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TRANSCRIPTIONAL REGULATION OF A WHEAT PATHOGENESIS-RELATED GENE PROMOTER

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Plant pathogenesis-related (PR) proteins are a family of pathogen-inducible proteins involved in defence. Induction of defence responses is triggered by a complex network of signal transduction processes resulting in the rapid activation of defence gene expression. A number of secondary signal molecules such as salicylic acid (SA), methyl jasmonate (MeJA) and ethylene act to amplify and regulate defence responses as demonstrated by studies on mutants and exogenous treatments with these chemicals. In several plant species, SA is able to trigger systemic acquired resistance (SAR). Transcription of distinct classes of PR genes has been reported to be activated by specific signal molecules. Moreover some PR proteins show differential tissue-specific expression, regulated by developmental cues.

To gain a better understanding of the expression pattern of defence genes we characterized a genomic wheat clone encoding a wPR4 protein in terms of transcriptional activity of its 5' non coding region which contains several cis-acting domains found in other defence genes. We constructed a plant transformation vector carrying the 1700 bp 5' untranslated region of wPR4e upstream of the β -glucuronidase (GUS) reporter gene and evaluated the constitutive and inducible trascriptional activity of the putative promoter in transgenic tobacco plants.

To evaluate the involvement of various signal transduction mechanisms several independent tobacco stable transformants were obtained by *A. tumefaciens* transformation which were tested for GUS expression upon leaf treatments with SA (100 uM) or MeJA (100 uM) and after wounding. GUS assays confirmed that the *wPR4e* promoter is able to drive constitutive expression of the reporter gene. Besides, all the tested treatments resulted in strong induction of GUS expression.

The above results confirm that activation of at least one wheat PR4 gene follows both SA- and JAdependent pathways, while Arabidopsis PR4 genes are specifically induced through JA signaling.

To study the spatial distribution of GUS activity driven by the *wPR4e* promoter at different development stages of growth, GUS activity was determined in tobacco seedlings and full grown plants. A reproducible constitutive trascriptional activation, which appears governed by developmental cues, was observed in stems, roots, flowers and seeds.

The potential role of PR4 genes in resistance to pathogens was also investigated in wheat by determining the wPR4e gene expression in response to *F. culmorum* infection and treatments with SAR chemical inducers, namely SA and MeJA. We also addressed the question whether the expression of these genes in wheat was inducible after wounding. The involvement of wPR4e in defence responses triggered by

different signal transduction pathways was evaluated by RT-PCR analysis of its expression in different wheat tissues.