

# DIFFERENCES IN RESISTANCE TO LOW TEMPERATURES SHOWN BY CLOVER VARIETIES

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(WITH TWO FIGURES)

## Introduction

Differences shown by clover varieties in ability to withstand low temperatures are of great economic importance in the northern part of the United States. In this region the relative hardiness of a variety will often determine whether or not it is suitable for a given locality and whether or not it will endure average winter temperatures. The temperature limits for many crop plants can be determined best by field trials. Under field conditions, however, it is impossible to regulate or to control many conditions under which such tests are carried out and consequently the importance of a number of factors cannot be determined. Temperature is one of these factors. By mechanical control of temperatures it is possible to determine the response of varieties to different temperatures and the conditions under which the freezing of plant tissue occurs.

The aim of this work was to determine if relative hardiness of clover varieties could be determined under conditions where temperature could be controlled. Tests were carried out at different stages of growth with a number of varieties of clover and a few varieties of alfalfa. Differences in exposure of plants were eliminated as far as possible by air circulation. Care was taken to obtain uniform conditions during periods of hardening and exposure to low temperature. The following points were considered in this group of tests: (1) Effect of low temperature on germination of clover seeds before planting; (2) Effect of low temperatures on the germination of seeds which had imbibed various amounts of water; (3) Effects of low temperatures on plants of different ages; (4) Differences in resistance to low temperatures shown by a number of varieties in the seedling and mature stages of growth.

## Results

### EFFECT OF LOW TEMPERATURES ON THE VIABILITY OF CLOVER SEEDS

Clover seeds which had been exposed to laboratory conditions and had a water content of less than 15 per cent. by weight were subjected to temperatures ranging from 0 to  $-48^{\circ}$  C. for 30 minutes. Germination tests before and after exposure to the lowest temperature were made, but showed no loss of viability from freezing. Dry seeds or those having low moisture

content are not injured by low temperatures ordinarily obtainable in the laboratory.

Low temperatures showed more harmful effects on the viability of clover seeds with higher moisture content. Seeds of Minnesota Medium Red clover with a germination of 90 per cent. were placed in distilled water for three to four hours at 37° C. At the end of that time they had imbibed approximately 90 per cent. of water, calculated on the dry weight basis. The results in table I show the percentage of germination after exposure to temperatures ranging from 0° C. to — 40° C.

TABLE I

INFLUENCE OF LOW TEMPERATURE ON GERMINATION OF MOIST CLOVER SEEDS

TEMPERATURE IN DEGREES C.	GERMINATION AFTER EXPOSURE IN PER CENT.
0	85.0
- 2	74.0
- 4	53.0
- 6	33.0
- 8	26.0
-10	9.0
-15	8.5
-20	5.5
-25	4.0
-30	0.0
-40	0.0

In another trial fully imbibed seeds were placed over a desiccating agent ( $H_2SO_4$ ) of various known concentrations and the moisture partially removed. These were then exposed to various temperatures for 30 minutes and germination tests again determined. Germination fell off rapidly when the moisture content became greater than 15 per cent., as is shown by table II.

TABLE II

INFLUENCE OF MOISTURE CONTENT ON INJURY BY FREEZING

MOISTURE CONTENT IN PERCENTAGE	GERMINATION PERCENTAGE AFTER EXPOSURE FOR 30 MINUTES TO			
	- 10° C.	- 20° C.	- 30° C.	- 40° C.
89.75	9.0	5.5	0.0	0.0
25.	85.0	43.0	10.5	9.0
15.	85.0	55.0	50.0	43.0
13.	90.0	90.0	90.0	90.0
12.	90.0	90.0	90.0	90.0
10.	90.0	90.0	90.0	90.0

In fig. 1 are shown the effects of varying moisture contents on the killing points of clover seedlings. It is evident that as soon as the seed begins to imbibe moisture, it becomes much less resistant to low temperature.

#### EFFECT OF LOW TEMPERATURES ON PLANTS OF DIFFERENT AGES

The apparatus used for hardening and freezing seedling and mature plants consisted of a constant temperature chamber with electrical control. Low temperatures were secured by refrigeration and could be controlled within a range of .5 degrees C. Equal temperatures throughout the chamber are secured by means of an electric fan. Etiolation of the plants was prevented by the use of artificial light over the hardening chambers.

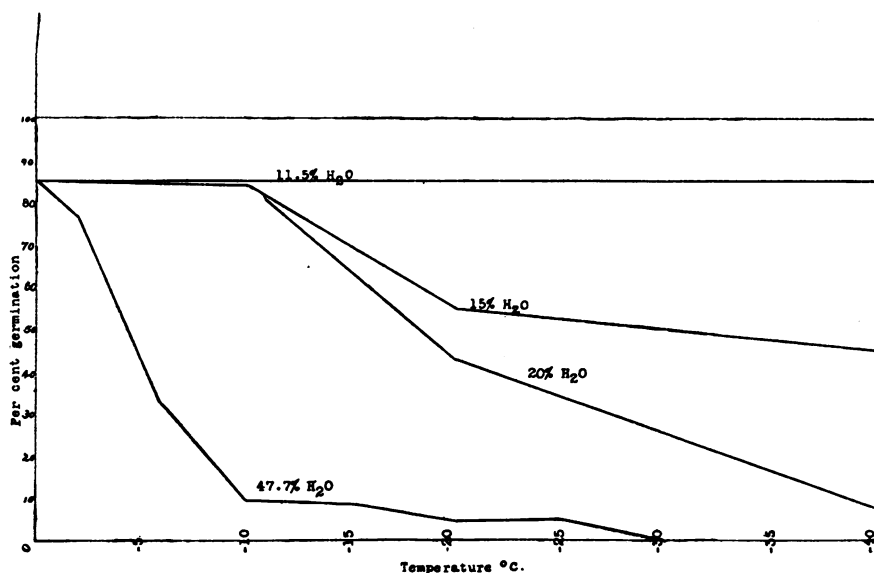


FIG. 1. Killing temperatures of clover seed (Minnesota Med. Red) with different moisture contents.

The necessity of having the plants for a given test at the same age is apparent, as there is considerable variation in the resistance to low temperatures of plants of the same variety at different ages. Difference in this respect is indicated by comparing plants of Minnesota Medium Red clover at various ages when exposed to low temperature, table III and fig. 2.

The plants seem quite resistant when emerging in the seedling stage and are most susceptible to frost injury when about three weeks old at the time of forming the first pair of permanent leaves.

Placing the plants at temperatures slightly above 0° C. for one or two days before freezing increased to a marked extent the ability of the plants

TABLE III  
AGE AND RESISTANCE TO FREEZING

AGE	PERCENTAGE OF PLANTS KILLED AT $-3^{\circ}\text{C}$ .
7 days	0.0
10 "	17.0
14 "	46.6
21 "	100.0
90 "	0.0

to endure freezing temperatures, growth during this hardening period being materially checked. By so treating the plants it was possible to use lower temperatures and accordingly bring out more striking differences in cold resistance and in the rate of acquiring hardness.

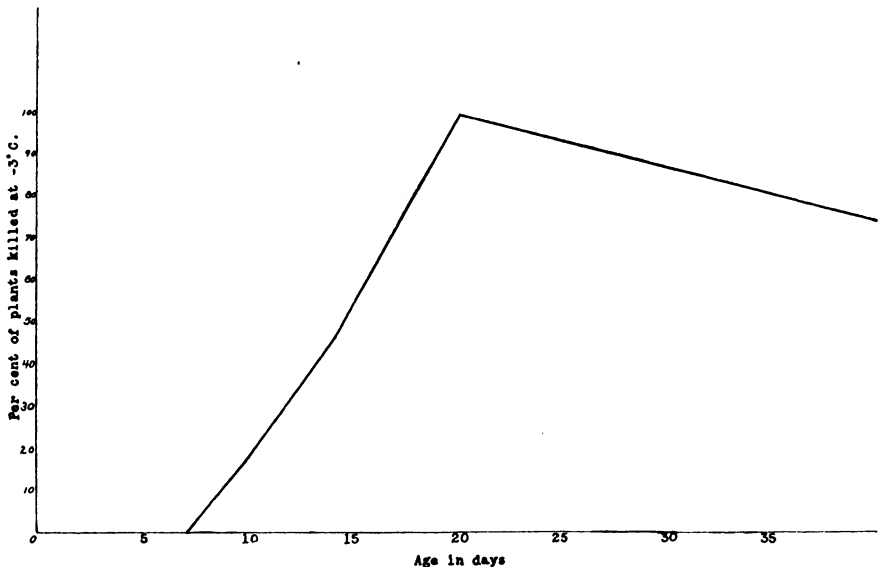


FIG. 2. Variations in low temperature resistance by clover plants of different ages (Minnesota Med. Red).

#### DIFFERENCE IN LOW TEMPERATURE RESISTANCE AMONG VARIETIES

Results obtained by hardening clover seedlings and then subjecting them to freezing temperatures showed considerable difference in cold resistance among the common varieties. Seeds were obtained through the courtesy of Dr. A. J. PIETERS, of the Office of Forage Crop Investigations, Bureau of Plant Industry. Only four varieties were used to simplify the trial. Duplicate tests gave results as shown in table IV.

TABLE IV  
VARIETAL RESISTANCE OF CLOVERS TO FREEZING

VARIETY	PERCENTAGE ALIVE AFTER EXPOSURE TO			
	-4° C.	-5° C.	-6° C.	-7° C.
Altaswede .....	100	100	85	30
Michigan .....	97	94	83	30
French 2213 .....	86	80	52	19
Italian 54779 .....	92	64	70	15

The seedlings were seven days old when hardened. Freezing was observed at higher temperatures when the plants were in contact with water. Freezing of seedlings in vials showed injury to occur first on the root end of the seedling.

With more mature plants the results were similar to those obtained in the seedlings. In general the northern varieties were more resistant and responded more rapidly to hardening. In tables V and VI are given data from two of many sets of clovers showing the approximate percentage of killing. The plants used were three weeks old and hardened for about 48 hours at 2° C. before trial.

TABLE V  
VARIETAL RESISTANCE OF CLOVERS TO FREEZING

VARIETY	PERCENTAGE KILLED AT -4° C.
Minnesota Med. Red Clover .....	50.
Ohio Red 1687 .....	60.
French Red 1233 .....	60.
French Red 1247 .....	65.
Bohemian 1250 .....	80.
Idaho Red 1287 .....	92.
Hungarian Red 1237 .....	95.
Chilean Red 1285 (one plant in 25 unin- jured) .....	96.
French Red 1291 .....	100.
Australian 1655 .....	100.

In all cases where plants were not completely killed the injury was evident first on the outer leaves, whereas crowns were less easily injured.

**TABLE VI**  
**VARIETAL RESISTANCE OF CLOVERS TO FREEZING**

VARIETY	PERCENTAGE KILLED AT $-4^{\circ}$ C.
Minnesota Med. Red Clover .....	40.
Oregon Red 2148 .....	50.
French Red 1294 .....	55.
Ohio Red 1687 .....	60.
Idaho Red 1287 .....	65.
Chilean Red 1239 .....	80.
Chilean Red 1605 .....	93.
Tennessee .....	95.
Australian Red 1655 .....	100.
French Red 1291 .....	100.
Bohemian Red 1250 .....	100.
Chilean Red 1285 .....	100.
Hungarian 1237 .....	100.

### Summary

1. The viability of clover seeds containing a moisture content of less than 15 per cent. of the dry weight is unaffected by temperatures as low as  $-48^{\circ}$  C. for short periods.
2. The germination of clover seeds with high moisture content is greatly impaired by low temperatures, falling off very rapidly in seeds with a moisture content above 25–30 per cent.
3. Clover plants of the same variety vary with age in respect to killing temperatures. Susceptibility to injury is most pronounced at about three weeks of age, or at the time of formation of the first pair of permanent leaves.
4. There is considerable difference among varieties in ability to withstand low temperature. In general European or southern varieties proved less resistant than those grown in northern areas of the United States. This variation was present both in the seedling and mature plants.

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