A STUDY OF THE EFFECT OF LIGHT AND PHOTOSYNTHESIS ON THE RESISTANCE OF SEEDLING WHEATS TO HIGH TEMPERATURE

by

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INTRODUCTION

Drought is probably the greatest limiting factor in crop production in the semi-arid regions of the world. Occurring in these regions are the principal wheat producing areas of the United States, Canada, Argentina, Australia, and Russia. The plants in these regions must struggle against such adversities as high temperatures, high evaporation, low humidity, hot winds, and erratic rainfall. Aamodt and Johnston (1936), in citing from "Research on Drought in Russia", stated that drought in Russia, followed by famine and disease, caused the loss of millions of lives in 1921. This plague paralleled in almost every way the drought disaster of 1892. Hardly a year passes but that one of these major wheat producing areas suffers seriously from drought. According to Westbrook (1934) it was estimated that the total loss from the 1934 drought in the United States was $5,000,000,000.

Drought may be considered to be either edaphic or atmospheric. Edaphic drought is characterized by a deficiency of soil moisture for the normal development and growth of the plants therein. In atmospheric drought, which is caused by hot dry winds, the temperature usually rises so high that plants are injured and severe dessication may result. Only the plants' reaction to high temperature will be considered in this paper. As the main factor in atmospheric drought is high temperature, the term "atmospheric drought under controlled conditions" may be used to designate the effect of the heat chamber.

Since conditions of high temperature do not occur in each of the semi-arid regions every year, plant research regarding drought resistance would be greatly aided if artificial conditions approximating or equaling natural conditions of drought could be produced. Several investigators have devised methods of inducing artificial drought under controlled conditions. Aamodt (1935) concluded that knowledge regarding drought resistance and other characters is limited because of the difficulty of artificially producing the characteristics, that such equipment as heat chambers will continue to play an important role in studies of these characters.

Kreizinger (1938) discovered that alfalfa plants tested in the morning in a heat chamber were injured to a greater degree than plants treated in the afternoon, even though the conditions for the test and the materials used were the same for both trials. Laude (1939) reported that this diurnal cycle of heat resistance was found in corn, wheat, barley, and sorghum in addition to alfalfa. Heyne and Laude (1940) decided that light had a marked influence on the resistance of seedling plants to high temperatures. These results indicate that photosynthesis may be one of the mechanisms involved in drought resistance. This thesis deals mainly with the problem of determining the effect of light and carbon dioxide on the heat tolerance of seedling plants of wheat a and the probable relationship of these factors to the mechanisms of drought resistance in the plants.

REVIEW OF LITERATURE

Use of High Temperature Chambers

Although there has been considerable study on the problem of drought resistance, relatively few investigators have employed the use of artificial drought chambers to aid in the acquisition of more knowledge concerning this complex character. This problem deals with the resistance of plants to high temperature, which is a form of atmospheric drought under controlled conditions. Heat chambers have thus been devised so that studies relating to drought may progress more rapidly and under controlled conditions.

Krassnosselsky-Maximov (1931) employed artificial, hot, dry wind on wheat and oats to simulate the condition called wind burn. It was found that plants were injured more at the flowering stage than at stages of milky ripeness or waxy ripeness. Krassnosselsky-Maximov and Kondo (1933) subjected cereals and other plants to artificial, hot, dry wind. They showed that stages of development played the greatest role in the susceptibility of plants to dry wind, which was the flowering and heading stages for cereals. It was further shown that a deficient soil moisture caused the plants to become hardened to atmospheric drought.

It was concluded by Berkley and Berkley (1933) that the thermal death point of a plant seems to depend upon its age and the duration and conditions of exposure. It was found that the lethal temperature of cotton ranged from 40° to 84° Centigrade. They defined thermal death point as that temperature which will kill protoplasm immediately at a given relative humidity.

Shirley (1934) devised a drought chamber for testing drought resistance of seedling conifers. This "drought machine" consisted of an illuminated chamber with a revolving table through which dry air was forced. He found that results in the drought machine correlated well with those obtained in the field. Later experiments by Shirley (1936) and Shirley and Meuli (1939) agreed well with the earlier results. Shirley (1936) concluded that resistance to excessive heat increases with increasing age of plant tissue and size of plant tissue.

Aamodt (1935) built a machine for testing the resistance of plants to artificial atmospheric drought. He found that wheat varieties known to be drought susceptible in the field showed more injury from artificial drought than those varieties known to be drought resistant. According to Aamodt and Johnston (1936), three factors were of major importance in drought resistance, viz., the ability to evade early periods of drought, capacity of rapidly developing root systems early, and greater ability to endure drought without permanent injury.

Hunter, Laude, and Brunson (1936) conducted artificial heat and drought tests on inbred strains of corn. Fourteen-day-old seedlings were treated at a temperature of 140° F. and a relative humidity of about 30 percent for 6.5 hours. Resistant lines under field conditions showed little or no injury while lines susceptible to firing in the field showed heavy injury in the controlled tests. Several trials gave consistent results.

Bayles, Taylor, and Bartel (1937) tested the reaction of eight varieties of spring wheat to heat by placing pots of each variety on a revolving table in a current of hot air. A close relationship between the performance in the field and under artificial hot winds was apparent.

According to Schultz and Hays (1938) in reviewing a paper by Peto (1937), a diurnal effect was found when artificial drought injury was tested during early and late stages of plant growth. This diurnal condition was primarily the result of period variations in sunlight.

Schultz and Hays (1938) compared the resistance of plants in both seedling and sod stage in a drought machine with behavior under field conditions. Very good agreement was obtained with artificial drought trials as compared with field data. They concluded that artificial tests of drought resistance may be used to indicate those species or varieties of forage which can be best expected to survive under natural atmospheric drought.

Heyne and Laude (1940) subjected 20-day-old corn seedlings of a number of strains to heat for five hours at 130° F. and a relative humidity of 20-30 percent. The reactions of the strains to artificial drought correlated very well with behavior of the same strains under drought conditions in the field. They further found that the heat resistance of seedling plants of corn was considerably increased by exposure to light for as short a period as one hour.

Effect of Sunlight on Plants

It has long been known that plants utilize the energy from the sun's rays in the process of photosynthesis. It is also known that the plant is the only means of utilizing the radiant energy and that this process is a very inefficient machine. The problem of how the plant transforms kinetic energy from the sun into potential energy is as yet unknown.

Physicists think of light as radiant energy visible to the human eye and including all the primary colors as well as their various shades and which can be separated by means of a glass prism. Sunlight is commonly thought to include the ultra violet and infra red rays, which are invisible to the human eye.

It would be very impractical to review all of the literature in this paper regarding light and its effect upon plants. Ramaley (1933) presented a working bibliography of studies relating to the effect of day length and artificial illumination as affecting growth of seed plants. Miller (1938) also has reviewed the literature thoroughly in regard to effect of light on plants.

As has been mentioned previously, the most important effect of light is in the process of photosynthesis.

However, it may affect the plant in a number of ways.

Arthur (1930) stated that the visible region of sunlight is the most important in the process of photosynthesis. Sheard, Higgins, and Foster (1930) found that growth and development are enhanced by the ultra violet and infra red portions of the spectrum. The portion of maximal energy, which is the green portion, is inhibitory to germination and growth. They further discovered that light had just the opposite effect upon the development of chlorophyll. Ultra violet and infra red rays hindered development of chlorophyll while the yellowish-green, green, and greenish-blue seemed to stimulate chlorophyll development. Sayre (1928) stated that wave lengths of radiant energy longer than 680 $m\mu$ are not effective in the formation of chlorophyll in seedlings of wheat, corn, and oats. He believed that the effectiveness of radiant energy increased with wave length up to about 680 $m\mu$ and then ended abruptly.

In studying effect of light on seedlings in relation to available nitrogen in carbon, it was found by Reid (1929a) that light does not favor the growth of seedlings from low protein, starchy seeds unless extra nitrogen is supplied, that light favors assimilation of nitrates, and that light favors the process of thickening of cell walls in all types of seedlings.

Reid (1929b) reported in another paper that exposure of light during normal length of days in May and June band an inhibitory effect on the growth of stem and hypocatye, but a stimulatory effect upon the growth of leaves and folioceous catyledons.

Miller (1938) stated that a number of workers have studied the influence of light upon the absorption of certain ions. It was found that in the case of Nitella light greatly increased the absorption of certain ions.

It has been found by Green (1894) that light exercised a destructive influence upon diostase, the deleterious effect being caused by the rays from violet end of the spectrum. It is generally agreed that all enzynes are sensitive to light, especially ultra violet light.

Spoehr (1915) found that light caused an increase in the rate of respiration. He believed this to be due to higher oxidative power of air during the hours of illumination.

It is believed by Miller (1938) that light may increase transpiration in one or all of three ways: (1) Light may cause higher temperature of leaves, (2) light may cause greater permeability of the protoplasmic membrane, (3) light may cause inhibitional changes in the cell wall colloids so as to render the cell wall more permeable to water.

Vickery et al. (1937) found, in working with tobacco leaves in light and dark, that there was a synthesis of organic solids of considerable amount in light. In the dark there is a decomposition of organic solids into volutile products.

Physiological Aspects of Drought Resistance

As the study of the resistance of seedling plants to high temperature is closely allied with tolerance to atmospheric drought, it would be well to review some studies regarding the internal nature of drought resistance.

The nature of drought resistance, according to Vassiliev (1929), is first of all determined by the internal character of the plant itself. He believed that a study of these characters was a means leading to the knowledge of the properties of drought resistance and guiding the breeder as to the choice of peculiarities that may be valuable in selecting drought resistant plants.

Tysdal (1933), working with factors influencing the hardening process in alfalfa for resistance to freezing, found that light influenced hardening nearly as much as temperature. He noted that both period and intensity of light were factors. According to Dexter (1933) conditions which cause accumulation or conservation of carbohydrates and other food reserves favor the hardening of plants. Illumination helped markedly in hardening. He further found that the removal of carbon dioxide from the air which the plants received or the placing of plants in the dark greatly decreased, and in some cases entirely prevented hardening to cold. He concluded that conditions, which increase photosynthesis and decrease respiration and the growth of vegetative parts, are the basis of resistance and hardening.

Vassiliev (1931) believed that the mobile fraction of carbohydrates in the plant regulates the life processes of the plant. He further stated that the accumulation of soluble carbohydrates by a plant is a means of increasing its drought resistance. In later experiments, Vassiliev and Vassiliev (1936) stated that carbohydrates aid markedly in regulating the osmotic pressure of the plant cell. Carbohydrates also play the role of protector in preventing coagulation of the protoplasm when influenced by harmful factors. The authors believed that an accumulation of hemicellulose during the stage of water loss is a means of resistance and is a natural reaction of the wheat plant toward drought.

Kondo (1931) found that conditions of growth previous to the experiment determines to a large extent the degree of resistance the plant has in withstanding dehydration. He also believed that the stage of development may play an important role in a plant's ability to resist drought.

Newton and Martin (1930) proved that bound water content was a dependable index of drought resistance among cultivated wheats and some grasses.

MATERIAL AND METHODS

Equipment

The heat chamber used in this experiment to simulate drought conditions in the field consisted of an insulated room 6' x 5'4" x 9'. The heat was produced by blowing air through a steam radiator and on into the chamber through vents in the wall. The relative humidity was increased by the escape of steam from a nozzle into the air stream, and decreased by fresh air drawn in from the outside. The temperature was also decreased in this manner. The air stream was kept somewhat constant by allowing the old air to escape when fresh air was being brought into the chamber. A series of baffles and dampers controlled the path of the air and were regulated by thermostat and humidistat, thus controlling the temperature and relative humidity. The velocity of air flow can be regulated by a damper but for these trials the velocity was set at 81.5 feet per second. A turn table five feet in diameter is located in the center of the room and is driven by an electric motor at a velocity reduced to about 1.2 revolutions per minute.

The chamber may be lighted by four 250-watt bulbs if desired. Three panes of glass separate the bulbs from the chamber so that the heat from the light bulbs will not influence the controlled heating of the chamber.

To study the effect of photosynthesis on resistance of plants to heat, a carbon dioxide eliminator was devised. This was patterned after a similar eliminator used and described by Miller (1910) in working with _Helianthus annuus_.

The carbon dioxide eliminator consisted of an electric motor running an air pump, which forced the air through four different solutions into two bell jars and then out into the atmosphere. The solutions through which the air passed in order are two solutions of 30 percent sodium hydroxide, one saturated solution of barium hydroxide, and one solution of concentrated sulfuric acid. The first two serve in eliminating the CO_2, the third indicates whether or not all the CO_2 has been removed and the fourth eliminates the moisture in the air. The bell jars were connected in series and are immersed in a water bath with an oil covering so that no air can escape or enter except through the proper tubes. One pot of plants was placed under each bell jar. In Plate I is shown the carbon dioxide eliminator. This test involving the elimination of the carbon dioxide will hereafter be referred to as the carbon dioxide test.

Material

The plant material consisted entirely of Tenmarq wheat. This variety was selected as it is one of the best adapted varieties for Kansas and is grown widely throughout the state. It was also intermediate among varieties tested as to drought resistance in the seedling stage.

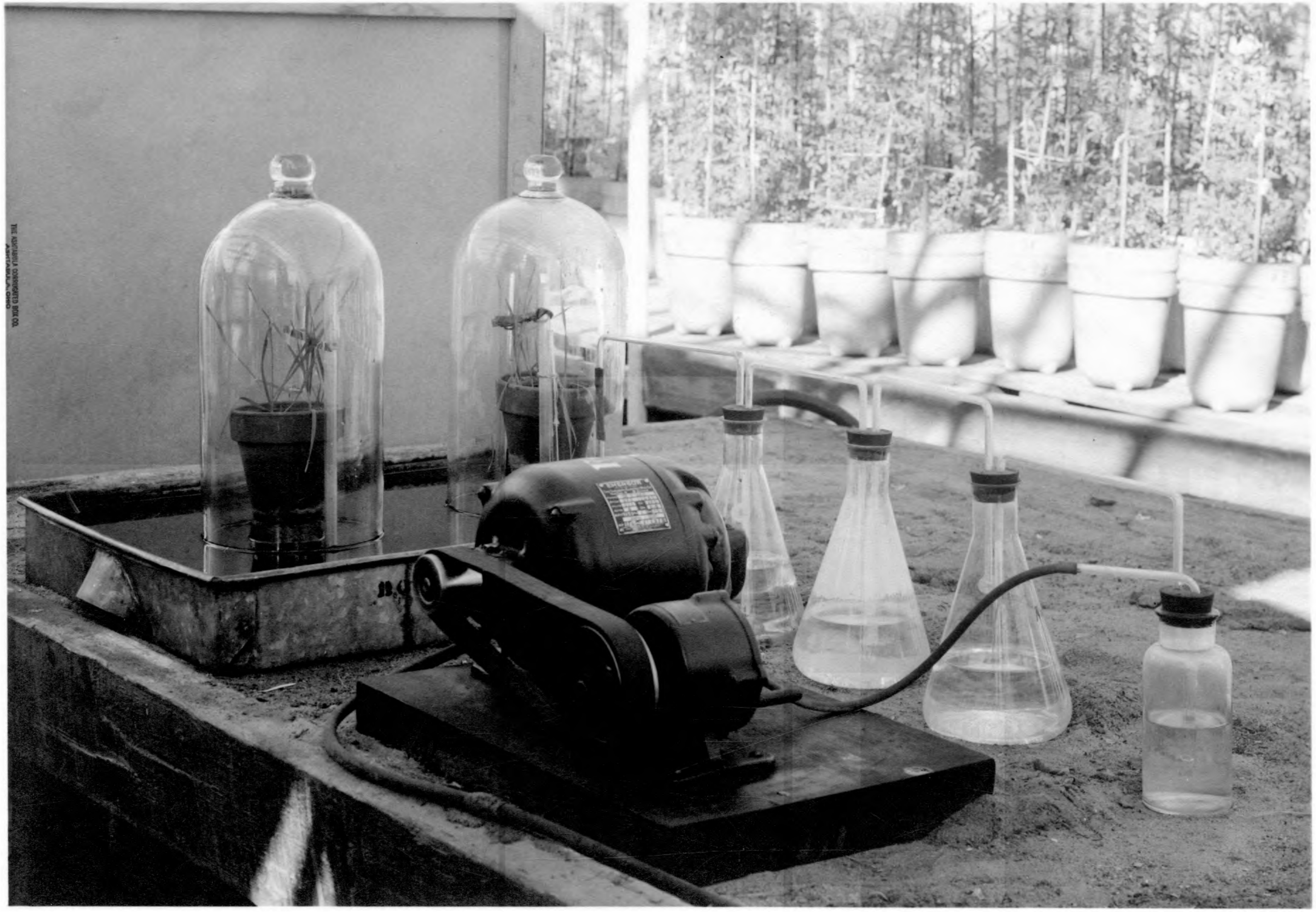
The soil used was a good uniform bottom land soil brought in from the Agronomy Farm. Nine kernels were planted in each four-inch, unglazed, clay pot and the seedlings later thinned to five per pot. In the two light studies, there was a replication of four pots for each treatment. For the carbon dioxide study, only duplicate pots were used as the carbon dioxide eliminator would handle only two pots at one time. The plants were grown at optimum conditions in the greenhouse, and plants were well watered before each trial.

For the tests run during the summer, 14-day-old wheat seedlings were used. Suneson and Peltier (1934) have shown that young winter wheat seedlings still dependent upon the

EXPLANATION OF PLATE I

Apparatus designed to eliminate CO_2 from the bell jars. Plants are therefore placed in the sunlight in a CO_2 free atmosphere.

Plate I



endosperm varied considerably in cold resistance from those plants more advanced in growth. Heyne and Laude (1940) showed that 20-day-old corn seedlings were no longer dependent upon any material in the endosperm. It has also been shown by unpublished data from the Kansas Agricultural Experiment Station that 19-day-old wheat seedlings were no longer dependent upon the endosperm for food. For this reason 21-day-old wheat seedlings were used for all material tested during the fall and winter.

The dark boxes used for the test were cardboard boxes of uniform size, each holding four pots. These were sealed so that no light could enter and placed over the pots at intervals according to the design of the experiment.

Experimental Methods

Seedling plants of Tenmarq wheat were placed in the dark boxes at 8:00 P.M. of the day preceding each trial. The following day the pots were removed from the dark boxes at intervals, according to the design of the experiment, the first treatment being always removed at 8:00 A.M. For the carbon dioxide test run during the summer of 1939, only six pots were used in one trial. At 8:00 A.M. of the day of the trial, two pots were removed from the dark boxes and placed in the carbon dioxide eliminator, and two pots were placed in the sunlight under natural greenhouse conditions. The remaining two pots were left in the dark box until 1:00 P.M. At this time all pots were watered thoroughly but not excessively and transferred immediately to the heat chamber. They remained in the heat chamber for a period of five hours at a temperature of 120° F. and a relative humidity of 35 percent. The experiment was repeated 12 times.

It was decided that the glass of the bell jars in the carbon dioxide eliminator might not allow some rays of light to pass through to the plants. Another treatment was therefore added and this experiment was repeated 10 times. The same procedure was followed as before except that eight pots were used in one trial. They were all placed in dark boxes at 8:00 P.M. of the day preceding the test. At 8:00 A.M. of the day following, six pots were removed from the dark box. Two pots were placed in carbon dioxide eliminators; two pots were placed under bell jars which were open at the bottom allowing free circulation of air; and two were placed under normal greenhouse conditions in the sunlight. At 1:00 P.M. the eight pots were watered and placed in the heat chamber for a period of five hours at a temperature of 130° F. and a relative humidity of 35 percent.

During the summer of 1939 a temperature of 120° F. was high enough to cause differential killing. However, during the fall and winter the level had to be increased to 130° F. A possible explanation for the difference in the levels required is that the whitewash on the greenhouse during the summer may have exercised a screening effect of some of the beneficial light rays, thereby leaving the plants more susceptible to heat. There was no whitewash on the greenhouse during the fall and winter. Also, the plants tested in the summer were 14-day-old plants while those tested in the fall and winter were 21 days old. It may be that the older plants had an accompanying increase in resistance to heat.

The light-interval tests were designed in a similar manner. The long interval experiment had a total of six treatments, replicated four times and repeated twenty times. Plants were again placed in dark boxes at 8:00 P.M. preceding the test. The day of the trial four pots were removed at intervals of one hour beginning at 8:00 A.M. so that the treatments consisted of five, four, three, two, one hour of light and no light. As the plants were removed from the dark boxes, they were placed under normal sunlight in the greenhouse. They were treated for a period of five hours at 120° F. and a relative humidity of 35 percent in the heat chamber.

The short interval test was similar to the long interval except that 15 treatments were used, replicated four time. The treatments consisted of no light, 10 minutes, 20 minutes, 30 minutes, 40 minutes, 50 minutes, 60 minutes, 75 minutes, 105 minutes, 2 hours, $2\frac{1}{2}$ hours, 3 hours, 4 hours, and 5 hours of exposure to sunlight. The pots were then treated in exactly the same manner as the long light interval test except that 130° F. was needed to cause differential killing. The experiment was repeated six times.

After having been in the heat chamber for a period of five hours, the plants were transferred to the greenhouse and placed under normal greenhouse conditions. As soon as the soil in the pots had cooled to nearly normal, the plants were watered thoroughly. The fourth day after treatment the percent of injury was determined. This measure was based upon the percent of leaf area injured and dessicated by the heat treatment. This measure depended upon an estimate of the observer, but after a little practice, considerable accuracy was attained as to comparable readings. The twelfth day after treatment the percent of survival readings were recorded. Each pot was considered a unit and no attempt was made to record the injury of individual plants.

Statistical Methods

All the experiments were designed so that statistical analysis could be applied. As the analysis of variance has proved to be the most precise, flexible, and readily usable method of analysis of data from experiments involving two or more variables, it was used in analyzing these data. The difference between pots in the same treatment was considered as error as replications of the treatment should have the same reading except for uncontrolled variations and chance. All second and third order interactions involving pots were also considered as error. Variation due to trials, treatments, and the interaction between trials and treatments was found in all cases.

There has been considerable work done recently in regard to the transformation of percentage data to increase the validity of generalized standard errors and especially analysis of variance. It was decided to transform the data by use of the formula used by Clark and Leonard (1939). In this formula each estimate of p is replaced by $\sin^2 \theta$. θ is therefore $= \frac{1}{2} \cos (1-2p)$. The derivation is as follows:

$$p = \sin^2 \theta$$
$$\sin^2 \theta = \tfrac{1}{2}(1-\cos 2\theta) = p$$
$$2p = 1 - \cos 2\theta$$
$$\cos 2\theta = 1-2p$$
$$2\theta = \cos^{-1}(1-2p)$$
$$\theta = \tfrac{1}{2}\cos^{-1}(1-2p)$$

The sample calculation is as follows:

$$p = 60\% = .60$$
$$2p = 1.20$$
$$1-2p = -.20$$
$$\cos^{-1} = 101.54$$
$$\tfrac{1}{2}\cos^{-1} = 50.77^{\circ}$$

This type of transformation is designed to change discrete into continuous data.

The data on the 72 observations of the first carbon dioxide test were transformed and analysis of variance run on the transformation. The analysis of variance was also run on the original percentage data and the results compared It was decided that the accuracy was increased very little by the transformation and this added accuracy was not worthy of the time involved in transforming the data. Perhaps the reason for so much similarity was that there were a great number of observations at both extremes of the percentage scale. Transformed data is also discontinuous at these extremes so that little accuracy is gained. It was decided to discontinue the transformations and make the remainder of the analysis on the basis of the original percentage data. The tables of both original and percentage data, and analysis of variance tables for both are given in the experimental results.

EXPERIMENTAL RESULTS

Effect of Long Intervals of Light upon Resistance of Wheat Seedlings to High Temperature

The first experiment was made mainly for the purpose of verifying the results of other workers who have shown that light was a major factor in the resistance of plants in the seedling stage to high temperatures. The data obtained from the long light interval study show clearly that the resistance of plants to high temperatures is affected by light. However, the data were treated statistically by the use of analysis of variance. The average percent injury and percent survival are shown in Tables 1 and 2, respectively. These data are the average of four pots of each treatment. The data for the 20 trials are given and the mean for each treatment, which is the average of 80 observations. The summaries of the analysis of variance for both injury and survival and the calculations of the level of significance for both are shown in Tables 3 and 4, respectively. Variations due to treatments, trials, and the interaction between treatments and trials are clearly significant in all cases as they exceed the one percent point for all variables and

Table 1. Long interval light test, average injury of four pots in percent, 1939.

No.	Trial	Treatment: 5 hrs. light	4 hrs. light	3 hrs. light	2 hrs. light	1 hr. light	no light
1	June 7	26	32	20	10	33	61
2	June 8	35	50	25	6	51	91
3	June 9	29	25	33	21	24	60
4	June 14	69	73	83	81	80	80
5	June 15	5	8	6	10	20	48
6	June 16	25	19	30	33	25	51
7	June 20	9	9	14	18	20	31
8	June 21	44	40	46	38	39	88
9	June 22	71	73	79	83	83	96
10	July 6	24	4	5	9	24	90
11	July 7	3	9	8	6	8	76
12	July 8	0	8	4	3	1	83
13	July 10	3	11	5	4	24	70
14	July 11	1	1	3	4	16	84
15	July 12	4	0	4	8	11	89
16	July 13	3	3	3	9	10	84
17	July 14	0	0	0	3	8	85
18	July 15	16	21	15	16	9	46
19	July 16	4	5	8	8	10	36
20	July 17	16	5	8	8	10	39
	Mean	19.25	19.66	19.82	18.70	25.19	69.37

Mean of all individuals = 28.66

Table 2. Long interval light test, average percent survival of plants in four pots, 1939.

No.	Trial	Treatment 5 hrs. light	4 hrs. light	3 hrs. light	2 hrs. light	1 hr. light	no light
1	June 7	100	100	90	100	100	70
2	June 8	95	95	100	100	90	20
3	June 9	100	95	100	100	100	60
4	June 14	45	65	45	50	65	60
5	June 15	100	100	100	100	90	80
6	June 16	100	100	100	95	95	90
7	June 20	95	100	100	100	90	85
8	June 21	90	65	60	85	75	15
9	June 22	30	20	20	30	15	0
10	July 6	90	100	100	95	85	23
11	July 7	100	100	100	95	100	68
12	July 8	100	90	100	100	100	45
13	July 10	100	85	100	100	80	15
14	July 11	100	100	100	100	90	30
15	July 12	100	100	100	100	100	29
16	July 13	100	100	100	100	100	43
17	July 14	100	100	100	100	100	30
18	July 15	95	95	100	100	95	90
19	July 16	100	100	100	100	100	89
20	July 17	100	100	100	100	100	90
	Mean	92.00	90.50	90.67	92.50	88.50	51.46

Table 3. Summary of the analysis of variance for injury for the long interval light test.

Variation due to		df	Sum of squares	Mean square	F value Calc.	5% pt.	1% pt.
Treatments		5	161,361.513	32,227.230	141.033	2.24	3.07
Trials		19	173,065.493	9,108.710	39.861	1.64	1.99
Treatments x trials		95	60,113.122	632.770	2.769	1.14	1.19
Error							
Pots	3)						
Treatments x pots	15)	360	82,263.408	228.509			
Trials x pots	57)						
Trials x treatments x pots	285)						
Total		479	476,802.536				

Standard error of a single determination = $\sqrt{228.509} = 15.113$

Standard error of the mean for each treatment = $\frac{15.113}{\sqrt{80}} = \frac{15.113}{8.944} = 1.689$

Standard error of a difference = $\sqrt{2} \cdot 1.689 = 2.388$

Level of significance for 5% point = 2.388 x 1.967 = 4.697

Table 4. Summary of the analysis of variance for survival for the long interval light test.

Variation due to		df	Sum of squares	Mean square	F value Calc.	5% pt.	1% pt.
Treatments		5	104,188.53	20,837.706	72.44	2.24	3.07
Trials		19	161,620.91	8,506.364	29.57	1.64	1.99
Treatments x trials		95	58,633.55	617.195	2.15	1.14	1.19
Error							
Pots	3)						
Treatments x pots	15)	360	103,554.37	287.651			
Trials x pots	57)						
Treatments x trials x pots	285)						
Total			427,997.36				

Standard error of a single determination = $\sqrt{287.651} = 16.96$

Standard error of the mean for each treatment = $\frac{16.96}{\sqrt{80}} = \frac{16.96}{8.944} = 1.896$

Standard error of a difference = $\sqrt{2} \cdot 1.896 = 2.681$

Level of significance for 5% point = 2.681 x 1.967 = 5.274

greatly exceed the one percent point in the case of treatments for both injury and survival. The level of significance for the means of each treatment indicates that statistically there is no difference between treatments of two, three, four, and five hours of light. However, those plants treated with only one hour of light were injured significantly more than those treated with more light. The plants treated with no light are very significantly different from all other treatments.

The level of significance for survival shows that the treatment of no light is very significantly different from each of the other five treatments but that there is no significant difference between any two of the other five treatments.

Figure 1 is a graphic presentation of the data for injury, showing the sharp drop in percent injury in the first hour of light. This is also borne out in Fig. 2 which shows a similar rapid rise in percent survival during the first hour the plant is exposed to sunlight.

It was observed that most of the individual observations for injury fell into the lower classes and for that reason it was deemed advisable to show a histogram of injury (Fig. 3) and of survival (Fig. 4). This indicates a very shrewd distribution of the data. However, as the data indicate that

Fig. 1. Effect of long intervals of light on injury.

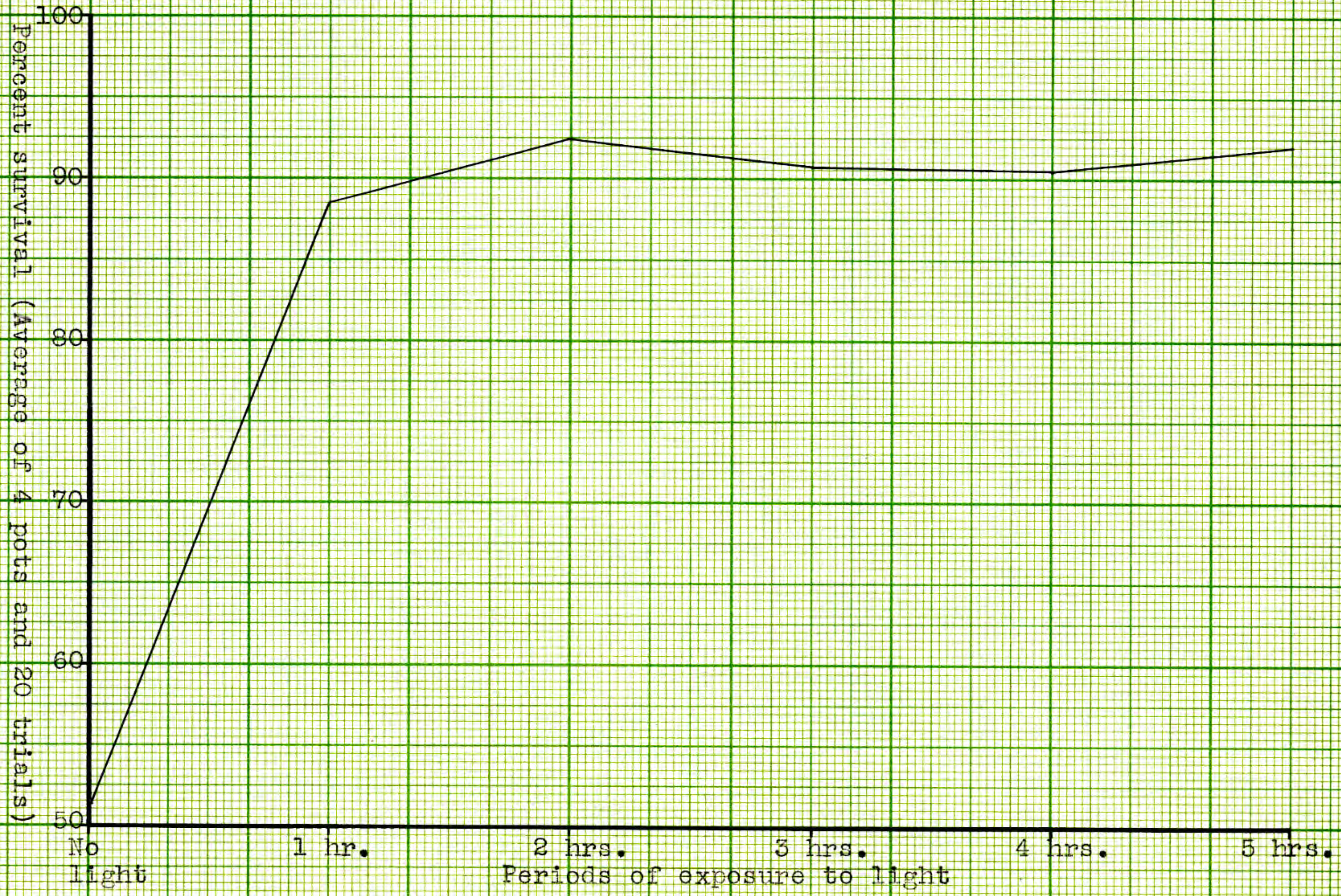


Fig. 2. Effect of long light intervals on survival.

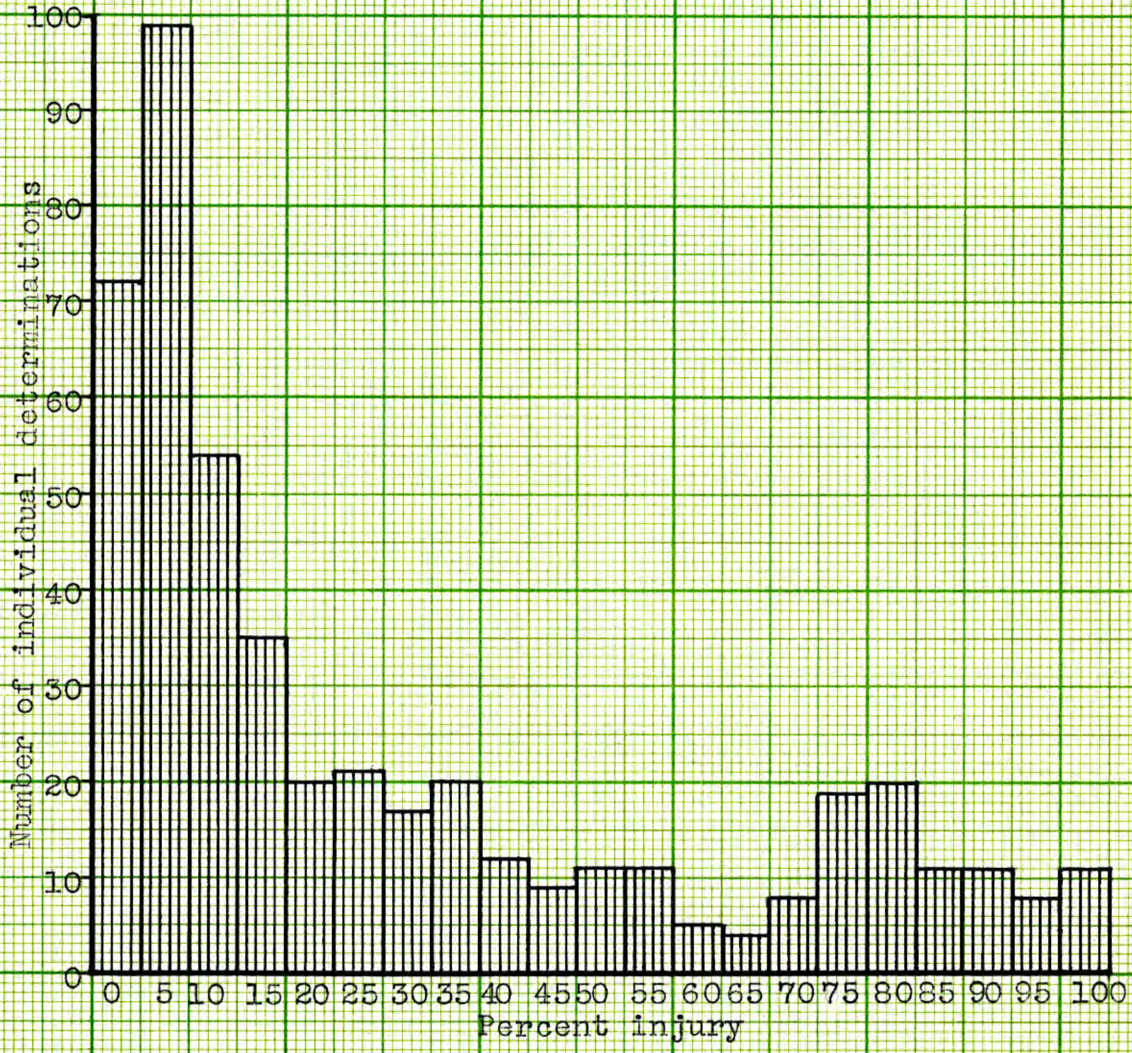


Fig. 3. Histogram of percent injury (478 individual determinations).

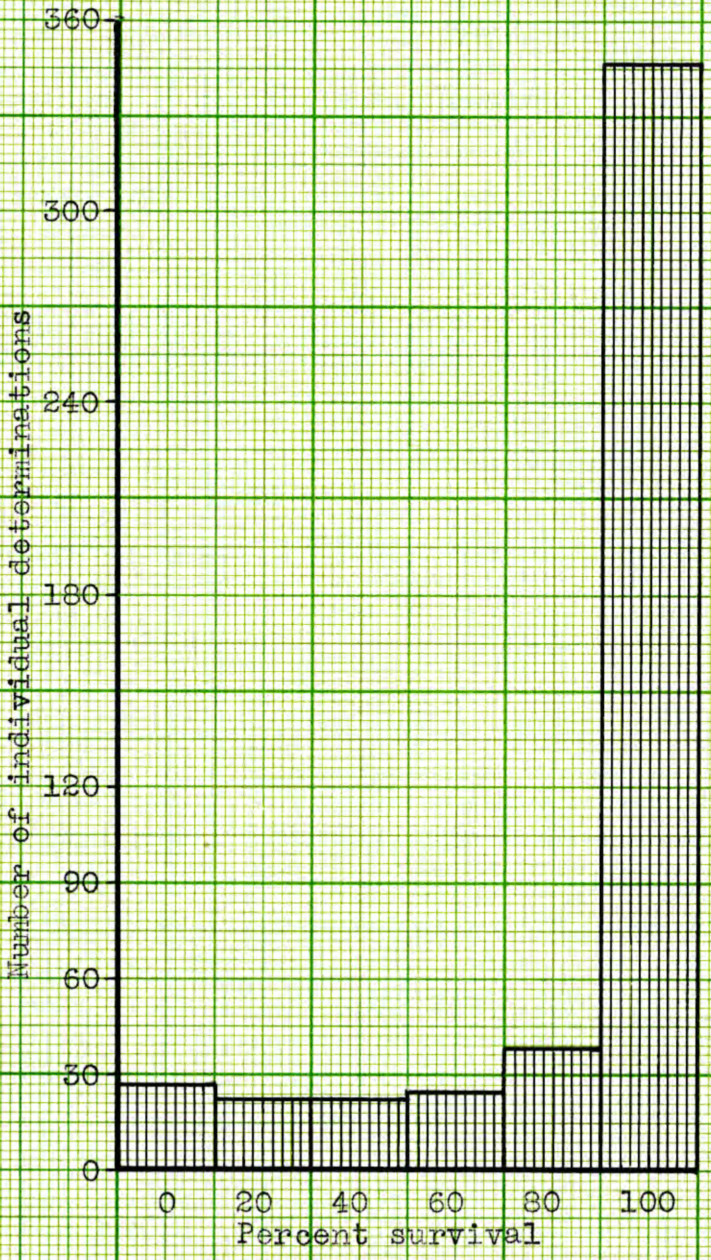


Fig. 4. Histogram of survival (478 individual determinations).

two hours of sunlight seems to increase the resistance of the plant to high temperatures as much as five hours, it is easy to explain. There would naturally be more individual observations in the lower classes of percent injury and in the higher classes of percent survival as all the individuals for four treatments would fall in these classes. The individuals of the one hour treatment are somewhat intermediate, which leaves those individuals with no light to occupy the other extremes on the percentage scale.

A scatter diagram of percent injury and percent survival is shown in Fig. 5. These seems to be a strong negative correlation between survival and injury as would be expected. This was borne out by a calculated coefficient of correlation of -.762, which is very significant.

Effect of Short Intervals of Light upon the Resistance of Wheat Seedlings to High Temperatures

As was stated previously, most of the change in both percent injury and percent survival came within the period of the first two hours the plants were exposed to light. Also, as this change was extremely rapid, it was decided to use shorter intervals of light for the first two hours to determine the reaction in that period. These data for injury and survival are shown in Tables 5 and 6, respectively.

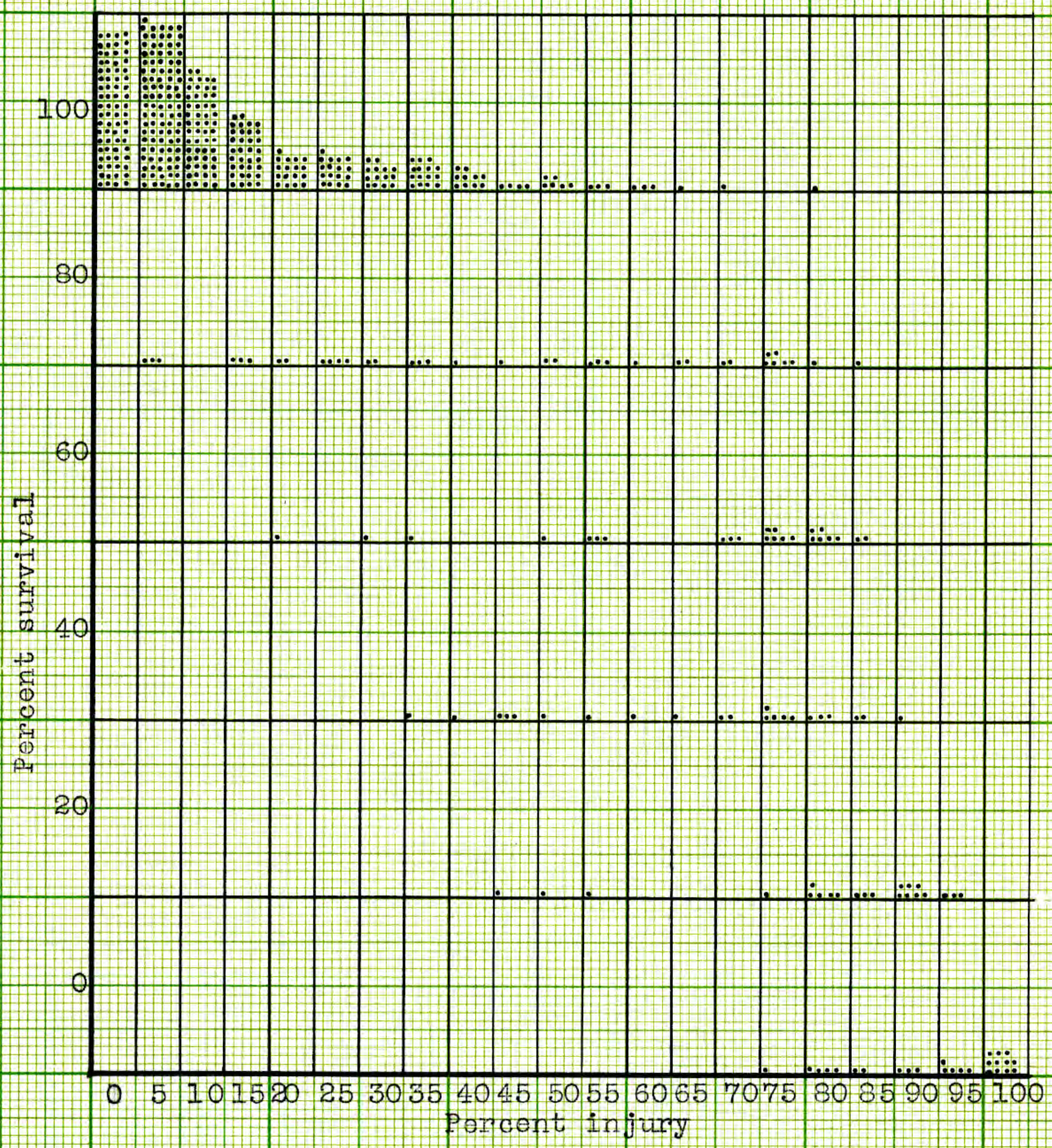


Fig. 5. Scatter diagram of injury and survival (478 individual determinations).

Table 5. Short interval light test, average percent injury of plants in four pots.

Treatment period of exposure to light	Trial						
	Dec. 1	Dec. 2	Dec. 6	Dec. 8	Dec. 9	Dec. 11	Mean
5 hours	45	35	50	76	39	50	49.58
4 hours	50	35	50	74	35	60	50.63
3 hours	50	35	53	61	35	60	49.38
$2\frac{1}{2}$ hours	50	35	55	66	30	64	50.00
2 hours	50	43	73	79	40	58	58.88
105 minutes	55	58	73	71	35	75	61.04
90 minutes	58	53	80	74	35	75	62.29
75 minutes	63	59	65	83	40	78	64.38
60 minutes	84	51	94	84	40	75	71.25
50 minutes	81	68	84	96	35	93	76.04
40 minutes	96	64	89	86	40	74	74.79
30 minutes	93	83	94	96	41	78	80.63
20 minutes	96	98	93	88	75	84	88.75
10 minutes	100	100	95	91	90	81	92.92
No light	100	100	100	96	100	93	98.54

Table 6. Short interval light test, average percent survival of plants in four pots.

Treatment period of exposure to light	Trial						
	Dec. 1	Dec. 2	Dec. 6	Dec. 8	Dec. 9	Dec. 11	Mean
5 hours	100	100	100	55	90	100	90.83
4 hours	100	100	100	65	100	100	91.67
3 hours	100	100	95	85	95	95	95.00
2½ hours	100	100	95	60	100	85	90.00
2 hours	100	100	60	55	75	90	80.00
105 minutes	95	70	70	40	100	70	74.17
90 minutes	100	90	55	35	100	75	75.83
75 minutes	75	75	90	5	65	60	58.33
60 minutes	35	85	5	10	90	55	46.67
50 minutes	40	75	35	0	100	25	45.83
40 minutes	5	70	20	5	95	95	48.33
30 minutes	25	30	5	0	70	75	33.33
20 minutes	10	10	15	15	65	50	27.50
10 minutes	0	0	15	5	15	65	16.67
No light	0	0	0	0	0	35	5.83

Again the figures given represent the average of four pots for that particular treatment. The mean for each treatment is also shown. Summary tables of the analysis of variance are shown for both injury and survival in Tables 7 and 8. Again, all variables tested are highly significant as compared to the F values for the one percent point.

The level of significance for the means of each treatment shows a value of 4.56 percent. Upon examination of the means for each treatment the difference between a treatment of ten minutes and no light is statistically significant. There is no statistical difference between ten minutes of light and twenty minutes although the difference between the two lacks only .39 percent of being significant. A significant difference is found between treatments of 20 and 30 minutes and also between 30 and 40 minutes. Those plants exposed to 50 minutes of light are somewhat out of line with the other treatments. Between treatments of 40 and 50 minutes or of 40 and 60 minutes there is no significant difference. However, the difference is statistically significant for treatments of 50 and 60 minutes.

Upon examination of the data, the December 11 trial seems to be out of line from the others. The only explanation for this seems to be that there was greater uncontrolled variation in this trial than in the other trials, which

Table 7. Summary of analysis of variance for injury for the short interval light test.

Variation due to		df	Sum of squares	Mean square	F value Calc.	5% pt.	1% pt.
Treatments		14	90,995.139	6,499.653	95.70	1.75	2.19
Trials		5	45,717.223	9,143.445	134.63	2.25	3.09
Treatments x trials		70	6,109.861	87.284	1.29	1.16	1.24
Error							
Pots	3)						
Pots x treatments	42)	270	18,337.550	67.917			
Pots x trials	15)						
Pots x treatments x trials	210)						
Total		359	161,659.723				

Standard error of a single determination = $\sqrt{67.917} = 8.024$

Standard error of the mean for each treatment = $\frac{8.024}{\sqrt{24}} = \frac{8.024}{4.89} = 1.640$

Standard error of a difference = $\sqrt{2} \times 1.640 = 2.319$

Level of significance for 5% point = 2.319 x 1.969 = 4.566

Table 8. Summary of analysis of variance for survival for the short interval light test.

Variation due to		df	Sum of squares	Mean square	F value Calc.	F value 5% pt.	F value 1% pt.
Treatments		14	287,993.333	20,570.952	38.22	1.75	2.19
Trials		5	94,586.667	18,917.333	35.15	2.25	3.09
Treatments x trials		70	101,880.000	1,455.429	2.70	1.16	1.24
Error							
Pots	3)						
Pots x treatments	42)	270	145,300.000	538.148			
Pots x trials	15)						
Pots x treatments x trials	210)						
Total		359	629,760.000				

Standard error of a single determination = 23.19

Standard error of the mean for each treatment = $\frac{23.19}{\sqrt{24}} = \frac{23.19}{4.89} = 4.742$

Standard error of a difference = $\sqrt{2} \cdot 4.742 = 6.705$

Level of significance for 5% point = 6.705 x 1.969 = 13.202

probably would have been minimized or eliminated by running more trials. A significant difference occurs between the exposure of light for 60 minutes and the next three treatments of 75, 90, and 105 minutes. Between any two of these three latter treatments, however, there is no statistical difference. As the intervals have increased here, it can be assumed that the effect of light upon resistance is diminishing. No significant difference occurs between 2 hours and 90 minutes nor 2 hours and 105 minutes. However, there is a significant difference between treatments of 75 minutes and 2 hours and between 2 hours and $2\frac{1}{2}$ hours. The difference between treatments of two and one-half, three, four, and five hours of light is not statistically significant. It can be concluded from this that light has little or no effect on the resistance of the plant to high temperatures after the plants have been exposed to light for a period of two and one-half hours.

The long interval test showed no difference between two hours and three hours of light. The latter test shows, however, that light of two hours and three hours are significantly different in affecting the plants' resistance to high temperature. During the last week in November, there were five cloudy days. A possible explanation for this discrepancy in regard to two and one-half hours of exposure to light is that the plants tested were low in resistance due to lack of sunlight when young. Therefore, sunlight might materially aid the plant in its resistance for a longer period of time than during the summer when there were few cloudy days.

As shown in Table 8, the level of significance for the five percent point for survival is 13.20. As the level of significance for injury is about one-third of that for survival, it may be that the factors within the plant are not the same for recovery and survival as those for resistance to injury. A more plausible explanation may be that less accuracy in calculating the level of significance for survival was obtained, thereby increasing the numerical value of the level of significance.

There is no significant difference between the means of treatments of no light and 10 minutes, between 10 minutes and 20 minutes, and between 20 minutes and 30 minutes in regard to survival. However, the differences are statistically significant between no light and 20 minutes of exposure to light and between 10 minutes of light and 30 minutes. In other words, the interval of significance for survival seems to be 20 minutes, while for injury it was 10 minutes. There is a slight discrepancy among treatments of 40, 50, and 60 minutes of exposure to light, but the difference is not significant. Treatments of 75 minutes are also not significant from the 40, 50, and 60 minute treatments, but it lacks very little. A statistical difference is found between 75 and 90 minutes but none occurs between 90, 105, or 120 minutes. The difference between the treatments of 105 minutes of sunlight and 2½ hours is significant. There is no significant difference in survival of wheat seedlings exposed to two, two and one-half, three, four, or five hours of sunlight. This is excepting the difference between two hours and three hours, which is statistically significant.

It should be remembered when considering significance that it is only an arbitrary point in the probability scale selected either by the investigator or by common usage. In analyzing these date the five percent point was used to denote significance. Probabilities are continuous between certainty that an event will not happen to certainty that the event will happen. Therefore, a value slightly below the level of significance has almost the same probability as a value slightly above the level. As the five percent point seems to be used by the majority of investigators and as some point must necessarily be chosen as the level of significance, it was used in the analysis of these data.

Figure 6 illustrates graphically the decrease in

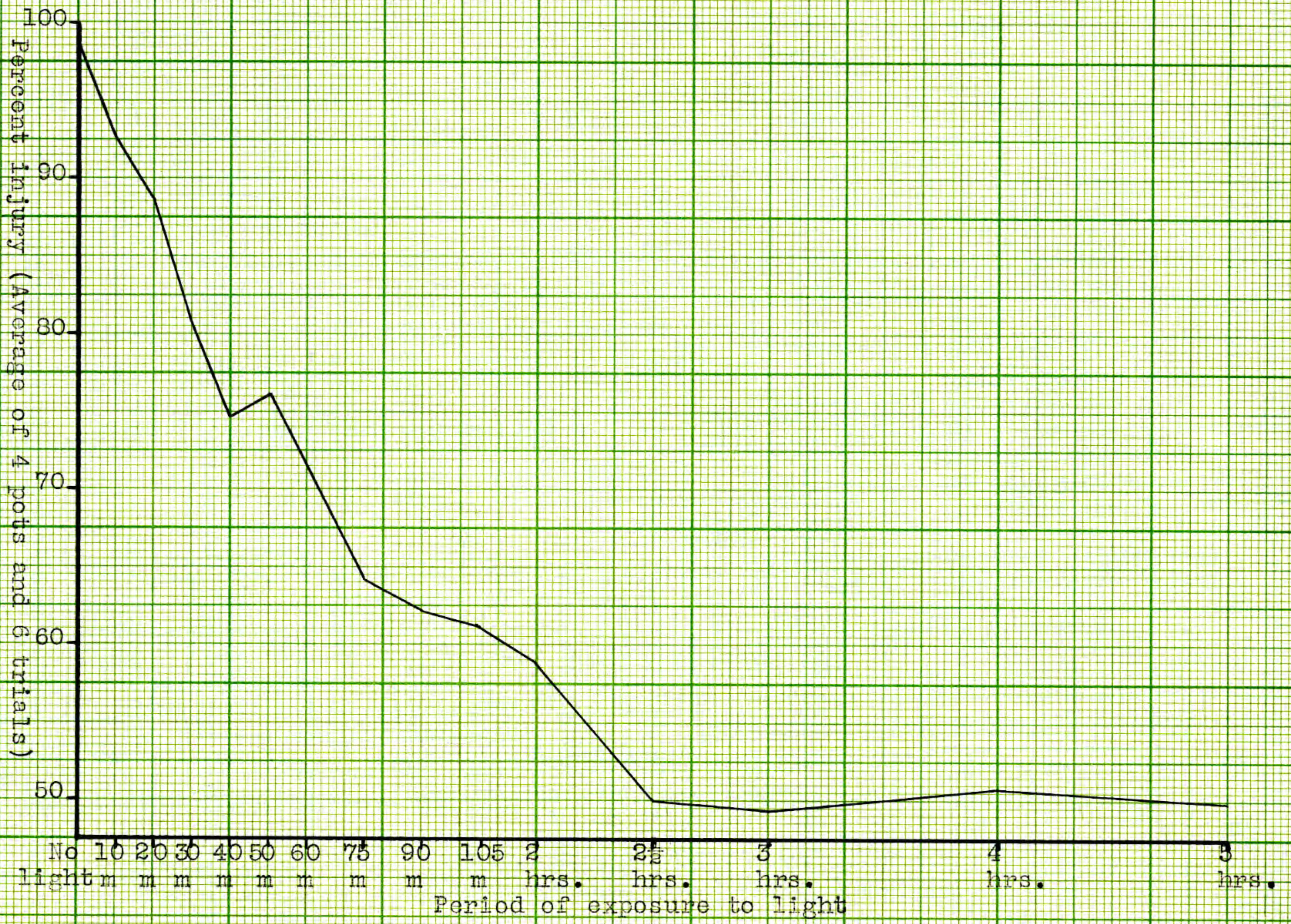


Fig. 6. Effect of short intervals of light upon injury

percent injury with an increase in exposure to light. In Fig. 7 is shown the increase in percent survival with an increase in the length of time the plants are exposed to light. These graphs show clearly that light is effective only for the first two and one-half hours of exposure. Also, the light has a greater and more rapid effect the first hour than the second hour and a still lesser effect the third hour. It is also indicated by the graphs that the hardening of the plants is gradual, although the acceleration decreases after the first hour.

The Effect of Photosynthesis on the Resistance of Wheat Seedlings to High Temperatures

It was concluded from the light interval test that light was a major factor in hardening plants to high temperatures. It would seem that photosynthetic products in the plant might account for the increase in resistance with an increase in exposure to light. Following this hypothesis, it was decided to design an experiment whereby the plant would receive light under comparatively normal conditions and still not carry on photosynthesis. Elimination of the carbon dioxide seemed the best possible means to attain this end. The carbon dioxide eliminator was set up and 12 trials run during the summer. These data are shown in Table 9 for

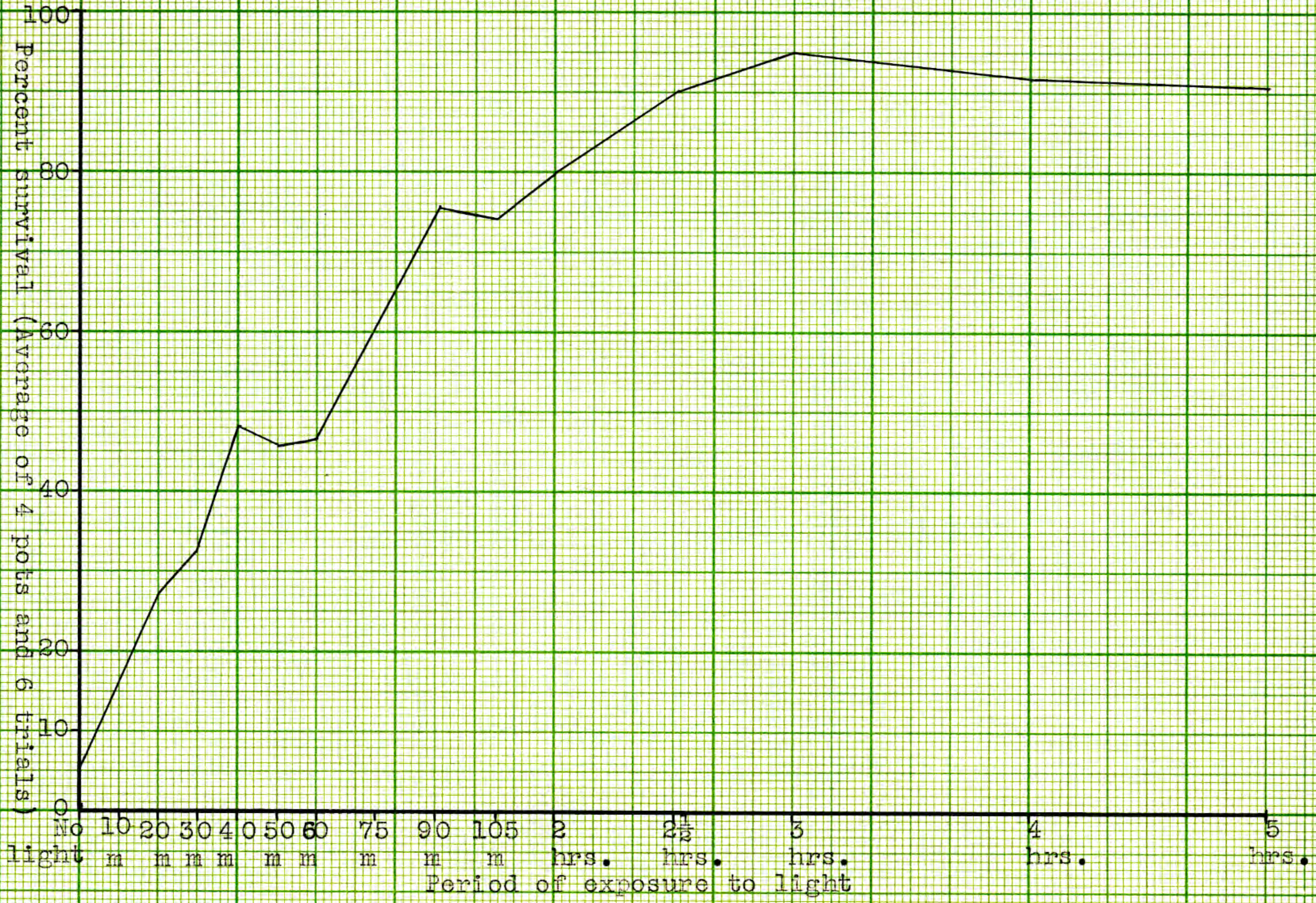


Fig. 7. Effect of short intervals of light upon survival.

injury and survival. The figures given are the original readings taken on each pot with the mean for each treatment.

As was mentioned in the material and methods, these percentage data were transformed into degrees by use of the formula $p = \sin^2\theta$ and are reported in Table 10. Analysis of variance was computed on both the transformed data and the original data. Table 11 shows the summary of the analysis of variance for injury using the original percentage data. Table 12 is the summary of the analysis of variance for injury using the transformed data. All variables were significant and those for treatments highly significant in both transformed and original data. However, the F value of the original data was 84.08 for treatments which is considerably higher than the F value of 58.52 for the transformed data. The level of significance is lower for the transformed data than for the original data (7.93 percent as compared with 8.66 percent). As the range of transformed data is narrower also (from 0°-90°) there is little if any added advantage in using transformations in the analysis of these data.

In Tables 13 and 14 are found the summaries of the analysis of variance in regard to survival for the original data and transformed data, respectively. As we are more interested in treatments than the other variables, they will be compared. The F value of the original percentage data is

Table 9. Carbon dioxide test in which only three treatments were used.

Trials 1939	Dark Pot I	Dark Pot II	CO_2 Free Pot I	CO_2 Free Pot II	Light Pot I	Light Pot II
			Percent injury			
June 20	60.0	50.0	35.0	45.0	20.0	25.0
June 22	100.0	95.0	80.0	100.0	85.0	75.0
June 23	80.0	100.0	100.0	70.0	65.0	80.0
July 6	75.0	85.0	55.0	55.0	5.0	5.0
July 7	80.0	85.0	10.0	5.0	10.0	0.0
July 8	100.0	90.0	90.0	10.0	10.0	5.0
July 10	85.0	95.0	40.0	10.0	5.0	0.0
July 11	95.0	85.0	10.0	15.0	0.0	5.0
July 12	85.0	65.0	100.0	40.0	10.0	0.0
August 19	100.0	100.0	98.0	100.0	100.0	80.0
August 22	98.0	70.0	98.0	95.0	35.0	30.0
August 26	100.0	100.0	100.0	90.0	45.0	55.0
Mean	86.58%		60.46%		31.25%	
			Percent survival			
June 20	80.0	60.0	100.0	100.0	100.0	100.0
June 22	0.0	20.0	40.0	0.0	20.0	60.0
June 23	60.0	0.0	0.0	0.0	40.0	40.0
July 6	40.0	40.0	60.0	60.0	80.0	100.0
July 7	60.0	20.0	100.0	100.0	100.0	100.0
July 8	0.0	40.0	20.0	80.0	100.0	100.0
July 10	40.0	20.0	60.0	100.0	100.0	100.0
July 11	0.0	0.0	100.0	80.0	100.0	100.0
July 12	60.0	80.0	0.0	80.0	100.0	100.0
August 19	0.0	0.0	0.0	0.0	0.0	60.0
August 22	0.0	40.0	0.0	0.0	60.0	60.0
August 26	0.0	0.0	0.0	20.0	60.0	60.0
Mean	28.33%		45.83%		76.67%	

Table 10. Carbon dioxide test in which only three treatments were used.

Trials 1939	Dark Pot I	Dark Pot II	CO_2 Free Pot I	CO_2 Free Pot II	Light Pot I	Light Pot II
			Degree injury			
June 20	50.77	45.00	36.28	42.13	26.57	30.00
June 22	90.00	77.08	63.43	90.00	67.22	60.00
June 23	63.43	90.00	90.00	56.79	53.73	63.43
July 6	60.00	67.22	47.87	47.87	12.93	12.93
July 7	63.43	67.22	18.43	12.93	18.43	0.00
July 8	90.00	71.57	71.57	18.43	18.43	12.93
July 10	67.22	77.08	39.23	18.43	12.93	0.00
July 11	77.08	67.22	18.43	22.73	0.00	12.93
July 12	67.22	53.73	90.00	39.23	18.43	0.00
August 19	90.00	90.00	81.81	90.00	90.00	63.43
August 22	81.81	56.79	81.81	77.08	36.28	33.21
August 26	90.00	90.00	90.00	71.57	42.13	47.87
Average	72.66		54.83		30.57	
			Degree survival			
June 20	63.43	50.77	90.00	90.00	90.00	90.00
June 22	0.00	26.57	39.23	0.00	26.57	50.77
June 23	50.77	0.00	0.00	0.00	39.23	39.23
July 6	39.23	39.23	50.77	50.77	63.43	90.00
July 7	50.77	26.57	90.00	90.00	90.00	90.00
July 8	0.00	39.23	26.57	63.43	90.00	90.00
July 10	50.77	26.57	50.77	90.00	90.00	90.00
July 11	0.00	0.00	90.00	63.43	90.00	90.00
July 12	50.77	63.43	0.00	63.43	90.00	90.00
August 19	0.00	0.00	0.00	0.00	0.00	50.77
August 23	0.00	39.23	0.00	0.00	50.77	50.77
August 26	0.00	0.00	0.00	26.57	50.77	50.77
Average	25.72		40.62		68.46	

Table 11. Summary of analysis of variance for injury using the original percentage data obtained from the carbon dioxide test in which three treatments were involved.

Variation due to		df	Sum of squares	Mean square	F value Calc.	5% pt.	1% pt.
Treatments		2	36,779.361	18,389.681	84.08	3.26	5.26
Trials		11	35,527.486	3,229.771	14.77	2.07	2.79
Treatments x trials		22	17,453.306	793.332	3.63	1.87	2.42
Error							
Pots	1)						
Pots x treatments	2)	36	7,873.500	218.708			
Pots x trials	11)						
Pots x treatments x trials	22)						
Total		71	97,633.653				

Standard error of a single determination = $\sqrt{218.708} = 14.78$

Standard error of the mean for each treatment = $\frac{14.78}{\sqrt{24}} = \frac{14.78}{4.89} = 3.022$

Standard error of a difference = $\sqrt{2} \cdot 3.022 = 4.273$

Level of significance for 5% point = 2.028 x 4.273 = 8.666

Table 12. Summary of analysis of variance for injury using transformed data from the carbon dioxide test in which three treatments were used.

Variation due to		df	Sum of squares	Mean square	F Value Actual	F Value 5% pt.	F Value 1% pt.
Treatments		2	21,420.417	10,710.209	58.52	3.26	5.26
Trials		11	22,822.970	2,074.815	11.33	2.07	2.79
Treatments x trials		22	8,155.679	370.713	2.03	1.87	2.42
Error							
Pots	1)						
Treatments x pots	2)	36	6,588.256	183.007			
Trials x pots	11)						
Treatments x trials x pots	22)						
Total		71	58,609.322				

Standard error of a single determination = 13.53

Standard error of the mean for each treatment = $\frac{s}{\sqrt{n}} = \frac{13.53}{4.89} = 2.767$

Standard error of a difference = $\sigma_{\bar{x}} \sqrt{2}$ = 2.767 x 1.414 = 3.913

Level of significance for the 5% point = 3.158 x 2.028 = 7.935

Table 13. Summary of analysis of variance for survival using original percentage data obtained from the carbon dioxide test in which three treatments were involved.

Variation due to		df	Sum of squares	Mean square	F value Calc.	5% pt.	1% pt.
Treatments		2	28,744.453	14,372.227	95.93	3.26	5.26
Trials		11	48,727.778	4,429.798	29.26	2.07	2.79
Treatments x trials		22	27,472.214	1,248.737	8.24	1.87	2.42
Error							
Pots	1)						
Pots x treatments	2)	36	5,450.000	151.389			
Pots x trials	11)						
Pots x treatments x trials	22)						
Total		71	110,394.445				

Standard error of a single determination = $\sqrt{151.389} = 12.31$

Standard error of the mean for each treatment = $\frac{12.31}{\sqrt{24}} = \frac{12.31}{4.89} = 2.517$

Standard error of a difference = $2.517 \cdot \sqrt{2} = 3.559$

Level of significance for 5% point = 2.028 x 3.559 = 7.218

Table 14. Summary of analysis of variance for survival using transformed data from the carbon dioxide test involving three treatments.

Variation due to	df	Sum of squares	Mean square	F value Actual	5% pt.	1% pt.
Treatments	2	22,589.0656	11,294.5328	29.30	3.26	5.26
Trials	11	37,363.3443	3,396.6676	8.81	2.07	2.79
Treatments x trials	22	9,922.1824	451.0083	1.17	1.87	2.42
Error						
Pots 1)						
Treatments x pots 2)	36	13,875.2421	385.423			
Trials x pots 11)						
Treatments x trials x pots 22)						
Total	71	83,749.8344				

Standard error of a single determination = 19.63

Standard error of the mean for each treatment = $\frac{s}{\sqrt{n}} = \frac{19.63}{4.89} = 4.014$

Standard error of a difference = $\sigma_{\bar{x}}\sqrt{2} = 4.014 \times 1.414 = 5.676$

Level of significance for the 5% point = 5.676 x 2.028 = 11.511

about three and one-half times as large as that of the transformed data. In addition, the level of significance for the transformed data was 11.51 and only 7.218 for the original percentage data. In all cases when compared to the means of treatments, the differences are significant. However, the original percentage data has a range from 0 to 100 percent, which would allow a greater level of significance to equal a smaller level of significance in the transformed data which has a range only from 0 to 90 degrees. In the survival tests, the level of significance for the transformed data is higher than that of the original percentage data, indicating that the accuracy in this case was decreased by a transformation of the data.

In Fig. 8 is shown a graphic representation of the means for the different treatments of both injury and survival for both the original data and the transformed data. This also shows clearly that there is little difference between the original and transformed data.

It was concluded that with these data, the accuracy of the generalized standard error was increased very little, if any, and the increased validity was not worth the expenditure of time in transforming the data. It was then decided that the remainder of the data would be analyzed on the basis of the original percentage readings.

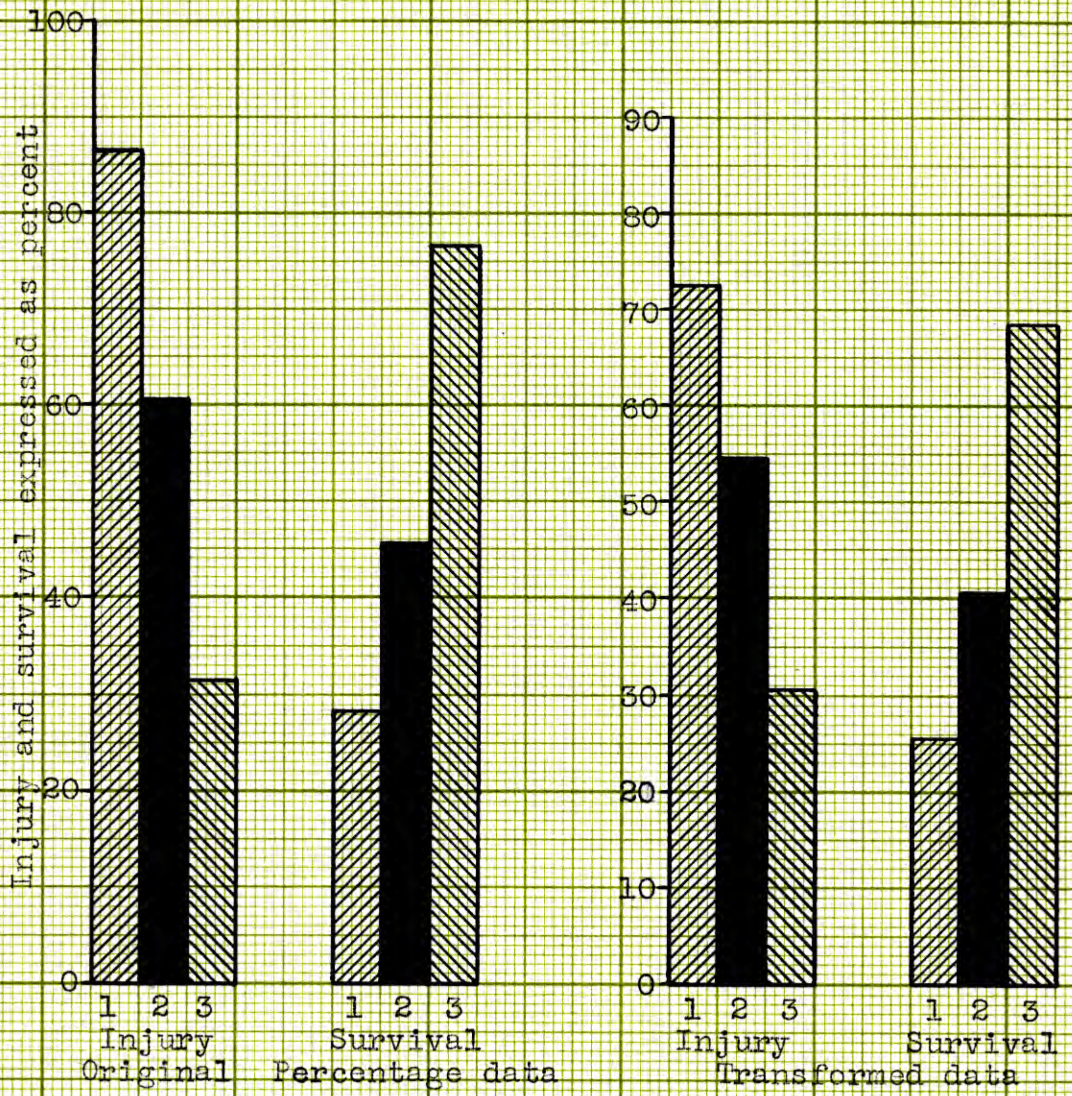


Fig. 8. Bar diagram showing percent injury and survival for both original and transformed data.

Figures 9 and 10 show the histograms for injury and survival, respectively. Again, there is an accumulation of individual observations at the limits of the range. However, here are shown three distinct peaks in the histograms of both injury and survival. This probably is the result of the three treatments even though the means for the treatments do not fall very close to the three modes. The reason for this discrepancy is that there was considerable variation between the levels of trials as is shown by the F values for trials in all the analysis of variance summary tables.

According to the hypothesis that photosynthesis products cause a plant to be resistant to high temperatures, those plants placed in the carbon dioxide eliminator for a period of five hours should theoretically be as susceptible to heat as those remaining in the dark for that length of time. However, this was not true as shown by the means for each treatment of both survival and injury. The differences between each of the means was highly significant in both injury and survival when compared to the level of significance for the five percent point. This, however, can be explained by the fact that the CO_2 of respiration is undoubtedly being used before it reaches the atmosphere. Miller (1910) stated, "The carbon dioxide liberated by the seedling by respiration during daylight is, under these conditions,

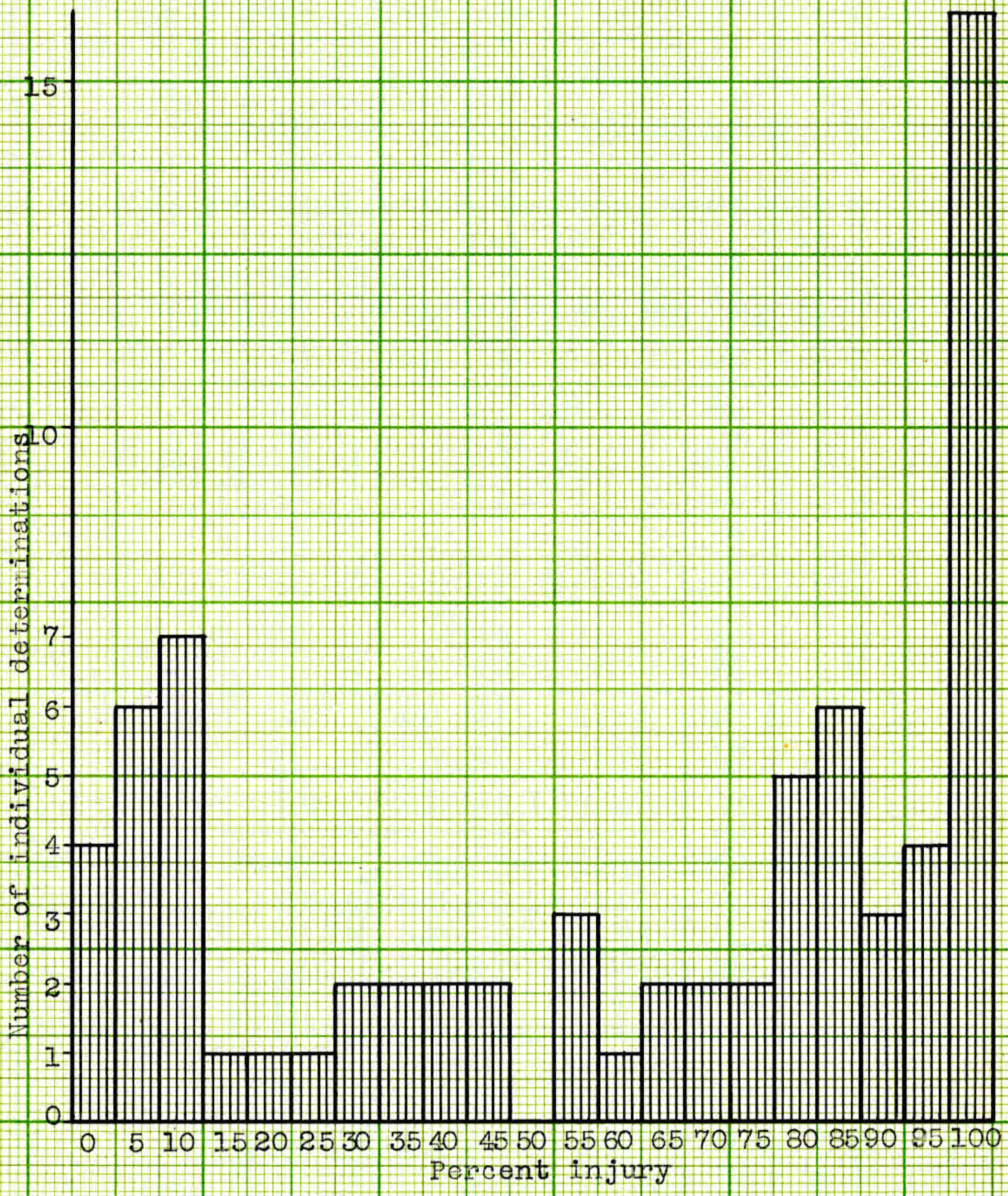


Fig. 9. Histogram of percent injury (72 individual determinations).

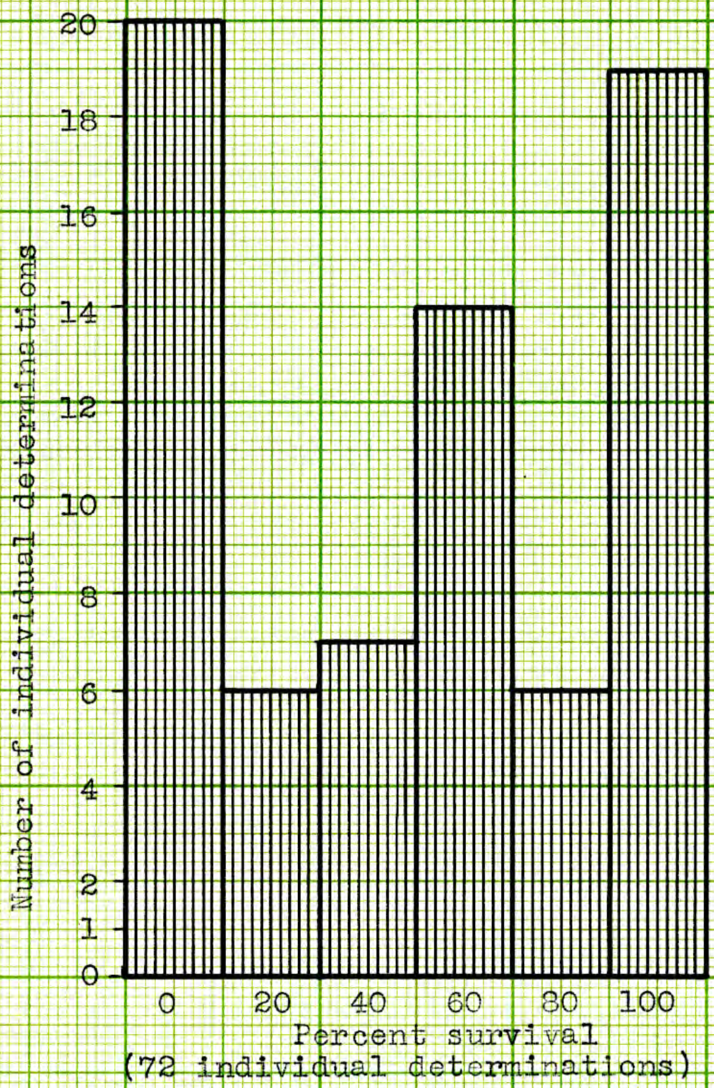


Fig. 10. Histogram of percent survival.

probably used in photosynthetic processes before it leaves the plant. There seems to be no possible means of preventing the CO_2 thus liberated from being utilized by the plant in daylight." These "conditions" referred to by Miller in the above quotation are conditions similar to those of plants in the carbon dioxide eliminator. Also, as shown by the short light interval test, a period of 10 minutes light imparts much resistance to plants as there is no way of placing the plants in the carbon dioxide eliminator without allowing atmospheric air to fill the bell jars. This may be available for photosynthesis for a short period. It is not known how long it takes the air to become free from carbon dioxide as it is a problem of dilution rather than complete elimination. Therefore, if photosynthesis is one of the mechanisms of resistance, the plants in the carbon dioxide eliminator would be expected to be more resistant to high temperatures than those in the dark.

As plants in the carbon dioxide eliminator and those in light were not under quite comparable exposures to light, it was decided to insert a fourth treatment into the experiment. In order for rays of light to reach the plants in the eliminator, they must pass first through the glass in the greenhouse and then through the glass of the bell jars. The rays of light hitting those plants placed under normal greenhouse conditions had only to pass through the layer of glass on the greenhouse. The fourth treatment consisted of two pots of plants placed under bell jars so as to allow free circulation of air around the plants but so that no sunlight could reach the plants except through the bell jars. By this method it could be determined whether or not there was a screening effect exercised by the bell jars. In Table 15 is shown the original percentage data for both injury and survival for each pot and the mean for each treatment. The experiment was repeated 10 times, the data being reported in Table 15.

Tables 16 and 17 show the summaries of analysis of variance and the levels of significance for injury and survival, respectively. Again, the level of significance for survival is three times that of injury. The level of significance of 4.019 for the five percent point indicates that the differences between the means of injury of dark and carbon dioxide treatments are highly significant. There is a greater difference between the means of those plants placed in the carbon dioxide eliminator and those placed in the open bell jars. Also, the difference is very highly significant between plants receiving normal sunlight in the green house and those having no carbon dioxide. There is also a significant difference between the means of plants placed

Table 15. Original percentage data obtained from the carbon dioxide test in which four treatments were used.

Trial No.	1939	Dark Pot I	Dark Pot II	CO_2 Free Pot I	CO_2 Free Pot II	Open jars Pot I	Open jars Pot II	Light Pot I	Light Pot II
					Percent injury of each pot				
1	Dec. 2	90	100	60	60	40	40	45	45
2	Dec. 4	100	100	75	100	30	30	45	45
3	Dec. 5	85	95	60	60	40	40	40	35
4	Dec. 6	100	100	85	85	60	60	70	70
5	Dec. 8	100	100	70	95	45	45	50	50
6	Dec. 9	100	100	90	85	35	35	45	45
7	Dec. 11	100	100	50	45	40	45	30	40
8	Dec. 15	100	100	55	45	40	40	40	40
9	Dec. 21	60	60	70	90	35	35	40	40
10	Dec. 27	100	100	100	100	85	60	90	70
Mean		94.5		74.0		44.0		48.75	
					Percent survival of each pot				
1	Dec. 2	40	0	100	100	100	100	100	100
2	Dec. 4	0	0	60	0	100	100	100	100
3	Dec. 5	80	20	80	100	100	100	100	100
4	Dec. 6	0	0	40	60	100	100	100	100
5	Dec. 8	0	0	80	20	100	100	100	100
6	Dec. 9	0	0	20	40	100	100	100	100
7	Dec. 11	0	0	100	20	100	80	80	40
8	Dec. 15	0	0	80	100	100	100	100	100
9	Dec. 21	80	100	80	20	100	100	100	100
10	Dec. 27	0	0	0	0	20	80	20	60
Mean		16.0		55.0		94.0		90.0	

Table 16. Summary of analysis of variance for injury using the original percentage data obtained from the carbon dioxide test in which four treatments were involved.

Variation due to		df	Sum of squares	Mean square	F value Calc.	F value 5% pt.	F value 1% pt.
Treatments		3	33,118.438	11,039.479	2781.64	2.84	4.31
Trials		9	8,770.313	974.479	24.55	2.16	2.91
Treatments x trials		27	6,790.937	251.516	6.34	1.75	2.25
Error							
Pots	1)						
Pots x trials	9)	40	1,587.494	39.687			
Pots x treatments	3)						
Pots x trials x treatments	27)						
Total		79	50,267.182				

Standard error of a single determination = $\sqrt{39.687} = 6.29$

Standard error of the mean for each treatment = $\frac{6.29}{\sqrt{20}} = \frac{6.29}{4.472} = 1.407$

Standard error of a difference = $\sqrt{2} \cdot 1.407 = 1.989$

Level of significance for 5% point = 1.989 x 2.021 = 4.019

Table 17. Summary of analysis of variance for survival using original percentage data obtained from the carbon dioxide test in which four treatments were involved.

Variation due to		df	Sum of squares	Mean square	F value Calc.	F value 5% pt.	F value 1% pt.
Treatments		3	79,215	26,405	66.85	2.84	4.31
Trials		9	25,225	2,802.778	7.10	2.16	2.91
Treatments x trials		27	22,235	823.519	2.08	1.75	2.25
Error							
Pots	1)						
Pots x trials	9)	40	15,800	395.00			
Pots x treatments	3)						
Pots x treatments x trials	27)						
Total		79	142,475				

Standard error of a single determination = $\sqrt{395} = 19.87$

Standard error of the mean for each treatment $\frac{19.87}{\sqrt{20}} = \frac{19.87}{4.472} = 4.443$

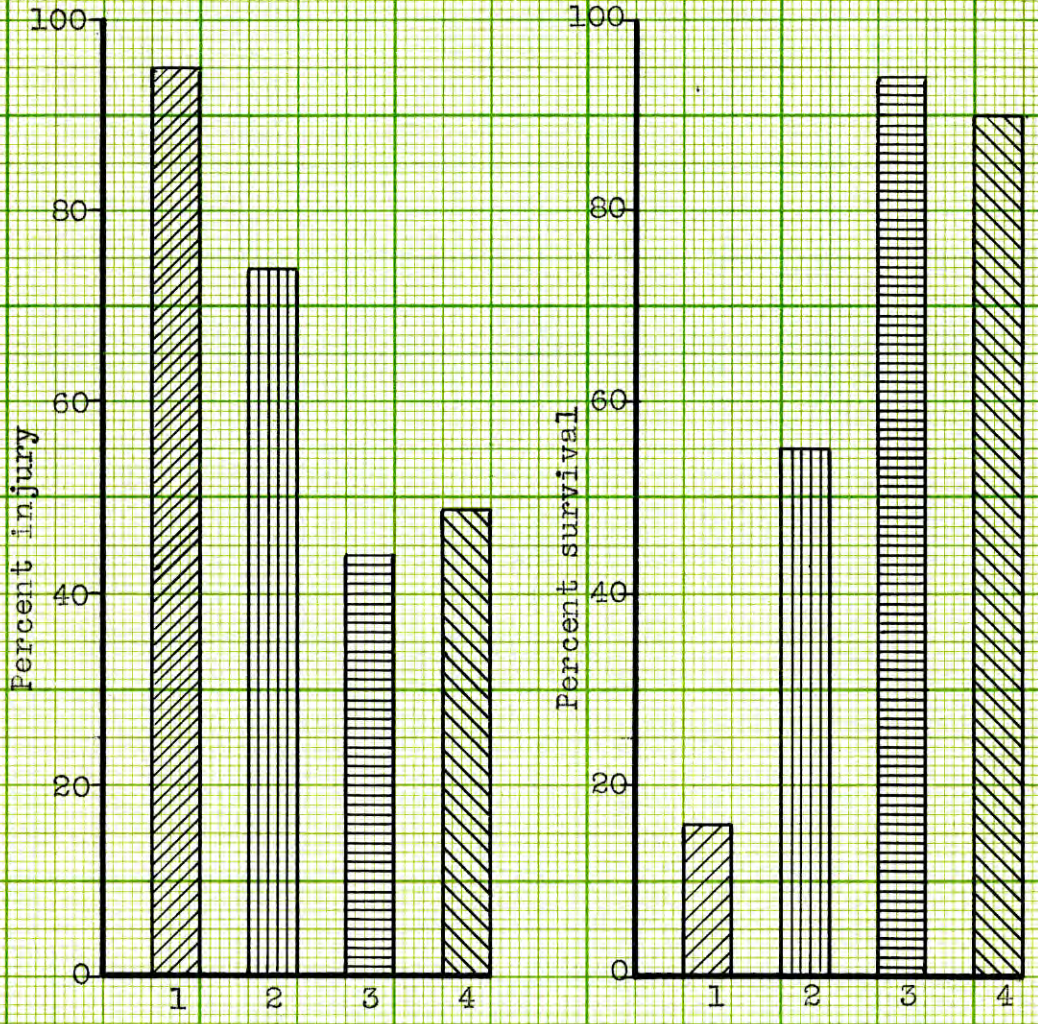
Standard error of a difference = $\sqrt{2} \cdot 4.443 = 6.282$

Level of significance for 5% point = $6.282 \times 2.021 = 12.696$

in open bell jars and those plants placed in the light under natural greenhouse conditions. According to the hypothesis, these two treatments should have no significant difference in their effect upon the resistance of wheat seedlings to high temperatures. The temperatures of the air surrounding the plants in all treatments were recorded soon after it was noticed that the plant in the open bell jars were injured less than those in the natural light in the greenhouse. However, the temperatures between any of the treatments varied not more than two degrees, so probably the hardening influence of the open bell jars is not a temperature factor. There was no way of measuring the relative humidity of air surrounding the plants in the various treatments. However, there was no condensate on the walls of the open bell jars so it was assumed that the relative humidity within the open bell jars and the atmosphere of the greenhouse were essentially the same. Perhaps a possible explanation would be that there was a concentration of carbon dioxide in the bell jars but as it is heavier than air this explanation is unlikely. In the last two trials, condensate was noticed on the bell jars of carbon dioxide eliminator. This might change the light relationship in the last two trials as the rays of light must pass through drops of water in addition to glass in order to reach the plant. However, this occurred in only two trials and probably had little effect on resistance. The temperature of the greenhouse varied considerably during the treatment and probably caused the condensation.

The level of significance for the five percent point for survival was 12.696. Upon examination of the means of each treatment in regard to survival, it is found that there is no significant difference between plants in the normal light and those in the open jars, although the survival was somewhat greater for plants in the open jars. There is a significant difference between the means of plants treated in the dark and those treated in the carbon dioxide eliminator. The difference between the means of the carbon dioxide free treatment and the open jars, the carbon dioxide free treatment and the direct light in the greenhouse, and the treatment in the dark compared with the open jars and the light are all statistically significant. All the calculated F values for injury greatly exceed the one percent point, indicating high significance. Both treatments and trials are highly significant in the analysis of variance for survival. The F value for the interaction between treatments and trials exceeds the five percent point but not the one percent point.

Figure 11 illustrates graphically the means of the



1 - Treatment of no light
2 - Treated in CO_2 eliminator
3 - Treated in open bell jars
4 - Exposed to normal greenhouse sunlight

Fig. 11. Bar diagram showing percent injury and survival of plants exposed to artificial drought after receiving treatments 1, 2, 3, and 4.

treatments for both injury and survival. Plate II shows a representative trial of the carbon dioxide tests.

DISCUSSION

It has been stated by Maximov (1929) that the study of drought resistance is probably as complex a problem as that involving any other plant character. Drought resistance not only must be considered from the standpoint of the plant's reaction to high temperatures, low humidity, and other atmospheric factors but also from the standpoint of soil factors, and the plant's own physiological and morphological adaptation to such adverse conditions. The means that a plant employs to avoid a too intensive loss of water are varied, numerous, and different for different species.

A knowledge of the mechanisms of drought resistance would pave the way for satisfactory methods of determining drought resistance under controlled conditions, thereby materially aiding the plant breeder in his selection of drought resistant varieties. In order to accomplish this aim, the problem of drought resistance must be split into its various subdivisions and the sections studied separately. For example, it has been found by Heyne and Laude (1940) and corroborated by the work discussed in this paper, that light greatly hardens plants to high temperatures. This

EXPLANATION OF PLATE II

All four pots were placed in the heat chamber at a temperature of 130° F. and 35 percent humidity for a period of five hours. Treatment preceding trials was as follows:

Pot 1 - placed in sunlight in the greenhouse for 5 hours.

Pot 2 - placed in sunlight in open bell jars for 5 hours.

Pot 3 - placed in CO_2 eliminator for 5 hours.

Pot 4 - plants received no light since day preceding the trial.

Plate II

indicates that a lack or deficiency of sunlight decreases the tolerance to heat in at least wheat and corn. Tumanov (1929), however, found that plants grown in the shade were more resistant to edaphic drought than plants grown in the direct sunlight. Caldwell (1913) showed that plants growing under shady conditions reduced the water content of the soil to a greater degree than plants grown in direct sunlight. This property would enable them to resist soil drought for a longer period. On the strength of these investigations it may be concluded that sunlight is advantageous to the plant in resisting high temperatures but harmful to its evasion of edaphic drought. These factors might tend to counteract one another and the action of sunlight obscured if the subdivisions were not studied separately.

Similarly, plants grown under humid conditions were found by Caldwell (1913) to deplete the soil moisture to a greater degree, thereby resisting edaphic drought for a longer time. On the contrary, Berkley and Berkley (1933), working with cotton, found that for an exposure of one minute, the cotton plant was killed in moist air at 65° C. In dry air 84° C. was the lethal temperature required. Again there seems to be an opposing reaction when both atmospheric and edaphic drought are considered.

In considering the effect of light on the tolerance of wheat seedlings to high temperatures, it might be assumed the factor in resistance was the photosynthetic products formed. A dearth of carbon dioxide should therefore produce the same effect as lack of sunlight. This seemed to be the case, as shown by the tests with carbon dioxide reported in this manuscript. Although the plants treated in the carbon dioxide eliminator were injured significantly less than those in the dark, the carbon dioxide of respiration would be available to those plants in the eliminator for photosynthesis. Also, the carbon dioxide could not be eliminated directly but was diminished by a process of dilution.

As shown by these tests and previous experiments, there is little doubt but that photosynthesis is partly the cause of resistance in seedling plants of wheat and corn. This seems to be only a partial explanation, as the greatest resistance was garnered by the plant during the first hour of exposure to light and probably the first period of ten minutes aided the plant more than any other equal period the plant was exposed to sunlight. It is doubtful if photosynthesis can act to such an extent as to cause this rapid increase in resistance. It was observed by Osterhout and Haas (1918) and Spoehr and McGee (1923) that the rate of photosynthesis of plants, which have remained in the darkness for a period and then exposed to light, is slow at first and then steadily increases to the maximum. This would further tend to prove that photosynthesis was not responsible for the resistance gained by wheat seedlings during the first hour of exposure. Andrews (1925) stated that temporary starch appeared in the chloroplasts as early as six minutes after exposure to sunlight. Sugar is formed more quickly than starch so it would be safe to estimate that three minutes of exposure would cause the synthesis of sugar. Even in the face of this information, it is doubtful that photosynthesis is the sole cause of resistance to atmospheric drought.

If photosynthesis is not responsible then the best probable solution left is through the phenomena called photocatalytic action. According to Miller (1938), Trumpf discovered that periods of exposure of sunlight of only one or two minutes produced marked changes in the appearance of etiolated plants.

Priestly (1925) has confirmed the work of Trumpf in showing that light exposure of one or two minutes per day, even to a relatively weak artificial light, will fail to produce any signs of chlorophyll in the expanded leaf tissue while effectively removing most of the characteristic effects of etiolation. This evidence seems to dispel the contention that photosynthetic products are concerned in these changes during such brief periods. Further, the experiments of Trumpf were carried on at such low temperatures that growth could not take place, yet the effect of the brief exposures of light still occurred. Of course, the experiments of Trumpf and Priestly were concerned only with morphological changes of etiolated plants. However, it is well within the realm of possibility that resistance may be affected in a similar manner.

The exact chemical action stimulated by light is yet unknown. Priestly (1925) suggested that it was "a photocatalytic action upon fatty or lipoid substances which has the result either of releasing them from the surface of the protoplast into the wall or setting them free from combination within the wall, with the result that they slowly diffuse through the aqueous substratum of the cellulose wall and finally accumulate at the surface of the shoot in the cuticle." The theory advanced by Priestly may explain the morphological changes which occur in the etiolated plants, but it is hardly the answer to the problem of heat tolerance.

The physiological reaction of the plant to light has been studied in regard to a number of characters. Perhaps the most obvious from the standpoint of its application to the question of drought tolerance is permeability. Miller (1938), citing Lepeschkin, stated that light increased the permeability of the pulvinal cell of legumes resulting in a decrease in volume and turgor. Darkness caused a decrease in permeability of the protoplasm bringing about an increased turgor and volume. The speed with which these reactions occur is not known but it is entirely possible that they are affected readily by light. Darkness would therefore cause a more luscious, succulent plant which probably would be injured to a greater extent by high temperatures than those plants in the light.

The effect of light on proteins should perhaps also be considered. Miller (1938) concluded, after the review of literature on the subject, that light had little or no effect on protein synthesis provided there was a sufficient supply of sugars present in the cells. As this would reflect back upon the problem of photosynthesis, it in no way concerns the photocatalytic response of plants.

No method of attacking this problem can be suggested at the present time. The photocatalytic response seems to be so delicate that the many variables involved can not be controlled with the present equipment.

SUMMARY AND CONCLUSIONS

The effect of light and photosynthesis on the resistance of wheat seedlings to high temperature was studied. The results obtained agreed very well with previous work in regard to the influence of light.

Three main tests were studied: (1) the effect of long intervals of light upon the resistance of seedling plants of wheat to high temperatures; (2) the effect of short intervals of light on the heat tolerance of wheat seedlings; and (3) the effect of the lack of carbon dioxide on the resistance of seedling plants of wheat to atmospheric drought under controlled conditions.

Testing 14-day-old seedlings in the heat chamber for a period of five hours at 120° F. and a relative humidity of 35 percent showed that the resistance was increased the greatest during the first hour of exposure to light. After two hours of light there was no increase in the resistance of seedlings with a corresponding increase in the exposure to light. This was confirmed by the short interval light test in which 21-day-old seedlings were tested at 130° F. and a relative humidity of 35 percent.

Plants placed in a specially devised carbon dioxide eliminator reacted to high temperatures in a similar manner as plants kept in the darkness preceding the heat treatment although the difference between them was significant.

A statistical study was made of the individual readings obtained in tests involving the elimination of carbon dioxide to determine the advisability of transforming percentage data. It was concluded that with these data the accuracy of the generalized standard error was not increased sufficiently to compensate for the extra labor involved in making the transformations.

It was concluded from these studies that light and carbon dioxide have a marked effect upon the resistance of seedling plants of wheat to high temperatures. These results indicate that the products of photosynthesis are instrumental in causing plants to be resistant to high temperatures, although other mechanisms are probably involved.

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