

## P 35 Analysis of wheat (*Triticum aestivum* L.) freezing tolerance based on expression of the freezing tolerance-related genes

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### Objectives

This study was aimed to overcome the unstable variables inherent to environmental conditions in the field tests to analyze freezing tolerance in wheat. The various methods including Western analysis, Ion-leakage test and Frost test were used to estimate freezing tolerance in 5 selected wheat cultivars showing different ranges of tolerance to freeze. The results will be used for molecular markers for freeze tolerance in wheat.

### Materials and Methods

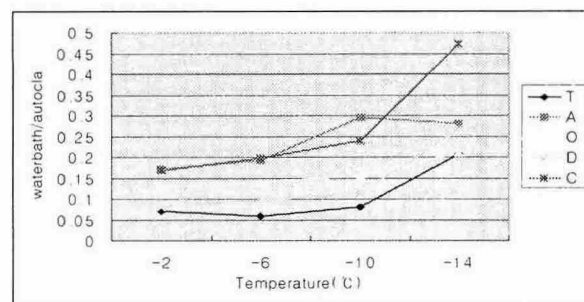
1. Materials: 5 cultivars of wheat treated for cold acclimation at 6/4°C in 9/15hrs (day/night)
2. Methods: Ion Leakage Test, Western, Frost test



**Figure 1.** Western analysis of the apoplastic proteins from cold acclimated wheat leaves probed with CLP antiserum.

### Results and Discussion

The ion leakage test showed increasing levels of freezing tolerance as increasing duration of exposure to low temperatures for acclimation and also different levels of freezing tolerance in 5 species of wheat examined. Western analyses using anti-CLP and anti-TLP serum also indicated a strong correlation between the amounts of the antifreeze proteins accumulated in extra-cellular space and freezing tolerance. Based on ion leakage and frost test applied, it is possible to confirm the fact that a cold acclimation enhances a freeze tolerance or different level of freezing tolerance in each species of wheat. Besides we could observe that a longer period of cold acclimation lead to a higher level of freeze tolerance as well as the amounts of antifreeze protein accumulated in apoplast according to western analysis. Quantitative RT-PCR and *in situ* immunolocalization are being performed. The previous data with barley will also be compared to what observed with wheat. Taken all these methods together, it may be possible to estimate freezing tolerance with accuracy.



**Figure 2.** Ion leakage test at CA 40day after cold acclimation. T (Tapdong), A (Alchan), O (Ol), D (Dahong), C (Chongae)