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## EXPRESSION OF A PLASTID LIPOXYGENASE DURING TOMATO RIPENING

I. VERDESCA, A. LEONE, G. ZACHEO

CNR, Institute of Sciences of Food Production (ISPA) - Section of Lecce

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Fruit ripening is a complex, genetically programmed process that culminates in changes involving colour, texture, flavour, and aroma of the fruit flesh. Due to the economic importance of fruit crop species these processes are studied extensively at both biochemical and genetic levels.

As ripening progresses, the expression of many genes has been shown to be initiated or up-regulated, among these, lipoxygenases (LOX, EC 1.13.11.12) are demonstrated to play a key role in the biosynthesis of volatile compounds. LOXs are ubiquitous non heme iron containing dioxygenases that catalyse the oxygenation of polyunsaturated fatty acids (PUFAs), containing a cis,cis-1,4 pentadiene moiety, to produce unsaturated fatty acid hydroperoxides, initiating the synthesis of a group of compounds collectively called oxylipins. The products of the LOX pathway have a role in a series of processes such as plant response to injuries, like wounding and herbivore attacks, or to other physiological and environmental stimuli. Some of these molecules are volatile compounds, such as aldheydes and alcohols, which are important constituents of the flavour and aroma of many plant products. In this context LOX have a primary role in influencing the organoleptic characteristics of fruits and vegetables. In addition LOXs are of great interest in food science because their products can have deleterious effects on nutritionally important compounds, such as vitamins, because of the co-oxidation reaction resulting in the bleaching of carotenoid pigments.

Tomato ripening involves changes in plastid structure and an increase of the carotenoid content, which influence the organoleptic and nutritional value of the fruits. The transformation of chloroplasts into chromoplasts is distinguished by the breakdown of the photosynthetic apparatus and a massive synthesis and deposition of carotenoids. It has been suggested that LOX may play a role in the disruption of chloroplast thylakoid membranes that occurs during the chloroplast-to-chromoplast transition during ripening. Tomato LOX are extensively studied, they consists of a family of at least five genes, during ripening individual LOX isoforms are differentially regulated and may have different functions (Griffiths *et al.*, 1999).

We carried out a biochemical and molecular characterization of a tomato fruit lipoxygenase, in order to clarify the possible involvement of a plastid-targeted enzyme (TomloxC) in volatile compounds synthesis during fruit ripening. Expression of TomloxC was analysed by semi-quantitative RT-PCR experiments performed on LOX mRNA from tomato fruits at three ripening stages (unripe, turning, ripe). The results revealed that TomloxC is expressed in ripening fruits with a maximum at the turning stage. TomloxC transcripts were completely absent in tomato leaves.

LOX activity was measured spectrophotometrically in soluble fraction and in chromoplasts purified from pericarp tissue of fruits at the three stages. A higher enzymatic activity, with optimal pH 6.0, was found in chromoplasts purified from turning fruits.

The products obtained from the reaction of plastid tomato LOX and linoleic acid were also analysed.