

P-34. CHARACTERIZATION OF THE OXIDATIVE STRESS OCCURRING AT THE ONSET OF FRUIT RIPENING: DOES IT PLAY A REGULATORY ROLE?

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Ripening is the final stage of fleshy fruit development. It starts at the moment of fruit softening and/or coloring (called veraison in grapes), when embryos are almost mature and mother plants invest in making the fruit attractive to mammals for seeds dispersal. Fruit ripening is thus characterized by the accumulation of flavors, sugars and pigments which ultimately represent an important issue for human diet and are therefore focus of many scientific researches.

Grapes is a non-climacteric fruit in which abscisic acid seems to play the major role in promoting ripening of hard green berries^{1,2}. The mechanism through which abscisic acid signaling takes place has not been described yet, probably because of the complexity of veraison, which represents the moment when many endogenous signals and exogenous stimuli occur simultaneously and are integrated into a developmental program. This concurrence of events make it more difficult to discriminate specific pathways and to link them in a cause-effect relationship.

Our work has focused on the characterization of the oxidative stress occurring transiently at veraison with the aim to ascertain its possible role in ripening control. First of all, we have measured *in vitro* the hydrogen peroxide (H₂O₂) content of grapes skin and flesh collected at different stages along fruit development. We found a transient accumulation of H₂O₂ only in the skin samples, which started at veraison and reached a maximum two weeks later. Confocal microscopy confirmed the tissue specificity of this burst and showed that H₂O₂ accumulates in the cytosol. Whilst the origin of H₂O₂ increased content is not clear, skin cells likely restore its physiological levels through the induction of ROS scavenger activities, mainly catalase. The second important finding of this work concerns the identification of the regio-specific peroxidation (at position 13) of the linolenic acyl fatty chains of mono- (MGDG) and digalactosyldiacylglycerols (DGDG) in the samples prepared from grape berry skins. This observation somehow suggests that strictly regulated, enzyme-driven, processes are operating in the chloroplast, where galactolipids are specifically localized. The enzymatic origin of galactolipid peroxidation is further supported by the transcript and protein expression analyses of a putative chloroplastic 13-Lipoxygenase. The fate of these oxidized forms of galactolipids has not been so far understood and leaves room for interesting speculations.

In conclusion, we describe an oxidative stress in the skin of grape berries at the commencement of ripening, characterized by H₂O₂ in the cytosol on one side, and galactolipid 13-peroxidation on the other side. Both are consistent with a regulatory function, as H₂O₂ could modulate cell transcription through redox-based mechanisms, whereas chloroplasts, which undergo a transition to non-photosynthetic organelles, need to communicate with the nucleus.

1. Gambetta et al., *Planta*, 2010 232:219-34.

2. Sun et al., *BMC Plant Biol.*, 2010, 10:257.