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THE EFFECT OF LIGHT, SOIL TEMPERATURE, AND
SOIL MOISTURE ON HIGH-LIME CHLOROSIS

by

Lauren M. Burch

A thesis submitted in partial fulfillment
of the requirements for the degree

of

MASTER OF SCIENCE

in

Soil Science

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Logan, Utah

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INTRODUCTION

Chlorosis of plants from a lack of available iron is one of the most common plant nutrient problems of the calcareous soils of the west. The disease is characterized by a yellowing of the plant leaves and is accompanied, in severe cases, by a partial root death and premature defoliation (3). In addition to reducing growth, the disease greatly reduces the quality and yield of plants.

Many types of plants are affected by iron chlorosis. In Hawaii and Porto Rico rice, sugar cane and pineapple are susceptible, while in California and Arizona, citrus trees are seriously affected. In Utah apples, peaches, plums, prunes, apricots, pears, grapes, raspberries and many ornamental plants are affected (27). The problem of iron chlorosis, therefore, is of great importance to agriculture in the west.

Chlorosis has been studied for more than one hundred and fifty years, but until recently, little progress has been made toward finding the solution to the problem. Although these past studies have not solved the problem of iron chlorosis, they have shown many factors to be closely related to the occurrence of the disease.

These factors include an unbalanced ratio of available manganese to iron in the growth medium (9, 23, 29); chlorotic leaves are high in potassium, nitrogen in the form of ammonia (21), and in ferric iron and are low in total calcium and ferrous iron in comparison with green leaves (12, 13, 14, 17, 25, 26).

The climatic factors of light, soil temperature and soil moisture also appear to be of fundamental importance in chlorosis. It is a common observation that fruit trees are more chlorotic during the early spring when the temperature of the soil is low and the moisture level high. Many investigators have noted that chlorosis tends to be most severe in the poorly drained portions of fruit orchards when water tends to accumulate. However, the work on the effect of climatic conditions on chlorosis has been limited largely to observations.

The purpose of this investigation was to study, under controlled conditions, the effect of light, soil temperature and soil moisture on a lime-induced chlorosis.

REVIEW OF LITERATURE

Because of the extensive literature on the subject of chlorosis, it is felt that the presentation of a complete literature review is not advisable. The review of literature will be limited, therefore, to the factors directly connected with this research, namely, light, soil temperature and soil moisture.

Light

Gericke (5), in 1925, observed that the deficiency or absence of iron had a more harmful effect on plants growing in bright sunlight than it had on those growing under shaded conditions. From this observation, he concluded that the plant's need for iron increases with the plant's exposure to light. Gericke thought that this increased need for iron was due to a more rapid rate of growth in the light; for although the plants exposed to bright sunlight were much smaller than those grown in the shade, they were more mature.

Ingalls and Shive (10) found that the pH increased in the fluid extracts of stems and leaves during the day and decreased at night. They observed that plants having relatively low pH values for tissue fluids absorbed very small quantities of iron, but the absorbed iron under conditions of low pH remained soluble and hence presumably was available for chlorophyll formation. In species having the pH of tissue fluids near or above the precipitation point of iron, iron tended to accumulate in the tissues in an insoluble form and as such could not be used in the synthesis of chlorophyll. One inter-relationship between light and chlorosis, therefore, would appear to be

as follows: The pH of the plant tissue fluids increase as the light intensity increases. With the increase in pH, iron precipitates in the plant tissues and becomes unavailable, and chlorosis develops as a result of a deficiency of iron.

In addition to affecting the pH of the plant tissue fluids, light seems to play an important role in the oxidation-reduction system of the plant. Hopkins and others (9) found that solutions of iron in the form of ferric citrate were reduced to the ferrous form by sunlight. The effect of light in this reduction seems to be a catalytic one, and Hopkins concludes that the state of oxidation of iron is not constant but variable with light intensity, permeability of leaf tissues to light, the amount of manganese, the permeability of the tissues to oxygen and carbon dioxide and the rate of photosynthesis and respiration. Although the problem is very complex because of the number of unpredictable factors, Hopkins (9) believes that, "Light is of great importance in influencing the effect of iron and manganese on plant growth, and that its main role consists of its control of the oxidation potential."

Temperature

At the present time little information is available concerning the influence of temperature on chlorosis. It has, however, long been common knowledge that chlorosis is often most severe in the early spring, especially after a sudden temperature decrease. Jones (11) noticed that the leaves of gardenia plants growing in the greenhouse in winter became chlorotic when the gardenia pots were placed in the cooler sections of the greenhouse. When these chlorotic gardenias were placed near the steam pipes, the leaves soon regained their healthy green color. Further

investigations by Jones showed that the temperature of the soil is a definite factor in inducing chlorosis.

Chlorosis seems to occur under high soil temperatures as well as under low soil temperatures. Wann (27) found that chlorosis of grapes was most severe during the midsummer period of high light intensity and high temperatures. The explanation that usually accompanies this observation is that the high temperature oxidizes soluble ferrous iron in the soil to the ferric form which is precipitated in the soil as hydrated ferric oxide. Iron thus becomes unavailable to the plant, and chlorosis results from iron deficiency.

Hofler (8) reports that according to observations in Austria, humid springs, followed by sudden summer heat are a factor in the development of chlorosis on the high-lime soils of that region. Hofler cites the observations of other German and Austrian scientists that abnormally cold periods early in the year followed by normal or excessively warm temperatures produce a marked chlorosis of plant leaves.

Moisture

The influence of moisture and aeration on iron availability is another phase of chlorosis about which little is known. Haas (7) observed that on calcareous soils the most vigorous trees were found on soil in which the moisture equivalent values either decreased or remained relatively unchanged with increasing depth. Under these conditions the pH of the soil was lower, and the soil aeration was improved. Gile and Carrero (6) reported that the availability of iron in calcareous soils was slightly greater near the optimum moisture content of the soil than at higher water levels, but it remained for Burgess and Pohlman (2) in Arizona to show that the method of irrigation is associated with

chlorosis. They were able to show that the common practice on irrigated land of maintaining soil moisture close to the field capacity level much of the time is one of the principal causes of chlorosis. These Arizona investigators demonstrated that by permitting the soil to approach the wilting point before irrigation and then applying a heavy irrigation of five or six inches of water, the chlorotic condition could be markedly reduced. In spite of the fact that this work has been duplicated by others, there is much controversy over the reason for the results obtained. The principal benefits from this controlled irrigation were attributed to the improvement of soil aeration and promotion of a more favorable pH. Most investigators are in agreement that the lowering of the oxygen pressure of the soil promotes the reduction of ferric iron to ferrous and increases soil alkalinity. On the other hand, increasing the oxygen pressure creates aerobic conditions which tend to oxidize ferrous iron to ferric iron. However, these aerobic conditions tend to reduce soil pH, and all in all conditions for plant growth are greatly improved.

Reuther and Crawford (19) observed that in summer practically no chlorosis occurred on any plots regardless of moisture treatments, but that during the winter months and early spring, chlorosis increased markedly on the wet plots and only slightly on the normal moisture plots. However, the trees grown on wet plots made more growth and had larger leaves and more dry weight than the trees grown on the dry plots. Further investigations by Reuther and Crawford (20) showed that chlorosis was most severe in the winter when the oxygen concentration was highest. They also noted that over-irrigation increased chlorosis.

From their studies they concluded that, "There is an inverse relation between the moisture content of irrigated calcareous soils and the concentration of oxygen in the soil atmosphere, and a direct relation to the carbon dioxide concentration." From their work and the findings of Whitney and Gardner (26) they suggest that the hypothesis that a calcareous soil becomes more alkaline as the moisture content is increased may not be true and should be studied further.

GENERAL PROCEDURE

The general procedure was to grow bean plants in a high-lime soil placed in deep pots submerged in water baths thermostatically controlled at desired temperatures. Provisions were also made for regulating light conditions on the plants and moisture availability in the soil. Diagram 1 shows the experimental design used in the greenhouse experiments. The design of the outdoor experiment is shown in diagram 2.

The soil selected was a highly calcareous soil in which a wide variety of plants became chlorotic. The soil was from the site of chlorosis field studies located east of Fifth East Street in Logan, Utah, between Seventh and Eighth North Streets. The soil* was brought in moist from the field, mixed, fertilized with 16-20-0 fertilizer and placed in 15-inch deep iron pots. These pots had previously been treated with Kodakcoat paint. Care was taken to prevent the soil from drying during the potting operations.

Each water bath was thermostatically controlled and maintained within approximately $\pm 2^{\circ}\text{C}$. of the experimental temperature. Temperatures were selected at various points over the range of 14 to 36°C .

* The soil description was as follows: CaCO_3 , 35%; moisture equivalent, 29%; pH, 7.8; organic matter, 4.7%; readily available potassium, 425 p.p.m.; acetate soluble phosphorus, 46 p.p.m.

** Color	2537	2967	3132	3342	3650	4078	4359	4660	4960
Blue	8	27	16	13	50	51	58	60	39
Red	0	0	0	1	0	0	0	0	0
Color	5461	5780	6234	6500	6900				
Blue	15	3.5	0	0	15				
Red	0	6.5	76	95	87				

Visible Light Rays - 4078 - 6900

Ultraviolet Light Rays - Below 4078

DIAGRAM I. ARRANGEMENT OF CONSTANT TEMPERATURE BATHS, POTS AND LIGHT TREATMENTS IN THE GREENHOUSE.

DIAGRAM 2. ARRANGEMENT OF CONSTANT TEMPERATURE BATHS, POTS AND LIGHT TREATMENTS IN THE OUTDOOR EXPERIMENT.

The higher temperatured baths were heated by glass encased heaters, composed of 25 feet of coiled chrome wire of .664 ohms per foot resistance. The water in the baths was agitated by electric pumps in the heated tanks and by a steady flow of cooling water in the unheated tanks. The water baths in the greenhouse were stirred by means of steel paddles, powered by an electric motor and a system of pulleys.

Bean plants of the Great Northern variety were used as test plants. The seeds were germinated in peat moss and then transplanted to the soil in the pots--five plants being left in each pot.

Different light and moisture conditions were provided for the plants in each bath. The light conditions employed in the greenhouse included normal daylight and normal daylight plus 7 hours of artificial illumination. The light conditions employed in the outside experiments included full summer sunlight, and sunlight through red and blue plastics of known light transmission values.⁴² The colored, reenforced plastic material was made by the Dobeckum Company of Cleveland, Ohio.

Soil moisture in the greenhouse experiments was controlled through the use of soil tensiometers and Bouyoucos blocks. In the outdoor experiments moisture was controlled by weighing the pots at frequent intervals. The moisture levels used in the greenhouse included irrigation with maximum soil moisture tensions of 75, 100, 250, 600-700 centimeters of water and near the wilting point.

Observations on degree of growth and chlorosis of the plants were made at weekly intervals. Upon the completion of each experiment, the plant tops were harvested, and the stem length and terminal leaf meas-

urements were recorded. The leaves were then separated from the stems, and both stems and leaves were washed in .01 N HCL and distilled water. The tops were dried in a drying oven for 48-72 hours at 65-75° F. After the dry weights were recorded, the plant material was ground in a porcelain mortar and pestle. Separate chemical analyses were then conducted on leaves, stems and, in one case, roots.

Two-gram samples of ground plant material were digested with a one to one nitric-perchloric acid mixture, evaporated until colorless and diluted to a final volume of 200 cc. From these basic solutions aliquots were taken and analyzed colorimetrically for the following: manganese by the periodate method (4); phosphorus by the reduction of phosphomolybdate with amidol according to the procedure of Allen (1); potassium and calcium by the procedure reported by Reitemeyer (18); iron by the 1-10 orthophenanthroline procedure as discussed by Smith (22). An A. C. Fisher type colorimeter was used for the determinations.

EXPERIMENTAL

Experiment I

In January of 1947, a preliminary greenhouse experiment was undertaken to study the influence of soil temperature and light duration on the growth and chlorosis of beans.

Five concrete tanks located in the botany greenhouse were wired with heaters and thermostats. The five tanks were divided in half by a black curtain, which prevented any light transmission. Immediately above each tank and on the west side, a 500 watt bulb was mounted to provide extra illumination. Eight soil pots were placed in each bath, four pots receiving normal winter daylight with the other four pots receiving normal winter daylight plus 7 hours of extra illumination. The temperatures used were as follows: 15, 20, 25, 30 and 35 degrees Centigrade.

The heaters and stirring apparatus were started several days prior to the actual planting of the bean plants in order to permit the soil in the pots to adjust to the designated temperature. On January 16, bean seedlings were planted in the soil pots, four plants to each pot, and on January 22 the light variable was introduced. The plants were watered as needed with tap water.

Observations were first recorded on February 1. The plants receiving additional light were making the most rapid growth and in general were lighter in color than the plants receiving less light. Growth appeared greatest in the 25 and 30 degree tanks followed by the 35, 20 and 15 degree tanks in that order. Chlorosis was most noticeable in the 35 degree pots.

On February 21 the plants were harvested, and final observations were taken. The plant tops were then washed and dried according to the previously described procedure. The roots were removed from the soil, washed and dried in the oven. The average values for plant chlorosis, dry weight of tops and roots, are shown in table 1.

At the time of harvest, the 35 degree plants were the most chlorotic, followed by the 30, the 15, 20 and 25 degree plants. There was no significant difference between the length of day and the degree of chlorosis for any temperature.

The plants grown in increased light produced considerably more dry weight than the plants receiving only normal winter daylight. The greatest dry weight was produced in the intermediate temperatures, and growth was greatly reduced in both temperature extremes (table 1). The effect of length of day and temperature on the dry weight of bean tops is shown graphically in figure 1.

The root weights followed the same general trend in the increased light, but under normal winter daylight, the most extensive root system was produced under the lowest temperatures and the most reduced root system under the highest temperature (figure 2). There was a possibility, however, that some roots may have been lost during the harvest.

A separate chemical analysis was conducted on the plant tops and roots for total iron, phosphorus, manganese, calcium and potassium. The averages for the results obtained are given in table 2.

The total iron content of the bean plants could not be correlated with the degree of chlorosis nor with temperature. The analysis did

APPENDIX

Table 1. Relationships between soil temperature and light, and the growth and degree of chlorosis of bean plants

Treatment	*Chlorosis rating	Av. dry weight bean tops	Av. dry weight bean roots
		gms.	gms.
Soil 15° C			
Short day	1.5	2.97	0.714
Long day	0.0	3.96	0.808
Soil 20° C			
Short day	1.5	4.08	0.584
Long day	1.3	6.87	1.187
Soil 25° C			
Short day	0.9	5.15	0.547
Long day	0.9	8.42	1.225
Soil 30° C			
Short day	1.9	4.42	0.504
Long day	1.0	7.42	0.995
Soil 35° C			
Short day	3.5	3.52	0.282
Long day	3.1	3.46	0.562

* 0 indicates no chlorosis; 1. slight yellowing; 2. general yellowing of leaves; 3. leaves bright yellow color; 4. leaves bright yellow with some necrosis.

~~FIGURE 1.~~ LIGHT AND SOIL TEMPERATURE IN RELATION TO DRY WEIGHT OF BEAN TOPS

~~FIGURE 2.~~ LIGHT AND SOIL TEMPERATURE IN RELATION TO DRY WEIGHT OF BEAN ROOTS

Table 2. The iron, manganese, phosphorus, calcium and potassium content of bean tops and roots as related to light and temperature *soil*

(analyses are reported on a dry weight basis)

Bean tops						
Treatment	Chlorosis rating	Fe	Mn	P	Ca	K
		P.P.m.	P.P.m.	%	%	%
Soil 15° C						
Short day	1.5	639	71.5	0.22	2.08	5.15
Long day	0.8	215	69.7	0.23	2.20	6.75
Soil 20° C						
Short day	1.5	232	88.6	0.29	2.61	5.99
Long day	1.3	207	72.3	0.27	2.06	6.16
Soil 25° C						
Short day	0.9	375	74.6	0.29	2.33	6.72
Long day	0.9	222	82.0	0.30	2.47	6.97
Soil 30° C						
Short day	1.9	387	69.1	0.32	2.39	5.84
Long day	1.0	214	69.8	0.29	2.23	5.79
Soil 35° C						
Short day	3.5	613	60.0	0.29	2.65	6.20
Long day	3.1	283	63.3	0.26	2.65	6.20
Bean roots						
Soil 15° C						
Short day	1.5	2484	0.18	0.16	1.61	--
Long day	0.8	2260		0.14	1.19	1.74
Soil 20° C						
Short day	1.5	2968		0.19	1.50	0.52
Long day	1.3	1748		0.15	2.28	0.81
Soil 25° C						
Short day	0.9	1395		0.16	1.48	1.33
Long day	0.9	1908		0.14	2.09	0.78
Soil 30° C						
Short day	1.9	2353		0.19	1.65	--
Long day	1.0	3193		0.18	1.67	--
Soil 35° C						
Short day	3.5	1935		0.13	1.50	--
Long day	3.1	2350		0.17	1.42	--

show, however, an extremely large iron accumulation in the roots of the plants. The iron content of roots was from 4 to 10 times greater than the iron content of the tops.

The phosphorus content of the plants showed a more consistent relationship with all treatments containing approximately the same content of phosphorus. This amount seemed more than adequate for optimum growth; therefore, any variations in yield were in all probability not caused by a lack of phosphorus.

The calcium content of both tops and roots varied little with either changes of temperature or light. The potassium content in the bean plants varied considerably with temperature, with the maximum accumulation occurring in the 25 degree temperature in tops and the 30 degree temperature in the roots. These results, however, did not follow any regular pattern and there was no indication of the high potassium to calcium ratio often reported in chlorotic leaves. These results do not disprove the existence of such a ratio inasmuch as the analysis was based on the whole plant and therefore included both chlorotic and normal tissue.

Taken as a whole, the data did not show any consistent difference between the length of day and the nutrient uptake of bean plants, nor did differences of temperature seem to influence the content of plant nutrients. In general the roots were lower in calcium, potassium and phosphorus than the tops, but the roots contained much more total iron than the tops (table 2).

Experiment II.

In the summer of 1947 a study involving the effect of soil temperature, light intensity and light quality on high-lime chlorosis was undertaken. Five concrete tanks, located north of the agronomy greenhouses, were wired for heaters and solenoid valves to control temperature. The water was agitated by electric pumps in the heated baths and by a constant flow of cold water in the unheated baths.

Each bath was divided into three parts to provide different light conditions. The south section was left exposed to the full rays of sunlight. Above the northeastern section of each tank a blue plastic shelter was erected, and above the northwestern section of each tank a red plastic shelter was constructed. These shelters were 2 1/2 by 2 1/2 by 4 feet and were ventilated at the top and bottom by a slanted opening. These openings were constructed so that direct sun rays could not be reflected inside the shelters. The red and blue shelters were separated by a layer of the red and blue material which prevented any appreciable passage of light from one shelter to the other.

Three deep pots containing the high-lime soil were suspended in the water bath under each light treatment. The same temperatures that were used in the greenhouse were employed in the outdoor treatment. These temperatures were 15, 20, 25, 30 and 35 degrees centigrade.

Since a possibility existed that some of the chlorosis observed in the greenhouse experiment was caused by excessive moisture, moisture was controlled as accurately as possible by weighing the pots at weekly intervals. Soils were brought to field capacity once a week, and small quantities of water were added as needed between these periods.

On August 9, the soil was brought in from the field, mixed and fertilized and potted August 11. The temperature controls were started on August 12. On August 15, 7 bean seedlings were planted in each of the 45 pots, and on August 20 the stirring apparatus was started. The plastic shelters were installed on August 29.

Observations recorded on September 6 indicated that growth was most rapid in the red light, intermediate in the blue light and least rapid in full sunlight. Only the 15-degree plants showed any marked chlorosis; however, all the plants at that temperature were chlorotic.

The plants were harvested October 2, and final observations were taken. Chlorosis seemed to be most severe in the 15 degree temperatures with some chlorosis in evidence in the 35 degree temperatures. The plants growing in red light in the 20 and 30 degree temperatures showed a slight yellowing of the leaves, but all other plants were normal in color. From the evidence available at the time of harvest, chlorosis appears to be more influenced by temperature variations than by light quality or light intensity.

Because of the marked differences in stem length, leaf size and yield, these parts of the plant were measured separately. Then the leaves were separated from the stems, washed according to the regular procedure and dried in the oven. Table 3 shows the degree of chlorosis, the average terminal leaf measurement, the average length of stems and the average dry weight of the plants. These data are shown graphically in figures 3 and 4.

Table 3. Relationships between soil temperature and light, and the growth and degree of chlorosis of bean plants

Treatment	*Chlorosis rating	Terminal leaf measurement		Av. stem length	Av. dry weight
		length	width		
		(mm.)	(mm.)	(mm.)	(gms.)
Soil 15° C					
Red light	2	71	46	182	0.773
Blue light	1	52	36	162	0.389
Sunlight	3	52	33	119	0.760
					1.922
Soil 20° C					
Red light	1	78	57	368	1.328
Blue light	0	68	48	272	1.069
Sunlight	0	51	37	162	1.748
					4.145
Soil 25° C					
Red light	0	73	58	442	1.674
Blue light	0	63	47	265	0.939
Sunlight	0	57	42	193	2.427
					5.040
Soil 30° C					
Red light	0.5-1	70	55	334	1.511
Blue light	0	64	47	251	0.864
Sunlight	0	59	45	163	2.250
					4.625
Soil 35° C					
Red light	2	70	53	250	1.152
Blue light	1	57	41	228	0.719
Sunlight	0-1	57	42	163	2.033
					3.904

* 0 indicated no chlorosis; 1. slight yellowing; 2. general yellowing of leaves; 3. leaves bright yellow color; 4. leaves bright yellow with some necrosis.

FIGURE 3. LIGHT AND SOIL TEMPERATURE IN RELATION TO THE TOTAL DRY WEIGHT OF BEAN TOPS

FIGURE 4. THE RELATION BETWEEN LIGHT, SOIL TEMPERATURE AND AVERAGE LENGTH OF STEMS

The greatest dry weight was produced in full sunlight, followed by the red light and blue light. The longest stems were produced under red light, blue light and full sunlight in that order. Red light produced far larger leaves than either the blue light or full sunlight. (Figure 5).

Table 4 shows the calcium, potassium, phosphorus, manganese and total iron content of the leaves for this experiment. The data do not show any consistent correlation with the degree of chlorosis, but they do show several interesting trends. The phosphorus accumulation in the leaves varied little with light treatments, but when the phosphorus content for all light treatments was plotted against temperature, a parabolic curve resulted with the maximum at 25 degrees. The calcium content, on the other hand, showed no variation with either light or temperature treatments. In general the potassium accumulation was highest in the 15 degree plants and was consistently lower in the higher temperatures. As in Experiment I, the manganese content of leaves was extremely variable showing no consistent pattern with either light or temperature differences. The total iron content of the bean leaves was also variable with greater differences often occurring between replications than between treatments; however, the plants growing in the intermediate temperatures, 20 and 25 degrees, showed the least iron accumulation.

A complete stem and root analysis was not undertaken, but the total iron content of a few stem samples was determined to see if iron had any tendency to accumulate in the stem tissues. The analysis showed very little iron to be present in the stems of bean plants.

FIGURE 5. LIGHT AND SOIL TEMPERATURE IN RELATION TO LENGTH OF TERMINAL LEAVES OF BEANS

Table 4. The iron, manganese, phosphorus, calcium and potassium content of bean tops as related to light and soil temperature conditions

(analyses are reported on a dry weight basis)

Treatment	*Chlorosis rating	Av. Fe P.P.m.	Av. Mn P.P.m.	Av. P %	Av. Ca %	Av. K %
Soil 15° C						
Red light	2	189	---	.14	---	---
Blue light	1	204	0	.20	2.81	4.78
Sunlight	3	168	224	.19	3.49	3.25
		<u>561</u>		<u>.53</u>		
Soil 20° C						
Red light	1	157	208	.22	3.10	2.83
Blue light	0	184	187	.26	3.07	3.35
Sunlight	0	140	117	.21	2.79	2.54
		<u>481</u>		<u>.69</u>		
Soil 25° C						
Red light	0	184	129	.21	3.06	2.56
Blue light	0	188	121	.26	3.23	2.83
Sunlight	0	142	125	.25	2.98	2.28
		<u>514</u>		<u>.72</u>		
Soil 30° C						
Red light	0.5	211	117	.19	3.17	2.03
Blue light	0	305	158	.22	2.76	2.40
Sunlight	0	185	104	.27	2.94	2.87
		<u>701</u>		<u>.68</u>		
Soil 35° C						
Red light	2	518	169	.14	3.23	1.74
Blue light	1	339	179	.19	3.52	2.67
Sunlight	.5	142	167	.20	3.38	2.05
		<u>799</u>		<u>.53</u>		

* 0 indicates no chlorosis; 1. slight yellowing; 2. general yellowing of leaves; 3. leaves bright yellow color; 4. leaves bright yellow with some necrosis.

Experiment III

A greenhouse experiment was next designed which would include light, soil temperature and soil moisture variations with two replications. One additional concrete tank was added to the general plan of Experiment I making a total of six experimental temperature baths. The temperature variables were reduced to three--17, 25, and 33 degrees centigrade--while the light variables were left unchanged; the long day and short day conditions used in the original design were retained. The new variable, moisture, was introduced in an effort to determine the relative importance of the various climatic factors in inducing iron chlorosis. Four moisture levels were superimposed on the temperature and light treatments. These moisture levels include irrigation when the tension on the tensiometers reached a maximum of 100, 250, and 600 centimeters of water. The fourth moisture level was permitted to approach the wilting point before irrigation was applied. This point was found to be approximately 18,000 ohms resistance to electrical current passage through a Bouyoucos block. The Bouyoucos bridge was used to measure this resistance.

Fresh soil from the same source used in the first experiments was brought in, treated as before, and potted November 30. The young seedlings were planted December 7. These plants were harvested prematurely because of severe fumigation damage and a breakdown of the stirring apparatus used in maintaining constant temperature.

New seedlings were planted on December 20 and observations were recorded at weekly intervals. By January 13, it was evident that light

was the limiting factor in growth, but little difference could be found in the amount of chlorosis produced by either a long day or a short day. Traces of yellowing were showing on the high moisture treatments in the low temperature bath (17 degrees). The most significant color difference, however, was occurring in the soils that were permitted to reach the wilting point before irrigation. All of the plants under this low moisture level, regardless of temperature, were showing a very dark, healthy green color in the leaves.

By January 26, the difference between the extremely dry treatments was very significant. The intermediate dry treatments, 600 centimeters of water, were a significantly lighter color, but showed no sign of chlorosis. The intermediate wet and wet treatments showed considerable chlorosis in the 17 degree baths and slight chlorosis in the 25 degree baths. These differences in degree of mottling seemed to be the same in both the long and short day light treatments, but growth rates were greatly increased by the extra illumination of the long day.

On February 5, the plants were removed from the tanks and the final observations recorded. Some chlorosis was apparent in all the 17 degree treatments except those in which the plants were subjected to extreme drought. The plants under this low moisture condition had a dark green color. The most extreme yellowing occurred in the low light, high moisture treatments. This observation would at first glance indicate that chlorosis is increased by reduced light as well as by high moisture and low temperature. However, this observation does not hold for the other temperatures, and a better explanation would seem to be that these plants,

because of their reduced growth, had a lower transpiration rate and thus remained wet for longer periods than the plants growing under the long day.

The degree of chlorosis seemed to be most severe under high moisture and low temperature with some chlorosis occurring under high moisture and high temperature. Only traces of chlorosis were evident in any treatment under the 25 degree temperatures. No plants became chlorotic under conditions of extremely low moisture under any light or temperature treatment, but the plants subjected to moisture tensions of 600 cm. of water were a significantly lighter green. The lighter color noticed in these plants, however, did not show any symptoms of lime-induced chlorosis.

After the final observations were recorded, the plants were harvested according to the procedure used in Experiment II. Table 5 shows the data for the degree of chlorosis, length of stems and total dry weight of plant tops for the experimental conditions. The yield data are shown graphically in figures 6 and 7. From the curves in figure 6, it is evident that the greatest stem elongation occurred under the long day and at a soil temperature of 25 degrees, followed by the long day and 33 degree soil temperature. In general the greatest elongation occurred at the intermediate moisture level, 250 cm. of water, and the least elongation of stems was produced by the extremely low moisture levels. Figure 6 indicates that light was the limiting factor in stem elongation under the short day light treatment.

The dry weight curve, figure 7, followed the same trend as the curve for stem length with the exception that under the long day treat-

Table 5. ^y The relation ^R between ^{shoots} light, ^{soil} temperature, and ^{soil} moisture and ^{light and} high-lime chlorosis
the degree of chlorosis of bean plants

Max. moisture tension	Light conditions	*Chlorosis rating	Av. stem length	Av. dry weight
cm. of H ₂ O			cm.	gms.
Soil 17° C				
100	long day	1.00	93	1.803
250	" "	0.50	78	1.302
600	" "	0.50	76	1.532
wilting point	sho " "	dark green	42	0.930
100	short day	3.50	51	0.985
250	" "	0.75	31	0.526
600	" "	1.00	32	0.904
wilting point	" "	dark green	35	0.639
Soil 25° C				
100	long day	0.75	126	2.537
250	" "	1.00	159	3.471
600	" "	0.00	108	1.780
wilting point	" "	dark green	78	1.093
100	short day	0.75	55	1.242
250	" "	0.25	48	0.838
600	" "	0.50	44	0.544
wilting point	" "	dark green	34	0.630
Soil 33° C				
100	long day	1.50	108	1.605
250	" "	0.25	129	1.961
600	" "	0.00	78	0.764
wilting point	" "	dark green	55	0.528
100	short day	1.50	54	1.190
250	" "	1.50	52	0.845
600	" "	0.50	56	1.045
wilting point	" "	dark green	30	0.440

* 0 indicates no chlorosis; 1. slight yellowing; 2. general yellowing of leaves; 3. leaves bright yellow color; 4. leaves bright yellow with some necrosis.

FIGURE 6. LIGHT, SOIL TEMPERATURE AND SOIL MOISTURE IN RELATION TO THE LENGTH OF STEMS OF BEAN PLANTS

FIGURE 7. LIGHT, SOIL TEMPERATURE AND SOIL MOISTURE IN RELATION TO THE TOTAL DRY WEIGHT OF BEAN TOPS

ment, more total dry weight was produced under the 17 degree soil temperature than under the 35 degree soil temperature.

The leaf samples were ground and digested according to the procedure described on page 12. The samples were then analyzed chemically for calcium, potassium, total iron, phosphorus and manganese. These data are reported in table 6.

The calcium content of the leaves remained virtually constant regardless of temperature or light variations. This same pattern was observed in Experiments I and II. Potassium as before, was extremely variable with variations between replications often greater than variations between treatments. As in the other experiments, there was no indication of a low calcium to potassium ratio. The phosphorus content of the bean leaves followed the same parabolic curve described in Experiment II indicating that phosphorus is affected more by temperature changes than by moisture or light differences. In all experiments, the maximum phosphorus accumulation occurred at 25 degrees. The total iron content of the bean leaves varied considerably with both temperature and moisture but little with light conditions. In general the total iron content was significantly greater in the low temperature, 17 degrees, for all variations of light and moisture. In the long-day treatment the minimum iron accumulation tended to occur at the 25 degree temperature level, but in the short-day treatment, the minimum iron accumulation occurred at the highest temperature. As in the other experiments, the manganese content was extremely variable, but the lowest manganese concentration seemed to occur at the 25 degree temperature for all treatments.

Table 6. The potassium, calcium, phosphorus, iron and manganese content of bean tops as related to light, temperature and soil moisture conditions

(analyses are reported on a dry weight basis)

Max. moisture tension	Light conditions	*Chlorosis rating	Av. K	Av. Ca	Av. P	Av. Fe	Av. Mn.
cm. of H ₂ O			%	%	%	p.p.m.	p.p.m.
Soil 17° C							
100	long day	1.00	6.37	2.39	.22	157	129
250	" "	0.50	6.73	2.70	.20	240	92
600	" "	0.50	4.15	2.42	.19	176	117
wilting pt.	" "	dark gr.	4.29	2.56	.21	205	104
100	short day	3.50	5.05	2.03	.20	238	117
250	" "	0.75	6.61	2.37	.24	255	129
600	" "	1.00	4.73	2.20	.16	199	48
wilting pt.	" "	dark gr.	6.32	2.16	.27	202	67
Soil 25° C							
100	long day	0.75	5.37	2.15	.46	140	92
250	" "	1.00	5.17	2.08	.56	145	67
600	" "	0.00	5.37	2.27	.33	168	79
wilting pt.	" "	dark gr.	6.00	2.42	.32	109	58
100	short day	0.75	4.90	2.20	.37	153	48
250	" "	0.25	6.29	2.37	.32	198	45
600	" "	0.50	6.15	2.44	.31	175	56
wilting pt.	" "	dark gr.	5.16	2.22	.28	170	38
Soil 33° C							
100	long day	1.50	5.68	2.52	.28	163	107
250	" "	0.25	5.56	2.81	.31	135	67
600	" "	0.00	5.73	2.66	.28	145	45
wilting pt.	" "	dark gr.	6.68	3.12	.25	170	93
100	short day	1.50	5.07	2.73	.31	146	75
250	" "	1.50	6.39	3.02	.27	120	123
600	" "	0.50	4.44	2.73	.28	146	93
wilting pt.	" "	dark gr.	6.63	2.29	.31	87	133

* 0 indicates no chlorosis; 1. slight yellowing; 2. general yellowing of leaves; 3. leaves bright yellow color; 4. leaves bright yellow with some necrosis.

Experiment IV

Experiment IV was designed primarily to verify the results obtained in Experiment III. A second purpose of the experiment was to try to determine whether the great difference in color between the extreme dry and the intermediate dry treatments was due to a nitrogen deficiency or to other causes.

The experimental design was modified to include tensionmeters in the extreme dry treatments. The purpose of these additional tensionmeters was to give a more accurate moisture reading when the dry pots were irrigated. This is advantageous because the Bouyoucos blocks are not very effective at high-moisture levels. The temperatures were modified slightly in an attempt to exaggerate the chlorosis symptoms. The temperatures for this experiment consisted of 15, 25 and 34 degrees centigrade. The same soil used in Experiment III was used in this experiment, and the pots were undisturbed except for a 600 pound per acre addition of Uramon (42-0-0) fertilizer. Each soil was given the same moisture and temperature treatment which it received in the previous experiment, but the pots were again randomized within temperature baths.

Great Northern bean seedlings were planted on March 2 and were given several days to become established. Temperature controls were started on March 8, and on March 10 the light and moisture controls were established. The delay in starting the moisture controls was used to permit the young plants in the extremely dry treatments to become better established before subjecting them to extreme drought.

From the start, growth was hindered by a root rot disease which had become established in the soil of some of the pots. Transplanting new plants into these diseased soils was not successful.

The observations taken April 3 showed a moderate chlorosis in the 15 degree-high-moisture (100 cm.) treatment. Plants in the dry soil held at 15 degrees were a dark green color. Plants under the 15 degree high-moisture, short-day treatment showed extreme yellowing, with a few leaves appearing nearly white. The plants growing under the 25 and 34 degree temperatures all showed normal green growth. There was no indication of the lighter green color noticed in the 600 centimeters of water tension plants in the previous experiment, indicating that perhaps the color difference had been due to a lack of nitrogen. Growth was extremely variable because of the root rot disease; however, growth of unaffected plants seemed to vary but little with moisture.

On April 11, the conditions remained unchanged at the 15 degree temperature level under the long-day; but under the short-day, the high-moisture treatment showed signs of recovery. The lower, older leaves retained the severe chlorosis symptoms; but the younger, upper leaves appeared normal in color. The high-moisture treatment under the long-day showed slight chlorosis symptoms, but all plants under both lengths of day were normal in color.

The plants growing under the short-day light treatment were approaching maturity at a faster rate than those growing under the added light. The rate of growth appeared greatest at 25 degrees for all light conditions, followed by the 15 degree temperature under the short-day.

On April 16 final observations were recorded, and the plants were harvested. On this date all of the chlorotic plants showed either complete recovery or definite signs of improvement on the young leaves. The growth was extremely variable, but in general the longest stems occurred under the wetter treatments in the 25 degree temperature. The results from this experiment indicated that some factor other than temperature or moisture was causing the improvement of the chlorotic plants. The most probable explanation for the improvement would seem to be either an increased light intensity or an increased air temperature.

DISCUSSION

The experiments show that the combination of a high soil moisture level and a low soil temperature are the ^{environmental} climatic conditions most conducive to the occurrence of lime chlorosis. ~~Neither~~ Quality, intensity ^{and} ~~nor~~ duration of light appear to be of ^{less} major significance in the occurrence of the disease, ^{than either soil temperature or moisture} however, duration of light was the only light condition tested sufficiently to draw definite conclusions. The study of light intensity was limited to one experiment, and since that experiment was conducted rather late in the summer season, it is doubtful that the effects of full summer light intensity were obtained. ^{Light quality has a greater influence on plant structure than it} ~~does not seem to bear any definite relation to the degree of chlorosis,~~ but it does have a significant effect on the structure of the plant.

High soil moisture levels seem to be the most significant factor in lime chlorosis, but low soil temperatures also induce the condition.

The data are in agreement with the field observations of Reuther and Crawford (19, 20), Thorne (24), Gile and Carrero (6), and Burgess and Pohlman (2).

The results tend to contradict the findings of Ingalls and Shive (10), ~~Watts (27)~~, Gericke (5), and Hopkins (9). These investigators cite evidence to show that light intensity may be the most significant of the climatic factors. Gericke, however, stated that the greater occurrence of chlorosis in plants growing under high light intensity may be caused by the increased rate of growth under those conditions. Gericke, therefore, believed that the plants exposed to high light intensity outgrew their iron supply faster than the plants growing under

shaded conditions. Hopkins' work was done in Porto Rico where chlorosis seems to be caused by ^a ~~the~~ high ratio of manganese to iron in the soil. There is little evidence to show that the iron-manganese ratio is a cause of chlorosis in Utah. Ingalls and Shive concluded from their data that the iron requirement for plants varies with the light intensity. They were working with culture solutions, however, and were probably testing only the chemical solubility of iron. ^{Since high light intensity} ~~Warm at the Utah station~~ during summer months is also usually accompanied by observed chlorosis of grapes to be most severe during the midsummer period of high soil temperatures. ^{high soil temperatures, corresponding to field observations do} ~~ice of high light intensity and high soil temperature.~~

^{the present} The results of this experiment indicate that much of the lime-chlorosis in the irrigated regions of the west could be alleviated by the proper use of irrigation water. In some poorly drained sections affected by the disease, drainage, if economically feasible, should help by removing excess water in the spring of the year. This removal of water would, in turn, improve the situation by permitting a faster increase in soil temperature. The most satisfactory irrigation practice seems to be one using heavy irrigations with medium-long periods between irrigations. ^{Deep rooted cover crops such as alfalfa or clover may also be helpful in chlorosis}

In addition to the chlorosis observations, the experiments produced some interesting growth relationships. These results on yield and elongation of plants verify the observations of several plant physiologists as summarized by Miller (16). Miller states that in general the growth of plants increases with the duration of light exposure.

^{In the present} ~~study, growth differences from varied levels of light intensity were only distinct when the plants were grown under a fixed illumination.~~ The yield data for the outdoor experiment on light quality and intensity also follow the pattern of earlier investigations. From Meyer and Anderson's (15) discussion of light quality and intensity, it appears that the outdoor experimental results follow the general pattern of light intensity more closely than they do light quality. Meyer and Anderson

Control by removing excess moisture from soil following and by bringing the soil rather uniform to the existing point on a shaded field of time them would be regulation a close relationship orchard

summarize several reports which indicate that the total dry weight and thickness of stem and leaf, all increase with an increase in light intensity. Maximum height of plants and maximum leaf area, however, occur at light intensities considerably below those of full summer sunlight.

The contradictory reports in the literature as well as the inconsistencies in the present series of experiments indicate the complexity of the problem. Soil taken from chlorotic locations often grow normal plants when mixed and placed in pots. On several occasions, the same pot was found to contain both normal and chlorotic plants. Furthermore, the chemical analysis showed no correlation between the degree of chlorosis and the ^{content of ions in plants that are} ~~concentration of ions~~ usually associated with the disease. ~~The last experiment in the series followed the same chlorosis pattern as the preceding experiments until approximately two weeks before the harvest; then for no apparent reason the plants began showing signs of recovery. Since the soil moisture and temperature levels remained constant throughout the experiment, the improvement must have been due to some uncontrolled factor--perhaps air temperature or bacterial activity.~~

The discrepancy between the first experiment and the later studies can perhaps be explained. In the first experiment, chlorosis was most severe under conditions of high temperature than under low soil temperatures. There was a tendency for the soil at the high temperature to dry out rapidly at the surface. Since no means for measuring the moisture content was included in the experimental design, these pots were in some cases over-irrigated and partially waterlogged at the bottom. In the

later experiments moisture was controlled through the use of tensio-meters and Bouyoucos blocks. Under these controlled conditions, chlorosis was more severe at the low soil temperatures than at the high.

here In the event additional experiments similar to those reported are conducted, several suggestions may prove helpful. Since the movement of the soil from its original location seems to reduce the tendency of the soil to produce chlorotic plants, it might be profitable to sample the soil in horizons and to include the same general layering of soil in the pots that was present in the field. Growing a preliminary crop in the pots may help the soil regain its original structure.

An outdoor experiment involving the intensity of light should be conducted using fewer temperatures and more replications. An attempt should be made to reduce the variability of light intensity by using cheesecloth shades over part of the plants exposed to full sunlight. Various moisture levels should also be included in the design.

CONCLUSION

A high-soil moisture level together with low-soil temperature is the condition most conducive to the development of high-lime chlorosis. Neither quality, intensity nor duration of light seem of major importance in the occurrence of the disease. The order of importance of the individual climatic factors appears to be: (1) a high-soil moisture level, (2) a low-soil temperature, (3) a high-soil temperature, (4) light.

SUMMARY

1. The effect of ^{climatic} ~~climatic~~ conditions on a high-lime chlorosis is an aspect of the problem which has received little attention. The investigation was designed to study the importance of the ~~climatic~~ ^{factors} conditions: light, soil temperature and soil moisture on a lime-induced chlorosis.
2. Bean plants were grown in a high-lime soil placed in 15 inch-deep ~~iron~~ pots. The pots were suspended in water baths at various constant temperatures ranging from 15 to 35 degrees centigrade. Light variations used included: full summer sunlight, full summer sunlight through red and blue plastics of known light transmission values, normal winter day in the greenhouse, and normal winter day in the greenhouse plus 7 hours of artificial illumination. Moisture levels were controlled through the use of tensiometers and Bouyoucos blocks. Moisture variations included irrigation at maximum soil tensions of 75, 100, 250, 500-700 centimeters of water, and near the wilting point. Records were made of the degree of chlorosis of plants at regular intervals. The yield of plants and their mineral content was determined.
3. The results indicate that a high-moisture level together with low soil temperature is the condition most conducive to the development of high-lime chlorosis. Neither quality, intensity nor duration of light seem of major importance in the occurrence of the disease. The order of importance of the individual climatic factors appears to be: (1) a high soil moisture level, (2) a low soil temperature, (3) a high soil temperature, (4) light.

5. The data indicate that much of the lime-chlorosis in the irrigated sections of the west could be alleviated by the proper use of irrigation water.

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