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TOWARDS THE UNDERSTANDING OF THE OZONE-INDUCED SIGNAL TRANSDUCTION PATHWAYS IN ARABIDOPSIS

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Plants are constantly exposed to a variety of biotic and abiotic stresses and react to these challenges with rapid biochemical and molecular changes that trigger physiological adaptations. Ozone (O_3) , a component of photochemical smog, represents an oxidative stress to living organisms, damaging crops and forests and its concentration has increased dramatically during the past few decades. The plant's response to O₃ resembles components of the pathogen defence response, including the production of reactive oxygen species (ROS), the induction of hypersensitive response (HR) and the development of systemic acquired resistance (SAR). Along this line, the characterization of Arabidopsis transcriptional responses to ozone and pathogen treatments distinguished between shared responses of ozone and pathogen exposure and transcripts specifically regulated by ozone (reviewed in Baier et al. 2005). With the aim to study the oxidative signalling mediated by acute O_3 fumigation in plants, we have carried out microrray analysis to monitor the alterations in gene expression during and after O₃ fumigation in the in the tolerant Arabidopsis Col-0 ecotype (Tosti et al. in press). The O₃ –mediated transcriptional profile is complex, as new genes (i.e. reticuline oxidase) and pathways, other than those already reported as ozone-sensitive, appear to be involved. Interestingly, our microarray analysis revealed that most of the WRKY genes (i.e WRKY22, 25, 33) and some of the genes of the MAPK cascade (i.e. AtMPK3) induced transcriptionally by pathogens, NO or light stress were also induced by ozone. Thus, all these stresses may, at least partially, trigger the same signalling event. Yet, the W-box motifs resulted to be the only cis-element over-represented in the promoter region of up-regulated genes during O₃ treatment. Genes containing the W- promoter element are likely targets of WRKY, including WRKY themselves. Many W-box elements were also identified in the promoter region of most *RLKs* up-regulated during and after O₃ treatment. WRKYs can be substrates of MAPKs and WRKY can regulate RLK expression upon challenging plants with pathogens (Andreasson et al. 2005, Du and Chen, 2000). Thus, we are currently testing the hypothesis that, among the response signals induced by 0₃ in plants, there is a MPK-dependent regulation of WRKYs and that, in turn, WRKYs are involved in the initial activation and/or maintenance of RLK expression during O₃ treatment. To this purpose, we have initiated a preliminary genetic characterization of A.thaliana mutant lines harbouring T-DNA insertions on key 03-induced RLK genes, to be tested along with *wrky* and *mpk* mutant lines for their responsiveness to O_3 . By analysing the transcriptional profiles and physical/functional interactions of candidate O₃ –responsive genes and proteins in single and multiple mutant lines, we expect to gain more insights into the 0_3 -mediated cascade events.

References Andreasson et al (2005) EMBO J 24.2579-2489. Baier et al. (2005) Plant Cell Environment 28 :1012-1020. Du & Chen (2000) Plant J 24.837-847 Tosti et al (in press) Plant Cell Environment