COLD RESISTANCE OF WINTER WHEAT AS RELATED TO NUTRIENT CONDITIONS OF THE SOIL

by

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INTRODUCTION

The phenomenon of winter hardiness has long been observed in plants. The amount of injury resulting from exposure to a given low temperature varies greatly from plant to plant or even on the same plant at different times. This phenomenon is governed by a complex group of factors of both internal and external nature. Quisenberry and Clark (40), Hayes and Garber (19) and others have demonstrated the complexity of the actual causes of hardiness by the inheritance of this characteristic. Temperature, light, moisture and mineral nutrition are vital external factors controlling the winter hardiness of plants which have been investigated by Tysdal (51), Suneson and Peltier (48), Salmon (43), Dexter (10), Laude (30), Wilhelm (53) and many others. None of these external factors has a simple relationship to cold resistance but investigations of all these environmental factors have been the chief source in discovering and understanding some of the internal causes of cold resistance.

The importance of this problem is well established by a perusal of surveys for the first forty years of this century. Quisenberry (39) reports that an average of 11 per cent of the acreage of winter wheat was abandoned from 1901 to 1928, which was largely due to winter killing. Bayles and Taylor (2) state that wheat losses from 1909 to 1937 were only 1 per cent in some winters while in others as much as 60 per cent was destroyed, with an average of 10 per cent. Quisenberry and Clark (40) stress the importance of the problem in stating that losses in wheat due to winter killing are nearly as great as from all the diseases combined. This large, but fluctuating, economic loss caused by low temperature in fall, winter or spring has for many years stimulated interest and investigation by many scientific workers in many countries, as to the basic cause and effect of this injury. It is hoped that as a result of research into these basic causes intelligent plant breeding and effective methods of control could be derived to alleviate this great yearly drain on our agricultural economy. The efforts of science during the last fifty years have not been without effect, although the basic cause of winter hardiness of plants still remains but slightly understood. The reduction of economic loss has been substantial due to introduction and improvement of winter hardy varieties, cultivation methods and date of planting. That this problem is of vital importance is further emphasized by the report of the Food and Agricultural Organization of the United Nations (57) which points out that before the war about one-half of the world's population was subsisting at a level of food consumption which was not high enough to maintain normal health, allow for average growth of children or furnish enough energy for daily work.

This study was undertaken to determine some of the possible effects of the nutrient level of the fields in which winter wheat is grown upon the resistance of the crop to low temperature. Laude and Metzger (31) found that winter wheat grown on fertile soils survived cold better than on infertile soil. Additional information is needed as to the role of various nutrients in preparing plants for winter conditions. Whether this difference in survival between fertile and infertile soil is attributable to a difference in the protoplasm of plants grown in different nutrient levels or to a difference in opportunity for the plants to rejuvenate and re-establish themselves after freezing is considered in this research problem.

Since many factors involved in winter killing are beyond the control of man, any modification of the environmental conditions, such as the availability of nutrients, may go far in affecting internal characteristics controlling the ability of the plant to resist cold and thus make a contribution of some value toward the final solution of the problem.

REVIEW OF LITERATURE

The resulting injury to plants after exposure to cold temperature has been studied by numerous investigators. The general subject has been reviewed by many authors (5, 41, 36, 32, 18) but the literature concerned with the nutrient level of plants in relationship to winter hardiness is much restricted as compared to other aspects of the problem.

It seems fairly well accepted by most men working on the problem that cold resistance in plants is founded in some physiochemical properties of the protoplasm which are not fully understood; nor has a practical means of measuring these properties yet been devised, although high correlations between protoplasmic characteristics and winter hardiness have been found by various workers. The prodigous amount of literature on this subject makes it necessary to limit the scope of this review and therefore only those references are cited which are concerned with cold resistance in relation to fertility level of the soil.

In their study of the influence of fertilizers in protecting corn against freezing, Magistad and Truog (35) concluded that application of fertilizers increases the osmotic pressure of the sap in young corn plants, which in turn lowers the freezing temperature of the plant from one to two degrees centigrade. They found that "in every case the unfertilized plant was frozen as much or more than the fertilized one."

Holbert (21) also investigated cold resistance in corn. He exposed the plants to freezing temperatures under field conditions by means of a portable refrigeration machine and observed that plants growing in the soil to which fertilizers had been applied were more resistant to cold than the plants of the same strain growing in untreated soil. Some strains susceptible to cold when grown in unfertilized soil were killed in the young plant stage, by exposure for a few minutes to a temperature of 33 to 34 degrees F. These same strains showed no visible injury when grown in the same soil supplied with a hill-drop application of a 5-15-5 fertilizer at the rate of 100 pounds per acre, when exposed for four hours to a temperature of 30 degrees F. Holbert noted further that even when there was no visible injury at the time of freezing later inspection at harvest time revealed that the ears of the unfertilized plants had not increased in weight after the time of freezing, whereas, ears from the fertilized plants, exposed to the same freezing temperature, had increased in weight after freezing and were almost comparable in weight to the fertilized ears not exposed to freezing temperature. His conclusions that corn plants are more resistant to cold, both in the young plant stage and in the maturing stage when grown on more productive soil, is in agreement with the work of Magistad and Truog.

Schribaux (45) fertilized three different soils and found that the percentage of cereal plants killed was two to three times as great in the untreated soils as on the fertilized plots. The increased resistance to cold, he concluded, was due to nitrogen, phosphorus or potassium but did not assign the resistance to any one element. He noted potassium had increased resistance of the plants to cold to the greatest extent.

Wilhelm, (53, 54, 55), using sand cultures and nutrient solutions, has extensively studied the effect of nitrogen, phosphorus and potassium on the hardiness of wheat, oats, barley, rye, tomatoes and beans. The nutrients were supplied as high, normal, low and minus quantities of these elements. In his work on barley, he noted that plants receiving an abundance of nitrogen and phosphorus were the least resistant to cold. Tests on beans, tobacco and tomato plants revealed that those plants receiving low quantities of a nitrogen-phosphorus-potassium fertilizer received the most pronounced injury, while a nutrition rich in potassium increased the resistance of the plants to cold. In general, he found that high quantities of potassium were associated with the greatest survival after exposure to freezing temperatures; low and normal nitrogen availability were most conducive to resistance to cold; low and normal quantities of phosphorus also gave the greatest resistance. The lowest survival of plants usually resulted when these elements were absent.

Lacis (29) noted that potato vines fertilized with potassium were more resistant to frost than those vines in plots without fertilization.

In experiments on the winterkilling of vegetable crops in market gardens, Wallace (52) fertilized one-half of an area of winter onions with 300 pounds of K_2SO_4. The usual winterkilling occurred in the untreated areas while those receiving the potash made large root systems and developed into healthy plants.

Boswell (3), working with cabbage, found that heavy applications of nitrogenous fertilizer in the plant bed produced large, succulent plants which winterkilled badly but under field conditions the fertilizers had no apparent influence.

Using leaf lettuce, head lettuce, cauliflower, and cabbage, Rosa (41) observed that plants grown on poor soils (sand and various amounts of compost) were smaller and grew more slowly but were more resistant to cold than plants grown on better soils. He also found that plants having a liberal quantity of moisture were less hardy than plants receiving a minimum quantity of moisture. Rosa concluded that "any treatment materially checking the growth of plants increases cold resistance."

Dunn (13) grew cabbages under high and low levels of available nitrogen, phosphorus and potassium but did not change the degree of hardiness of the plants.

Lott (34), studying the correlation of chemical composition in regard to hardiness in brambles, increased hardiness by removing the first two crops of shoots from the raspberry bushes.

Collison and Harlan (6) noted in their study of the relationship of nutritional treatment to winter injury of Baldwin apple trees, that fertilizers in general reduced winter injury and that nitrogen fertilizer alone gave a greater decrease than a complete fertilizer. Geslin (15), working with fruit trees, concluded that the divergence among individuals of a variety to susceptibility to cold was due to differences in nutrition. Winter injury in the trunks of some apple trees was experimentally produced by Tingley, Smith, Phillips and Potter (49) by a fall application of a nitrogenous fertilizer.

Wohack (56) noted a general frost protective effect from the use of potassium even when the potassium was applied after the first frost in the fall and concluded this effect to be due to the general strengthening of the plant which allowed it to overcome more easily the frost injury.

On lemon seedlings, Gocholashvili (17) found that untreated check plots lost 69 per cent of their leaves due to cold temperatures, while those receiving an application of manure, nitrogen, phosphorus and potassium lost only 29 to 35 per cent of their leaves. When Kerch slag was used in place of manure with a complete fertilizer, the loss of leaves due to low temperature was reduced 23 to 25 per cent.

In their review of the hardiness problem in fruit trees, Dorsey and Bushnell (12) noted that excessive fertilization produced an abundant, succulent, tender growth very susceptible to cold. If, on the other extreme, a deficiency of nutrients existed this was also detrimental to hardiness.

In their study of the effect of commercial fertilizers on fruit trees, Cooper and Wiggans (7) found that nutrients, and incidentally vigor, affected winter mortality of buds only as the stage of maturity of the buds was influenced.

Knowlton and Dorsey (24) induced slightly greater hardiness in the buds of peach trees by application of a nitrate fertilizer.

Crane (8) observed that fertilizations with nitrate of soda caused buds of peach trees to be more susceptible to winter injury.

The following fertilizer treatments were applied to vineyards of grapes by Gladwin (16): nitrogen, phosphorus, potassium and lime; nitrogen, phosphorus and potassium; nitrogen and phosphorus; nitrogen and potassium; and phosphorus and potassium. The resulting differences in injury or killing were not greater than should be expected due to experimental error.

Kopitke (27), in investigating the effect of potassium salts on the hardening of coniferous seedlings, noted several changes due to application of potash fertilizers. First, the fertilizer promoted the accumulation of simple and invert sugars in the tissue of the seedlings; secondly, there was an increase in the content of total solids, an increase in osmotic pressure, and a decreased freezing point of the sap. All these changes indicated a marked improvement in the ability of nursery or planting stock to withstand frost. He concluded that "a balanced ratio of nutrients and especially an adequate supply of available potash appears to be the most important requisite for the production of frost resistant stock."

In his study of the effect of potassium salts on the winter hardiness of barley, Yasuda (60) observed that a deficiency of potassium inhibits the formation of sugar and the plants become less hardy. A high application of potassium under low temperatures increases the sugar content of the plants which consequently become hardier. Under conditions of an ordinary greenhouse, the high application of potassium induces rapid growth of plants, thereby causing a decrease of sugar content. The effect of potassium for increasing the sugar content becomes noticeable two or three days after its application.

In studying the effect of nitrogenous fertilizers on Kentucky bluegrass, Carroll and Welton (4) noted no difference in resistance to cold of fertilized and unfertilized grasses when exposed in the unhardened condition. After Kentucky bluegrass was hardened, the grass receiving the nitrogenous fertilizer was less resistant to cold than the unfertilized plant.

Kimball (22) found that herbaceous plants treated with $NaNO_3$ were the least resistant to cold.

Dexter (9) observed that winter wheat grown under excess and minimum nitrogen supplies in solution varied in ability to harden. When grown at 2 degrees C., without light, those plants high in nitrogen did not harden while those low in nitrogen hardened well in the dark. In addition to winter wheat, plants of winter rye, winter barley, winteroats and cabbage grown under minimum nitrogen conditions hardened while the same plants grown in surplus nutrient conditions and low in carbohydrates due to the surplus of nitrogen, did not harden. In testing the winter hardiness of weeds, Dexter (11) also found that quack grass fertilized with nitrogen was less hardy than unfertilized quack grass.

Using Dexter's electrical conductivity method of testing for hardiness, Megee (37) found no difference in the hardiness of alfalfa roots grown with applications of phosphorus and potassium applied on May 29, a nitrate fertilizer applied on September 24th, or untreated roots. Ahlgren (1) concludes that losses due to winter injury are likely to be less when alfalfa is grown on a fertile soil rather than a soil of moderately low fertility.

Elliot and Wolfe (14) found, in their study of the relationship of fertilizers to Hessian fly injury and winterkilling of wheat, that, under Virginia conditions, manure markedly prevented winterkilling as determined by yields compared to check plots. Phosphorus (decidely deficient in Virginia soils) was, they concluded, the element most essential to increased yield and decreased amount of winterkilling.

Kuksa (28) found that an application of phosphorus and potassium to a soil containing a normal amount of nitrogen provided conditions in which the hardiness of winter wheat was markedly increased; the total sugar content of the plant being increased correspondingly. He concluded, further, with respect to phosphorus and potassium, that phosphorus should be applied at heavier rates than potassium in the fertilizer. The most effective ratio for application of these elements was four to one. Under these conditions the survival of plants increased 95 per cent and the sugar content rose to 26.3 per cent. When nitrogen was included in the fertilizer, particularly in large amounts, resistance of the plants to cold notably decreased; the percentage survival of the plants was reduced to 12 and the sugar content also decreased to 19.5 per cent. Muksa recommended that when nitrogen is applied it should be accompanied and exceeded by rates of phosphorus and potassium. He agreed with Ellet and Wolfe (referred to above) and Laude and Metzger (31) as to the beneficial effect of manures, alone and in combination with mineral fertilizers, on increasing hardiness.

Laude and Metzger (31) tested the hardiness of wheat seedlings that had been grown in a low fertility soil to which was added low, medium, and high applications of phosphorus both with and without applications of manure. They also tested plants grown on a fertile soil to which was added a high application of phosphorus with and without manure. Their results indicated that a high rate of phosphorus application in a low fertility soil increased the hardiness of the wheat so that the plants were nearly as resistant as those grown on a fertile soil. However, if phosphorus was applied in small quantities to the low fertility soil it slightly decreased the hardiness of the plants. They concluded that wheat plants grown on fertile soils were markedly more resistant to cold than those grown in non-fertile soils.

Saveljev (44) increased the frost resistance in wheat and rye hybrids by the application of phosphorus and potassium. Testing the plants at 10 to 17 degrees C., for twenty-four hours he found a much higher survival of plants receiving these elements. Winter hardiness in the plants was augmented even more when the same amount of fertilizer was applied in three installments, namely, after the last cultivation of fallow, at sowing and during tillering, than if the whole amount was applied at any one of these dates.

Longnecker and Gray (33) found that an application of 500 pounds of 4-12-8 fertilizer reduced the winter injury in fields of winter wheat, rye, oats and barley.

Worzella and Cutler (59) noted in their experiment that wheat seedlings grown on low and medium levels of fertility differed but little in their cold resistance. Those seedlings grown on high levels of fertility were large and succulent and suffered the greatest injury on exposure to low temperature.

In a study on the effect of fertilizers on winter hardiness in clover, Koperzinskii (25, 26) found that phosphates and potassium sulphate, either alone or combined, and particularly following liming, increased winter hardiness in the plants. He felt these fertilizers promoted the stability of proteins through hydrolysis and depolymerization in which unstable protein compounds present were converted into simpler but more stable forms. Phosphates increased respiration energy, while the potassium sulphates did not, and this was thought under certain conditions to be adverse to cold resistance in plants. Potassium chlorates, on the contrary, decreased winter hardiness in the plants due mostly to the toxic effect of chlorine ions penetrating plant tissues in large amounts.

Levitt (32), from his critical review of the effect of mineral nutrition on frost resistance of plants, concluded that generally excess nitrogen reduces the hardiness of plants; a deficiency increases hardiness. However, if the deficiency of nitrogen becomes too great hardiness is reduced. The effects of other elements are less striking, but in general a deficiency reduces the hardiness of plants. He states that:

> Excess potassium has often been stated to increase hardiness, but in many cases this really involves only a slight lowering of the freezing point of plants incapable of hardening. In others, overwintering may be affected but not the frost resistance. Yet some cases are on record in which excess potassium appears to increase true frost resistance.

MATERIALS AND METHODS

The plants used in this experiment were obtained from two sources: part of the plants were taken from the wheat fertilizer experiment plots of Kansas State College Agronomy Farm, the remainder from flats initially started in the greenhouse and then removed to the east side of the greenhouse where they hardened under natural conditions.

The soil used for the flats was an eroded subsoil obtained from the northwest section of the Agronomy Farm. After screening the subsoil, about 1/10 by weight of sand was added to each flat to improve the physical condition of the soil.

Plants used in these experiments were provided from five nutrient conditions prepared by adding nutrients as follows:

(1) A complete fertilizer of nitrogen, phosphorus and potassium.

(2) A fertilizer of nitrogen and phosphorus.

(3) A fertilizer of phosphorus only.

(4) A fertilizer of nitrogen only.

(5) The untreated soil.

In the garden flats, nitrogen was added as NH_4NO_3 at the rate of 250 pounds per acre; phosphorus in the form of $CaH_2(PO_4)_2$ at 100 pounds per acre and potassium as KCl at 100 pounds per acre, while on the Agronomy Farm plots, nitrogen was added at only 100 pounds per acre, phosphorus at 50 pounds and potassium at 25 pounds per acre. Into the three flats, representing each of the five fertility levels, six rows of grain containing 35 seeds were planted at a depth of ½ to 1 inch. Under optimum conditions of moisture and temperature in the greenhouse the seeds germinated and obtained a rather uniform heighth of three to four inches by the end of the first week after planting, at which time the flats were removed from the greenhouse to the garden. There the flats were grouped into a compact unit, where sunlight and drainage were well equalized, and were banked with sand to reduce border effects on the plants.

The fall weather conditions were conducive to rapid growth and strong tillering. The flats of the different fertility levels soon showed signs of differentiation which appeared in two general groups: (1) the nitrogen-phosphorus-potassium, nitrogen-phosphorus, and phosphorus flats had larger plants, greater number of tillers and were of a darker green coloring; (2) the nitrogen alone and the untreated flats were smaller, produced fewer tillers per plant and had a lighter green coloring. The plants grown in the flats were smaller and tillered less than those in the field plots probably due to the late planting date of the flats and the more sharply defined nutritional limits set by the deficient subsoil supplemented with added nutrients.

All the plants for any one experiment were removed from the soil at the same time and from the same source (either garden flat or Agronomy Farm). Care was taken to retain the root system without injury so far as possible. The soil was removed from the roots by shaking and washing and tillers were counted. The plants were then transplanted into a flat in half of which there was a fertile soil and in the other half a nonfertile subsoil. The flats were placed in the freezer room and subsequently removed to the greenhouse where they were retained under good conditions for recovery and growth of the wheat.

Five plants from each nutrient condition were transplanted into the fertile soil and five into the nonfertile soil. Thus each replication of the experiment contained 10 plants from each of the five fertility levels. The position of the plants representing the five fertility levels within the flat was rotated in subsequent replications to equalize any possible border effect.

In each replication of the experiment it was desired to expose the plants to such temperature and for such period of time in the cold room as would kill the least hardy of the group and would injure little if any of the most hardy. This proved to be a very difficult problem. Differences in hardiness appeared to be associated with the stage of development of the plants in early experiments compared to later experiments. Wide fluctuations in temperature, moisture and light over short periods of time during December and January probably induced changes in degree of hardening.[1]

The problem was further hampered by inability to evaluate at an early date after freezing, the degree of injury or winter hardiness before subsequent replications were exposed to freezing temperature. Under these circumstances, three methods of evaluation of resistance to cold were used: (1) the percentage of plants killed (2) the degree of injury as expressed by an estimate of vigor and growth of the surviving plants and (3) the degree of cold resistance evaluated by the percentage of tillers surviving on the plants.

The cold chamber used in these studies consisted of an insulated room 10x9x8 feet. Freezing temperatures were induced by means of a mechanical refrigeration unit regulated by a mercury controlled thermostat which, with an electric fan, for maintaining uniform movement of air, retained the fluctuation of the temperature within the chamber to 1 degree F. No attempt was made to control the humidity within the chamber,

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[1]Winter hardiness of wheat fluctuates greatly over short periods of time due to: (1) temperature: Suneson and Peltier (48) found high daily temperature with high radiation very conducive to hardening in winter wheat during November and early December. A subsequent period of colder temperature resulted in progressive hardiness. Peltier and Kiesselbach (38) and Suneson and Peltier (47) found that a continuous cold period, rather than alternating temperature, was more effective in hardening grain. (2) moisture: Klages (23), Salmon (42, 43), Hill and Salmon (20) and Suneson and Peltier (48) have noted the variability in hardiness due to moisture conditions and (3) light: Dexter (10) observed that wheat seedlings, shaded during half the day, were not as hardy as those exposed to full light; he was unable to harden them if kept entirely in the dark. Tumanov (50) (as cited by Levitt) concluded that low temperature alone is incapable of hardening winter grains but that light must be supplied.

but it was assumed that the humidity would be comparable to that out-of-doors, for the days on which the experiments were conducted.

EXPERIMENTAL RESULTS

Nutrient Deficiency Tests of Wheat Plants

Observation of the plants in the garden flats, as they developed and entered their semi-dormant stage during December, revealed a clear-cut differentiation between fertility levels with respect to size of plants, number of tillers and color of plants. To test what appeared to be an obvious deficiency of plant nutrients, a chemical plant tissue testing kit was utilized. The kit employed in the tests was the "Purdue Rapid Soil and Plant Tissue Testing Kit", developed by Purdue University. Tissue tests were conducted for nitrogen, phosphorus and potassium on plants from each of the five fertility levels with replications of the test by a man experienced in the use of the kit in addition to those made by the writer.

Plants from the five fertility levels selected from the wheat fertilizer test plots on the Agronomy farm were also tested. However, the visual differentiation between plants from these various fertility levels was not so readily observed.

No marked deficiency of any of the three elements was indicated by the chemical tests. There were slight differences in the quantity of these elements available as shown by the color of the chemical tests but all three elements were either high or very high in their rating.[2]

Whether the observed differences in the plants were due to a deficiency of other essential or minor elements required by the plant for normal growth was not determined.

Transplanting Technique

The plan of the experiment provided for digging the plants from the field or from flats, removing the soil from the roots by shaking and washing and finally transplanting them into the two soil conditions where they were frozen. The loss of plants due to this procedure was evaluated in order to differentiate between transplanting and low temperature losses. Consequently, frequent check flats were run to determine if the mortality of the plants due to the process of transplanting were significant.[3] These check flats consisted of plants similar as to size, source and transplanting procedure but were not exposed to freezing temperatures.

Listed in Table 1, in addition to four check flats, are several flats which either were not exposed to sufficiently low temperature or not exposed long enough to cause differential killing and which therefore may be used to estimate transplanting losses.

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[2] The rating used for these elements in this tissue testing method was given as: very low, low, medium, high, and very high.

[3] Salmon (43) found a difference in the resistance to cold of a single plant or a clump of plants, the latter being more hardy. In testing the hardiness of plants relative to the disturbance of the root system he found no significant difference between plants whose roots had been thoroughly shaken free of soil and plants which retained the soil as much as possible in the transplanting process.

It is apparent that the transplanting losses were negligible, amounting only to one-third of one per cent.

Table 1. Percentage of seedlings lost due to handling and transplanting processes.

Flat designation	Transplanting date	Number of plants transplanted	Failure to survive Number	Failure to survive Per cent
Check	12/4/48	30	0	0
Flats ABC	12/11/48	60	0	0
Flats DE	12/14/48	40	1	2
Flat # 1	12/18/48	50	0	0
Check	12/20/48	50	0	0
Flat # 3	12/20/48	50	0	0
Flat # 4	12/21/48	50	0	0
Check	1/15/49	50	1	2
Check	2/11/49	50	0	0
Flat # 25	2/18/49	50	0	0
Flat # 26	2/18/29	50	0	0
Flat # 29	2/19/49	50	0	0
Totals		630	2	.31

Soil Temperatures During Freezing Tests

That the moisture content of the soil may have an important relation to the degree of winterkilling of winter grains has been rather strongly attested by the work of several investigators.[4] In these experiments the method of equalizing moisture conditions was to saturate the soil thoroughly several days before use so that at the time of transplanting the soil was at an optimum friable condition. The physical condition and the organic matter content of the fertile soil were widely different from the subsoil. The moisture holding capacity would therefore vary in the two sub-divisions of each flat and control to a small degree the temperature conditions within that division.

Since a single flat constituted a replication of the experiment in itself, the possibility of border plants being exposed to a lower temperature more quickly, and so for a longer period of time, was considered. To determine the temperature to which the

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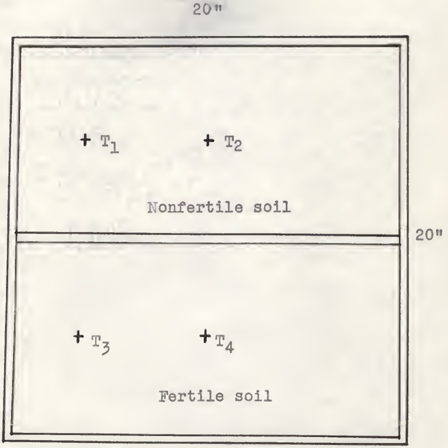
[4] Klages (23) in studying the relation of soil moisture content to resistance of wheat seedlings to low temperature, concluded that as the activities of life growing processes increase, due to more optimum moisture conditions, the plants become less resistant to temperature extremes and the percentage killed was higher. Klages modified this idea for field conditions, "there is every reason to believe that there is comparatively less killing on the higher soil moisture under field conditions." Salmon (42, 43) noted that a wet soil is warmer than a dry soil of the same type as long as the ground is not frozen due to the specific heat of the water and the heat of fusion of the moisture contained in the soil. When, however, the soil is frozen directly opposite results occur. This is due to a difference in thespecific heat of water and thermal conductivity of ice and water. Ice has one-half the specific heat of water and approximately three times the thermal conductivity of water. Hill and Salmon (20) observed that plants growing in a dry soil were injured more severely than plants in a wet soil due to the specific heat of water and consequent lag in temperature. Levitt (32) points out that low environmental moisture usually increases frost resistance of a plant by retarding growth activities, while high soil moisture content promotes a succulent growth and reduces hardiness, but this should not be confused with the references above which noted the effect of moisture on the general temperature of the soil.

crown and roots of the plant would be exposed four thermometers to a flat were used, one occupying the position of the border plants and another the central plant area of the flat. This was duplicated in both fertile soil and nonfertile subsoil. If the specific heat of the fertile soil differed appreciably from that of the nonfertile subsoil this would also be determined. The relative positions of the four thermometers are shown in Figure 1. Regardless of the above possible conditions, however, the plant positions of the five fertility levels were rotated in different replicates to obviate any error from this source.

Little difference in the temperatures prevailing at different locations in the flats was found indicating that the plants occupying border positions were at little if any disadvantage compared to plants located in other parts of the flat. An illustration of the comparative soil temperatures observed while exposed to -4 degrees F. are given in Table 2.

Figure 2 illustrates by graph (as average temperature of the four thermometers) the drop in soil temperature when the cold chamber was thermostatically controlled at -4 degrees F. The graph explains why wheat seedlings in a moist soil may survive freezing conditions while plants in a dry soil do not. The leveling off of the temperature curve, near the freezing point, is due to the heat released by the water molecules in their change of state from a liquid to a solid. This release of heat (called heat of fusion) delays the fairly consistent temperature drop up to that point and is referred to by some



Legend:

T_1 Thermometer number 1.

T_2 Thermometer number 2.

T_3 Thermometer number 3.

T_4 Thermometer number 4.

Figure 1. Relative positions of thermometers in flat to determine soil temperature.

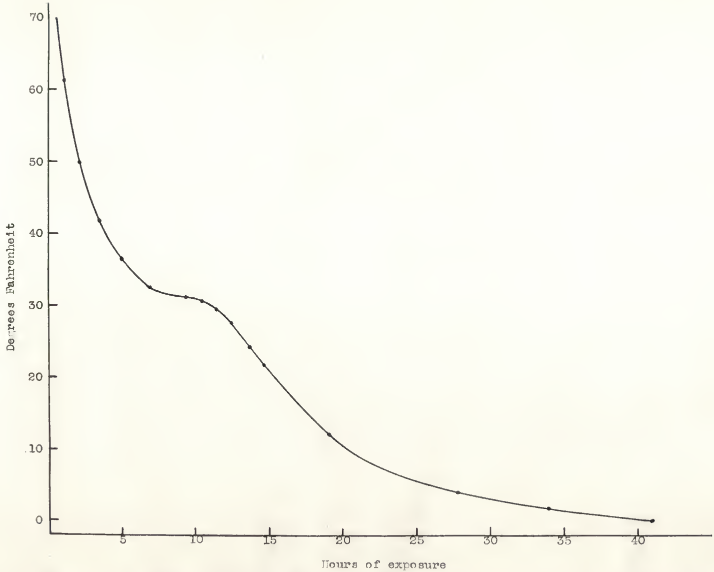


Figure 2. Reduction in soil temperature during 42 hours exposure at - 4°F.

investigators as the temperature 'lag' (see footnote 4). A soil containing a large quantity of water will have a slow temperature change near the freezing point and seedlings are given protection in a wet soil for a short time by the heat released from the water. After the moisture of a soil is frozen the seedlings are more subjected to low extremes of temperature because ice more readily conducts cold than a dry soil.

Table 2. Soil temperatures at 4 different positions in a flat during a 42 hour freeze. Temperature of cold chamber -4 degrees F.

Hours exposure	Thermometer				Average temperature
	1	2	3	4	
0	73.5	73.5	73.5	73.5	73.5
1	60.5	61.5	61.5	61.0	61.0
2	49.5	49.0	49.5	49.5	49.5
3 1/4	41.0	41.0	41.5	42.0	41.5
5	35.0	35.5	36.5	36.5	36.0
7	31.5	32.0	33.0	33.0	32.5
9 1/2	31.0	30.5	31.5	31.5	31.0
10 1/2	29.5	29.5	31.5	31.5	30.5
11 1/2	28.0	29.0	29.5	31.0	29.5
12 1/2	25.5	27.5	27.0	29.0	27.5
13 3/4	22.0	25.0	23.0	25.5	24.0
14 3/2	20.0	22.5	20.5	23.0	21.5
19 1/4	12.5	12.0	11.5	13.0	12.0
28	3.5	3.5	4.0	5.0	4.0
34	3.0	1.5	1.5	2.5	2.0
42	1.5	-1.0	0.0	0.5	0.0

Cold Resistance as Evaluated by Survival of Tillers

The results of experiments 1 to 6 are tabulated in Table 3. In this group of experiments the degree of damage to the plants, caused by exposure to freezing temperature, was expressed in terms of percentage of tillers that survived in each group of plants. The survival percentages have since been transformed into arc sines and are reported in that form in the table.[5] The tillers of the plants in each flat were counted before the plants were frozen at -4 degrees F. for a 24 hour period and again 21 days after freezing. The plants for these 6 experiments were obtained from the garden flats in which the nutrient conditions were sharply defined by the addition of various fertilzers.

Marked differences in percentage of tillers that survived were found between plants grown on different fertility levels, while no difference in survival existed between plants transplanted in the fertile soil or nonfertile soil. The following summarization may be made in regards to these data asto differences of plants grown in the five nutrient conditions: (1) the cold resistance of plants grown on soil treated with nitrogen-phosphorus, as expressed by the percentage of tillers surviving, was significantly greater than the cold resistance of all plants from other fertility conditions except phosphorus; (2) the cold resistance of plants grown in soil to which phosphorus was added was significantly greater than the cold

[5] Snedecor (46) recommends that data expressed as percentages derived from small samples be transformed to arc sines for greater accuracy in statistical analysis.

Table 3. Percentage survival of tillers of plants grown under different nutrient conditions and transplanted into a fertile soil and a nonfertile soil and frozen at -4° F.

	Nutrient condition in which plants grew									
	Nitrogen Phosphorus Potassium		Nitrogen Phosphorus		Nitrogen		Phosphorus		No treatment	
	Soil into which plants were transplanted									
Experiment	FS*	NS**	FS	NS	FS	NS	FS	NS	FS	NS
1	50.8	51.3	62.7	49.6	51.1	49.0	56.8	62.0	30.0	47.6
2	47.9	62.7	55.9	64.2	54.7	47.3	58.0	53.7	45.0	40.1
3	30.0	30.0	50.8	62.7	45.0	26.6	46.7	45.0	42.4	39.2
4	49.6	57.1	40.4	50.8	31.5	25.2	45.3	41.5	50.8	50.8
5	18.4	29.2	66.4	38.4	31.5	23.1	48.2	45.0	18.4	42.4
6	38.6	30.0	38.6	38.6	30.0	16.7	32.8	41.8	35.2	35.2
Total	234.3	260.3	314.8	304.3	243.8	187.9	287.8	289.0	221.8	255.3
Soil Mean	39.2	43.4	52.5	50.7	40.6	31.3	47.9	48.1	36.9	42.5
Nutrient	41.3		51.6		35.9		48.0		39.7	

* FS Fertile soil.
**NS Nonfertile soil.

resistance of plants grown in nitrogen treated soil or untreated soil but was not significantly greater than cold resistance of plants from soil treated with nitrogen-phosphorus-potassium; (3) plants from soil treated with nitrogen-phosphorus-potassium, untreated soil or nitrogen treated soil were not significantly different.

Results of the analysis of variance are given in Table 4.

Table 4. Analysis of variance for experiments 1 to 6.

Factors	Degrees of freedom	Sums of squares	Variance	Calculated F	Table readings of F (P=.05)	(P=.01)
Between nutrient conditions	4	1,939.98	484.99	5.77 **	2.61	3.83
Between soils	1	0.75	0.75	0.01 *	4.08	7.31
Interaction nutrient soil X	4	414.56	103.64	1.23 *	2.61	3.83
Between experiments	5	2,999.72	599.94	7.15 **	2.45	3.51
Interaction experiments X soil	5	48.07	9.61	0.11 *	2.45	3.51
Remainder	40	3,354.51	83.86			
Total	59	8,757.59				

Least significant difference "between nutrient conditions" 7.54

*Nonsignificant
**Highly significant

If the survival of the plant is influenced by its nutrient condition after freezing then an "F" value of significance should have been obtained in the analysis for variance for the factor "between soils". The information thus far obtained from these 6 experiments indicates that the cold resistance of winter wheat is noticeably altered by the nutrient condition in which the plants are grown. Once the plants are frozen it appears that their survival is dependent to a greater degree upon protoplasmic characterisitics of the plant cell than upon external factors. The difference in survival apparently is not due to the ability of the plants to re-establish themselves due to available nutrients.

Cold Resistance as Evaluated by Vigor and Growth

The results of experiments 7 to 12 are given in Table 5. The plants for these 6 experiments were obtained from fertility plots on the Kansas State College Agronomy farm. All the plants in these experiments were frozen at -4 degrees F. but for different lengths of exposure. Experiments 7 and 8 were frozen 30 hours, 9 and 10 for 36 hours and 11 and 12 for 42 hours. The greater damage caused by long exposure may be readily noted in the vigor and growth estimates of Table 5. The ratings as given for vigor and growth are for transplanted plants in both fertile and non-fertile soil. This rating was combined because of the slight differences in plants of the two soils.

Plate 1 illustrates the retardation of growth, and therefore the degree of injury, due to the various lengths of exposure as compared to check flat A which wasnot frozen, flat B which was frozen 30 hours, flat C 36 hours and flat D 42 hours. Plate II

Table 5. Rating of vigor and growth of plants from different nutrient conditions after being frozen at -4 degrees F.

Experi-ment	Hours frozen	Nutrient condition in which plants grew					
		NPK	NP	N	P	No treatment	Average
7	30	90	90	60	80	60	76
8	30	80	80	65	65	50	68
9	36	45	45	25	35	25	35
10	36	50	50	30	40	30	30
11	42	40	30	10	30	20	26
12	42	40	30	10	20	30	26
Totals		345	325	200	270	215	
Mean		57.5	54.1	33.3	45.0	35.8	

Least significant difference "between nutrient conditions" 6.67.

also illustrates the degree of injury sustained from an additional six and twelve hours exposure periods (flat C and D, respectively). From this plate it may also be observed that the nutrient condition of a plant after it has been frozen has little effect on the survival of the plant. Plants in the nonfertile soil (upper portion of each flat) were not injured more severely and survived as well as the plants in fertile soil.

An analysis of these data gave a very highly significant "F" value of 21.98 for variance between the five nutrient conditions. The least significant difference for this factor at the 5 per cent level of probability was 6.67. With this difference required for significance it will be noted that (1) the difference

EXPLANATION OF PLATE I

Retardation of growth of winter wheat caused by low temperature injury.

Flat A. Check, unfrozen.
Flat B. Exposed for 30 hours, at -4° F.
Flat C. Exposed for 36 hours, at -4° F.
Flat D. Exposed for 42 hours, at -4° F.

Row 1. Plants grown in NP nutrient condition.
Row 2. Plants grown in N nutrient condition.
Row 3. Plants grown in P nutrient condition.
Row 4. Plants grown in NPK nutrient condition.
Row 5. Plants grown in untreated soil.

PLATE I

EXPLANATION OF PLATE II

Environmental effects on vigor and growth of transplanted plants following freezing.

Nonfertile soil upper portion of flat.
Fertile soil lower portion of flat.

Row 1. Plants grown in NP nutrient condition.
Row 2. Plants grown in N nutrient condition.
Row 3. Plants grown in P nutrient condition.
Row 4. Plants grown in NPK nutrient condition.
Row 5. Plants grown in untreated soil.

PLATE II

in injury between plants grown on soil treated with nitrogen-phosphorus-potassium or nitrogen-phosphorus was nonsignificant; (2) plants grown on soils treated with nitrogen-phosphorus-potassium or nitrogen-phosphorus were injured significantly less than plants from soils treated with phosphorus, nitrogen or untreated soils; (3) plants from the phosphorus nutrient condition were injured significantly less than plants from nitrogen treated soil or untreated soil.

Cold Resistance as Determined by Percentage of Plants Killed

In another small set of experiments the comparative cold resistance was evaluated by the percentage of plants that were killed by freezing. Plants in the garden flats used in these experiments were not transplanted as were those in the two previous series of experiments but were frozen in the same nutritional condition in which they were grown without disturbing the root system. Each nutrient level was represented by a full flat of plants. The results of these experiments are given in Table 6.

Table 6. Percentage of plants, grown in five different nutrient conditions, killed by low temperature.

Experiment	Nutrient condition in which plants grow: Nitrogen Phosphorus Potassium	Nitrogen Phosphorus	Nitrogen	Phosphorus	No treatment
15	9.0	24.3	67.0	68.0	60.0
16	11.0	6.0	98.0	98.0	81.0
Average	10.0	15.1	82.5	83.0	70.5

A marked superior hardiness of plants from soil treated with nitrogen-phosphorus-potassium or nitrogen-phosphorus was again found over plants from the remaining fertility levels. The absence of potassium in the nitrogen-phosphorus soil treated flats did not alter the cold resistance of those plants possibly because of the unusually high amount of potassium found in most Kansas soils under natural conditions.

Effect of Nutrients on Development of Young Wheat Plants

The effects of nitrogen, phosphorus and potassium in increasing or decreasing hardiness are not fully understood. Different explanations have been given most of which note the relationship of these elements with sugar content, protein concentration, carbohydrate synthesis or hydration of colloids. Tillering and development of the plant in relation to hardiness were considered in this study as expressions of the influence and thus possibly as a partial explanation of the role of these nutrients.

Several investigators (23, 47, 56) have studied the relation of hardiness of winter wheat at various stages of development. The general conclusions of most of the research was that wheat seedlings are least resistant to cold when the plant food supply shifts from the endosperm to absorption through the root system. After this low resistance of the transition period cold resistance increases.

In the experimental work of this problem the five fertility levels used greatly modified the development of the young wheat plants. The average number of tillers for plants from the five nutrient conditions studied are presented in Table 7. Highly significant differences in number of tillers of plants grown under different treatments were found as shown by the F value in the analysis of variance. The relation between number of tillers per plant and cold resistance is illustrated in Figure 3. This figure indicates the correlation between number of tillers per plant and hardiness when hardiness was evaluated by a rating of vigor and growth of plants following freezing (from Table 5).

The average weights in grams for groups of 10 plants grown in the five nutrient conditions are given in Table 8. An analysis of variance of these data gave a highly significant F value for differences in weight of plants from different nutrient levels. The plants from the five fertility levels ranked in decreasing weight as follows:

(a) Plants grown on nitrogen-phosphorus-potassium treated soil.
(b) Plants grown on nitrogen-phosphorus treated soil.
(c) Plants grown on phosphorus treated soil.
(d) Plants grown on nitrogen treated soil.
(e) Plants grown on untreated soil.

Table 7. Average number of tillers of winter wheat plants grown in different nutrient conditions.*

Experiment number	Nutrient condition				
	NPK	NP	N	P	No treatment
3	6.7	6.9	7.5	6.9	6.3
4	9.4	6.9	7.1	8.5	7.1
6	7.5	7.3	8.7	7.5	7.6
8	8.6	6.6	7.6	8.2	6.6
9	9.5	6.3	7.2	8.4	7.5
12	9.5	7.2	7.1	8.2	7.6
13	9.7	7.6	7.2	8.3	6.7
14	9.7	7.9	7.6	7.9	5.6
15	8.7	7.5	--	6.6	6.2
16	8.7	8.3	7.0	5.9	5.5
17	3.8	3.9	2.4	2.6	2.0
18	4.1	3.7	2.2	2.5	2.0
19	3.8	3.8	2.7	3.9	2.3
20	3.9	3.5	2.5	3.5	2.3
21	4.0	3.9	2.1	3.3	2.1
22	3.6	3.7	2.2	3.2	2.0
23	4.1	4.0	2.4	3.8	2.1
24	3.8	3.6	2.4	3.5	2.4
25	9.2	9.4	6.8	7.7	5.8
26	9.6	9.9	6.2	7.3	5.9
27	9.9	10.6	6.7	8.1	6.0
28	9.9	9.5	6.3	8.5	6.3
29	11.3	9.0	8.1	8.8	6.3
30	10.2	8.8	7.0	8.8	6.8
Total	179.2	160.3	127.0	151.1	121.0
Average	7.46	6.67	5.52	6.29	5.04

Least significant difference "between treatments" 0.096.

*Plants used in experiments 17 to 24 were grown in flats, the remaining plants are from field plots.

Table 8. Weight of plants grown in different nutrient conditions.*
(Weight in grams for ten plants)

Experiment number	Nutrient condition				
	NPK	NP	N	P	No treatment
1	28.8	31.0	24.0	26.3	20.8
3	23.4	21.2	32.1	21.2	18.5
4	39.4	22.6	22.0	30.3	16.2
6	28.2	24.7	25.3	23.1	17.8
8	29.2	19.9	23.4	28.0	16.9
9	28.1	18.9	22.7	28.0	17.0
12	30.7	19.3	23.8	21.2	15.1
13	31.6	20.7	24.9	23.9	13.3
14	28.0	19.0	19.0	16.4	12.3
15	30.1	18.0	17.1	15.5	12.8
16	23.0	19.3	16.9	13.9	--
17	6.9	6.4	3.5	3.7	4.2
18	7.3	6.4	3.5	3.4	3.7
19	6.2	5.8	4.3	5.3	3.7
20	6.6	5.0	3.7	4.3	3.2
21	5.8	6.1	3.6	4.5	3.3
22	5.8	5.4	3.3	3.3	2.9
23	5.3	6.1	3.9	5.9	3.5
24	5.2	5.1	3.4	5.0	3.5
25	22.5	24.1	13.5	17.7	12.2
26	22.1	24.5	13.2	16.8	11.2
27	20.6	26.2	12.5	18.1	10.4
28	23.6	23.1	11.1	19.3	13.2
29	25.0	23.0	16.4	18.3	11.7
30	20.3	21.6	13.4	15.0	12.6
Total	503.7	423.4	360.5	388.4	259.9
Average	20.1	16.9	14.4	15.5	10.8

Least significant difference "between treatments" 1.89

*Plants used in experiments 17 to 24 were grown in flats, the remaining plants are from field plots.

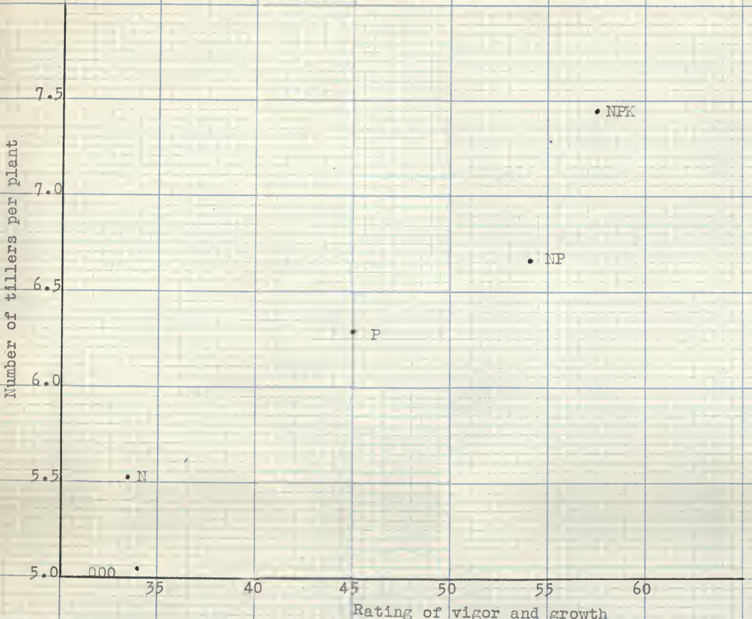


Figure 3. Correlation of hardiness, as expressed by vigor and growth after freezing and number of tillers per plant after freezing.

Nitrogen-phosphorus plants and phosphorus plants were not significantly different in weight nor were plants of phosphorus treated soil and nitrogen treated soil. Figure 4 indicates the relation of weight of plant to hardiness evaluated by vigor and growth rating of plants.

It will be noted from these two figures that in general, plants grown under good nutritional conditions had the greatest resistance to cold and were also the most fully tillered and largest in size, number of tillers, and cold resistance while plants from nitrogen treated soil and untreated soil were smallest in size, number of tillers and had the least cold resistance. This aspect of hardiness is well summarized by Levitt[6] "it is not the developmental stage itself which is of paramount importance, but certain internal characteristics normally associated with it. These may be morphological (cell-size) or physicochemical (carbohydrate or moisture content, cell sap concentration)."

This correlation noted between the cold resistance and the size and number of tillers would suggest that hardiness of winter wheat is due in large part to internal characteristics which are strongly influenced by the nutrient condition in which the plant is grown. The nutrient condition is relatively unimportant to the recovery of the plant from freezing but becomes important again in subsequent growth of the crop.

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[6] J. Levitt, Frost Killing and Hardiness of Plants, p. 144.

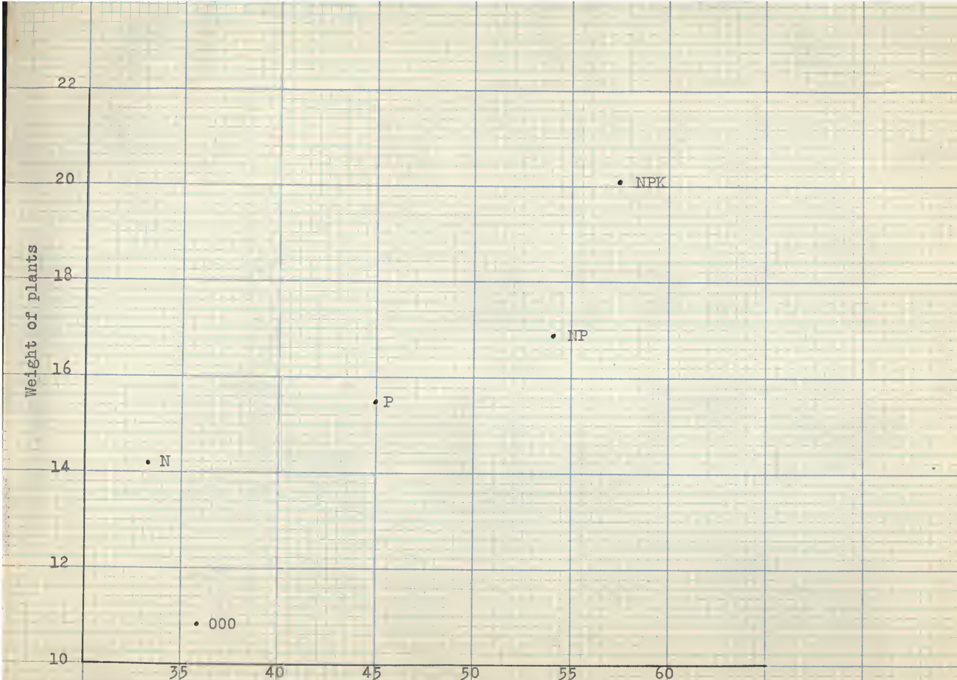

Figure 4. Correlation of weight of plants with hardiness as evaluated by vigor and growth after freezing.

A practical aspect of this problem suggests itself: that in winter wheat areas, where winter injury and killing are serious problems, fall application of a complete fertilizer may reduce appreciably the loss where soil nutrient deficiencies exist.

SUMMARY

During the fall and winter of 1948-49 an investigation was made on the effects of nutrients on cold resistance of winter wheat. Plants were grown on soils treated with nitrogen-phosphorus-potassium, nitrogen-phosphorus, nitrogen, phosphorus and on untreated soil.

Plants from the five nutrient conditions were observed to differentiate as to number of tillers, size and color. Plant tissue tests for nitrogen, phosphorus and potassium gave no indication of a deficiency for these elements.

Tests to determine the mortality of plants caused by their removal from field or flats and transplanting into a different soil revealed a negligible loss amounting only to one-third of one per cent.

The effect of variable soil temperatures to plants tested due to the position occupied in the flat and difference in temperature between fertile soil and nonfertile soil was considered. Temperature of the soil varied but slightly at different positions in the flat or between soils and was considered insignificant in its degree of injury to the plants.

Cold resistance was evaluated for plants from the five nutrient conditions, grown in flats, by percentage of tillers surviving. Plants from the nitrogen-phosphorus treated soil were most resistant. Plants from phosphorus treated soil were fairly high while nitrogen-phosphorus-potassium plants were low in resistance compared to later tests. Plants from untreated soil and nitrogen treated soil were least resistant.

Cold resistance of plants from the five nutrient levels was also evaluated by the vigor and growth of the plants after freezing. Plants from nitrogen-phosphorus-potassium and nitrogen-phosphorus treated soils had the greatest hardiness. Plants from phosphorus treated soil were intermediate in hardiness and plants grown on nitrogen treated soil or untreated soil had the least hardiness.

The percentage of plants killed was also used as the basis for evaluating hardiness. In this series of experiments the plants were not transplanted as were the plants in the two previous series of tests. Plants grown in a nitrogen-phosphorus-potassium treated soil and a nitrogen-phosphorus treated soil had markedly fewer plants killed by freezing temperature than plants grown on a phosphorus treated soil, a nitrogen treated soil or an untreated soil.

A correlation was noted between size of plant with hardiness and between number of tillers per plant with hardiness.

It is suggested that hardiness of winter wheat is due in large part to internal characteristics which may be strongly influenced by the nutrient condition in which the plant is grown.

The nutrient condition of winter wheat is relatively unimportant to the recovery to the plant after freezing but becomes important again in the subsequent growth of the plant.

A possible practical aspect of this problem is suggested. In winter wheat areas, where winter injury and killing are a serious problem, a fall application of a complete fertilizer may reduce greatly the loss where soil nutrient deficiencies exist.

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