68	188.59
2	BTX 622	1.0	1.94	49.00	39.08	105.41	115.32
3	1082 H16	1.0	2.04	44.54	33.96	132.22	125.05
4	OK 632	1.0	2.05	45.76	36.95	95.81	97.74
5	ROKY 8	1.0	2.08	41.35	35.40	81.07	94.42
6	Redlan	1.0	2.08	49.28	31.67	75.44	71.30
7	85-3062	1.0	2.20	40.66	30.20	130.74	146.72
8	85-3026	1.0	2.23	59.08	29.92	142.85	160.50
9	ROKY 40	1.0	2.28	44.00	29.00	90.35	81.63
10	Wheatland	1.0	2.73	41.40	28.92	152.24	168.33
LSD(0.05)		NS	*	NS	NS	*	*

* Significant at the 0.05 level of probability using the OSL

\# Score chlorosis score
Chlt chlorophyll content
TDM total dry matter

^ Soln solution

NS Non-significant using the LSD

TABLE VI

CHLOROSIS SCORE, CHLOROPHYLL CONTENT AND DRY MATTER OF 10 SORGHUM CULTIVARS GROWN IN THE GREENHOUSE ON TELLER AND WOODWARD LOAM

	Score#		Chlt		TDM	
Cultivar	Teller loam	Wd* loam	Teller loam	Wd loam	Teller loam	Wd loam
			...ug cm^{-2}..		.....g.....	
ROKY	1.0	2.05	42.37	37.86	181.25	210.50
BTX 622	1.0	2.20	30.79	26.41	337.88	275.38
Plainsman	1.0	2.33	36.05	34.32	209.88	255.13
OK 632	1.0	2.35	39.52	23.53	288.75	424.25
85-3026	1.0	2.38	35.17	24.50	266.00	328.25
1082H16	1.0	2.48	37.25	23.33	242.00	319.13
Redlan	1.0	2.50	42.92	28.69	349.63	305.63
85-3062	1.0	2.56	39.59	25.88	218.38	128.13
Wheatland	1.0	2.58	42.59	25.35	225.88	245.00
ROKY 40	1.0	2.58	38.30	21.78	367.00	293.00
LSD(0.05)	NS	NS	NS	16.04	NS	NS

NS non significant at the 0.05 level of probability
\# Score chlorosis score
Chlt chlorophyll content
TDM total dry matter
* Wd Woodward

TABLE VII

CHLOROSIS SCORE, GRAIN WEIGHT, CHLOROPHYLL CONTENT AND PERCENT WEIGHT REDUCTION OF TEN SORGHUM CULTIVARS GROWN IN THE FIELD ON CAREY LOAM AND WOODWARD LOAM

	Score#			Chlt	Grain weight		
	Carey loam	Wd+ loam		Wd loam	Carey loam	Wd loam	
Cultivar	1987	1987	1988	1988	1987		%WR*
				$\mu g\ cm^{-2}$	.....g.....		
Plainsman	1.02	1.90	1.54	48.35	19.44	6.89	64.5
1082H16	1.00	2.48	1.83	40.28	19.07	9.56	50.0
85-3062	1.08	2.55	2.13	39.61	17.53	6.86	60.8
ROKY 8	1.20	2.63	2.13	39.48	22.09	8.20	62.2
Wheatland	1.04	2.64	1.92	44.29	26.61	10.85	59.2
ROKY40	1.09	2.64	2.48	36.22	27.81	9.95	64.2
BTX 622	1.11	2.64	2.21	33.37	21.69	1.60	92.6
85-3026	1.18	2.73	2.08	38.14	21.88	5.23	76.1
OK 632	1.06	2.74	1.92	40.49	24.18	12.20	49.5
Redlan	1.24	3.00	2.46	41.48	28.29	6.37	78.0
LSD(0.05)	NS	0.48	NS	NS	NS	4.08	

\# Score chlorosis score
Chlt chlorophyll content
\+ Wd Woodward loam
Cl carey loam
* %WR weight percent reduction
%WR = (weight from Cl - weight from Wd)/weight from Cl
NS Non-significant at the 0.05 level of probability

Based on Fe-deficiency ranking (grouping) as criteria for evaluation of sorghum cultivars, Plainsman had the lowest average score (1.99) over all experiments (Table III) almost equal to that which Williams et al. (1982) reported for the same cultivar in a similar experiment (growth chamber study) and classified it as tolerant to Fe deficiency. In fact, Plainsman's field mean score

(1.72) was the lowest among all. Wheatland and Redlan, susceptible, had average scores over all experiments (2.53 and 2.44) smaller than 3.0 and 3.8 reported for the same entries in the experiment conducted by Williams et al. (1982), where they were ranked susceptible. Pau (1987) also found them susceptible to Fe deficiency on Quinlan soil in a growth chamber potted experiment. '1082H16', 'ROKY 8', and 'OK 632' from the Regional Sorghum Yield Nursery could be classified as more moderately susceptible than others. Because the rating scale used was restricted by a maximum value of 5.0, a more descriminative scale or method of evaluation may be required to differentiate susceptible individuals up to that level. Progress would require developing techniques capable of stressing plants at a high level both in nutrient solutions and Fe-deficient soils. Such a problem has been observed in developing soybean resistance to Fe-induced chlorosis (Cianzio and Fehr, 1980; Fehr et al., 1985; Prohaska and Fehr, 1981). For example as a technique for further differentiation of resistant soybean genotypes, Fairbanks et al. (1987) obtained high chlorosis levels in the growth chamber with Fe-deficient soils by increasing their matric potential near 0 and fertilizing them with 300 mg of P as sodium orthophosphate (Na_2HPO_4) per Kg of soil.

Effect of Fe Deficiency on Leaf Chlorophyll Content, Total Dry Matter and Grain Weight of Sorghum Plants

Table IV shows the mean squares for chlorosis scores, chlorophyll content and total dry matter or grain weight of sorghum plants grown in different media. The effect of Fe availability on chlorosis

scores, chlorophyll content (greenhouse and growth chamber) and grain weight (field) of sorghum plants was significant for all experiments, but the effect of nutrient solution on plant biomass was only significant for the growth chamber study. Significant chlorosis, Chlt, and grain weight differences between soil type (Fe-deficient and Fe sufficient) or nutrient solution treatment means averaged over all genotypes confirm that these responses were not equal in either two soils or solutions and that chlorosis indeed resulted from a deficiency of Fe (Table VIII). Figures 2 & 3 illustrate that for all cultivars, leaf chlt was reduced by Fe deficiency in both the greenhouse and growth chamber. However, a significant nutrient solution x genotype interaction was found for leaf chlorophyll content in the growth chamber since cultivars did not have the same relative chlorophyll content magnitude in the two growth solutions (Table IV and Figure 3).

In the greenhouse and 1988 field studies leaf chlt of varieties grown in the -Fe solution and Fe-deficient soil were negatively correlated (r = -0.68 & -0.56, respectively) with chlorosis scores assigned to them. The following models were constructed to explain the relationships using linear regression (Figure 4 & 5).

$$GHchlt = 74.94 - 19.90 * GHscore$$

$$Fchlt = 60.45 - 7.79 * Fscore$$

where

GHchlt and Fchlt are the mean responses of the populations of plant leaf chlt for greenhouse

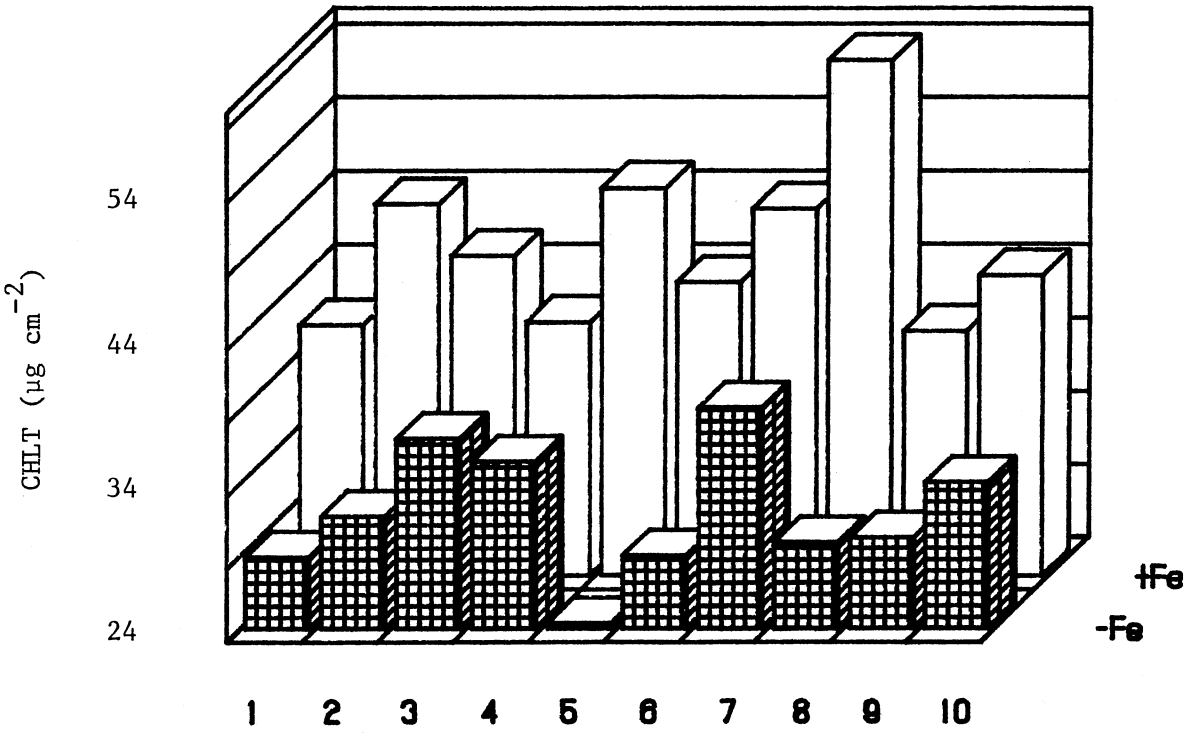


Figure 2. Effect of Fe Deficiency on Leaf Chlorophyll Content (Chlt) of Sorghum Plants Grown in Nutrient Solution.

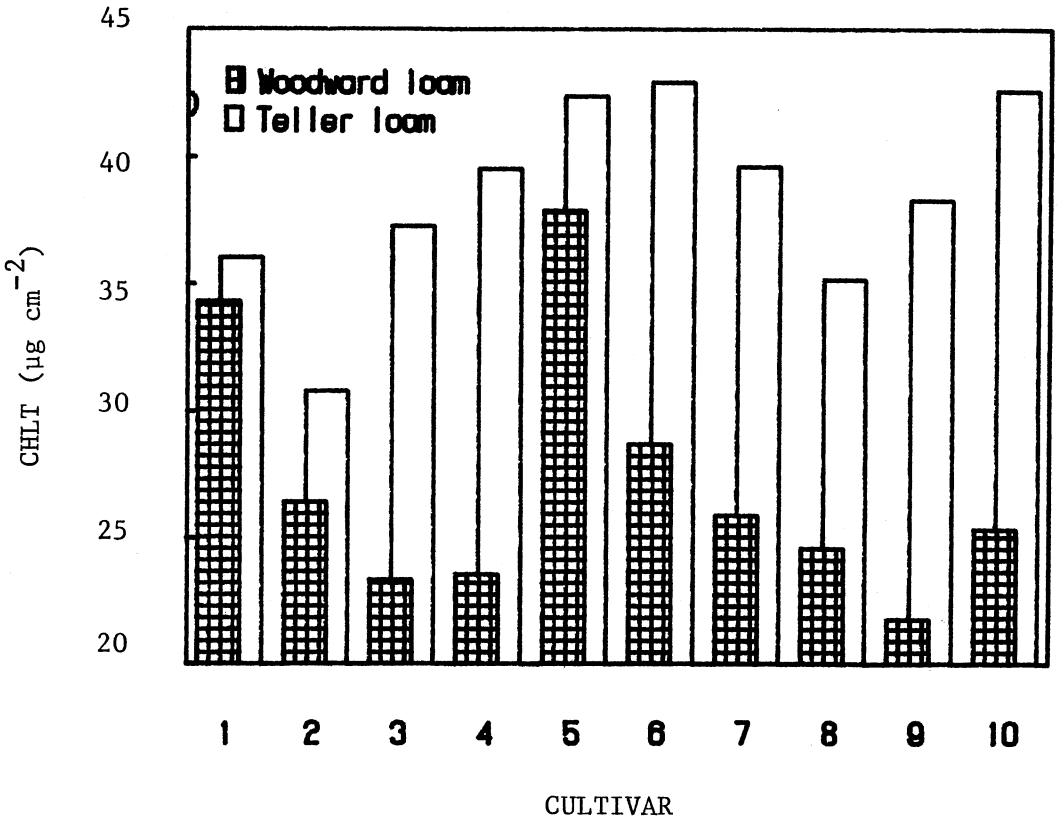


Figure 3. Leaf Chlorophyll Content (Chlt) of Sorghum Plants Grown in the Greenhouse.

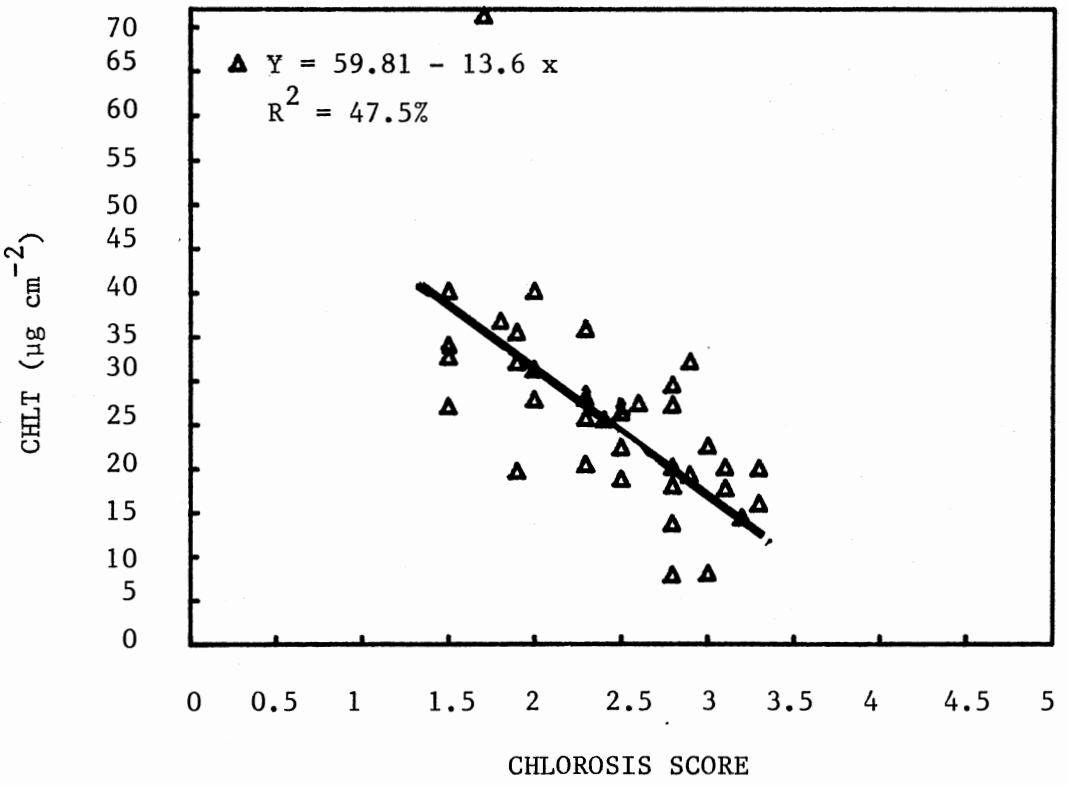


Figure 4. Correlation of Leaf Chlorophyll Content (Chlt) with Chlorosis Scores in the Greenhouse Experiment.

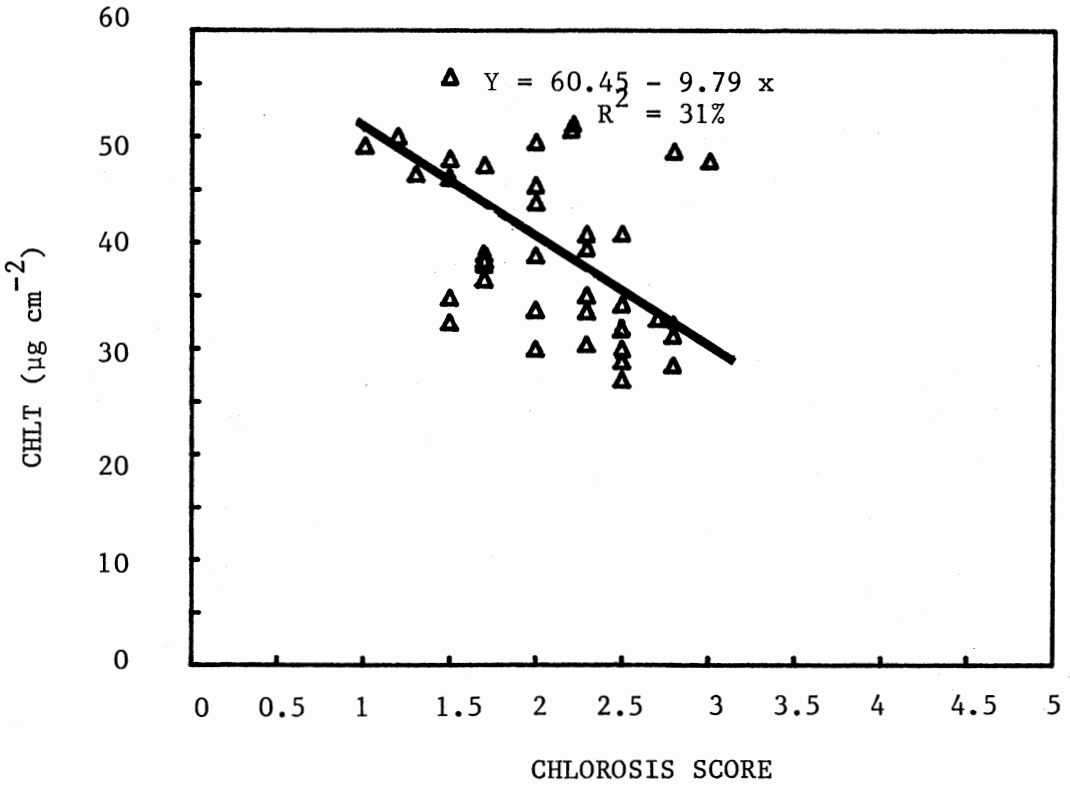


Figure 5. Correlation of Leaf Chlorophyll Content (Chlt) with Chlorosis Scores in the 1988 Field Experiment.

scores, GHscore, and field scores, Fscore,

respectively.

These relations indicate that visual chlorosis ratings can be substituted for chlorophyll analyses which require sophisticated equipment and are time consuming. The time imput needed would certainly be less using the visual rating method to select Fe efficient cultivars; However, Cianzio et al. (1979) remarked that the subjectivity involved in visual ratings should be a great concern for some research, mostly when different environments and personnel are used. Other correlations involving total dry weight, chlt and grain weight with growth chamber and field scores were not significant ($r < 0.26$).

Table VIII and Figures 6 & 7, show that plant dry weight was not generally affected by the Fe deficiency. Unexpectedly, many Fe-stressed (chlorotic) plants weighed more than healthy ones when they were grown either in nutrient solutions or in soil (greenhouse). Unfortunately, plant dry weight was not evaluated in the field. This unexpected result leaves one thinking that reduction of plant dry matter in Fe-stressed medium reported by some research work could be associated with other factors besides the Fe deficiency. Many researchers (Al-Showk et al., 1986; Wallace et al. 1976; Watanabe et al., 1965; Pau, 1987) have found the Fe-deficiency associated with ion balance in the plant or soil solution. Identifying a plant growth stage beyond which intensive Fe chlorosis will cause a decrease in plant dry matter and a reduction in grain weight would help streamline screening effort.

Chlorotic plants in the field were stunted, short, weak and had small panicles. This explains the drastic significant decrease in plant grain weight observed for almost every genotype (Table VII & Figure 8) grown on Woodward loam soil. Percent reductions in grain weight were not significantly correlated with chlorosis scores from either field, greenhouse or growth chamber. It is expected that the

TABLE VIII

MEAN CHLOROSIS SCORE, CHLOROPHYLL CONTENT, PLANT BIOMASS OR GRAIN WEIGHT FOR GROWTH CHAMBER, GREENHOUSE, AND FIELD EXPERIMENTS AVERAGED OVER 10 SORGHUM CULTIVARS

Environment	Growth Medium	Chlorosis Score	Chlt^	TDM or Grain Wt
		0-5	ug/cm2	mg
Growth Chamber	-Fe Solution	2.13	33.86	128.54
	+Fe Solution	1.00	45.96	118.58
	LSD (0.05)	*	*	NS
Greenhouse	Woodward Loam	2.40	27.17	278.44
	Teller Loam	1.00	38.45	268.66
	LSD (0.05)	0.19	4.25	NS
Field#	Woodward Loam	2.57	--	7.77
	Carey Loam	1.08	--	22.93
	LSD (0.05)	0.15		2.83

--not estimated
* Significant at the 0.05 level of probability using the OSL
^ Chlt chlorophyll content
TDM total dry matter
Chlorosis score and grain weight evaluated

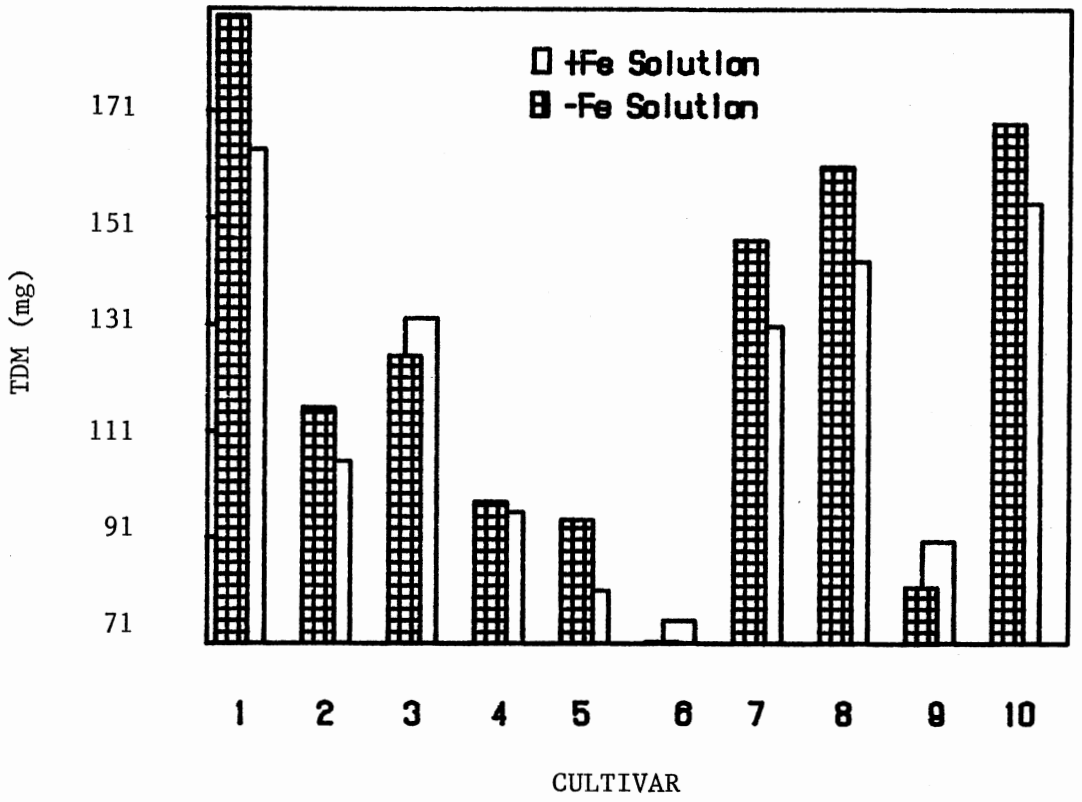


Figure 6. Total Dry Matter (TDM) of Sorghum Plants Grown in the Growth Chamber.

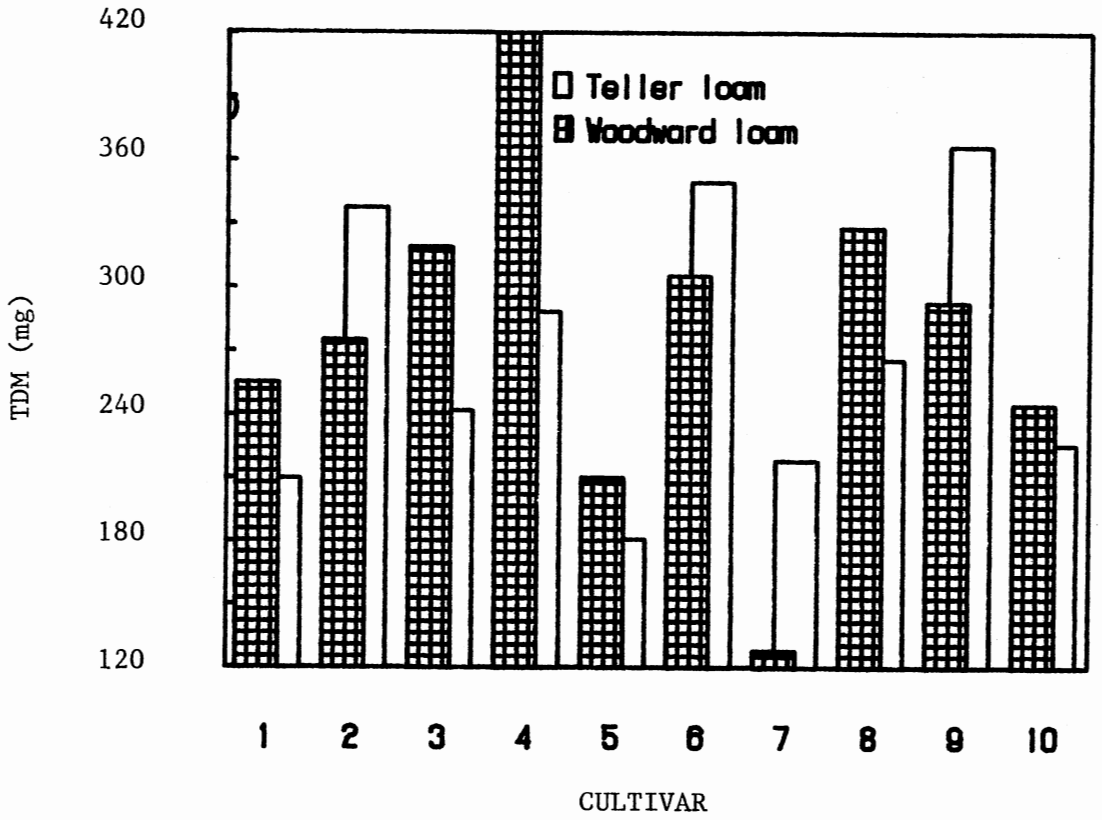


Figure 7. Total Dry Matter (TDM) of Sorghum Plants Grown in the Greenhouse on Woodward and Teller loam.

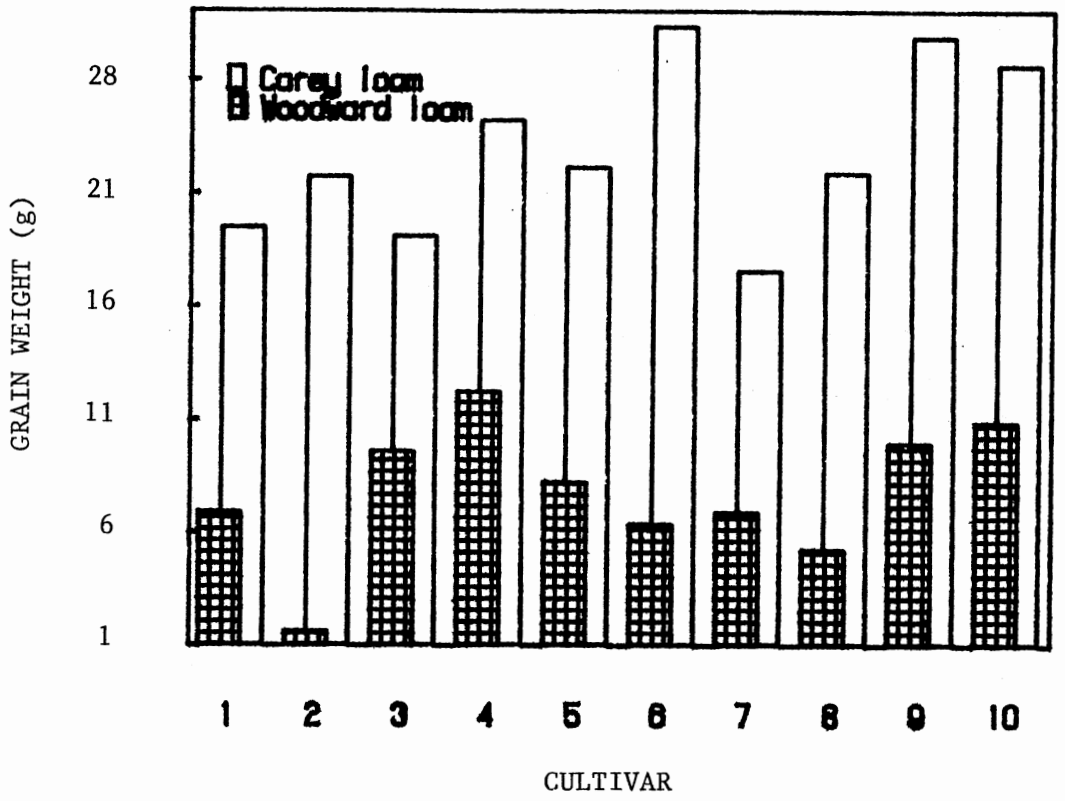


Figure 8. Grain Weight of Sorghum Plants Grown in the Field on Woodward and Carey loam.

difference between the yield of resistant and susceptible sorghum cultivars will continue in the future and yield losses due to Fe-deficiency severity will be highly related to levels of symptoms.

CHAPTER V

SUMMARY AND CONCLUSION

The ten sorghum genotypes in experiments conducted in the growth chamber, greenhouse and field did not exhibit wide ranges of chlorosis symptoms in both -Fe solution and Fe-deficient soil (Woodward loam) when screening for Fe efficiency, making their general classification as resistant, intermediate, moderately intermediate, or susceptible very difficult. However, Plainsman, Wheatland and Redlan for which previous information was known, had a tolerance or susceptibility consistent with that of other investigators. Further work is required to define selection procedures of sorghum cultivars for Fe efficiency. The nutrient solution would be one of the best screening media, because nutrient concentration and supply can be easily managed. Parameters other than growing performances are needed to assess plants for their ability to grow under Fe-deficient conditions or additional measurements and analyses on roots of individual plants are needed. However, problems of Fe-deficiency ratings may arise because of root entanglement and competition for available Fe in case of stress. In the experiment, Fe-efficient genotypes may have influenced the inefficient ones through the root interaction in nutrient solution. This can be averted in the future by growing fewer plants per container, replacing nutrient solutions every day in the container or growing each genotype in an individual container.

Visual chlorosis scores in the growth chamber significantly correlate with those from the field. This significant correlation will help predict field Fe-deficiency performances of new sorghum cultivars of the Regional Sorghum Yield Nursery, therefore shorten the screening procedure.

Fe-deficiency conditions contributed to the decrease in leaf chlorophyll content of plants grown both in nutrient solution and on Woodward loam. Significant linear relationships were obtained between the greenhouse and field Fe chlorosis scores, and leaf chlorophyll content of the associated plants suggesting a substitution of visual ratings for chlorophyll analyses. Visual ratings should be used when time is limited and large number of plants needed to be screened. Leaf chlorophyll analyses would be better for screening on a small scale if one wants to avoid subjectivity involved in visual ratings. Plant total dry matter was not significantly affected by Fe-stress conditions in either nutrient solutions or soil, contrary to what many researchers had reported. Further investigation should focus on identifying at which growth stage a decrease in total dry matter of an iron-stressed sorghum plant would occur and subsequently lead to a grain yield loss. Perhaps letting plants go longer in the greenhouse or nutrient solution would give substantial relations.

Plant grain weight was reduced in the Fe-deficient soil, but the grain weight reductions of cultivars (50-90 per cent) were not significantly correlated with any set of Fe-chlorosis scores. There is a need for relating these losses to levels of Fe-deficiency severity in order to apply an effective screening procedure for Fe-efficient sorghum cultivars. Nutrient and soil screening results in

the controlled environment and greenhouse to evaluate sorghum genotype differences in their abilities to grow and develop under Fe-deficient conditions should be correlated with field results for both dry matter production and grain yield to make selection procedure efficient.

LITERATURE CITED

Al-Showk, A. M., R. L. Westerman and D. L. Weeks. 1986. Iron chlorosis in soybeans. J. Plant Nutr. 9:355-371.

Ambler, J. E., and J. C. Brown. 1972. Iron stress response in mixed and monocultures of soybean cultivars. Plant Physio. 50:675-678.

Banin, A., and J. Navrot. 1972. Pattern of iron distribution in the soil-plant system and its possible relation to iron chlorosis. Commun. Soil Anal. 3:177-182.

Bates, G. W. 1982. Parallels in plant and human iron nutrition. J. Plant Nutr. 5:269-276.

Bernado, L. M., R. B. Clark, and J. N. Maranville. 1984a. Nitrate/Ammonium ratio effects on nutrient solution pH, dry matter yield, and nitrogen uptake by sorghum. J. Plant Nutr. 7:1389-1400.

Bernado, L. M., R. B. Clark, and J. N. Maranville. 1984b. Nitrate/Ammonium ratios on mineral element uptake by sorghum. J. Plant Nutr. 7:1401-1414.

Brown, J. C., and R. S. Holmes. 1956. Iron supply and interaction factors related to lime-induced chlorosis. Soil Sci. 82:507-519.

Brown, J. C., and W. E. Jones. 1962. Absorption of Fe, Mn, Zn, Ca, Rb, and phosphate ions by soybean roots that differ in their reductive capacity. Soil Sci. 94:173-179.

Brown, J. C. and W. E. Jones. 1976. A technique to determine iron efficiency in plants. J. Soil Sci. Soc. Amer. 40:398-405.

Byron, D. F., and J. W. Lambert. 1983. Screening soybeans for Fe efficiency in the growth chamber. Crop Sci. 23:885-888.

Cain, J. C. 1954. Chlorosis blueberry in relation to leaf pH and mineral composition. Proc. Am. Soc. Hort. Sci. 64:61-70.

Chen, Y. and P. Barak. 1982. Iron nutrition of plants in calcareous soils. Advances in Agronomy. 35:217-240.

Cianzio, S. R., W. R. Fehr, and I. C. Anderson. 1979. Genotypic evaluation for iron-deficiency chlorosis in soybeans by visual score and chlorophyll concentration. Crop Sci. 19:644-646.

Cianzio, S. R., and W. R. Fehr. 1980. Genetic control of iron-deficiency chlorosis in soybeans. Iowa State J. Res. 54:367-375.

Clark, R. B. 1982a. Iron deficiency in plants grown in the Great Plains of the U.S.. J. Plt. Nutr. 5:251-268.

Clark, R. B. 1982b. Growth of sorghum and corn in mineral nutrition studies. J. Plt. Nutr. 5:1039-1057.

Clark, R. B., Y. Yusuf, W. M. Ross, and J. W. Maranville. 1982. Screening for sorghum genotype differences to iron deficiency. J. Plt. Nutr. 5:587-604.

Clark, J. B., and R. D. Gross. 1986. Plant genotype differences to iron. J. Plt. Nutr. 9:471-491.

Cochran, W. G., and G. M. Cox. 1957. Randomized complete design. In Experimental Design. 2nd ed. John, Wiley and Sons, New York, USA.

Colgrove, M. S., and A. N. Roberts. 1956. Growth of the Azalea as influenced by ammonium and nitrate. Proc. Am. Soc. Hort. Sci. 68:522-535.

Datin, C. L., and R. L. Westerman. 1982. Effect of phosphate and iron on grain sorghum. J. Plant Nutr. 5:703-714.

Dekok, P. C., A. Hall, and R. H. E. Inkson. 1979. Active iron in plant leaves. Ann. Bot. 43:737-740.

Esty, J. C., A. B. Onken, L. R. Hossner, and R. Matheson. 1980. Iron use efficiency in grain sorghum hybrids and parental lines. Agron. J. 72:589-592.

Fairbanks, D. J., J. H. Orf, W. P. Inskeep and P. R. Bloom. 1987. Evaluation of soybean genotypes for iron-deficiency chlorosis in potted calcareous soil. Crop sci. 27:953-957.

Fehr, W. R., D. M. Froehlick, and D. S. Ertl. 1985. Iron-deficiency chlorosis of soybean cultivars injured by plant cutoff and defoliation. Crop Sci. 25:21-23.

Inskeep, W. P. and P. R. Bloom. 1985. Extinction coefficients of chlorophyll a and b in N, N-Dimethylformamide and 80% acetone. Plant Physiol. 77:483-485.

Inskeep, W. P. and P. R. Bloom. 1986. Effect of soil moisture on soil pCO2, soil solution bicarbonate, and iron chlorosis in soybeans. Soil Sci. Soc. Am. J. 50:946-952.

Jonhson, G. and B. B. Tucker. 1982. Soil Testing and Calibrations. Fact sheet 2225. Coop. Ext. service. Div. of Agr., Oklahoma. State Univ., Stillwater.

Kannan, S. 1980a. Correlation influence of pH reduction on recovery from iron chlorosis in sorghum varieties. J. Plt. Nutr. 2:507-516.

Kannan, S. 1980b. Differences in iron stress response and iron uptake in some sorghum varieties. J. Plt. Nutr. 2:347-358.

Kannan, S. 1983. Cultivar differences for tolerance to Fe and Zn deficiency: A comparaison of two hybrids and their parents. J. Plt Nutr. 6:333-337.

Lindsay, W. L. and W. A. Norvell. 1978. Development of DPTA soil for zinc, iron, manganese and copper. Soil Sci. Soc. Am. J. 42:421-428.

McDaniel, M.E., and J.C. Brown. 1982. Differential iron chlorosis of oat cultivars - A review. J. Plt. Nutr. 5:545-552.

McKenzie, D. B., L. Hossner, and R. J. Newton. 1984. Sorghum cultivar evaluation for iron chlorosis resistance by visual scores. J. Plt. Nutr. 7:877-685.

McLean, E. O. 1982. Soil pH and Lime Requirement. In Method of Soil Analysis. Part 2. 2nd ed. Agronomy 12:215-217.

Mengel, K., and G. Guertzen. 1986. Iron chlorosis on calcareous soils. Alkaline nutritional condition as the cause for the chlorosis. J. Plt. Nutr. 9:161-173.

Mortvedt, J. J., and P. M. Giordano. 1971. Response of grain sorghum to iron sources applied alone and with fertilizer. Agro. J. 63:758-761.

Morvedt, J. J., and P. M. Giordano. 1973. Grain sorghum response to iron in a ferrous sulfate-ammonium thriosulfate-ammonium polyphosphate suspension. Soil Sci. Am. Proc. 37:951-955.

Nance, E., and F. Gray. 1978. Detailed soil survey Southern Great Plains Field Station, Woodward, Oklahoma. Research Report. p. 774.

Nofziger, D. L., J. R. Williams, H. J. Barreto, and D. B. Reynolds. 1986. Turbostat: A data management, statistics, and graphics system developed by graduate students in agronomy. Oklahoma State Univ..

Oserkowsky, J. 1933. Quantitative relation between chlorophyll and iron in green and chlorotic pear leaves. Plt. Physiol. 8:449-468.

Pau, Y. Y. 1987. Effects of soil moisture, phosphorus, and iron fertilization on iron chlorosis of sorghum. Oklahoma State Univ.. Master's thesis.

Prohaska, K. R., and W. R. Fehr. 1981. Recurrent selection for resistance to iron deficiency chlorosis in soybeans. Crop Sci. 21:524-526.

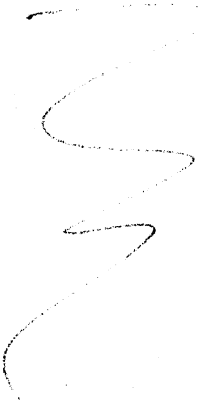
Wallace, A. A., E. M. Romney, and G. V. Alexander. 1976. Lime-induced chlorosis caused by excess irrigation water. Comm. Soil Sci. Plt. Anal. 7(1):47-49.

Watanabe, F. S., W. L. Lindsay, and S. R. Olsen. 1965. Nutrient balance involving phosphorus, iron and zinc. Soil Sci. Soc. Am. Proc. 29:562-565.

Williams, E. P., R. B. Clark, Y. Yusuf, W. M. Ross, and J. W. Maranville. 1982. Variability of sorghum genotypes to tolerate iron deficiency. J. Plt. Nutr. 5:553-567.

Ulrich, W. R., and A. Novacky. 1981. Nitrate-dependent membrane potential changes and their induction in lemna gibba GL. Plt. Sci. Lett. 22:217-227.

APPENDIXES



APPENDIX A

STATISTICAL ANALYSIS FOR THE GROWTH CHAMBER EXPERIMENT (GENERAL LINEAR MODELS)

Response #	Source of variation	DF	Type I SS	MS	F	PR>F
Chlorosis score (-Fe sol.)	Replication	4	10.616	2.654	56.19	0.0001
	Cultivar	9	2.475	0.275	5.82*	0.0001
	Error	34	1.606	0.047		
	Total	47	14.700			
Chlt	Replication	4	307.72	76.93	1.77	0.1230
	Solution	1	3166.82	3166.82	72.85*	0.0001
	Error a	4	173.88	43.47		
	Cultivar	9	973.27	108.14	1.52	0.1623
	Sol. x Cult.	9	2195.35	243.93	3.44*	0.0020
	Error b	56	3974.22	70.97		
	Total	83	10791.27			
T.D.M.	Replication	4	18350.32	4587.58	0.92	0.1230
	Solution	1	13519.09	13519.09	2.70*	0.0213
	Error a	4	20032.48	5008.12		
	Cultivar	9	601324.24	66813.80	27.47*	0.0001
	Sol. x Cult.	9	19704.61	2189.40	0.90	0.5302
	Error b	67	162939.60	2431.93		
	Total	94	835870.35			

# Sol. = Solution
Cult. = Cultivar
Chlt. = Chlorophyll Content
T.D.M. = Total dry matter
* Significant at the 0.05 level of probability.

APPENDIX B

STATISTICAL ANALYSIS FOR THE
GREENHOUSE EXPERIMENT
(ANOVA)

Response	Source of variation	DF	SS	MS	F ratio
Score	Block	3	0.165	5.486×10^{-2}	
	Soil type	1	39.223	39.223	222.29*
	Cultivar	9	0.575	6.392×10^{-2}	0.36
	Soil x Cult.	9	0.575	6.392×10^{-2}	0.36
	Error	57	10.058	0.176	
	Total	79	50.596		
Chlt	Block	3	454.7	151.60	
	Soil	1	2549.0	2549.00	28.32*
	Cultivar	9	892.5	99.16	1.10
	Soil x Cult.	9	583.1	64.79	0.72
	Error	57	5131.0	90.02	
	Total	79	9610.0		
T.D.M.	Block	3	157911.7	52637.24	
	Soil	1	1911.0	1911.01	0.13
	Cultivar	9	271371.9	30152.43	2.09
	Soil x Cult.	9	99920.5	11102.28	0.77
	Error	79	1351551.3		
			(REGRESSION ANALYSIS)		
Chlt vs chlorosis score	Regression	1	2103.6	2103.6	34.37*
	Residual	38	2325.9	61.2	
	Total	39	4429.5		
	Regression a = 59.81				
	Coefficient b = 13.60				
	R^2 = 47.49%				

* Significant at the 0.05 level of probability

APPENDIX C

STATISTICAL ANALYSIS FOR THE FIELD EXPERIMENTS (ANOVA)

1987 #

Response	Source of Variation	DF	SS	MS	F ratio
Chlorosis score	Location	1	219.48	219.48	60.89*
	Blocks	3	21.18	7.06	
	Error a	3	10.81	3.60	
	Cultivar	9	12.76	1.42	3.47*
	Loc. x Cult.	9	6.87	0.76	1.87
	Error b	54	22.03	0.41	
	Total	79	311.05		
Grain weight	Location	1	22970.74	22970.74	93.09*
	Blocks	3	1297.41	432.47	
	Error a	3	740.29	246.76	
	Cultivar	9	2806.30	311.81	1.59
	Loc. x Cult.	9	1601.89	177.99	0.91
	Error b	54	10592.52	196.16	
	Total	79	76556.87		
1988 #					
Score	Block	3	2.95	0.983	
	Cultivar	9	2.84	0.315	1.69
	Error	27	5.04	0.186	
	Total	39	10.82		
Chlt	Block	4	990.45	330.15	
	Cultivar	9	616.16	68.46	1.09
	Error	27	1703.22	63.08	
	Total	39	3309.84		
	(REGRESSION ANALYSIS)				
Chlt vs score	Regression	1	1037.39	1037.39	17.35*
	Residual	38	2272.45	59.80	
	Total	39	3309.84		
	Regression a = 60.45				
	Coefficient b = 9.79				
	R^2 = 31.34%				

# 1987 and 1988 Field data
Loc. = Location
* Significant at the 0.05 level of probability.

VITA 2

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Master of Science

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