

# Differences in barley *Dhn* genes expression in the plants exposed to short/long term drought stress

P. MIKULKOVÁ, L. HOLKOVÁ, M. BRADÁČOVÁ and O. CHLOUPEK

Water deficit is one of the major factors limiting plant growth and development and crop productivity (MALATRASI et al. 2002). Adaptation mechanisms of plants to drought stress conditions are based on many factors involving physiological and genetics traits as root system size, thickness of cuticula and the size of stomata. These factors result in different expression of stress genes on molecular level. Tolerance to abiotic stress is associated with accumulation of stress-induced proteins including LEA proteins.

Some of them -dehydrine- are accumulated under abiotic stress (drought, low temperature, osmotic stress, seed drying, salinity) and protect the cells from water deficit. It shows that intensity and start of expression of *Dhn* genes are genotypically dependent. In SUPRUNOVA et al. (2004) high correlations between the rates of relation expression level of these genes and the level of tolerance of tested barley varieties were detected.

In our experiments we studied a dependence of level of drought tolerance of different barley varieties on intensity of *Dhn1* gene expression under different drought stress conditions. Expression was evaluated in 5 barley varieties with different level of drought tolerance:

*Tadmor* (Tad) - drought tolerant variety, species *Hordeum spontaneum*, origin Syria, *Amulet* (Amu) - recommended to drought areas, CZ, *Bojos* (Boj), *Malz* (Mal) - universal varieties to every agricultural areas, CZ, *Jersey* (Jer) - low yield of grain in drought areas, NLD.

## Dehydration treatment for expression analysis

We exposed barley plants to 2 types of drought stress conditions:

1. in short-term drought stress condition plants were grown hydroponically in MS medium. Leaves tissues were

cutted from 10-day-old seedlings and placed onto dry filter paper. After 1, 3, 6, 12 and 24 hours of dehydratation the samples were taken for RNA analysis.

2. in long-term drought stress condition were two weeks old plants removed to solution of PEG (0,3 MPa) for simulation of drought conditions in soil. The control plants were growing in MS medium. After 3, 6, 12 and 24 hours and than after 4, 7 and 14 days of growth in PEG the samples were taken for RNA analysis.

Our results confirm supposition that the variety *Amulet* was adaptable to drought stress conditions at the level of expression of stress genes. More resistant varieties to drought (*Tadmor* and *Amulet*) initiated the expression of *Dhn* genes in cutted leaves later, but the intensity was higher (*Figure 1*) which can be explained by better defense mechanisms on molecular level. Resistant varieties were able to compensate water loss rate with active expression of dehydrins. Long term drought stress (PEG) is not so stress inducing for these varieties, therefore

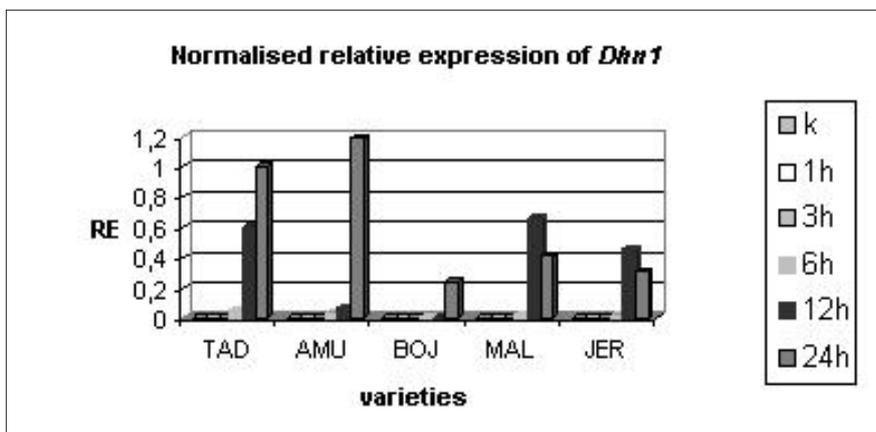


Figure 1: Evaluation of relative expression of *Dhn1* gene in leaves tissues was evaluated according to PFAFFL (2001). Quantification is based on Ct (threshold cycle) values that were normalised using the Ct value corresponding to a barley housekeeping gene for  $\alpha$  tubulin.

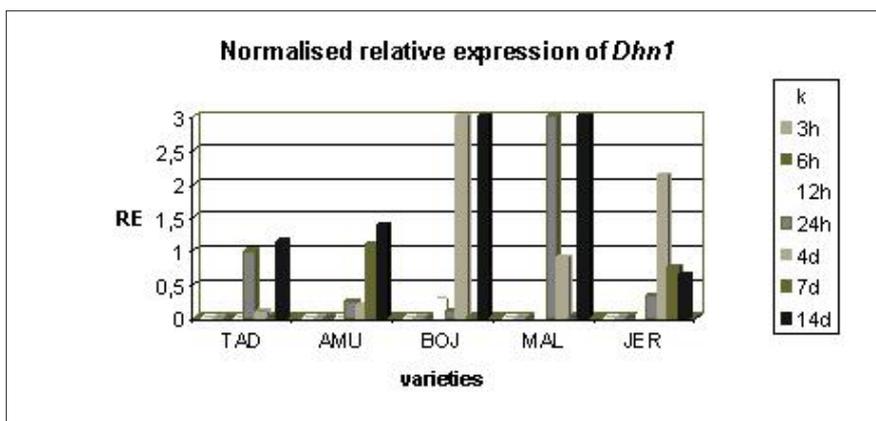


Figure 2: Evaluation of relative expression of *Dhn1* gene in plants growing in PEG solution

Autoren: P. MIKULKOVÁ, L. HOLKOVÁ, M. BRADÁČOVÁ and O. CHLOUPEK, Department of Crop Science, Breeding and Plant Medicine, Mendel University of Agriculture and Forestry, Zemedelska 1, CZ-613 00 BRNO

they were able to cope better with water deficit. Used cultivation conditions were probably more stressful for more sensitive varieties *H. vulgare* probably due to slower closing of stomata and higher cuticular transcription in the beginning of desiccation. All these factors lead to higher activity of *Dhn* genes in these sensitive varieties (Figure 2).

It shows that evaluation of *Dhn1* gene expression could be used for preliminary prediction of sensitivity of barley geno-

types to drought stress on genetic level. More tolerant genotypes show probably earlier start of expression of this gene during short strong stress and on the contrary delayed but more intensive expression during long-term mild stress.

## References

MALATRASI, M., T.J. CLOSE and N. MARMIROLI, 2002: Identification and mapping of a putative stress response regulator gene in barley. *Plant Molecular Biology* 50: 143-152.

PFÄFFL, M.W., 2001: A new mathematical model for relative quantification in real-time RT-PCR. *Nucleic Acids Res.* 29(9): E45-E45.

SUPRUNOVA, T., T. KRUGMAN, T. FAHIMA, G. CHEN, I. SHAMS, A. KOROL and E. NEVO, 2004: Differential expression of dehydrin genes in wild barley, *Hordeum spontaneum*, associated with resistance to water deficit. *Plant, Cell and Environment* 27: 1297-1308.

This work was supported by grants OF 3191 and VC 1M0570.