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Low Temperature Tolerance of *Panax quinquefolium*

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Abstract : One exotherm was detected in the intact ginseng seeds containing more than 35% water, but in seeds with 20% there was no exotherm. The shapes of exotherm were remarkably uniform without relation to water content above 35%. The temperature at the initiation of freezing varied from −3.5°C to −9.6°C with the different water content in the seeds, and the initial temperature of freezing delayed with the decrease of water content. The resistance damage at low temperature appeared in order of main body, rhizome, lateral root of 3-year-old yearling rhizome, and fine root of 3-year-old. Ginseng roots didn't receive any damage at −5°C for 24 hours. Otherwise they received serious damage below −10°C even for 5 hours' exposure. Hence, alternative low temperature gave more severe damage compared to constant low temperature. This result suggests that the possibillity of receiving injury at low temperature was higher during the thawing season of the early spring than in the winter.

Key words : *Panax quinquefolium*, seeds, root, exotherm, freezing.

Introduction

American ginseng (*Panax quinquefolium* L.) is native to southern Canada and survives low winter temperatures under natural mulches in deciduous woodlands and under applied mulches in cultivation.[1] A number of herbaceous perennials successfully overwinter in cold climates. For instance, fern rhizomes growing in deciduous forest floors in Hokkaido, Japan, resisted temperatures ranging from −5 to −17.5°C.[2] Strawberry crowns overwinter but may be injured by low temperatures of about −23°C, or by alternating warm and cool periods where the lower temperatures may range from about −4°C to −9°C.[3] We have been unable to find similar information for ginseng. Lewis and Zenger[4] reported low and variable seed survival in indigenous populations and low temperature injury at this time is possible. Hu *et al.*[5] have also reported very high death rates in natural populations but forwarded no explanation. Again low temperature injury could be inferred. We have observed instances where cultivated ginseng has not grown in the spring, implicating low temperature injury to the rhizome in the previous winter. Also, we have seen examples of low temperature injury to ginseng rhizomes after they were accidentally exposed to temperatures of −20°C. Because of these and other similar instances and the associated economic loss, we undertook studies to examine the low temperature tolerance of roots and seeds of ginseng.

Materials and Methods

Experiments were conducted with American ginseng (*Panax quinquefolium* L.) seeds and roots.

1. Seeds

Seeds were harvested in September and held in an outdoor stratification bed untill November in next year. They were then put in moistened towels in plastic bags and placed in a cold storage held at 4±2℃ and 50±5% relative humidity. Where seeds of different moisture content were needed in subsequent experiments, the required seed moisture content was obtained by leaving the seeds saturated by moisture at room temperature for specific intervals. To follow temperature changes in seeds during cooling tests in the freezer thermocouples were inserted into the seeds, without endocarps, to 1.5 mm. They were then placed inside small glass tubes and capped with aluminum foil. All samples were cooled to 0℃ for 1 hour before treatment. Supercooling nucleation points were determined by differential thermal analysis (DTA) with a constant freezing rate of 10℃ per hour. Exotherms were recorded on a multi-point potentiometric recorder.

Samples for the survival tests were removed from the freezer at the appropriate temperature and held at 2℃ for 24 hours before the ability to germinate was tested in the dark at 15℃ for 10 days.

2. Roots

Three-year-old field grown roots of American ginseng were obtained from the commercial grower on November 3. They were placed in plastic bags in which high humidity was maintained with moistened vermuculite. The bags were placed in storage at 4±2℃ and 50±5% relative humidity to satisfy the dormancy requirements of the roots. In late January and February in next year, DTA of the rhizome, cortex and pith was carried out as for seeds. Rhizomes were cut from whole roots, which in turn were separated into cortex and pith pieces about 5 mm wide, 10 mm long and 2 mm thick. Mean water content of the rhizome was 78%, of the cortex 76%, and of the pith 75%. Chlorenchyma temperatures were determined by DTA with a constant freezing rate of 10℃ per hour and also 1 mm beneath the surface near the center of the chlorenchyma, where the temperature averaged 0.2℃ higher during cooling phase than at the surface.

For survival test samples were removed from the freezer at the appropriate temperature, kept at 2℃ for 24 hours and then kept at room temperature for 24 hours before triphenyl tetrazolium chloride (TTC) tests. And the procedure for the refined TTC test is same as the Steponkus and Lanplear's method.[6]

Results and Discussion

Exotherms were found in the intact seeds containing more than 35% water, but in seeds with 20% there were no exotherms (Fig. 1). The exotherms of all the seeds showed one peak and their shapes were remarkably uniform without

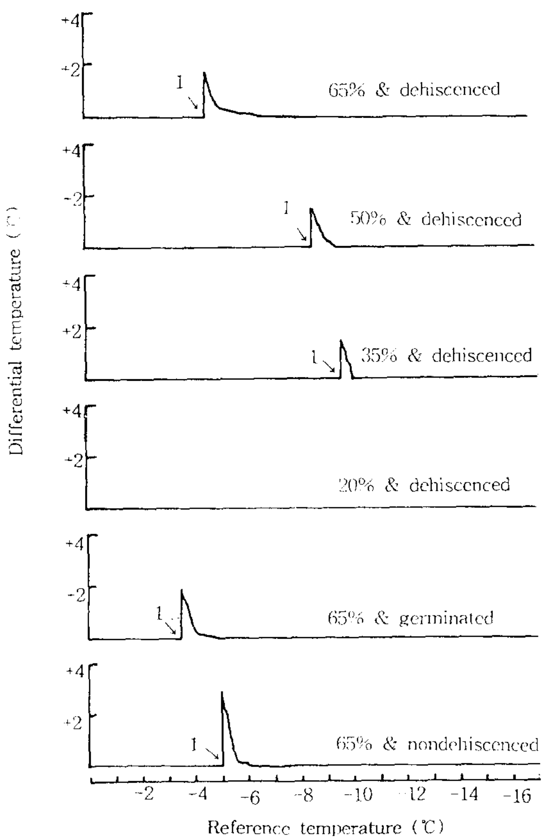

Fig. 1. Differential thermal analysis profile of ginseng seeds with moisture contents at various levels. 1 indicates the initial point at which freezing began.

relation to water content. Studies of supercooling have involved seeds and buds or xylem ray parenchyma of wood plants[7] Our general observations of freezing patterns in ginseng seeds showed only one exotherm that did not agree with the results of Bourque's experiment[8] where two exotherms were observed for lettuce seeds.

The temperature at the initiation of freezing varied from −3.5℃ to −9.6℃ with the different water content in the seeds. That is, the initial temperature of freezing delayed with the decrease of water content. Among the seeds with the same water content, initial temperature of the exotherm of the germinated seeds was higher than that of the nongerminated seeds, but in the nondehiscenced seeds was lower. These exotherms were not related to the injury of intact seeds and we suggest that it represents the freezing of extracellular or bulk water in the out layers, similar to extracellular freezing of other plant tissue.[9]

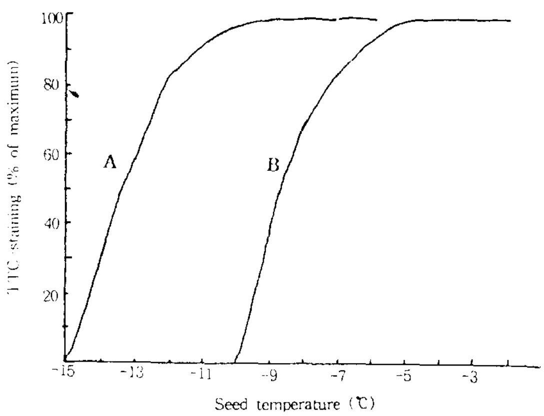

Fig. 2. Temperature dependence of the TTC-staining for nongerminated seeds (A) and germinated seeds (B). Seeds temperatures were measured at 1.5 mm below the surface.

Fig. 2 showed the death of the seed at low temperature with a constant freezing rate of 10℃ per hour. The temperature injured on staining of seeds differed with seeds condition. That is, in germinated seeds the temperature for 50% inhibition of staining was −8.8℃, while the comparable temperature was −14.2℃ for nongerminated seeds, 65% water content and dehiscenced.

In testing for the critical low temperature and its period of exposure for the death of seeds, except for the germinated seeds which died at 5 hours' exposure at −5℃, they did not receive any damage at low temperatures even after 24 hours at −5℃. On the other hand, at 10℃, the staining rate was increased with the decrease of water content. The seeds with coats did not receive any damage at low temperature even after 24 hours' exposure at −15℃ (Table 1). Our results agree with the results that seeds resistance to freezing stress appears to be related to water content.[10]

The exotherms of the rhizome, cortex and pith of ginseng roots presented in Fig. 3 showed that their exotherms had one peak and the shapes were uniform. However, the temperature at initiation of freezing rhizome was higher than the cortex or the pith, but they do not differ

Table 1. Staining percentage of ginseng seeds with moisture contents at various levels held at different low temperature and period of their exposure

Seed conditions		5℃		−10℃		−15℃	
Moisture content	Embro growth	5 hrs.	24 hrs.	5 hrs.	24 hrs.	5 hrs.	24 hrs.
65%	Dehiscenced seeds	100	100	20	0	0	0
50%	〃	100	100	70	50	60	20
35%	〃	100	100	80	60	70	30
20%	〃	100	100	90	80	80	50
65%	Undehiscenced seeds	100	100	100	100	100	100
65%	Germinated seeds	0	0	0	0	0	0

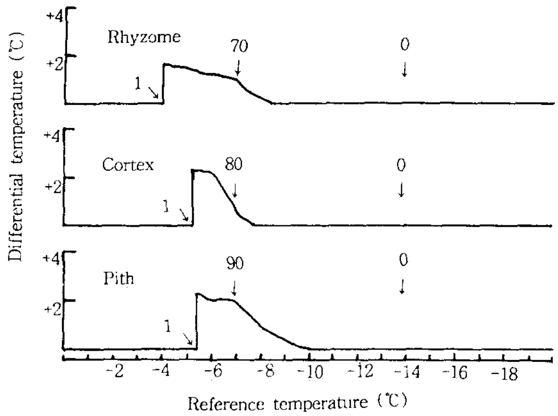

Fig. 3. Differential thermal analysis profile of rhyzome, cortex and pith of 3-yr-old ginseng roots. Figures and arrows show the staining percentage in samples removed at temperature indicated and 1 indicates the initial point at which freezing began.

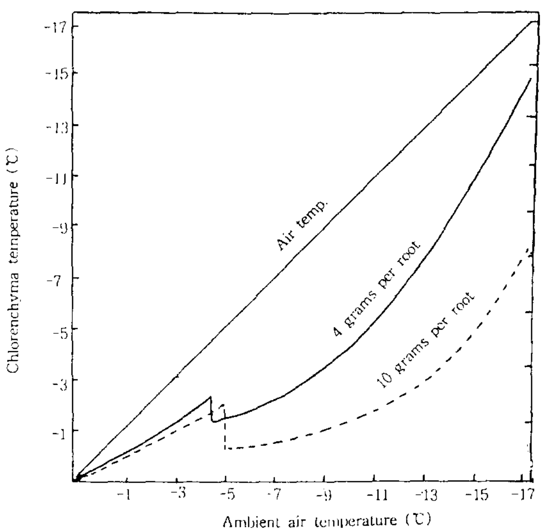

Fig. 4. Cooling curve, showing how the tissue temperature of ginseng root with different root size decreased as the air temperature was lowered.

between the cortex and the pith.

In the TTC test for the survival of the rhizome, cortex and pith, they were some different at the same temperature as the exotherm. That is, staining percentage of rhyzome was 70%, of the cortex 80%, and of the pith 90%. Quamme *et al.*[9] observed that during the controlled freezing as many as 4 exotherms were detected in apple twigs and injury to xylem and pith occurred when twigs were frozen below the initiation temperature of the fourth exotherm independently of the cooling rate. We guess that the exotherm may represent the freezing of a fraction of water which remains unfrozen well below the temperature at which the bulk of the fraction of water freezes.

Table 2. Injuries at the low temperatures on the parts of ginseng roots

Parts of whole root		Temperature (℃)					
		-10	-11	-12	-13	-14	-17
Rhyzome,	1-yr-old	-	-	+	++	++	++
Rhyzome,	3-yr-old	-	-	-	+	++	++
Fine root,	3-yr-old	-	+	++	++	++	++
Lateral root,	3-yr-old	-	-	-	+	++	++
Main body,	3-yr-old	-	-	-	-	-	++

Note : Visual rating of root injury - : none, + : moderate injury, ++ : severe injury.

Typical differential thermal analysis (DTA) profiles of whole roots with different root size are shown in Fig. 4. When the air temperature was continuously decreased, the chlorenchyma temperature in root weight 4 grams decreased to -4.5℃ and then rose to about -2℃. After the rise, the temperature decreased gradually to -10℃ and then decreased more rapidly. In small root, 10 grams, the chlorenchyma temperature decreaded to -5℃ and then rose to about -0.5℃. After the rise, the temperature decreased gradually to -13℃ and then decreased more rapidly. Exotherms appeared at chlorenchyma temperature from -0.5℃ to -2.5℃ and air temperatures in the compartment from -4.5℃ to -5℃. The injuries of visual rating at the low temperatures on the parts of whole roots are shown in Table 2. They did not receive any damage at -10℃ with a constant freezing rate of 10℃ per hour, but the fine roots of 3-year-old received damage to low temperature, -11℃ and the yearling rhizome was damaged at -12℃. The resistance damage at low

Table 3. Staining percentage of 3-yr-old ginseng roots with different root size held at different low temperatures and period of their exposure

Root size	-5℃		-10℃		-15℃		20℃/-3℃
	5 hrs	24 hrs	5 hrs	24 hrs	5 hrs	24 hrs	12 hrs/12 hrs
12~13 g/root	100%	100%	45%	0%	0%	0%	0%
3~5 g/root	100%	100%	40%	0%	0%	0%	0%

temperature appears in the order of main body, rhizome, lateral root of 3-year-old, yearling rhizome, and fine root of 3-year-old.

The staining percentage of whole ginseng roots under different low temperatures and their exposure times was shown in Table 3. They survived at -5℃ for 24 hours. But they received serious damage below -10℃ even for 5 hours' exposure. The ginseng roots all died from the treatment of 12 hours at -3℃, 12 hours room temperature and 12 hours at -3℃, even though they did not receive any damage in continuous exposure for 24 hours at -5℃. Hence, alternative temperature gave more severe damage compared to constant low temperature. The results suggest that the probability of receiving injury at low temperature is higher during the thawing season of the early spring than in the winter. So we must be more careful in managing ginseng fields during the thawing season to prevent the decrease of ginseng plant population by low temperature injury.

References

1. Currie, D. J. : Proceedings of the Second National Ginseng Conference, Jefferson City, Missouri, p. 16(1980).
2. Sato, T. : In: P. H. Li and A. Saki (eds) *Plant Cold Hardiness and Freezing Stress*, Mechanisms and Crop Implications, Vol. 2. Academic Press, New York, p. 447(1982).
3. Daubeny, H. A., Norton, R. A., Schwartze, C. D. and Barritt, B. H. : *Hort Science* **5**, 153 (1970).
4. Lewis, W. H. and Zenger, V. E. : *Amer. J. Bot.* **60**, 1483 (1982).
5. Hu, S. Y., Rudengerg, L. and Tredici, P. del : Study American ginseng, Rhodora, p. 82, 627 (1980).
6. Steponkus, P. L. and Lanphear, F. O. : *Plant Physiol.* **42**, 1423 (1967).
7. Burke, M. J., Gusta, L. V., Quamme, H. A., Weiser, C. J. and Li, P. H. : *Annu. Rev. Plant Physiol.* **27**, 507 (1976).
8. Bourque, J. E. and Stephen J. W. : *Plant Physiol.* **70**, 1571 (1982).
9. Quamme, H., Stushnoff, C. and Weiser, C. J. : *J. Amer. Soc. Hort. Sci.* **97**, 608 (1972).
10. Soepadmo, E. and Lee, David W. : *Nature* **269**, 325 (1977).