

## **OPHIOBOLIN A ACTIVATES DIFFERENT DEFENCE RESPONSES DEPENDING ON THE APPLIED DOSE IN TBY-2 CELLS**

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Ophiobolins are a class of secondary metabolites produced by some fungi (*Bipolaris spp.*, *Aspergillus*) pathogens of various plants of agronomic interest, such as rice, sorghum and maize. These compounds are reported to produce different effects in plants, such as inhibition of roots and coleoptiles growth, reduction of seed germination, changes in cell membrane permeability, but their mode of action is still unknown. Ophiobolin A has been mostly studied for its effect on calcium metabolism, being a natural irreversible calmodulin inhibitor.

Here we report a study concerning the effect of ophiobolin A on Tobacco Bright Yellow-2 (TBY-2) cells. We found that ophiobolin A triggers different responses depending on the applied concentration. At concentrations equal or higher than 10  $\mu$ M, ophiobolin A induces programmed cell death (PCD) in tobacco cells, showing different PCD markers such as DNA laddering, cellular shrinkage, micronuclei formation.

Intriguingly, the ophiobolin A- triggered PCD appears not to be mediated by an overproduction of reactive oxygen species (ROS). Indeed, in ophiobolin A- treated cells an increase in hydrogen peroxide production occurs only lately after PCD induction and ROS scavenging treatment reduces ROS level without preventing PCD. This behaviour differs from other kinds of PCD well characterized in the same cell culture (Vacca et al. 2004; Locato et al. 2008).

Ophiobolin A concentrations that do not affect cell viability arrest cell cycle in a reversible manner. In particular, 5  $\mu$ M ophiobolin A treatment induces cell block in S/G2 phase. Concomitantly, ophiobolin A freezes the activity of the poly ADP- ribose polymerases (PARPs), nuclear enzymes involved in DNA metabolic transitions, which normally increases during the exponential growth phase in plant cells (Pellny et al. 2009).

### REFERENCES

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