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Cold Acclimation in Oats and Other Plants

Dissecting Low Temperature Responses Using a Comparative Genomic Approach

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Abstract

Cold acclimation protects plants from temperate regions of the world from the deleterious effects of low and freezing temperatures. This is through a series of transcriptional, regulatory and metabolic changes that enable continued growth and survival. The focus in this thesis is to increase our understanding of the cold acclimation process and there by open the door to development of cold hardy oat (*Avena sativa*) varieties for the Nordic climate conditions. We started by sequencing 9,792 oat ESTs from a cDNA library prepared from pooled total RNA extracted from cold induced oat plants. These sequences were assembled into a UniGene ser of 2,800 sequences, 398 displayed homology to genes previously reported to be involved in cold acclimation. The CBF factor family have a key regulatory role during cold acclimation and in our UniGene set we found four oat CBF sequences.

To infer regulatory networks we developed a rule-based method, which combined data from microarrays with promoter sequences and known *cis*-elements. The method was tested on the cold acclimation process in *Arabidopsis* and could indentify both known and novel network connections. We also performed a comparative transcriptome study between rice and *Arabidopsis* during low temperature stress to explore the molecular differences between chilling sensitive and freezing tolerant plants. Interesting observations were that the dynamics of the response of key genes appears to be higher in *Arabidopsis* than in rice. Several important downstream genes encoding proteins with freezing protective activities in *Arabidopsis* are not present in rice or important cis-elements. Also stress mediated hormone signalling seem to be absent in rice. Together these observations partly explain why rice is unable to cold acclimate to the same extent as *Arabidopsi*s.

Finally we developed a TILLING (Targeting Induced Local Lesions IN Genomes) population in the oat consisting of 2,600 independent events. By random sequencing of two genes involved in the lignin (*AsPAL1*) and ß-glucan (*AsClsF6*) synthesis we estimated the mutation frequency in the population to be approximately 1 per 26,000 bp. This means that each gene is mutated ca 250 times looking at the entire population and assuming an average gene size of 2 kb. This TILLING population will now be an important tool for both breeding and genetic studies in oats.

Keywords: cold acclimation, oat, EST, microarray, transcriptome, chilling, freezing, rice, *Arabidopsis*, TILLING, mutation