THE EFFECT OF HARDENING ON RELATIVE COLD RESISTANCE OF WINTER WHEAT VARIETIES

by

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B. S., Montana State College, 1928

A THESIS

submitted in partial fulfillment of the

requirements for the degree of

MASTER OF SCIENCE

KANSAS STATE AGRICULTURAL COLLEGE

1930

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INTRODUCTION

That artificial refrigeration may be expected under most conditions to give results in agreement with known field hardiness has been satisfactorily demonstrated in Kansas and elsewhere. Certain exceptions have been observed however, which, while they tend to reflect some doubt as to the merits of the method, may also be construed to indicate that the relations are more complex than is commonly supposed.

The promulgation of the hardening off hypothesis by Hill and Salmon (7) to explain the erratic behavior of such hardy wheats as Minhardi, opened the way for the present study, a major portion of which is concerned with the effects of hardening on the relative cold resistance of thirteen winter wheat varieties.

REVIEW OF LITERATURE

Winterkilling, according to Salmon (16), may be due to heaving, smothering, physiological drought, or direct effects of low temperature on the plant tissue and protoplasm. Of these, the latter is regarded as by far the most important in the southern Great Plains.

The measurement of resistance to cold has been attempted in diverse ways. In point of extent, the uniform winter hardiness nurseries, as reported by Clark, Martin, and Parker (4) are unsurpassed and permit the making of comparisons each year regardless of the mildness of the winter. The importance of these nurseries is not to be minimized, but it should be pointed out that other supplementary sources of similar information are necessary for the testing of a host of other hybrids, selections, or introductions which show promise, and to establish certain ecological relationships, peculiar perhaps to limited areas and therefore obtainable only in those areas.

Indirect methods of measuring the cold resistance of winter grains have been varied. Early workers were attracted to the possibility of determining hardiness by some morphological or anatomical characters, and even such recent workers as Klages (11), and Vavilov (22) present their findings in this field. These workers, and many others before them, have reached the general conclusion that morphological characters apparently have no absolute relationship to winter hardiness.

Recently, physiological changes as related to hardiness have attracted a large number of brilliant workers. The work of Akerman (1) in showing that hardening is accompanied by an increased sugar content in the plant in a sense pioneered this field. Since that time, Newton (14) has contributed the most extensively in this field, concentrating most of his attention on hydrophylic colloids and their relation to cold resistance. In a recently reported study (15), he concluded that a reduction of the water content of the cell with a subsequent concentration of the colloids and sugars constituted the most important change in the quantitative relationship of the plant constituents in producing winter hardiness. The work of Martin (12) failed to establish the dependability under a range of conditions of the measures employed by Newton, and, as a result, he suggests artificial refrigeration as the most desirable method of approach. The recent work of Janssen (9 and 10) indicates that interest in physiological changes has not entirely waned, however.

Artificial refrigeration to determine cold resistance was perhaps first used by Akerman (1). He, Hill and Salmon (7), and Tumanov and Borodin (21) have all shown that results by this method correlate very well with those obtained under field conditions. Klages (11) and Hill (6) showed that soil moisture exerted a protective influence during freezing. Klages (11) reported a progressive loss of hardiness in seedling plants from one to four weeks old, but so far as is known, all other workers have elected to freeze plants sometime after the tillering stage. Martin (12) was unable to find a consistent difference in the hardiness of greenhouse grown wheat plants varying in age from six weeks to four months.

The importance of thorough hardening of some varieties for a proper expression of their relative hardiness was stressed by Hill and Salmon (7). Akerman (1) and Tumanov and Borodin (21) used only hardened plants in their work and made no special effort to study hardening.

Hubbard (8) and Davis (5), working with hybrid lines grown continuously in the greenhouse before freezing, observed a rather wide difference in the injury of plants frozen in the morning and those frozen in the evening. They suggested that the greater hardiness in the latter results from an accumulation of sugars during the day. In a survey of a considerable range of freezing lots, Salmon (17) was unable to verify their findings.

The reliability of certain practices used in the present study have been determined by Salmon (17). In plants dug from the field and transplanted into four inch clay pots, he found that the amount of soil clinging to the roots when potted did not influence the subsequent results in freezing, but that size of clump (the amount of aerial parts potted in a single pot) did introduce a factor for variability, the larger clumps showing the least injury. He has also shown that the error in estimating per cent injury as measured by the standard deviation of the difference between two independent estimates is very small.

Recent work, outside the realm of artificial refrigeration, has a vital part in the present study. Working with a pure line of Turkey wheat (Wis. Ped. No. 2), Janssen (10) found that there was an optimum date for seeding this wheat with regard to its ability to withstand winter injury in the field. He reports that plants sown on the most desirable date have a greater capacity for changing their proteins from a precipitable to a non-precipitable form than do wheats sown on the other dates. In this connection, he also found that "the degree of hardiness of the winter wheat plant at different stages of development, resulting from successive seedings made in the fall, could not in all instances be attributed to the degree of root and top development".

Bower (3), working with seven varieties of winter wheat, found that Minturki, if sown early in October, was outstandingly the most hardy, and that Red Cross was among the least hardy. In late November seedings which failed to emerge before spring, Red Cross had a decided advantage over Minturki. Red Cross and Harvest Queen are considered by Salmon and Throckmorton (19) to be one and the same variety. So far as is known, time of seeding has not been a subject of investigation in artificial refrigeration work.

The measurement of cold resistance in the spring is reported by Akerman (1). He employed both sugar analysis and artificial refrigeration methods and found that great fluctuations occurred at this season.

Govorov, according to Maximov (13), found that in moving hardened plants into a warm environment, spring types gave a sharp reduction in sugar content, losing over 50 per cent in five days, whereas winter types showed only a slight reduction and in some instances, no reduction. This is not in agreement with the findings of Bayles and Salmon (2) who found marked changes in injury reflected within 48 hours.

For a more extensive review of the early literature dealing with cold resistance in cereals, the reader is referred to Akerman (1), Martin (12), and Hill (6).

EXPERIMENTAL PROCEDURE

This paper gives the results of a series of artificial refrigeration experiments conducted at the Kansas Agricultural Experiment Station in 1929-30. The refrigeration equipment consists of a direct expansion carbon dioxide refrigeration plant and a freezing chamber 10 feet by 4 feet in size. For a more detailed description of the same, the reader is referred to Hill (6) and to Sellschop and Salmon (20).

Thirteen varieties of winter wheat--Fulcaster, Currell, Harvest Queen, and Kawvale of the soft wheats; Oro, Minhardi, Cooperatorka, Blackhull, Early Blackhull, Tenmarq, Kanred x Hard Federation, Prelude x Kanred, and Kanred of the hard wheats--were tested for relative cold resistance during the months of November and December. Kansas accession record numbers for each of the above varieties may be found in Table I.

Four major types of material, hereinafter designated as groups I, II, III, and IV, differing greatly in treatment previous to freezing, were used. Plants in group I were sown on October 3 and thinned to five plants per pot soon after emergence. These five plants were distributed around the pot about one inch from the edge. They were grown continuously in the greenhouse until frozen. Six freezing lots or 450 pots are included in this group.

Plants in group II were planted on October 5 and treated exactly like those in group I until moved outside of the greenhouse on October 21 and 22 to undergo natural hardening off. The pots were placed directly on the earth in a fenced enclosure east of the greenhouses, being grouped in blocks of 300 pots which, in certain cases to be mentioned later, constituted a unit of four freezing lots frozen consecutively. In grouping the pots, care was taken to avoid having more than one pot for any variety in any one freezing lot on the outside of the group of pots where exposure was greatest and where injury would be expected to be the greatest. All varieties assumed a prostrate position by November 3, the degree depending on the variety. The plants were well watered in the absence of rain so that the soil moisture content was in harmony with that in group I. Sixteen freezing lots or 1200 pots were used in this group.

The plants used in group III were dug from plats at the Agronomy Farm and transplanted into four inch clay pots for freezing. These plats were planted September 23 and 24. Previous experience in transplanting wheat plants for freezing has shown that the plants must be of uniform clump size and be transplanted so that exactly the same aerial portions remain above ground as in the natural state. Suitable precautions regarding these factors were observed. The amount of soil adhering to the roots does not seem to influence the subsequent cold resistance of the plant but no clump of plants entirely void of soil was used. Usually two plants constituted a clump but occasionally there were three. As a rule, plants in this group were kept outside the greenhouse from one to three days after digging before being frozen. They were well watered to prevent wilting. Fifteen freezing lots or 1125 pots are reported from this group.

Frost in the soil was not a serious factor in any of these lots except those frozen November 23 to 25. In that case, the frost penetration was about four inches, making it necessary to bring the material into the greenhouse to allow sufficient thawing to permit potting. The experience was laborious and not entirely satisfactory. After potting, these plants were again placed outside.

Each freezing lot consisted of 15 pots of Kanred and five pots of each of the other varieties. The pots were frozen in boxes to facilitate easier handling, each box containing one pot of each variety except Kanred, of which there were three pots. Random distribution in the boxes was afforded even though previous experience had failed to indicate any necessity for it.

Freezing periods were uniformly 12 hours, each one beginning and ending at eight o'clock, with such temperature variations as seemed justified by past experience and existing conditions. It is not always possible to judge the desired temperature in advance, but the heavy demand on the freezing equipment and the extra labor requirement necessary in running test pot determinations for correct temperatures did not seem to warrant an extensive use of this practice under the changing conditions encountered in this experiment. For that reason, it was sometimes found necessary to change the temperature between consecutive lots of a weekly unit, or in some other cases, to re-freeze a lot in order to produce differential results between varieties. Whether or not this re-freezing introduced serious complications is not known but that some hardening occurred as a result of this exposure was evident. Lots which were re-frozen are starred in the tables.

Freezing lot averages for each of the varieties is the smallest unit presented in this study. This unit was adopted because any finer measure would only add bulk to the story. It might be thought that a measure of the variability of individual pots might be desirable but such a measure based on a population of 5 pots only seems to be unjustified. Modern statistical methods were used in the present paper when it seemed proper to do so.

Plants in group IV were analagous to those in group II except they were left outside the greenhouse all winter, thereby being subjected only to natural freezing.

In addition to the studies just mentioned, special studies involving cold resistance in the spring of 1929 after growth had started and a study of the rate of loss of hardiness under greenhouse temperatures were made. Material for both of these was secured at the Agronomy Farm and potted for freezing.

Maximum and minimum greenhouse temperatures during November and December are given in Table I. These data were obtained with a Columbia recording thermometer and are presented in weekly intervals. Outside temperatures are shown in Table II.

A general characterization of the weather during the autumn season may be of interest. In October, the mean temperature was practically in agreement with the 71 year average but the rainfall was considerably above normal.

November was termed "the coldest in 42 years", the cold wave being after the middle of the month. Precipitation was a trifle below normal. December temperature, while variable, was about normal.

Table I. Greenhouse Temperatures
During November and December

Week ending	Maximum	Minimum
November 6	71	55
" 13	70	53
" 20	70	53
" 27	74	51
December 4	64	44
" 11	65	49
" 18	62	50
" 25	62	42
January 1	66	47

Table II. Daily Maximum and Minimum Temperatures

for Manhattan, Kansas, 1929

Date	October		November		December	
	Max.	Min.	Max.	Min.	Max.	Min.
1	74	43	55	28	40	19
2	82	50	58	31	25	12
3	77	52	54	41	45	8
4	67	55	53	31	62	20
5	76	51	63	31	59	33
6	78	47	56	32	48	24
7	75	40	45	34	38	16
8	60	49	46	33	42	20
9	63	49	46	40	41	33
10	71	51	56	36	52	37
11	64	56	68	27	44	37
12	74	41	56	33	40	33
13	82	43	35	31	57	38
14	82	49	47	32	40	33
15	80	47	58	24	45	40
16	88	54	40	29	43	34
17	87	51	53	33	44	16
18	82	62	47	33	17	6
19	66	58	40	28	19	1
20	70	46	35	22	22	1
21	64	34	26	21	28	2
22	56	37	30	-1	30	0
23	53	33	33	20	45	3
24	58	40	43	14	56	29
25	72	31	50	29	62	19
26	76	39	54	29	61	30
27	74	36	57	35	51	22
28	60	46	48	30	49	32
29	63	46	31	22	62	24
30	62	46	38	12	67	30
31	52	39			56	23

In order to get some notion of the speed with which soil may be frozen and consequently of the temperature to which plants are actually subjected when frozen artificially, some observations of soil temperature were made at various times during the winter. For this purpose, an alcohol thermometer was used, the bulb being buried to a depth of about two inches in the center of the pot. The data secured are presented in Table III. In general, it appears that the soil temperature drops rapidly during the first few hours and then more slowly as it approaches the temperature of the air in the refrigeration chamber. During the last three or four hours, it appears to be within three to four degrees of the lowest temperatures in the refrigeration chamber.

It will also be apparent that for any given test, the air temperature in the freezing chamber is, for a considerable portion of the freezing period, considerably higher than the minimum finally attained. In reporting the results, the latter temperature is the one recorded in the tables.

Table III. Soil and Air Temperatures of Chamber in Degrees Fahrenheit at Hourly Intervals During Freezing

Hours	Time in Machine	8 0	9 1	10 2	11 3	12 4	1 5	2 6	3 7	5 9	8 12
	Air		7	-6	-14	-20	-23			-24	-24
	Soil	30	25	12	- 4	- 9	-15			-20	-20
	Air		1.4		-14	-14	-14			-14	-14
	Soil	35	30		25	7	- 4			-11	-11
	Air						- 2		-9	- 9	- 9
	Soil	41					14		-4	- 6	- 6
	Air		16			- 6	- 9		-9		- 9
	Soil	37	30			5	- 2		-6		- 6
	Air		5				5	5			5
	Soil	37	28				12	9			9
	Air			10	10		10		10		10
	Soil	37		28	21		14		14		14

EXPERIMENTAL RESULTS

Effect of Hardening on Relative Cold Resistance

As previously pointed out, the study of hardening involved four groups of plants grown and treated in various ways. The plants of one of these groups were grown in the greenhouse until they were frozen. This group was frozen in six freezing lots, two each on November 17, December 18, and December 19. The minimum temperatures to which the plants were exposed, the date of freezing, and the estimated degree of injury are given in Table IV.

The reported injury for each of the several varieties is an average of five pots (25 plants) except in the case of Kanred, of which there were 15 pots. The data are further summarized in Table V in which the per cent injury, the deviation from Kanred, and the ratio of D/E by the point binomial method as suggested by Salmon (18) for each date are given. In applying the latter, each varietal pot frozen in each box in the freezing chamber was compared with the mean injury for the three Kanred checks in that box. It will be remembered that a freezing lot consisted of five boxes, each containing one pot of each of the varieties except Kanred, of which there were three pots.

Table IV. Per Cent Injury by Freezing Lots of Greenhouse Grown Plants (Group I)

Variety	Kan. No.	Nov. 7 14°F A.M.	Nov. 7 14°F P.M.	Dec. 18[a] 3°F A.M.	Dec. 18[a] 3°F P.M.	Dec. 19 6°F A.M.	Dec. 19 6°F P.M.	Average Injury	Average Rank
Prelude x Kanred	2628	71	46	80	96	72	83	74.7	5
Early Blackhull	483	82	81	90	88	79	87	84.5	11
Blackhull	343	62	61	85	96	84	97	80.8	9
Tenmarq	439	65	62	83	93	77	92	78.7	7
Oro	495	73	37	79	83	60	79	68.5	3
Cooperatorka	499	50	73	80	93	78	86	76.6	6
Kan. x Hd. Fed.	2627	89	79	88	88	83	77	84.0	10
Minhardi	2450	60	28	65	62	32	46	48.8	1
Kawvale	2593	53	24	85	88	74	79	67.2	2
Fulcaster	317	86	83	85	94	91	89	88.0	12
Harvest Queen	19	54	69	84	95	83	92	79.5	8
Currell	501	90	93	97	99	93	98	95.0	13
Kanred Checks	2401	63	69	75	87	52	74	70.0	4

a. Previously frozen on December 14 at 8°F without injury.

Table V. Comparison of Varietal Relationships in Greenhouse Grown Plants (Group I)

Kan. No.	Variety	Nov. 7 (14°F) % Injury	± Kanred	D/E	Dec. 18 (3°F)[a] % Injury	± Kanred	D/E	Dec. 19 (6°F) % Injury	± Kanred	D/E
2628	Prelude x Kanred	58.5	-7.5	1.0	88.0	6.8	3.5	77.5	14.5	4.0
483	Early Blackhull	81.5	15.5	2.5	89.0	7.8	2.0	83.0	20.0	3.0
343	Blackhull	61.5	-4.5	0	90.5	9.3	3.0	90.5	27.5	5.0
439	Tenmarq	63.5	-2.5	1.0	88.0	6.8	3.0	84.5	21.5	5.0
495	Oro	55.0	-11.0	1.0	81.0	-.2	0	69.5	6.5	1.5
499	Cooperatorka	61.5	-4.5	0	86.5	5.3	2.0	82.0	19.0	3.0
2627	Kan. x Hd. Fed.	84.0	18.0	3.0	88.0	6.8	3.5	80.0	17.0	2.5
2650	Minhardi	44.0	-22.0	1.0	63.5	-17.7	3.5	39.0	-24.0	3.5
2593	Kawvale	38.5	-27.5	2.0	86.5	5.3	1.5	76.5	13.5	2.0
317	Fulcaster	84.5	18.5	4.0	89.5	8.3	2.0	90.0	27.0	4.5
19	Harvest Queen	61.5	-4.5	1.0	89.5	8.3	3.0	87.5	24.5	4.5
501	Currell	91.5	25.5	5.0	98.0	16.8	5.0	95.5	32.5	5.0
2401	Kanred	66.0	0	0	81.2	0	0	63.0	0	0
	Mean	65.6			85.0			76.3		

a. Previously frozen on December 14 at 8°F without injury

In freezing the different lots, it was intended so far as possible to subject the plants to a temperature which would produce approximately the same amount of injury in each. It will be seen that this objective was only partially attained. Thus the lot frozen on December 18 was injured considerably more than the others and the one frozen on November 7 least of all. Nevertheless, it is apparent that considerable hardening of the plants took place between November 7 and December 19, otherwise exposure to a temperature of 6° F on December 19 would have produced much more injury than was recorded for those lots. This conclusion is substantiated by the data given for the December 18 lots which had been frozen four days earlier at 8° F without producing measurable injury. It seems clear therefore, that moderate hardening of winter wheat may occur in the absence of freezing temperatures. This point is of special interest in view of the fact that minimum temperatures in the greenhouse during this period varied from 44° to 55° F.

The response of the different varieties to the hardening off phenomena is of special interest. Column five of Table IV shows only three varieties, viz., Currell, Fulcaster, and Kanred x Hard Federation 2627, which differed from Kanred by a statistically significant amount in early November. The average injury of Minhardi and Kawvale was materially less than that of Kanred but results with these varieties are so variable (as shown by D/E) that little can be said as to their position in this regard. Of the remaining varieties, none differ from Kanred by a statistically significant amount but all of them except Early Blackhull killed less than Kanred which is contrary to what would be expected on the basis of their known field behavior.

On December 18 and 19, all of the varieties except Oro and Kawvale differed from Kanred by a significant amount. The evidence seems to be clear cut in showing that Kanred had the ability to increase its resistance to low temperature to a greater degree than did most of the other varieties.

The difference in injury between Minhardi and Kanred was slightly greater when frozen in December than when frozen in November and the difference was more consistent in the latter case as shown by the ratio of D/E. Whether the difference is a significant one however, cannot be determined with certainty because of the great variability in the earlier frozen lots.

Kawvale apparently was more cold resistant than Kanred early in the season and less resistant later but this conclusion also is in doubt because of the variability in the early lots. Fulcaster and Currell seemed to harden off at about the same rate as Kanred. Both of them, of course, were much less resistant than Kanred at both dates.

The superiority of Minhardi over Kanred in all of these tests is not in line with the findings of Hill (6) who concluded that thorough hardening by exposure to rather low temperatures is essential for the expression of the normal relation of this variety to Kanred. Hill (6) found the survival of Minhardi to be about thirty per cent less than that of Kanred when frozen without hardening, while in this paper, the former variety shows a superiority to the extent of twenty per cent. This extreme difference in results suggests that temperature is not the only and perhaps not the most important cause for difference in varietal behavior in different years since greenhouse temperatures have been similar each year, as also have been the freezing temperatures.

But Minhardi was not the only marked exception to previously observed relationships. Blackhull showed cold resistance equal to that of Kanred on November 7 but was distinctly inferior on December 18 and 19. This early test, so far as the writer has been able to ascertain, is the first one in which Blackhull showed cold resistance equal to that of Kanred in freezing trials at the Kansas Experiment Station. Hill (6) reported a constant inferior relationship between Blackhull and Kanred in his work.

The relative behavior of Blackhull and Early Blackhull is also of interest. The early selection is regarded as the least hardy of the two under field conditions and this relationship held for the first tests but not for those made on December 18 and 19. Field comparisons of these varieties may be of interest. In eight cooperative tests in south central Kansas observed in March, 1930, Salmon reported an average of twenty three per cent injury to Blackhull and thirty four per cent to Early Blackhull. Data from 21 winter hardiness nursery stations, furnished by Dr. Parker, show that in 1928-29, Early Blackhull was the least hardy of the 30 varieties tested, with Fulcaster and Blackhull averaging slightly better.

The first lots of group II, i.e., greenhouse grown plants placed outside October 21 and 22, were frozen on November 4 after the plants had assumed a prostrate habit of growth, and the last on December 25. The per cent injury for each of the freezing lots in this group, together with the minimum temperatures to which they were exposed are given in Table VI and summarized in Table VII. The data show in a convincing way the striking rate at which winter wheats are able to adapt themselves for resistance to low temperatures. Thus, the injury on December 24 and 25 when the plants were subjected to a temperature of -25° F was even less than on November 4 and 5 when the temperature was 7° F. The former temperature is regarded by Martin (12) as about the minimum which unprotected wheat plants have been known to endure. Attention is called to the fact that four lots were frozen consecutively at each period in this group, as compared with two lots in group I, and for that reason, the averages as given in Table VI are based on four such lots in each instance.

In general, the observed behavior of the varieties was very similar to that noted in group I. Thus for the first date of freezing, the injury to Currell, Fulcaster, and Kanred x Hard Federation in relation to Kanred is precisely the same as in group I, and that of Minhardi in relation to Kanred differs only in the fact that the difference between them is statistically significant. At the last date of freezing, Oro, Kawvale, and Kanred x Hard Federation 2627 did not differ significantly from Kanred. Minhardi was clearly superior to Kanred, and all other varieties were clearly inferior.

Table VI. Per Cent Injury by Freezing Lots of Hardened Plants (Group II)

Kan. No.	Nov. 4 A.M. 7°	Nov. 4 P.M. 7°*	Nov. 5-6 P.M. 7°	Nov. 5-6 A.M. 7°	Nov. 18 A.M. 5°	Nov. 18 P.M. 1°*	Nov. 19 A.M. 2°	Nov. 19 P.M. 2°	Dec. 2-3 P.M. -19°@	Dec. 2-3 P.M. -12°†	Dec. 4 A.M. -19°	Dec. 4 P.M. -19°	Dec. 24 A.M. -25°	Dec. 24 P.M. -25°	Dec. 25 A.M. -25°	Dec. 25 P.M. -25°	Average Injury	Average Rank
2628	96	96	99	97	22	53	30	44	89	16	76	98	96	90	91	99	74.5	5
483	96	99	98	96	82	75	42	50	91	28	72	96	96	84	88	99	80.7	10
343	98	97	95	91	39	52	52	46	92	23	80	99	90	87	90	100	76.9	6
439	99	100	98	93	43	66	54	63	92	32	76	96	94	91	90	98	80.3	9
495	91	96	99	91	44	12	11	24	80	26	68	87	81	73	71	96	65.6	2
499	96	97	100	93	24	68	43	62	85	22	70	100	97	92	86	100	77.2	7
2627	99	100	100	96	28	85	53	31	91	26	66	98	92	92	84	95	77.2	7
2450	64	65	77	67	9	67	14	5	77	14	66	88	84	74	70	94	58.4	1
2593	95	95	99	94	17	48	32	26	80	24	58	99	96	89	72	92	69.8	3
317	98	100	100	98	38	94	54	67	97	52	72	100	97	98	93	99	84.8	12
19	95	98	100	93	54	90	73	41	96	60	68	98	94	96	93	100	84.3	11
501	100	100	100	100	74	99	79	64	100	93	93	100	100	100	98	100	93.8	13
2401	96	97	97	89	32	80	12	18	80	21	65	93	89	83	86	98	71.0	4

* Machine trouble
@ Previously frozen on December 2 at -10° F without injury
† Frozen 24 hours

Table VII. Comparison of Varietal Relationships in Group II

Kan. No.	Variety	Nov. 4-5 % Injury	Nov. 4-5 ± Kanred	Nov. 4-5 D/E	Nov. 18-19 % Injury	Nov. 18-19 ± Kanred	Nov. 18-19 D/E	Dec. 3-4 % Injury	Dec. 3-4 ± Kanred	Dec. 3-4 D/E	Dec. 24-25 % Injury	Dec. 24-25 ± Kanred	Dec. 24-25 D/E
2628	Prelude x Kanred	97.0	2.2	2.7	37.3	4.5	1.0	69.7	4.8	4.0	94.0	5.1	4.7
483	Early Blackhull	97.3	2.5	2.3	62.3	29.5	5.7	71.8	6.9	3.3	91.7	2.6	4.0
343	Blackhull	95.3	.5	.7	47.3	14.5	1.7	73.5	8.6	5.3	91.7	2.6	4.3
439	Tenmarq	97.5	2.7	2.7	56.5	23.7	3.3	74.0	9.1	5.0	93.2	4.3	4.0
495	Oro	94.3	-.5	1.3	22.3	-10.5	1.0	65.3	.4	.3	80.2	-8.7	2.0
499	Cooperatorka	96.5	1.7	1.0	49.3	16.5	1.3	69.3	4.4	2.3	93.7	4.8	5.0
2627	Kanred x Hard Fed.	99.0	4.2	5.0	49.3	16.5	5.0	70.3	5.4	3.0	90.7	1.8	2.7
2450	Minhardi	68.3	-26.5	6.0	22.7	-10.5	4.0	61.3	-3.6	.7	80.5	-8.4	4.3
2593	Kawvale	95.8	1.0	1.7	30.8	-2.0	1.0	65.3	.4	1.3	87.2	-1.7	0
317	Fulcaster	99.0	4.2	4.3	65.3	32.5	4.7	80.0	15.1	6.3	96.7	7.8	5.3
19	Harvest Queen	96.5	1.7	1.0	64.5	31.7	5.7	80.5	15.6	5.3	95.7	6.8	6.7
501	Currell	100.0	5.2	5.7	79.0	46.2	6.0	96.5	31.6	6.7	99.5	10.6	6.7
2401	Kanred	94.8	0	0	32.8	0	0	64.9	0	0	88.9	0	0

The data for the field grown plants (group III) are given in Tables VIII and IX. The plants frozen on November 12 and 13 were injured so slightly that they were refrozen on November 21 and 22, having been kept in the greenhouse hardening room during the interval. The injury notes for November 21 and 22 as recorded in Table VII therefore, are for the same plants as are those for November 12 and 13. They are arranged in reverse order however. That is, the lot frozen on the forenoon of November 12 is the same as the one frozen on the forenoon of November 22, etc. The freezings of November 23 and 25 were not entirely satisfactory because of the frozen soil which made it necessary to partially thaw the plants in the greenhouse before potting. The potting was far from satisfactory.

It is clear that the plants in this group did not attain the same degree of hardiness as those in group II, as shown by the fact that more severe injury occurred when frozen on December 8 and 12 with a temperature of -9° F than occurred in group II frozen December 2 to 4 with a temperature of -19° F. This of course is not difficult to understand in view of the greater exposure of the plants in group II (pots set on the surface of the ground) as compared with plants in the field.

Table VIII. Per Cent Injury by Freezing Lots of Field Grown Plants (Group III)

Kan. No.	Nov. 12		Nov. 13		Nov. 21		Nov. 22		Nov. 23		Nov. 25		Dec. 8		Dec. 12	Average	
	A.M. 7°	P.M. 7°	A.M. 7°	P.M. 7°	P.M. 7°@	A.M. 7°@	P.M. 7°@	A.M. 7°@	P.M. -8°	A.M. -7°	A.M. -7°*	P.M. -8°	A.M. -17°*	P.M. -9°	A.M. -9°	Injury	Rank
2628	68	60	46	44	66	79	83	86	78	65	46	83	84	80	92	70.7	8
483	75	54	54	54	81	79	77	79	81	65	71	97	96	76	95	75.6	12
343	76	58	42	60	70	66	76	66	86	74	80	83	93	85	94	73.9	11
439	73	46	44	38	72	66	70	66	68	48	71	71	92	64	76	64.3	6
495	60	52	40	52	66	64	66	64	64	62	60	68	85	66	90	63.9	4
499	66	54	42	50	72	68	72	68	70	66	79	90	87	91	91	71.1	9
2627	66	50	40	46	68	72	68	72	73	70	75	73	87	76	86	68.1	7
2450	34	46	34	17	58	56	58	56	42	22	38	58	46	56	46	44.5	1
2593	58	52	40	34	62	66	62	66	71	66	62	85	66	72	73	62.3	2
317	66	60	50	46	72	86	75	72	71	56	74	93	85	84	95	72.3	10
19	52	44	44	32	74	70	70	72	68	56	54	81	84	72	88	64.1	5
501	66	70	56	58	84	83	94	95	88	50	60	82	93	100	100	78.6	13
2401	65	59	45	39	63	67	71	76	62	47	62	73	78	70	77	63.7	3

* Machine trouble
@ Same lots as those frozen on November 12 and 13 but arranged in reverse order

Table IX. Comparison of Varietal Relationships in Group III

Kan. No.	Variety	Nov. 12-13 % Injury	Nov. 12-13 ± Kanred	Nov. 12-13 D/E	Nov. 21-22 % Injury	Nov. 21-22 ± Kanred	Nov. 21-22 D/E	Nov. 23-25 % Injury	Nov. 23-25 ± Kanred	Nov. 23-25 D/E	Dec. 8 & 12 % Injury	Dec. 8 & 12 ± Kanred	Dec. 8 & 12 D/E
2628	Prelude x Kanred	54.5	2.3	1.7	78.5	9.2	5.0	68.0	6.7	2.7	85.3	10.0	3.4
483	Early Blackhull	59.3	7.1	4.0	79.0	9.7	5.7	78.5	17.2	4.7	89.0	13.7	5.0
343	Blackhull	59.0	6.8	2.7	69.5	.2	.7	80.7	19.4	5.3	90.7	15.4	5.4
439	Tenmarq	50.3	-1.9	.7	68.5	-.8	.3	64.5	3.2	1.7	77.2	1.9	2.7
495	Oro	51.0	-1.2	.7	65.0	-4.3	1.3	63.5	2.2	.7	80.3	5.0	3.0
499	Cooperatorka	53.0	.8	0	70.0	.7	2.0	76.3	15.0	5.3	89.7	14.4	5.4
2627	Kanred x Hard Fed.	50.5	-1.7	1.7	70.0	.7	.7	72.3	11.0	3.3	83.0	7.7	2.7
2450	Minhardi	32.3	-19.9	5.0	57.0	-12.3	4.7	40.0	-21.3	5.3	49.3	-26.0	4.6
2593	Kawvale	46.0	-6.2	2.0	64.0	-5.3	1.3	71.0	9.7	3.7	70.3	-5.0	3.4
317	Fulcaster	55.5	3.3	3.0	76.2	6.9	2.3	73.5	12.2	4.7	87.7	12.4	5.0
19	Harvest Queen	43.0	-9.2	2.3	71.5	2.2	2.0	64.7	3.4	1.7	81.3	6.0	5.0
501	Currell	62.5	10.3	4.7	89.5	20.2	6.0	70.0	8.7	3.3	97.7	22.4	5.6
2401	Kanred	52.2	0	0	69.3	0	0	61.3	0	0	75.3	0	0

The varietal relationships are about the same as in groups I and II. Thus in the early freezing tests, Currell, Fulcaster, and Minhardi bear exactly the same relation to Kanred as in the other two groups. The relation between Early Blackhull and Kanred is the same as in other groups except that in this case, the difference appears to be statistically significant. Kanred x Hard Federation killed practically the same as Kanred in this group. The relation of all other varieties to Kanred is essentially the same as in the other two groups.

For the last period of freezing, i.e., on December 8 and 12, the agreement with groups I and II is good except for Kawvale. In group III, Kawvale killed less than Kanred by a significant amount. It also killed less in group II and more in group I but in neither case is the difference statistically significant. Tenmarq, as in the other groups, killed more than Kanred but the difference in group III is only 2.7 times the probable error.

The intermediate dates of freezing for all lots gave results essentially intermediate in character between those of the first and last dates of freezing for the respective groups. Reference to this fact is made later, hence a detailed discussion of the results is not called for at this time.

Since the behavior of the varieties was much the same in the three groups for any given period, it seems that the results may be averaged. This has been done for the earliest and for the latest freezing lots of each group only and the results are presented in Table X.

From this data, it will be seen that when frozen in November, Minhardi killed less than Kanred and Early Blackhull, Kanred x Hard Federation, Fulcaster, and Currell killed more than Kanred by a statistically significant amount. All others were essentially equal to Kanred. For the freezings in December, Minhardi killed less than Kanred as before, and all others, excepting Oro and Kawvale, killed considerably more and by a significant amount. Oro and Kawvale survived to about the same degree as Kanred. On the basis of these results, Kanred increased in hardiness to a materially greater degree than did Prelude x Kanred, Blackhull, Tenmarq, Cooperatorka, and Harvest Queen. There is no evidence to show a marked difference in this respect as compared with Minhardi, Early Blackhull, Kanred x Hard Federation, Oro, Kawvale, Fulcaster, and Currell. The trend in hardiness with respect to Kanred is especially well shown in figures 1, 2, and 3 in which the per cent of injury in deviations from Kanred are shown graphically.

Table X. Average Injury by Periods of Groups I, II, and III

Variety	Nov. 4-5; 7; and 12-13 % Injury	± Kanred	D/E	Dec. 8-12; 19; and 24-25 % Injury	± Kanred	D/E
Prelude x Kanred	70.0	-1.0	1.9	82.3	6.6	7.0
Early Blackhull	79.4	8.4	5.1	87.9	12.2	7.0
Blackhull	71.9	.9	1.8	90.9	15.2	8.4
Tenmarq	70.4	- .6	1.8	85.0	9.3	6.6
Oro	66.8	-4.2	0	71.7	-4.0	1.1
Cooperatorka	70.3	- .7	.7	88.5	12.8	8.0
Kan. x Hd. Fed.	77.8	6.8	6.9	84.6	8.9	4.5
Minhardi	48.2	-22.8	7.2	56.3	-19.4	7.3
Kawvale	60.1	-10.9	1.0	78.0	2.3	1.1
Fulcaster	79.3	8.3	6.9	91.5	15.8	8.6
Harvest Queen	67.0	-4.0	.4	88.2	12.5	9.5
Currell	84.7	13.7	7.9	97.6	21.9	10.0
Kanred	71.0	0	0	75.7	0	0

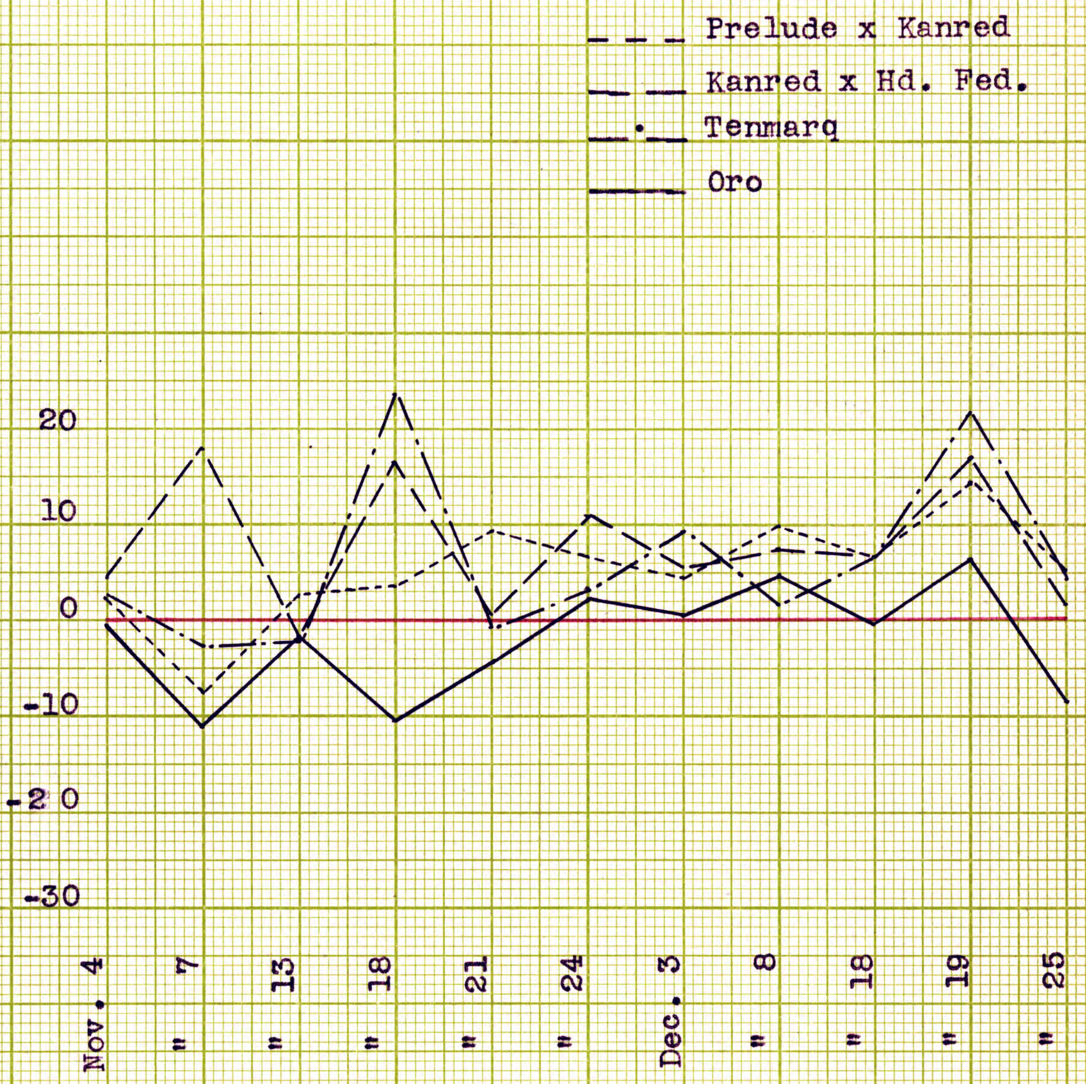

Fig. 1. Chronological deviations from Kanred in per cent injury.

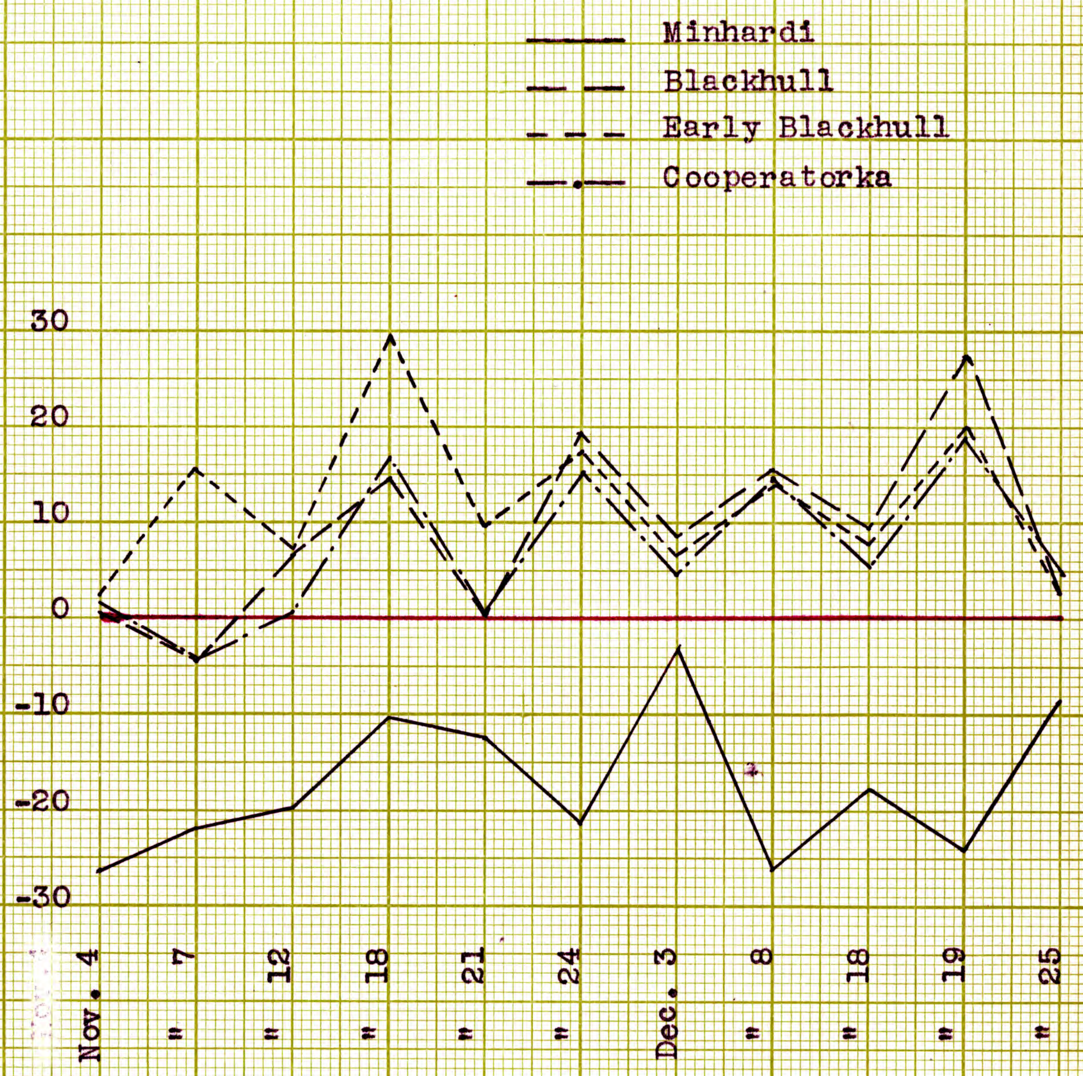

Fig. 2. Chronological deviations from Kanred in per cent injury.

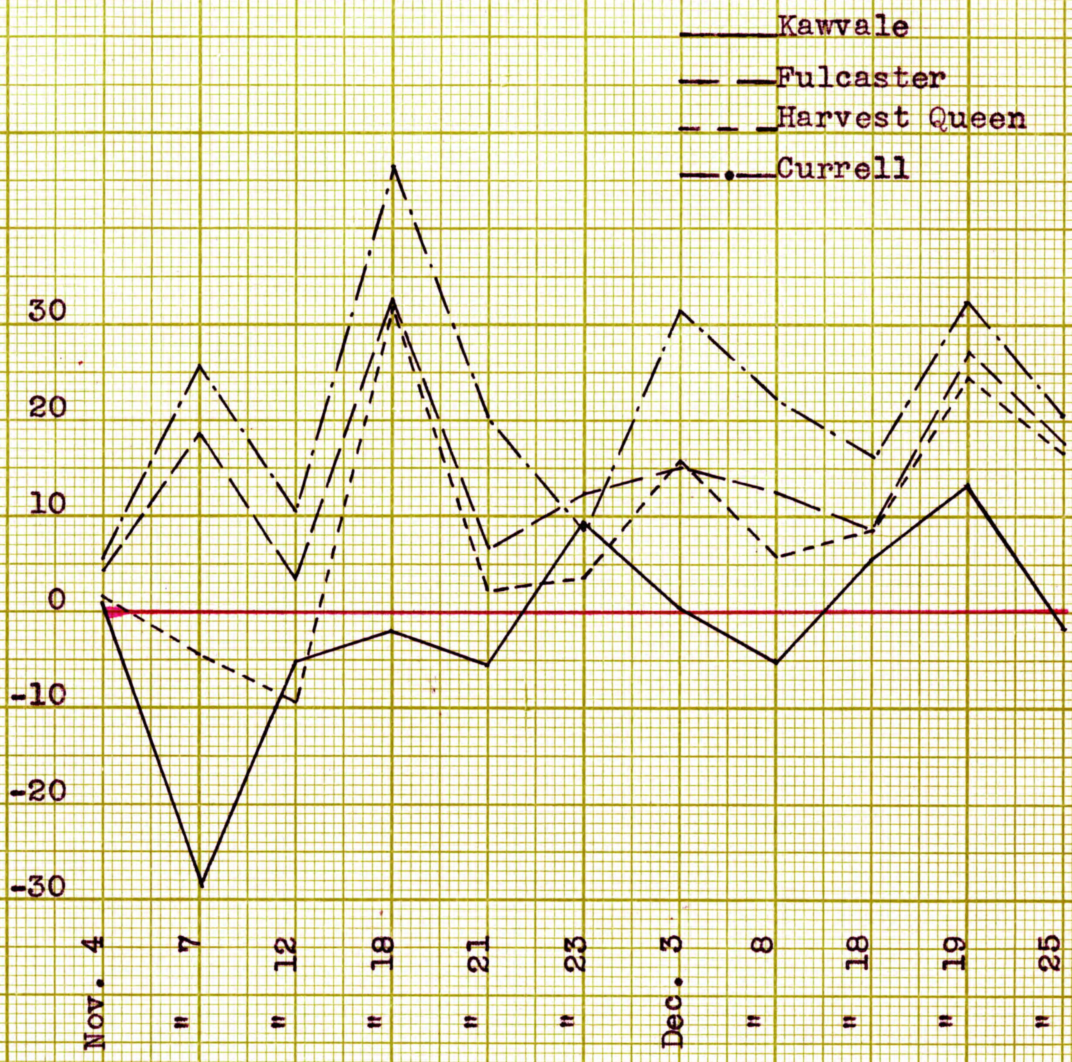

Fig. 3. Chronological deviations from Kanred in per cent injury.

These data are arranged chronologically without regard to group since, as has already been shown, the varieties tended to react alike in each group. In interpreting the graphs, it should be noted that deviations (from Kanred) tend to be minimized when the injury is great and also when the injury is slight. The wide deviations from Kanred in certain freezing lots and the slight deviations in others are in part due to this fact.

Of special interest in connection with the hardening off hypothesis is the fact that the greatest deviations from Kanred were secured in group I. This is shown clearly in Table XI which gives the deviations from Kanred for each variety for the first (November) freezing and for the last (December) freezing of each lot together with the range for each variety for each lot. It will be seen that more than half of the varieties (all but Early Blackhull, Kanred x Hard Federation, Minhardi, Fulcaster, and Currell) exhibit a greater range for group I than for group II or III. This result emphasizes the disagreement with the hypothesis suggested by Hill since, if exposure to low temperature is necessary for the full expression of winter hardiness, the greatest differences between varieties should have been secured in groups II and III.

Table XI. Comparative Group Changes in Varietal Relations

Variety	Group I			Group II			Group III		
	Nov.	Dec.	Range	Nov.	Dec.	Range	Nov.	Dec.	Range
Prelude x									
Kanred	- 7.5	14.5	22.0	2.2	5.1	2.9	2.3	10.0	6.7
Early Blackhull	+15.5	20.0	4.5	2.5	2.8	0.3	7.1	13.7	4.6
Blackhull	- 4.5	27.5	32.0	0.5	2.8	2.3	6.8	15.4	8.6
Tenmarq	- 2.5	21.5	19.0	2.7	4.3	1.6	-1.9	1.9	3.8
Oro	-11.0	6.5	17.0	-0.5	-8.7	8.3	-1.2	5.0	3.8
Cooperatorka	- 4.5	19.0	14.5	1.7	4.8	6.5	0.8	14.4	13.6
Kan. x Hd. Fed.	18.0	17.0	1.0	4.2	1.8	6.0	-1.7	7.7	9.4
Minhardi	-22.0	-24.0	2.0	-26.5	-8.4	18.1	-19.9	-26.0	6.1
Kawvale	-27.5	13.5	41.0	1.0	-1.7	2.7	-5.8	-5.0	0.8
Fulcaster	18.5	27.0	8.5	4.2	7.8	3.6	3.3	12.4	9.1
Harvest Queen	4.5	24.5	20.0	1.7	6.8	5.1	-9.2	6.0	3.2
Currell	25.5	32.5	7.0	5.2	10.6	5.4	10.3	22.4	12.1
Ave.			15.7			5.23			6.82

The point may be further emphasized in relation to Minhardi. This variety, it may be seen by reference to Tables V, VII, and IX, was included in eleven different freezing lots. In every case, it killed less than Kanred, and in every case but one, the difference is significant. In three of the lots, no material hardening as a result of low temperature could have taken place since they were frozen no later than November 13 and no cold weather had occurred up to that time.

It appears from the above that some modification of Hill's hypothesis may be necessary. The results presented here suggest that the true response of any given variety in relation to any other variety to freezing temperatures is a function of the seeding date or of the season of the year rather than of the temperature to which they are exposed. It seems to be not so much a function of the age of the plants as it is a function of the time of the year at which the plants emerge. It is of interest in this connection to note that Wanser (23) has reported distinct "minimum stimulating photoperiods" for the expression of jointing and heading stages in winter wheats, the same varying for different varieties. He likewise conceives of winter habit as resulting from an absence of a critical photoperiod stimulus for jointing. The present study seems to go still farther in suggesting a critical photoperiod for maximum expression of relative cold resistance, particularly in wheats such as Minhardi.

The importance of time of seeding in relation to relative cold resistance has also been suggested by other recent workers. Thus Janssen (10) found an optimum seeding date for a maximum expression of cold resistance in a selected line of Turkey, and Bower (3) found that Minturki survived best when seeded in early October, while Red Cross which had a low survival when planted in the early fall, was more cold resistant than Minturki when sown in November.

Certain results by Hill (6) also suggest the importance of time of seeding. In "flat" studies planted on October 8 he found that Minhardi was more cold resistant than Kanred in all tests, with both non-hardened and moderately hardened plants. His "pot" studies were seeded on October 17 and in these, Kanred was superior to Minhardi when non-hardened plants were frozen and the two varieties essentially alike when compared in a moderately hardened state. He recognized the disagreement between the two groups but seems to have missed a vital point when he combined the two seeding dates to give results which pointed to hardening by exposure to low temperatures as being responsible for the difference in favor of Minhardi in relation to Kanred. It so happened that over half of his hardened plants were seeded on the early date and practically all of the non-hardened comparisons were drawn from plants seeded on the later date. Salmon (17), working with greenhouse plants planted on November 4, found Kanred to be considerably more cold resistant than Minhardi.

Fortunately, studies by Salmon in progress during the time the present data was secured were comparable to groups I and II in every respect except that seeding had been eight days later. The behavior of Minhardi in particular deserves attention. In the early freezing tests, Minhardi was clearly superior to Kanred in non-hardened (greenhouse grown) comparisons, but was inferior in the hardened (outdoor material) group. The latter result is contrary to the findings in this paper and appears to have resulted from the difference (even though relatively slight) in seeding date.

A natural question and one of great importance has to do not only with the agreement between the results secured by artificial refrigeration and the survival under field conditions but also what methods should be used in the former to secure the best results. Some of the questions that may be considered here are (1) relative variability in the different tests, and (2) time of the year when the plants should be frozen. Table XII gives the standard deviation for the Kanred in each freezing lot, the figures given being the standard deviation for the mean of 15 pots. It will be seen that the standard deviation is least for group III, nearly as low for group II, and highest for group I. This perhaps is explained by the fact that group I was frozen at higher temperatures, consequently the insulating effect of the soil was greater and the variation in temperature in different portions of the freezing chamber was greater.

With regard to the second part, Table XIII has been prepared which gives the average injury for all groups regardless of time of freezing, the deviation from Kanred, and the rank. Consideration of the ranks as compared with those presented in Table X suggests that either ranking agrees fairly well with what is known of the behavior of the varieties under field conditions, excepting that a rank based on the early freezing results only presented in Table X does not agree with field results.

It appears therefore that in conducting a freezing test for any new variety, the test should not be made too

Table XII. Standard Deviation for Kanred in Each Freezing Lot As

A Measure of Lot Variability

Date	Group I Mean % Injury	S.D.	Date	Group II Mean % Injury	S.D.	Date	Group III Mean % Injury	S.D.
11-7 (A.M.)	62.7	6.63	11-4 (A.M.)	96.0	1.50	11-12 (A.M.)	65.3	2.92
" (P.M.)	69.3	5.65	" (P.M.)	97.0	.92	" (P.M.)	58.7	1.28
12-18(A.M.)	75.3	3.84	11-5 (P.M.)	97.3	1.04	11-13 (A.M.)	45.3	4.31
" (P.M.)	87.0	2.30	11-6 (A.M.)	88.7	2.68	" (P.M.)	39.3	3.59
12-19(A.M.)	51.7	5.41	11-18(A.M.)	31.7	6.96	11-23 (A.M.)	76.3	3.39
" (P.M.)	74.3	4.85	" (P.M.)	80.3	4.55	11-22 (P.M.)	71.0	3.02
			11-19(A.M.)	12.3	2.74	11-22 (A.M.)	66.7	3.45
			" (P.M.)	18.0	7.16	11-21 (P.M.)	63.3	1.54
			12-3 (P.M.)	80.3	2.49	11-23 (P.M.)	62.3	3.16
			12-2 (P.M.)	21.0	4.24	11-24 (A.M.)	47.3	3.05
			12-4 (A.M.)	64.7	2.08	11-25 (A.M.)	62.0	3.63
			" (P.M.)	93.3	.88	" (P.M.)	73.0	3.22
			12-24(A.M.)	89.3	1.85	12-8 (A.M.)	78.3	2.51
			" (P.M.)	83.0	3.01	" (P.M.)	70.0	2.45
			12-25(A.M.)	85.7	2.83	12-12 (A.M.)	77.3	3.49
			" (P.M.)	97.7	.64			
Average	70.0	4.98		71.0	3.44		63.7	3.10

Table XIII. Relative Injury by Freezing

Summary of All Data

Kan. No.	Variety	Average percentage injury: Group I	Group II	Group III	All Groups[a]	Rank	Deviation from Kanred	D/E
2628	Prelude x Kanred	74.7	74.5	70.7	73.5	5	5.4	8.94
483	Early Blackhull	84.5	80.7	75.6	80.2	11	12.1	12.80
343	Blackhull	80.8	76.9	73.9	77.3	10	9.2	10.04
439	Tenmarq	78.7	80.3	64.3	74.3	6	6.2	7.95
495	Oro	68.5	65.6	63.9	66.1	2	-2.0	0
499	Cooperatorka	76.6	77.2	71.1	75.3	7	7.2	8.28
2627	Kan. x Hd. Fed.	84.0	77.2	68.1	76.1	9	8.0	8.72
2450	Minhardi	48.8	58.4	44.5	50.7	1	-17.4	13.03
2593	Kawvale	67.2	69.8	62.3	66.5	3	-1.6	.77
317	Fulcaster	88.0	84.8	72.3	81.6	12	13.5	13.91
19	Harvest Queen	79.5	84.3	64.1	76.0	8	7.9	9.93
501	Currell	95.0	93.8	78.6	89.1	13	21.0	17.55
2401	Kanred	70.0	71.0	63.7	68.1	4	0	0

a. Weighted according to number of tests early in the fall. Furthermore, it would seem to be imperative that all varieties for a given comparison be seeded at the same date and at least desirable that the varieties be compared for more than one seeding date.

Table XIV gives the per cent of surviving plants in group IV, as determined by actual count on February 20 for 35 pots or 175 plants of each variety. It will be remembered that this group is comparable in every respect with group II except that the plants were left outside all winter and subjected to natural freezing only.

Rabbits caused considerable damage to all varieties in late December except Oro which they seemed to shun. The defoliation no doubt lowered the percentage of surviving plants but apparently affected all varieties (except Oro) alike. Another factor, drought, is believed to have caused the death of some plants which normally would have survived. The great fluctuations in temperature were conducive to a higher rate of evaporation than the attendant anticipated and consequently, the pots were found late in the winter to be very dry.

Despite these obvious sources of error, the behavior of the varieties in this environment as contrasted with artificial freezing seems to indicate a differential response.

Table XIV. Comparison of Natural Freezing Results With Those By Artificial Refrigeration

Variety	Spring Survival of Naturally Frozen Potted Plants % Surviving Plants	Rank	Rank Comparisons Group I	Group II	Group III
Prelude x Kanred	4.4	8	5	5	8
Early Blackhull	1.5	11	11	10	12
Blackhull	2.7	10	9	6	11
Tenmarq	12.6	7	7	9	6
Oro	33.1	2	3	2	4
Cooperatorka	1.1	12	6	7	9
Kan. x Hd. Fed.	17.1	3	10	7	7
Minhardi	53.6	1	1	1	1
Kawvale	14.3	6	2	3	2
Fulcaster	3.4	9	12	12	10
Harvest Queen	17.1	3	8	11	5
Currell	0	13	13	13	13
Kanred	15.2	5	4	4	3

This was particularly true of Harvest Queen and of Kanred x Hard Federation 2627, in which the survival was equal to that of Kanred and of Cooperatorka with a lower per cent of survival than Blackhull. Minhardi, it will be observed, was preeminently more cold resistant than any other variety. As may be seen later in Table XIX, giving results in spring freezing trials, Kanred x Hard Federation 2627 seems to reach its maximum hardiness in late winter so no doubt the relationship expressed here is accurate.

The exceptional behavior of certain varieties may be conceived of as a continuation of adjustments in varietal relationships throughout the winter but it is also possible to think of part of the difference at least as being caused by prolonged and repeated freezing and thawing. The cause cannot be fixed but the facts seem to be dependable since a population of 175 plants is large enough to minimize errors.

Effect of Location in Freezing Chamber

Since account had never been taken of the fact that two of the five boxes of each freezing lot have three exposed surfaces, whereas the three central boxes have only two exposures toward the refrigeration pipes on the walls of the chamber, it was felt that perhaps one cause for the rather high variability within varieties in a freezing lot might be found here.

Averages for the two end boxes were compared with those for the three inside boxes for eight freezing lots for which a definite record of location when frozen was available. This data is presented in Table XV. Apparently location in the chamber is not an important contributory cause to variability for in no case are the differences pronounced and the average for the two is essentially equal. It seems therefore, that the boxes in which the pots are frozen provide the adequate insulation against the uneven freezing which the construction of the chamber suggests.

Table XV. Effect of Location of Boxes in Freezing Chamber

Date	Group No.	Average Per Cent Injury End Boxes	Inside Boxes
Dec. 18 (A.M.)	I	79.8	83.1
" (P.M.)	I	90.5	88.1
Dec. 19 (A.M.)	I	69.0	71.9
" (P.M.)	I	85.2	79.6
Dec. 24 (A.M.)	II	91.9	92.7
" (P.M.)	II	88.9	86.9
Dec. 25 (A.M.)	II	86.2	84.9
" (P.M.)	II	96.7	98.3
Average		86.0	85.7

Day Versus Night Freezing

In comparing artificial freezing tests of segregating hybrid lines together with parent stocks, Hubbard (8) and Davis (5) have shown rather striking differences in favor of the lots put into the refrigerator in the evening and frozen during the night. They have advanced the hypothesis that the difference was due to an increase of sugars during the day and a subsequent translocation or utilization during the night. Salmon (17), employing a wide range of comparisons very much like the discussion to follow, failed to find any great day and night differences.

The present study is based on all strictly comparable, consecutively frozen lots from the three hardiness groups and from spring freezing trials to be discussed later. The data given in Table XVI are not concerned with the response of the several varieties but merely give the mean lot injury including all varieties.

From a study of this data, it is apparent that variations both ways occurred in all groups but that the mean for the two periods gives results opposite to those secured by Hubbard and by Davis. No explanation for this discrepancy is known but it may possibly be related to the fact that all material frozen by Hubbard and by Davis was grown in the south end of the greenhouse (both before and after freezing) where the temperature, as a rule, is about 10° F higher than in the north end. None of the material used in the present study was grown in the south end of the greenhouse.

At least it is clear that in making freezing tests, the possible difference between day and night freezing must be considered.

Table XVI. Day and Night Variations in Comparable Freezing Lots

Date	Group	Min. Temp.	Per Cent Injury Day	Frozen During Night	Difference
Nov. 7	I	14°	68.2	62.9	- 5.3
Dec. 18	I	3°	81.8	89.1	+ 6.3
Dec. 19	I	6°	70.7	81.9	+ 11.2
Nov. 5-6	II	7°	87.5	95.5	+ 8.0
Nov. 19	II	2°	38.3	37.0	- 1.3
Dec. 4	II	- 19°	73.0	96.0	+ 13.0
Dec. 24	II	- 25°	92.3	87.8	- 4.5
Dec. 24-25	II	- 25°	85.5	87.8	- 7.7
Dec. 25	II	- 25°	85.5	97.7	+ 12.2
Nov. 12	III	7°	63.7	54.8	- 8.9
Nov. 12-13	III	7°	44.5	54.8	+ 10.3
Nov. 13	III	7°	44.5	43.3	- 1.2
Nov. 21-22	III	7°	70.3	69.0	- 1.3
Nov. 22-23	III	7°	72.4	72.4	0.0
Nov. 22	III	7°	70.3	72.4	+ 2.1
Mar. 15	Spring	- 8°	73.5	79.1	+ 5.6
Mar. 15-16	Spring	- 8°	83.4	79.1	- 4.3
April 6	Spring	13°	84.3	68.2	- 16.1
Average			73.2	78.2	+ 5.0

Cold Resistance and Time of Heading

It seems to be a fairly general belief that the factors for resistance to cold in wheat are linked with those for late maturity. It seemed worth while to consider this subject inasmuch as practically the entire maturity range of Kansas wheat varieties were included in this study and since greater earliness with hardiness is one of the objectives sough in wheat improvement in this state.

In making the comparisons, the mean heading date was chosen as the time of maturity index since the ripening date in Kansas is often greatly disturbed by adverse temperatures. The rank of the 13 varieties from the hardiness study, together with their rank based on the average heading date for three crop years is given in Table XVII.

Table XVII. Comparison of Heading and Cold Resistance Ranks

Variety	Mean Heading Date	% Mean Injury	Comparison of Ranks Heading Date	Comparison of Ranks Cold Resistance
Prelude x Kanred	5-21	73.5	2	5
Early Blackhull	5-16.3	80.2	1	11
Blackhull	5-23.3	77.3	5	10
Tenmarq	5-23	74.3	10	6
Oro	5-27	66.1	11	2
Cooperatorka	5-29	75.3	12	7
Kan. x Hd. Fed.	5-23	76.1	3	9
Minhardi	6-1	50.7	13	1
Kawvale	5-24.7	66.5	7	3
Fulcaster	5-25.3	81.6	8	12
Harvest Queen	5-27	76.0	3	8
Currell	5-24	89.1	6	13
Kanred Checks	5-25.7	68.1	9	4

The correlation coefficient calculated from the ranks in this table was found to be -.452 $\pm$ .0015 which suggests a definite relationship.

There are a number of exceptions that merit special mention. Cooperatorka is distinctly later than Kanred and less winter hardy, both traits being undesirable from a Kansas viewpoint. Compared with this, Prelude x Kanred is conspicuously early and approaches the cold resistance of Kanred to a greater degree than has heretofore been noted in such an early wheat. The reader is referred to figure 1 for this comparison. The fact that the winter hardy parent used in this cross was an adapted variety may be singularly important.

In the soft wheat group, Kawvale, a recent product of selection, was outstandingly more hardy than the other soft wheats generally grown in Kansas, was earlier than Kanred, and was about equal to Kanred in cold resistance.

With these citations, it seems evident that a degree of cold resistance acceptable in Kansas may be combined with earlier maturity.

Loss of Hardiness from Exposure to Constant Greenhouse Temperatures

In order to gain some notion of the rate at which hardiness may be lost, once it is acquired, a few tests were made to determine the loss in consecutive 24 hour periods under practically constant greenhouse temperatures of about 50° F. For this purpose, 15 pots of field grown plants of each of five varieties were frozen in each of nine lots which had been kept in the greenhouse from 0 to 96 hours before freezing. The average per cent injury for each of the five varieties in each 24 hour interval is presented in Table XVIII.

The results are in agreement with those of Bayles and Salmon (2) who found that significant changes in hardiness occurred within 24 to 48 hours under greenhouse temperatures. It appears that a constant loss in hardiness may be expected up to 96 hours. In the instance where 24 hours inside produced greater injury than did 48 hours, it seems that the time of day when the plants were brought into the greenhouse had an effect. The former lot was the only one brought into the greenhouse in the evening.

The study was not extensive enough to permit reliable measurement of any varietal differences in response to this treatment. All seemed to respond in about the same way.

Table XVIII. Loss of Hardiness by Exposure to Constant

Greenhouse Temperatures

Variety	Plants dug on Dec. 5, Frozen 12 hours at -9° F, Hours inside before freezing				Plants dug on Dec. 27, Frozen 12 hours at -10° F, Hours inside before freezing				
	0	48	72	96	0	24	48	72	96
Kanred	73.7	82.0	94.7	91.3	72.0	97.7	91.7	98.0	99.0
Blackhull	89.5	94.7	97.0	99.0	98.3	99.3	100.0	99.7	100.0
Minhardi	51.0	62.0	69.7	73.0	66.7	86.7	78.0	78.0	94.3
Kawvale	72.5	85.3	96.3	97.0	84.3	99.0	97.7	99.7	99.3
Harvest Queen	80.0	91.7	98.0	97.7	99.0	98.3	99.7	99.3	100.0

Relative Hardiness of Selected Wheat Varieties After Spring Growth Begins

In order to get a more complete seasonal measure of the relative hardiness of several wheat varieties, some extensive tests were made in the spring of 1929. It was felt that the rate of loss of hardiness for these varieties under field conditions might also be determined at this time.

During the period March 15 to 21, five varieties concerned in this study were compared, each freezing lot consisting of 15 pots of Kanred and five pots of each of the other varieties. This period marked the beginning of active growth following the passing of frost from the soil. In all subsequent freezings, the mean of ten pots of each of six varieties were compared with the mean for 15 Kanred pots. The mean injury for the varieties together with the minimum temperature employed during each 12 hour freezing period is given in Table XIX. Several lots in which injury did not occur during the first freezing period are not included, even though injured in a second freezing, because it is felt that such data under the circumstances would only confuse the reader.

Table XIX. Average Lot Injury in Spring Freezing Trials

Date	Time of Freezing	Min. Temp. °F	Per Cent Injury: Kanred 2401	Oro 495	Kan. x Hd. Fed. 2627	Pre. x Kanred 2628	Kaw-vale 2593	Harvest Queen 19	Ful-caster 317	Currell 501
3-15	Day	-7.6	72	62	68	78	60			
"	Night	-7.6	75	73	73	75	71			
3-16	Day	-7.6	79	85	65	85	85			
3-21	Day	-7.6	92	92	97	97	89			
"	Night	8.6	72	70	89		62	84	72	97
3-27	Night	8.6	50	44	56		50	58	60	55
3-28	Day	3.2	73	77	73		80	90	86	98
4-6	Day	13.0	82	83	88		73	86	89	90
4-6	Night	17.6	68	67	72		67	66	62	77
4-14	Day	13.0	99	100	100		100	100	97	100
4-15	Day	15.5	86	77	81		80	93	89	94
4-16	Night	16.0	89	88	97		95	97	80	93
4-17	Day	15.5	84	80	81		86	81	69	72
Average, first four lots			79.5	78.0	75.8	83.8	76.5			
Average, last nine lots			78.1	76.2	81.9		77.0	83.9	78.2	86.2

Regarding the temperatures necessary to produce a desired degree of injury, it should be observed that they ranged from -7.6° F in early March to 16° F on April 16. This indicates a hardiness range about comparable with hardened material frozen on November 25, 1929, at the one extreme and less hardiness than was shown by the least hardy greenhouse plants reported in this paper on the other.

The results merit but little discussion inasmuch as the relative hardiness of one variety as compared with that of another showed no consistent relationship. This meant that neither hardiness nor loss of hardiness could be satisfactorily measured for the period. It seemed that the varieties tended to respond differently to the multitude of changing weather conditions during the spring. Even the first lots frozen are not in agreement with the relative hardiness studies reported elsewhere in this paper, so it is felt that even that early, the varieties were adapting themselves for growth in varying modes. Akerman (1) also concluded that spring measurements of hardiness are not satisfactory.

SUMMARY

Three groups of plants; (1) greenhouse grown continuously, (2) greenhouse grown until October 22 and exposed to outdoor conditions thereafter, and (3) field grown plants transplanted for freezing; were used in studying the effects of hardening, by exposure to low temperatures, on the relative cold resistance of winter wheats as measured by artificial refrigeration. The freezing tests were made in November and December.

The following varieties arranged in order of their average cold resistance in these studies were compared: Minhardi, Oro, Kawvale, Kanred, Prelude x Kanred 2628, Tenmarq, Cooperatorka, Harvest Queen, Kanred x Hard Federation 2627, Blackhull, Early Blackhull, Fulcaster, and Currell.

Relative cold resistance of the varieties was essentially the same in each of the groups at any given time.

Marked changes in the relative cold resistance of certain varieties were noted in comparing the early and late freezing lots. The most pronounced changes were observed in Kanred which ranked eighth and second respectively and Harvest Queen which ranked fourth and tenth respectively at the two extreme periods.

Exposure to low temperatures deemed necessary by Hill (1927) was not essential for a proper expression of the relative hardiness of Minhardi. This variety was more hardy than Kanred in all tests, irrespective of degree of hardening.

Changes in varietal relationships and the abnormal expressions of relative cold resistance sometimes observed in varietal comparisons are believed to result primarily from differences in time of seeding. The relation appears to be a function of time of the year rather than age of the plant.

Spring survival of potted plants was not in complete accord with artificial freezing results which suggests a continuous adjustment of varietal relationships throughout the winter.

Day versus night freezing trials did not show consistent differences in this experiment.

Early maturity and cold resistance are not generally associated in winter wheat. Prelude x Kanred 2628 appears to possess a highly desirable combination of these attributes.

Cold resistance is extremely variable after spring growth begins. Furthermore, the varietal relationships appear to be quite different from those observed during the late fall and winter.

Loss of hardiness under constant greenhouse temperatures was readily discernible in from 24 to 48 hours. This was previously observed by Bayles and Salmon (1928).

ACKNOWLEDGEMENT

The author wishes to express his sincere appreciation of the many and varied helps given by Professor S. C. Salmon in planning, executing, and summarizing the present problem. Perhaps the greatest among these, if they may be judged so, was the invaluable aid given in estimating freezing injury which, it is believed, added materially to the accuracy of the experiment.

Thanks are also due to Professor J. H. Parker for helpful suggestions at various times.

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