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Nanostructural differences in pectic polymers isolated from strawberry fruits with low expression levels of pectate lyase or polygalacturonase genes

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Our research group has obtained transgenic strawberry plants expressing antisense sequences of either a pectate lyase (APEL lines) [1] or a polygalacturonase gene (APG lines) [2]. Both genes encode ripening-specific endo-pectinases with a common target, deesterified homogalacturonans, but each enzyme act by a different mechanism and pH range. Ripe fruits from both transgenic genotypes were significantly firmer than control, being APG fruits on average 25% firmer than APEL fruits. Cell wall analysis of both transgenic genotypes indicated that pectin fractions extracted with CDTA and sodium carbonate were significantly modified in transgenic fruits [2,3]. To gain insight in the role of these pectinases in pectin disassembly during ripening, CDTA and Na_2CO_3 pectins have been analyzed by atomic force microscopy (AFM). APEL and APG CDTA pectins had similar contour lengths but both were significantly longer than control. Similarly, APG carbonate chains were longer than control, showing APEL carbonate chains an intermediate length. Furthermore, transgenic pectins displayed a more complex branching pattern and a higher number of micellar aggregates, especially in the sodium carbonate fractions of APG samples. Acid hydrolysis of carbonate pectins reduced the number of micellar aggregates. AFM analyses confirm that the inhibition of both pectinases reduces pectin disassembly, and also suggest that each pectinase acts on specific pectin domains. Particularly, polygalacturonase silencing induces more significant pectin modifications, nicely correlated with the firmer phenotype of APG fruits, than the down-regulation of pectate lyase.

[1] Jimenez-Bermudez et al. (2002) *Plant Physiol.*, **128,** 751-759 ; [2] Quesada *et al.* (2009a) *Plant Physiol.*, **150,** 1022-1032 ; [3] Santiago-Domenech *et al.* (2008) *J. Exp. Bot.*, **59,** 2769-2779.
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