



Prospection for genes expressed during wheat-Magnaporthe oryzae interaction

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Wheat blast was recorded for the first time in 1985 in Brazil. It is caused by the fungus Magnaporthe oryzae (previously M. grisea) which is known to be responsible for the most important disease of rice. Rice-Magnaporthe oryzae pathosystem is the genetic model for plant-pathogen interaction studies. In wheat, the fungus can infect wheat seedlings and adult plants but the most severe symptoms are observed in spikes which become bleached. The fungus prevents grain-filling or destroys the grain-bearing structures. Actually, wheat blast is the most important constraint to wheat production under Brazilian Cerrados conditions. Few wheat cultivars have been identified as resistant, depending on natural innoculum pressure. Since 2009, Embrapa coordinates a national program turned to the identification of resistance sources for blast disease and characterization of candidate genes related to defense responses. The PCR-based suppression subtractive hybridization (SSH) was conducted to search for early defense-related genes in a wheat candidate resistant genotype which exhibits contrasted response to Pyricularia infection in relation to Anahuac 75 (a highly susceptible cultivar). Two cDNA libraries were constructed after SSH, named forward and reverse. Two subtraction libraries, named forward and reverse, were generated by using cDNA synthesized with RNA from wheat spikes inoculated and RNA derived from mock-inoculated wheat spikes as the tester, respectively. A total of 1728 clones were sequenced. Concerning the 960 clones from the forward library, the mean EST size was 0.25 kb and they could be assembled into 91 contigs and 209 singlets. The 300 contigs and singlets were searched against the non-redundant nucleotide and protein databases in GenBank to predict the function of the corresponding genes. Seven per cent corresponded to ribosomal proteins while 6% corresponded to chloroplast genes. Twenty-nine (10%) had no significant homologue in Poaceae sequences and probably represent plant genes that previously were not annotated. Database searches revealed 100 (33%) contigs and singlets encode defense-related genes. Interestingly, two of them encode Ribulose-1,5-bisphosphate carboxylase oxygenase activase (RCA) genes. The levels of RCA transcripts and protein constrains the photosynthetic potential of plants under stress conditions. Function of other genes that are up-regulated following inoculation with wheat blast pathogen will be discussed.

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