

Bid in plants: characterization of plants expressing the animal pro-apoptotic protein Bid

Massimo Crimi, Anna Manara, Linda Fracasso, Zahra Imanifard, Diana Bellin.

Department of Biotechnology, University of Verona, Verona, Italy

Programmed cell death (PCD) is a crucial process in all the living organisms. Similarities and substantial differences are present in control, signaling and execution of this program in plants and animals.

The major regulators of the commitment to PCD in animal cells are the BCL-2 protein family. Homologous proteins are absent in plants. Among these, the BH3-only protein Bid is crucial for both life and death of the animal cells. We have constitutively expressed this pro-apoptotic protein, Bid from mouse, in *Arabidopsis thaliana* and in *Nicotiana tabacum* to explore the possible cross-talk between the apoptotic signaling pathway and the plant PCD pathway. This protein has been inserted in its full-length form, flBid, and not in its caspase-processed active form, tBid. For this reason, it requires an activation signal to exert its action on mitochondria that can be either a caspase-like processing activity or the binding of specific signal molecules (i.e. lysolipid molecules).

Transgenic plants show no differences with wild-type in growth and development. They were treated with different abiotic stress conditions to activate the PCD signaling cascade and to study the possible involvement of responses able to induce flBid activation within plant cells. No significant differences were observed when Bid plants and WT plants were treated either with NO or H₂O₂ as PCD inducer. Small differences are observed in necrotic death induction: Bid plants leaves appear to be more sensitive to oxidative stress when compared to the wild-type ones. Caspase-like activity on Bid protein is absent in these conditions. In root hair cells, we have observed a reduction in the number of apoptotic cells due to an increase of the necrotic ones after 24 hours from H₂O₂ treatment. These results indicated that full length Bid do not significantly change PCD response to H₂O₂ and NO stress in plants.