mRNA expression profiling and *in situ* hybridization assays in barley seeds of the asparagine synthetase gene *HvASN1* and its putative transcription regulator genes *HvbZIP53* and *HvBLZ1*

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In plants, the asparagine synthetase ASN enzymes catalyze the synthesis of asparagine from glutamine and aspartic acid, in an ATP-dependent reaction. In barley, the gene *HvASN1.1*, orthologous to the Arabidopsis *AtASN1*, is expressed in seeds both during maturation and upon germination. A phylogenomic analysis of the *ASN1* promoter in several species of the Gramineae tribe indicates a conserved region containing a G-box recognised by bZIP Transcription Factors (TFs; Jakoby *et al.*, 2002; Alonso *et al.*, 2009).
The *HvASN1*, *HvbZIP53* and *HvBLZ1* genes have been explored for their presence in seeds by RT-qPCR analyses at 5 different stages of seed development and at 0, 12, 24, 36 and 48 hours of seed imbibition. The expression kinetics of *HvASN1*, *HvbZIP53* and *HvBLZ1* are compatible with *ASN1* expression being regulated by these bZIP TFs and their heterodimers. This fact has been further supported by mRNA in situ hybridization analysis.

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