

Functional analysis of a TGA factor-binding site located in the promoter region controlling salicylic acid-induced *NIMIN-1* expression in *Arabidopsis*

J.P. Fonseca¹, M. Menossi¹, F. Thibaud-Nissen² and C.D. Town²

¹Departamento de Genética, Evolução e Bioagentes, Universidade Estadual de Campinas, Campinas, SP, Brasil ²The Craig Venter Institute (JCVI), Rockville, MD, USA

Corresponding author: J.P. Fonseca E-mail: zepedrof@gmail.com

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ABSTRACT. TGA factors play a key role in plant defense by binding to the promoter region of defense genes, inducing expression. Salicylic acid (SA) induces the expression of the gene encoding NIMIN-1, which interacts with NPR1/NIM1, a key regulator of systemic acquired resistance. We investigated whether the TGA2-binding motif TGACG located upstream of the NIMIN-1 gene is necessary for SA induction of NIMIN-1 expression. A mutated version of the NIMIN-1 promoter was created by site-directed mutagenesis. We generated T-DNA constructs in which native NIMIN-1 and mutated promoters were fused to green fluorescent protein and β-glucuronidase reporters. We produced transgenic Arabidopsis plants and observed NIMIN-1 promoter-driven green fluorescent protein expression in the roots, petiole and leaves. Constructs were agroinfiltrated into the leaves for transient quantitative assays of gene expression. Using quantitative real-time RT-PCR, we characterized the normal gene response to SA and compared it to the response of the mutant version of the NIMIN-1 promoter. Both the native NIMIN-1 construct and an endogenous copy of NIMIN-1 were induced by SA. However, the mutated promoter construct was much less sensitive to SA than the native NIMIN-1 promoter, indicating that this TGA2-binding motif is directly involved in the modulation of SA-induced NIMIN-1 expression in Arabidopsis.

Key words: NIMIN-1; TGA; Defense; Salicylic acid; *Arabidopsis*; Transient assays