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禾穀類의 耐寒性 檢定技術에 關한 研究

趙在衍 · 제이 디. 헤이스
作物試驗場 · 웰쉬植物育種試驗場

Investigation on Techniques for Evaluating Hardiness to Low Temperature in Cereals

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摘 要

1. 生長溫度가 낮을 때에 耐寒性이 中程度에 屬하는 品種들은 耐寒性의 向上을 보였으나 耐寒性이 極히 强하거나 極히 弱한 品種들은 生長溫度의 影響을 크게 받지 않는 傾向을 보였다.

2. 冷却溫度가 높고 또 冷却期間이 比較的 짧았을 때에는 2°C 에서 보다 5°C 에서 硬化가 잘 되었다.

3. 日長은 硬化前이나 硬化中 別로 影響하지 않았다.

4. 土壤濕度가 높을수록 耐寒性은 低下되었다.

5. 窒素施用은 耐寒性을 增加시켰으며 植物體의 乾物重을 增加시켰다. 乾物重과 耐寒性間에는 高度의 相關關係가 認定되었다.

6. 硬化期間은 길수록 有利하지만 試驗便宜上 4日程度가 適當하다고 判斷되었다. 高溫下에서의 軟化는 不過 1日~4日以內에 끝남을 알 수 있었다.

7. 耐寒性檢定에 適合한 冷却條件은 −8°C 에서 24時間程度이였다.

8. 品種에 따라서는 冷却處理 直後 그 被害가 나타나기도 하지만 回復力을 가지는 品種도 있으므로 處理後 一週日間 正常溫度에 두었다가 同被害程度를 調査함이 妥當한 것으로 推定되었다.

9. 全體的으로 보아 CD 80과 83의 耐寒性이 가장 높았고 다음은 Cappelle과 Maris Otter의 順序이였다. 4個의 秋播燕麥과 Jufy I은 中程度로 區分되었으나 3個의 春播燕麥은 極히 弱하였다. Peniarth는 Maris Otter와 對等하였고 S 147은 秋播燕麥中 가장 弱한 傾向을 나타내었다.

10. 水溶性炭水化物含量이 耐寒性에 어느 程度 關與하고 있음은 事實이나 이 自體가 耐寒性을 調節하는 關鍵은 아님을 알 수 있었다.

INTRODUCTION

In some years winterkilling causes substantial yield losses of cereals and the improvement of winterhardiness is, therefore, an important aspect of breeding in these crops. Breeders in many countries have tackled this problem from different aspects with varying degrees of success. Coffman[4] found that winterkilling of oats differred greatly from one variety to another and Finkner[6] reported that the transgressive segregation for increased winterhardiness was observed from progeny of the oat hybrids. These reports demonstrate the possibility of improvement through the exploitation of winterhardy genotypes in breeding programmes. However, winterhardiness of plants is not a simple character but a complicated physiological phenomenon and its expression is largely dependent on environmental factors such as temperature, photoperiod, air and soil moisture content, soil types, altitude and degree of snow cover (Dexter[5], Levitt[9),10)] and Olien[12]). Furthermore the crown, the most critical region concerned with the survival of the plant, is encased in leaf sheath and buried to varying depths below the soil surface.

In any breeding programme for the improvement of winterhardiness, selection under the natural field conditions is desirable. However, many winters are unsuitable for giving reliable information on winterhardiness of breeding materials. Because of this, several artificial freezing techniques have been developed to substitute field observation with laboratory selection (Amirshahi and Patterson[1], Rodger et. al.[14], Andrew[2], Bingham and Jenkins[3], Marshall[11], and Kretschmer[8]). Although many factors affect the expression of winterhardiness, these techniques have been mainly based on hardiness of plants to low temperature stress. Nevertheless these techniques generally give results in good agreement with field survival scores.

The objective of this study was to develop a freezing technique for evaluating hardiness to low temperature in cereals such as rye, wheat, barley and oats at relatively young developmental stages, using controlled environment facilities. The effects of growing and hardening temperature and photoperiod, duration of hardening, soil moisture content, and freezing temperature and duration of freezing on frost injury, have been extensively studied together with various means of assessing results. In addition, the effect of dehardening by exposing the hardened plants to a high temperature for varying periods was examined. The changes in water-soluble carbohydrate content (WSC) were measured under several of the treatment regimes to investigate the relationship between hardiness and the accumulation of WSC in the plants.

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MATERIALS AND METHODS

Twelve cereal varieties consisting of two ryes(CD 80 and 83), one winter wheat(Cappelle), one spring wheat(Jufy I), one winter barley (Maris Otter), one spring barley(Zephyr), four winter oats (Grey Winter, Peniarth, S. 147 and 172) and two spring oats(Condor and Manod), were used in most of the experiments. Twenty seeds of each variety were sown in either 38×51×8cm wooden boxes or 13cm diameter plastic pots filled with John Innes No. 1 compost. In the fertilizer experiment, the soils blended in a ratio of 1 loam soil, 1 sand, 1 fine gravel and 1 peat with no added nutrients were used and the fertilizers applied along the seeding drills before sowing.

The plants were grown in the unheated glasshouse in which temperature ranged from 0°C at night to 13°C in the daytime, or in the controlled growth room. This period is defined as the prehardening regime. In the experiment on effect of soil moisture content, 500cc of water was uniformly applied to each pot before planting and twice, two days and four days after planting, respectively. And then they were subjected to the five different watering schemes; nonwatering, every day, every other day, every 4th day and every 8th day watering. The plants were hardened in the 2°C or 5°C growth room for the periods of seven to ten days except for the experiment on duration of hardening. All the growth rooms were maintained at a light intensity of about 10,000 lux and under 8-hour photoperiod except for the experiment on photoperiod. The hardened plants which were aged 30 to 53 days old, were frozen in the dark freezing room at −7 to −8°C for 24 hours except in the experiment on the freezing conditions. After freezing, the plants were allowed to thaw gradually in the 2°C growth room for seven to 24 hours and then they were transferred to the 15°C glasshouse or outdoors for the assessment of frost injury. In order to minimize the gradient of temperature in the freezing room, the positions of boxes were interchanged within replicates during the freezing period at certain interval, if as necessary. When the plastic pots were used they were put into 46×31×15cm wooden boxes filled with granular peat before freezing to restrict heat loss to the soil surface.

Frost injury was assessed at three different times, namely two days, eight days and 15 days after freezing, except for the experiment on the effects of fertilizers in which only the first and last assessments were taken. The assessment was based on a 0–5 scale (Bingham and Jenkins[3]) in which 0 stands for no damage at all while 5 indicates complete killing. The mean scores taken from about 20 plants per replicate were used in the statistical analysis.

The samples for measurement of water-soluble carbohydrate content of plants were taken before and after hardening. The plants were cut at ground level, kept at about −15°C in a deep freeze cabinet until being dried in a freeze-drier. The dried samples were ground and 0.2g used for analysis of water-soluble carbohydrate content by the modified Yemm-Willis' method[17]. The data were transformed into angular values before the statistical analysis.

EXPERIMENTAL RESULTS

1. Effects of Growing and Hardening Temperatures

In this experiment four of the 12 varieties after germination in a 15°C glasshouse were grown under combinations of the following regimes prior to freezing at −8.3°C for 24 hours: a) Early growth at 10°C in environment chamber; b) hardening for 10 days at 2°C and 5°C.

The mean frost injury taken from different assessments under the above treatments are presented in Table 1. Over all treatments Cd 83 was found to be the most hardy and Conder the least with Cappelle and S 172 equal and intermediate. However there was varietal interaction in response to prehardening temperatures. Condor and Cd 83 showed little change in damage but the hardiness of Cappelle and S 172 was significantly higher under the lower temperature regime. On the other hand, no statistically significant effect of hardening temperature on hardiness was obtained.

In general, the first assessment values(taken two days after freezing) differred markedly from either the second (nine days after) or the third (16 days after). However, a highly significant variety × assessment time interaction was also exhibited. Cappelle demonstrated a strong ability to recover from the injury throughout the assessment period while Condor did not. On the other hand, Cd 83 and S 172 recovered substantially at the time of the second reading but after that no further changes were noticed. The assessment time also affected the damage depending on growing temperatures. Namely, the plants grown at 20°C showed continuous recovery as the time extended but those grown at 10°C recovered only until nine days after freezing.

Water-soluble carbohydrate was also estimated on parallel sets of treated seedlings. The results are presented in Table 2. Before hardening, the lower growing temperature(10°C) was much favorable for accumulation of WSC in the hardy varieties but the nonhardy variety Condor exhibited a reverse effect. The water-soluble carbohydrate content of both Cd 83 and S 172, on the basis of over-all means, were about 11% and higher than Cappelle or Condor. During the period of hardening WSC increased remarkably in all treatments but the effect of temperature was very different from that before hardening. The plants grown at 20°C accumulated generally higher WSC compared with those grown at 10°C, especially two oat varieties demonstrated more clearly. The lower hardening temperature was further effective on increase in WSC in all of the four varieties than the higher temperature. Furthermore, there was a highly significant prehardening temperature × hardening temperature interaction. Cd 83, Cappelle and S 172 showed greater difference between 2°C and 5°C when pre-grown at 10°C rather than at 20°C while Condor demonstrated the reverse effect. No evident relationship between the over-all varietal means of frost injury and those of % WSC was found.

Table 1. Mean frost injury taken from three different assessments under combinations of two prehardening and hardening temperatures.

Variety	Prehardening 10°C		Prehardening 20°C		Varietal Mean				Over-all
	Hardening				Prehardening		Hardening		
	2°C	5°C	2°C	5°C	10°C	20°C	2°C	5°C	
Cd 83	3.27	3.27	3.67	3.63	3.27	3.65	3.47	3.45	3.46
Cappelle	3.27	3.34	4.54	4.47	3.31	4.51	3.91	3.91	3.91
S 172	3.35	3.56	4.55	4.37	3.46	4.46	3.95	3.97	3.96
Condor	4.83	4.94	4.92	5.00	4.89	4.96	4.88	4.97	4.92
Mean	3.68	3.78	4.42	4.37	3.73	4.40	4.05	4.08	4.06
LSD(0.05) between treatments within variety					0.42		0.43		
LSD(0.05) between over-all varietal means									0.32

Table 2. Percentage of water-soluble carbohydrate content under combinations of two prehardening and hardening temperatures.

Variety	Before Hardening 10°C	Before Hardening 20°C	Before Hardening Mean	After Hardening: Prehardening 10°C		Prehardening 20°C		Varietal Means				Over-all
				Hardening				Prehardening		Hardening		
				2°C	5°C	2°C	5°C	10°C	20°C	2°C	5°C	
Cd 83	12.8	9.9	11.4	20.1	12.4	19.0	16.4	16.3	17.7	19.6	14.4	17.0
Cappelle	9.9	7.2	8.6	17.3	9.9	14.9	12.5	13.6	13.7	16.1	11.2	13.7
S 172	12.4	9.6	11.0	18.9	11.9	21.3	16.5	15.4	18.9	20.1	14.2	17.2
Condor	7.4	9.8	8.6	11.3	7.4	22.6	13.2	9.4	17.9	17.0	10.3	13.6
Mean	10.6	9.1	9.9	16.9	10.4	19.5	14.7	13.7	17.1	18.2	12.5	15.4
LSD(0.05) between treatments within variety				2.0				1.9		1.5		
LSD(0.05) between varietal means			1.0									0.9

2. Effects of Hardening and Freezing Temperatures and Durations of Freezing

The varietal mean frost injury assessed 16 days after freezing subjected to two hardening temperatures, three freezing temperatures and four durations of freezing is shown in Table 3. In addition, the varietal means of the injury related to three different times for assessment under a series of the above treatments are also presented.

The hardening at 5°C was more effective than that at 2°C. It was more clearly expressed under the combinations of the higher temperature and shorter duration of freezing. However, the spring varieties did not show pronounced effect of hardening temperatures. Out of the 12 varieties, two rye and four winter oat varieties were hardened far better at 5°C than at 2°C.

In general, the lower temperature caused more severe injury than the higher temperature but there was no noticeable difference between −7.2°C and −8.9°C. Only Zephyr showed marked differences between three freezing temperatures. This variety withstood better freezing stress at −8.9°C than at −7.2°C, especially when frozen for six to 12 hours. A highly significant interaction between temperatures of hardening and freezing was found. The hardening at 5°C resulted in a more marked difference between −5.6°C and −7.2°C compared with that at 20°C. The longer the duration of freezing, the more severe the frost injury occurred regardless of varieties. Relatively smaller difference in frost injury between freezings for 6 and 12 hours was generally obtained. Especially when frozen at −5.6°C all the varieties did not show any significant difference between these two durations.

Based on the over-all means of all the treatments (the last column in Table 3), Cd 80 and 83 were the most hardy and Zephyr, Condor and Manod were nonhardy at all as expected. Cappelle was significantly hardier than Maris Otter and any winter oat variety. The hardiness of Maris Otter was the same as Peniarth but greater than the other winter oats. S172 and Grey Winter belonged to the same group in hardiness. S147 was less hardier than any other winter oat variety. Jufy I did not differ from either Grey Winter or S147 in hardiness.

The time interval from freezing to assessment of frost injury seemed quite important as described in the first section. There was a highly significant difference among the assessment values obtained from the first, the second and the third dates of scoring, especially when freezing conditions were relatively more severe. Two wheat varieties and S 147 showed a strong ability to recover from the

Table 3. The varietal means of frost injury obtained 16 days after freezing in relation to hardening and freezing temperatures and durations of freezing and the mean scores taken at three different times.

Varieties	Hardening Temperature		Freezing Temperature			Freezing Duration				Assessment Time			
	20°C	5°C	−5.6°C	−7.2°C	−8.9°C	6hr	12hr	24hr	48hr	1st	2nd	3rd	Mean
Cd 80	2.24	1.40	1.10	2.14	2.23	1.09	1.47	2.12	2.61	1.75	1.79	1.82	1.79
Cd 83	2.33	1.51	1.15	2.23	2.38	1.32	1.53	2.23	2.60	1.83	1.90	1.92	1.88
Cappelle	2.87	2.58	2.27	2.98	2.94	1.81	2.51	2.91	3.68	2.98	2.74	2.73	2.82
Maris Otter	3.07	2.77	2.40	3.17	3.19	2.02	2.58	3.03	4.04	3.04	2.85	2.92	2.94
Peniarth	3.42	2.94	2.69	3.31	3.53	1.97	2.63	3.39	4.72	.842	3.07	3.18	3.03
S 172	3.53	3.01	2.73	3.40	3.68	2.14	2.57	3.56	4.82	3.11	3.20	3.27	3.19
Grey Winter	3.59	3.01	2.90	3.40	3.60	2.19	2.73	3.44	4.83	3.26	3.25	3.30	3.27
S 147	3.66	3.12	3.05	3.49	3.62	2.11	2.93	3.69	4.81	3.45	3.39	3.39	3.41
Jufy I	3.30	3.13	2.72	3.60	3.32	2.47	2.93	3.26	4.19	3.56	3.20	3.21	3.32
Zephyr	4.13	4.16	3.69	4.58	4.18	3.26	3.98	4.43	4.92	4.19	4.09	4.15	4.14
Condor	4.27	4.13	3.94	4.32	4.35	3.16	3.87	4.79	4.99	4.23	4.16	4.20	4.20
Manod	4.29	4.31	3.93	4.54	4.43	3.43	3.99	4.77	4.99	4.28	4.24	4.30	4.27
Mean	3.39	3.01	2.71	3.43	3.45	2.25	2.81	3.47	4.27	3.21	3.16	3.20	3.19
LSD (0.05) between treatments within variety	0.12		0.34			0.39				0.06		Between 2 over-all varietal means=0.12	
LSD (0.05) between varieties within treatment	0.17		0.21			0.24				0.16			

damage nine days after freezing while S 172 and Peniarth increased continuously the damage until the time of the final reading. Grey Winter remained relatively constantly throughout the assessment period. Maris Otter recovered slowly the damage at the second assessment but got increased at the third reading. Cd 80 and 83 increased slowly partial damage of the leaf tissues until the final reading. Lastly, the spring varieties recovered a little at the second assessment but returned to the initial degree of the damage.

3. Effects of Durations of Hardening

In order to find the proper duration of hardening, six durations; 12-hour, 1-, 2-, 4, and 8-day treat ments including the control were given to Cd 83, Cappelle, Maris Otter and Peniarth. The final assessment values of frost injury taken 16 days after freezing are only explained in this section since effect of the time interval was in general agreement with the previous sections.

All the winter cereals increased the hardiness as duration of hardening extended. The hardenings for four and eight days resulted in a marked increase in hardiness compared with the other treatments. However, no significant difference between these two durations of hardening was found. The hardening for less than four days did not give a sufficient result. A very similar result was also obtained from the preliminary experiment in which duration of hardening extended up to 16 days. Cd 83 was highly superior in hardiness over the other three varieties. The hardiness of Cappelle was greatly higher than that of Peniarth but slightly greater than that of Maris Otter. No statistical difference between Maris Otter and Peniarth was observed although the former was a little hardier than the latter. There was no noticeable variety x duration of hardening interaction.

Water-soluble carbohydrate content also increased continuously as duration of hardening extended regardless of varieties. All the hardening treatments were highly significantly different in % WSC each other except for between the hardenings for 12 and 24 hours. Cd 83 and Peniarth produced almost the same % WSC but the former increased greatly it when hardened for eight days compared with the latter. Similarly, Cappelle and Maris Otter gave an equal level of % WSC. The variety x duration of hardening interaction in change of % WSC was insignificant. A highly significant negative linear regression of frost injury of % WSC in all of four varieties subjected to six different durations of hardening, was found (Figure 1).

4. Effect of Photoperiod during Growing and Hardening

A series of two different photoperiods, 8- and 16-hour was given to Cd 83, Cappelle, S 172 and Condor during pre-hardending as well as hardening periods.

Effect of photoperiod on frost injury of cereals was not recognized. Hewever, there was an increasing tendency of frost injury when pre-hardened under the longer photoperiod in all of the four varieties. Cd 83 increased the hardiness a little when hardened under 8-hour photoperiod while the other varieties showed marginal increase under 16-hour photoperiod. Cd 83 was the most hardy and followed by Cappelle, S 172 and Condor. Cd 83 did not differ statistically from Cappelle. This might be due to relatively severe conditions of freezing in this experiment.

The statistical analysis of % WSC data also revealed that photoperiod during growing and

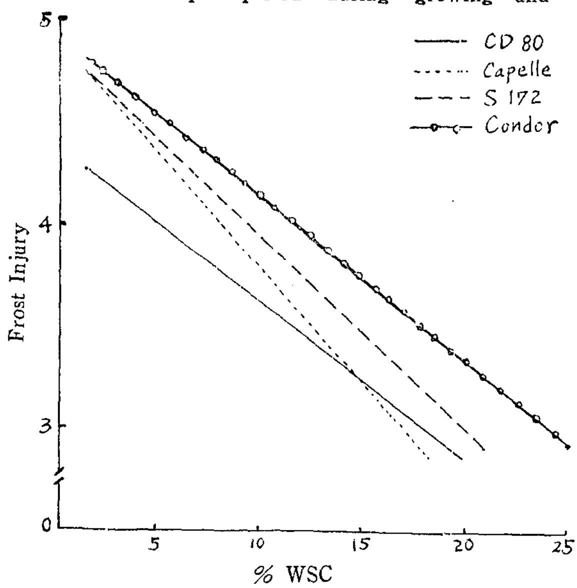

Fig. 1. Regression of frost injury on percentage of water-soluble carbohydrate content related to different durations of hardening in cereals.

hardening did not affect significantly % WSC. Nevertheless, the shorter photoperiod during prehardening seemed more favorable for increase in WSC than the longer one. Cappelle had significantly lower % WSC before being hardened than the other three varieties but the latters did not differ each other. On the average, WSC increased during hardening period by as much as about 11%. The photoperiod during hardening affected % WSC in very different way from that during growing. The 16-hour photoperiod increased %WSC only when prehardened under the longer photoperiod while 8-hour photoperiod did similarly when prehardened under the shorter photoperiod. It was surprising to find that Cappelle increased exceptionally % WSC under 16-hour photoperiod even though it was grown under the shorter photoperiod.

No clear relationship between the hardiness and

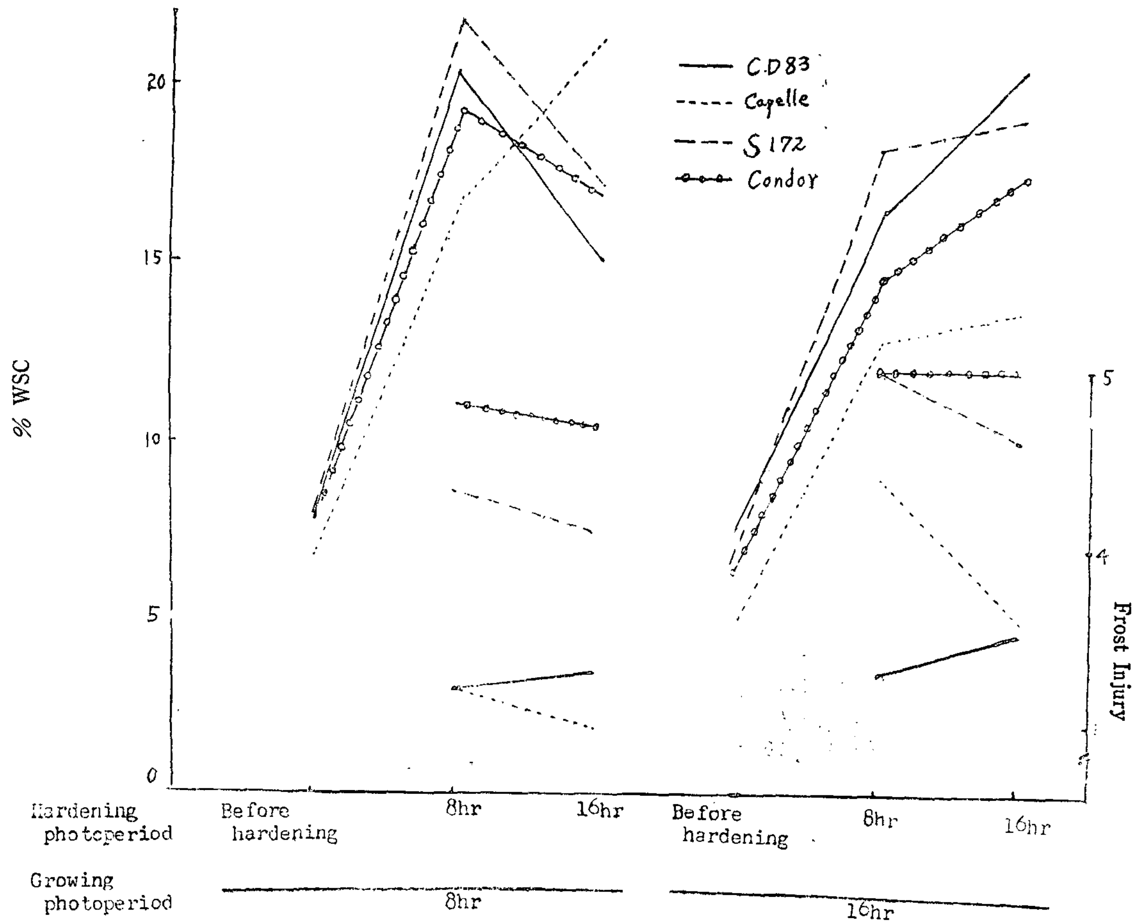

Fig. 2. Frost Injury and %WSC related to different photoperiods during growing and hardening in cereals.

%WSC when subjected to different photoperiods, was detected (Figure 2). In case of pre-growing under 8-hour photoperiod, cappelle and Cd 83 showed a positive relationship between these two values but two oat varieties produced a negative one. On the other hand, Cd 83 gave a negative relationship while Cappelle and S172 a positive one, when given 16-hour photoperiod during prehardening. This experiment is so premature to conclude effect of photoperiod on frost injury and WSC that further intensive studies are required.

5. Effect of Fertilizers

Four following levels of fertilizers were applied to the varieties: a nitrogen at a rate of 168kg/ha(N), b) nitrogen at 168kg/ha+phosphorus at 560kg/ha +potassiumat 560kg/ha(NPK), c) phosphorus and potassium 560kg/ha each (PK), and d) the control (No).

Because of the temperature gradient in the freezing room, one replicate differred considerably from another. The mean effects of the fertilizer treatments on frost injury and % dry matter (%DM) of the 37-day old seedlings sown at two different dates, are presented in Table 4.

Nitrogen application resulted in a marked decrease of frost injury compared with the other fertilizer treatments including the control plot. The effect of nitrogen depended upon variety. Two rye, Maris Otter and the nonhardy varieties did not give any effect of nitrogen but Cappelle, four winter oat varieties and Jufy I did. However, no significant difference between the N and NPK treatments was found in case of Peniarth, although the former was numerically effective for hardening than the other two treatments.

The percentage of dry matter taken from two

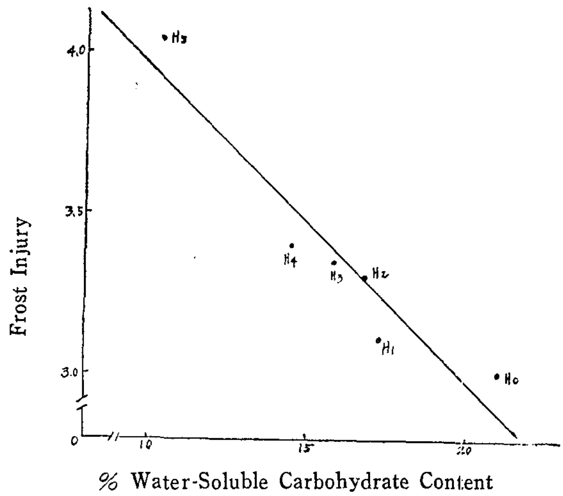

Fig. 3. Regression of mean frost injury on mean % dry matters of the seedlings in 12 *varieties in relation to the fertilizer* treatments. Numbers stand for varieties listed in Table 2.

Table 4. Mean effects of nitrogen (168kg/ha), phosphorus (560kg/ha) and potassium(560 kg/ha) application on frost injury and % dry matters of the 37-day old seedings in 12 cereal varieties sown at two different dates.

Varieties	Frost Injury					% Dry Matters				
	NPK	PK	N	No	Mean	NPK	PK	N	No	Mean
1. Cd 80	0.54	0.48	0.40	1.19	0.65	15.38	14.69	16.53	14.85	15.36
2. Cd 83	0.61	0.63	0.56	1.35	0.79	14.91	15.16	15.69	14.74	15.13
3. Cappelle	3.06	2.79	1.85	2.79	2.62	16.34	15.99	16.84	16.39	16.39
4. Maris Otter	4.16	4.06	3.60	4.44	4.07	13.23	12.34	13.24	12.32	12.78
5. Peniarth	4.03	4.30	3.18	4.18	3.92	14.95	14.16	14.90	13.87	14.47
6. S 172	3.91	4.09	2.95	4.06	3.75	14.40	14.31	15.44	14.72	14.72
7. Grey Winter	4.18	4.05	3.15	4.03	3.85	14.35	14.28	15.09	14.30	14.50
8. S 147	4.10	4.11	2.95	3.99	3.79	14.04	14.55	14.76	13.95	14.33
9. Jufy I	4.25	4.40	3.55	4.38	4.15	14.31	13.83	15.10	14.61	14.46
10. Zephyr	5.00	4.99	4.91	4.95	4.96	11.64	11.76	11.93	10.87	11.55
11. Condor	5.00	5.00	5.00	5.00	5.00	11.94	11.34	11.91	11.74	11.73
12. Manod	4.99	4.99	4.99	4.93	4.98	11.08	10.68	11.51	10.75	11.01
Mean	3.65	3.66	3.09	3.77	3.54	13.88	13.59	14.41	13.59	13.87
LSD(0.05)	Between treatments within variety=0.98 Between varieties within treatment=0.54 Between over-all varietal means=0.27					Between treatments within variety=1.20 Between varieties within treatment=0.95 Between over-all varietal means=0.47				

seedlings each increased greatly when nitrogen was applied. Nitrogen effect on %DM was highly significantly greater than any other treatments. However, the others did not show any effect. There was a distinguishable varietal difference in %DM on the basis of over-all means. According to the Duncan's new multiple range test, Cappelle produced the highest %DM. Two rye, four winter oat varieties and Jufy I belonged to the second group but S147 was the lowest among them so that it was distinguished from Cd 80. Maris Otter was the intermediate and Condor, Zephyr and Manod were classified as the lowest group.

The nitrogen effect on increase of %DM was highly negatively correlated with that on frost injury. Similarly, a highly significant negative linear regression of the varietal frost injuries on the corresponding %DM, was obtained (Figure 3).

6. Effect of Soil Moisture Content

The higher the soil moisture content, the greater the frost injury occurred. The plants without watering showed greater hardiness than the other four treatments. On the contrary, the plants watered every day were more severely damaged as compared with the other treatments. The magnitude of frost injury due to different watering schemes was more pronounced when hardened at 2°C rather than at 5°C. A highly significant positive linear regression of frost injury on % soil moisture content expressed as % oven-dry weight, was observed (Figure 4).

7. Effect of Duration of Exposure to High Temperature after Hardening

The following two sets of treatments were applied: a) the plants of Cd 83 Cappelle, Maris Otter and Peniarth which were hardened for seven days were exposed to 15°C for six different durations, 0, 6, 12, 24, 48 and 96 hours; b) Peniarth and S 147 hardened also for seven days were exposed to 20°C for four different durations, 0, 24, 48 and 72 hours.

All the varieties decreased the hardiness as duration of the exposure extended prior to freezing. The four-day exposure made plants dehardened markedly compared with the other exposure treatments. The two-day exposure resulted in significantly higher damage than the control. Based on the over-all varietal means, Cd 83 was the most hardy followed by Cappelle, Maris Otter and Peniarth. However, there were no statistical differences between Cappelle and Maris Otter and between Maris Otter and Peniarth. Dehardening of

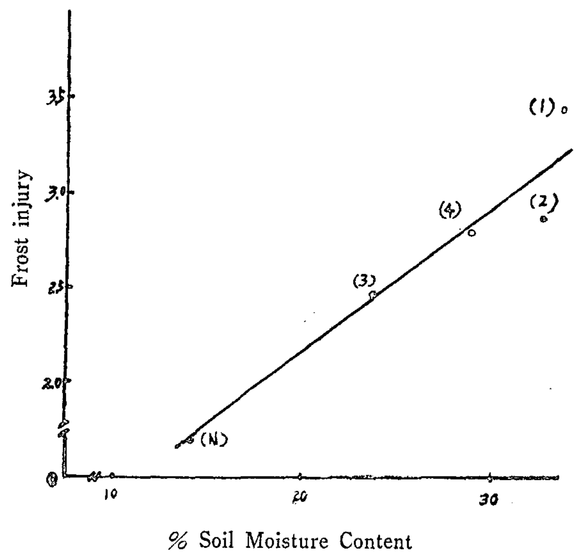

Fig. 4. Regression of mean mrost injury on mean soil moisture content expressed as % oven-dry weight in Peniarth hardened at 2°C and 5°C.

* (N), (1), (2), (4) and (8) indicate non-, every day, every other day, every 4th day and every 8th day waterings, respectively.

the plants took place rapidly when exposed to the relatively higher temperature, 20°C. Even one-day exposure brought about significantly higher damage than the control treatment.

Water-soluble carbohydrate content in the plants decreased gradually as the duration prolonged. Exposures for from six hours to four days caused a significant decrease in % WSC as compared with the control. However, no noticeable differences were obtained among the treatments exposed for six hours to two days. As a result, only the four-day exposure gave a significantly lower WSC than the other treatments. All the varieties demonstrated a similar change in % WSC under the treatments. Cappelle produced the lowest % WSC among four varieties and Peniarth did the highest. No marked differences between Peniarth and Maris Otter and between Maris Otter and Cd 83 were found. As a whole, frost injury was highly significantly correlated with changes in % WSC of the plants subjected to the different durations of exposure to high temperature (Figure 5). The magnitude of negative regression between these two values was considerably different from variety to variety. Cd 83 and Cappelle showed closer relationship than either Maris Otter or Peniarth.

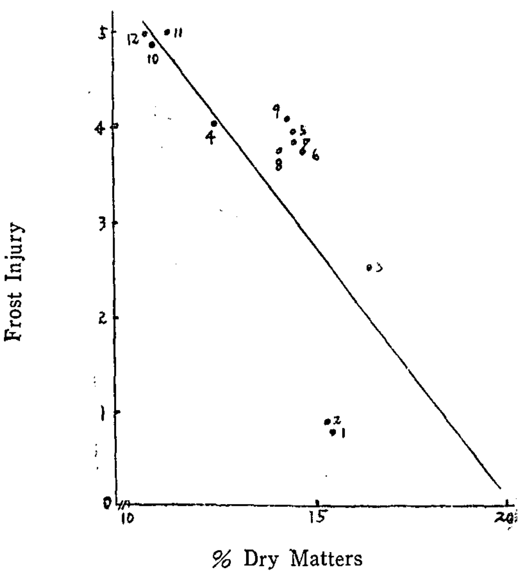

Fig. 5. Regression of mean frost injury on mean %WSC in four different varieties subjected to six different durations of exposure to 15°C.

* H_0, H_1, H_2, H_3, H_4 and H_5 represent non-, 6hr-, 12 hr-, 24 hr-, 48 hr- and 96 hr-exposures, respectively.

DISCUSSION

As a whole, Cd 80 and 83 were the most hardy followed by Cappelle and Maris Otter. Four winter oat varieties and Jufy I belonged to the intermdiate group while Condor, Manod and Zephyr were nonhardy at all. The hardiness of Maris Otter was almost equal to that of Peniarth, the most hardy winter oat variety used in this study. Out of the four winter oat varieties, S147 was the least hardy. It is interesting to know that wheat varieties behaved to freezing stress quite differently from other cereal varieties. Namely, Cappelle produced the highest %DM but showed relatively lower % WSC in spite of the higher hardiness. Jufy I was comparable with winter oat varieties in hardiness although the former has spring nature.

1. Prehardening Conditions

It was confirmed that temperature during prehardening is one of the most important environmental factors affecting hardiness of winter cereals. Relatively lower temperature promoted the hardiness and vice versa. However, the response to temperature varied from one variety to another. The extremely hardy or nonhardy varieties were not so much affected as the intermediate varieties in hardiness. On the contrary, effect of photoperiod during prehardening on frost injury of the plants was not statistically significant although the shorter photoperiod seemed a little favorable for hardening than the longer one. These results are in good agreement with Paulsen's result[13] except the fact that decreasing photoperiod resulted in decreased plant hardiness. This disagreement in effect of photoperiod on hardening might be attributed to genetic difference of the materials and difference in the freezing techniques employed. The varietal differences in response to photoperiod related to hardiness was reported by Hodgson[7] in alfalfa.

No doubt, soil moisture content affected plant hardiness to the considerable extent as reported by Tysdal[16], and Amirshahi and Patterson[1]. The lower the soil moisture content, the lower the frost damage occurred. The plants without watering for 39 days before freezing showed markedly higher hardiness than the other treatments while the plants which watered every day brought about significantly more severe injury. Therefore, it appears apparent that uniform control of soil moisture content is necessary in conducting any freezing experiment in order to obtain possible precise information. The fact that hardening at 2°C gave more pronounced differences among the different watering treatments than that at 5°C might be attributed to a relatively faster evaporation of water at the latter than at the former.

Application of nitrogen promoted greatly hardiness of the plants and increased % DM but the other fertilizer treatments such as NPK and PK did not have any evident effect. The effect of nitrogen could be explained with increase of total free amino nitrogen content as related to cold hardiness (Zech and Pauli[18]). The fact that the NPK treatment did not give the same effect as nitrogen might be resulted from chemical interaction among fertilizer elements which made plants at young growing stages difficult to absorbe them. However, the nitrogen effect depended upon varieties: two wheat and four winter oat varieties responded markedly to nitrogen application but two rye, two barley and two spring oat varieties did not. There was a distinguishable varietal difference in %DM on the basis of over-all means. These values were so significantly negatively correlated with the varietal mean frost injury that %DM of plants could be effectively used an easy criterion for evaluating the hardiness of plants, provided that further investigation confirm this relationship.

2. Hardening Conditions

Winter cereals increased gradually hardiness as duration of hardening extended to eight days. A similar result was obtained from the preliminary experiment in which the duration extended up to 16 days. From point of view of limited space in growth rooms and glasshouses, hardening for four days might be enough to acquire satisfactory hardiness before freezing. This result disagrees with the result of Amirshahi and Patterson[1] which revealed that one-day hardening was sufficient in winter oats. All the varieties decreased gradually the hardiness as duration of exposure to 15°C after hardening extended. The four-day exposure resulted in significantly higher damage compared the 0-, 6-, 12-, 24- and 48-hour exposures. Dehardening of the plants so rapidly took place when exposed to 20°C that even 24-hour exposure lowered greatly the hardiness. In general, hardening at 5°C was more effective on hardening than that at 2°C, especially when frozen at relatively higher temperature and for relatively shorter period. However, there was no statistical difference between these two hardening temperatures when followed by freezing at about −8°C for 24 hours. It seemed that photoperiod during hardening is not critical factor involved in hardiness since it did not show any noticeable effect.

3. Freezing Conditions

The lower the freezing temperature, the higher the frost injury was resulted in. However, between −7.2°C and −8.9°C there was no difference in frost damage of the plants. The longer the duration of freezing, the more severe the frost injury occurred. Between freezings for 6 and 12 hours, no marked difference was generally found. It seemed that the freezing for 6 to 12 hours was not sufficiently enough to damage the plant crowns grown in the 8 cm-deep wooden flat. The extremely hardy variety could withstand well even when frozen for 48 hours while the nonhardy varieties were almost killed under both the freezing stressess.

Based on the above results, the freezing at about −8°C for 24 hours is considered the most proper for evaluating winterhardiness of cereals under this controlled environment.

4. Time for Assessment of Frost Injury

It was observed that assessment time of frost injury after freezing affected degree of the hardiness to the considerable extent. Varietal response to time interval from freezing to scoring were different from variety to variety. As shown in Table 3, two wheat varieties exhibited strong ability to recover the damage at the time of the second reading compared with the first reading value taken two days after freezing. On the other hand, Peniarth and S 172 were significantly getting increased the damage as the interval prolonged. Effect of the time interval on frost injury was more clearly expressed when freezing conditions were relatively severe. It is assumed that assessment of frost injury immediately after thawing reflected hardiness of leaf tissues and the later assessment accounted for recovering ability of whole plant. It seemed apparent that it is necessary to allow plants to recover for at least one week before assessing the damage.

5. Relationship between Frost Injury and %WSC

The lower prehardening temperature was more effective on accumulation of %WSC in the hardy varieties but nonhardy variety showed a complete reverse effect. Photoperiod during prehardening did not affect change in %WSC although the shorter photoperiod seemed a little favorable for increase of %WSC. This result taken before hardening was good in agreement with the corresponding frost injury. However, after hardening the patter of effect changed. Plants grown at the higher temperature, 20°C produced more %WSC during hardening than those grown at 10°C. The lower hardening temperature appeared to be more effective on increase in %WSC than the higher one. The effect of hardening temperature was more clearly demonstrated when plants were grown at 10°C than at 20°C. Percentage of water-soluble carbohydrate content increased generally when the same photoperiod was given throughout prehardening and hardening. Highly significant negative regressions of frost injury on %WSC related to different durations of hardening and dehardening, were obtained. The evidence leaves no doubt that %WSC gradually increases during hardening and decreases during dehardening. Nevertheless, no significant parallelism between varietal hardiness and the corresponding %WSC was demonstrated. Therefore, it could be concluded that WSC in plants is associated with the hardiness to some extents but WSC is not primary factor affecting the hardiness. This conclusion completely agrees with the finding of Steponkus and Lanphear[15] which discussed the relationship between the total sugar content and cold hardiness in English ivy.

SUMMARY

1. The relatively lower prehardening temperature was more effective on increase of the hardiness of the intermediately hardy varieties than the higher one but either the extremely hardy or nonhardy varieties did not respond to the temperature as much as the intermediate types.
2. Five degree Centigrade was generally more favorable than 2°C on hardening of the plants, especially when frozen at higher temperature for shorter duration.
3. It appears that photoperiod during prehardening and hardening did not play so important role as temperature on the hardiness.
4. The higher the soil moisture content, the higher the frost injury occurred.
5. Application of nitrogen increased markedly the hardiness and % DM of the plants. Percentage of dry matter of young seedlings might be used as an easy and rough criterion for evaluating hardiness since there was a highly significant regression of varietal frost injury on the %DM.
6. Four days appeared to be enough for hardening of plants although the plants increased gradually the hardiness as duration of hardening extended. Dehardening of the plants at relatively higher temperature took place rapidly within one to four days.
7. Under this controlled environment, freezing at about −8°C for 24 hours seemed the best for the purpose of evaluating the hardiness to low temperature.
8. It is believed that assessment of frost injury should be done at least one week after freezing. Some varieties showed strong ability to recover from the damage as recovery period was extendeded.
9. As a whole, Cd 80 and 83 were the most hardy and followed by Cappelle and Maris Otter. Four. winter oats varieties and Jufy I belonged to the intermediate type while the other three spring varieties were nonhardy at all. Peniarth was comparable with Maris Otter in hardiness. S 147 appeared the least hardy among the winter oats varieties.
10. It is evident that water-soluble carbohydrate content is associated with the hardiness to some extent but not primary factor involved in hardiness.

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