

Fruit Science

Molecular Approaches for the Regulation of Fruit Ripening

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Abstract: -

Fruit ripening is a complex developmental process that converts immature fruit into an edible, flavorful, and nutritionally valuable product. It involves coordinated physiological, biochemical, and molecular changes such as modifications in texture, color, aroma, and sugar composition. These changes are regulated by hormonal signaling, transcriptional control, and epigenetic mechanisms. In climacteric fruits, ethylene plays a central role by activating ripening-related gene expression through specific biosynthesis and signaling pathways, whereas non-climacteric fruits depend more on other hormonal regulators despite retaining some ethylene signaling components. Key transcription factors, especially MADS-box proteins like RIN and NOR, regulate large-scale transcriptional reprogramming that drives the initiation and progression of ripening. Hormones including auxin, gibberellins, and abscisic acid further modulate the process by influencing gene expression and developmental timing. Ripening traits are associated with the coordinated expression of genes involved in cell wall modification, carbohydrate metabolism, pigment production, and aroma formation. Additionally, epigenetic mechanisms such as DNA methylation influence the accessibility of ripening genes to transcriptional regulators. Modern molecular approaches, including genetic engineering, genome editing, and post-harvest technologies, provide promising strategies to enhance fruit quality, extend shelf life, and reduce post-harvest losses.

Key words: Fruit ripening; Ethylene signaling; Transcription factors; Hormonal regulation; Gene expression; Epigenetic regulation; DNA methylation; Climacteric and non-climacteric fruits; Molecular approaches etc.

Introduction:

Fruit ripening is a highly coordinated physiological and biochemical process that converts immature fruit into a palatable, visually appealing, and nutritionally valuable product essential for both seed dispersal and human consumption. This transition involves

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extensive changes in texture, color, flavor, aroma, and nutritional composition. These modifications are controlled by tightly regulated hormonal signaling pathways, transcriptional regulation, and complex molecular networks that coordinate the activation and repression of ripening-related genes (Porika *et al.*, 2016).

Classification of Fruits Based on Ripening Behavior

Based on respiration patterns and ethylene production during ripening, fruits are broadly classified into **climacteric** and **non-climacteric** types.

Climacteric fruits such as tomato, apple, banana, and mango show a sharp increase in respiration along with a burst of ethylene production at the onset of ripening. This ethylene surge acts as a central regulatory signal that activates genes responsible for cell wall disassembly, pigment synthesis, sugar accumulation, and the formation of volatile aroma compounds. Disruption of ethylene biosynthesis or signaling significantly impairs ripening, highlighting its critical regulatory role (Porika *et al.*, 2016).

In contrast, non-climacteric fruits such as strawberry and grape do not exhibit a distinct rise in respiration or autocatalytic ethylene production. However, ethylene still influences the expression of certain ripening-related genes, indicating that components of

the ethylene signaling pathway are partially conserved across different fruit types despite differences in regulatory dominance.

Ethylene Biosynthesis and Signaling Pathway

Ethylene production is regulated by the rate-limiting enzymes **ACC synthase (ACS)** and **ACC oxidase (ACO)**. Ethylene perception occurs through membrane-associated receptors including **ETR1, ETR2, ERS1, ERS2, and EIN4**.

In the absence of ethylene, these receptors activate negative regulators such as **CTR1**, which suppress ripening-related signaling. When ethylene binds to its receptors, this inhibition is relieved, allowing downstream signal transduction through **EIN2** and **EIN3**. These components activate transcriptional programs that regulate the expression of ripening-associated genes and coordinate the progression of the ripening process (Liu and Zhang, 2004).

Transcriptional Regulation of Fruit Ripening

Fruit ripening involves extensive transcriptional reprogramming controlled by key transcription factors. Among these, **MADS-box transcription factors** play a major role in regulating fruit development and ripening.

Studies using tomato ripening mutants such as **ripening-inhibitor (rin)**, **non-**

ripening (nor), and colorless non-ripening (Cnr) have provided important insights into transcriptional control mechanisms. The **NOR** gene acts early in the regulatory hierarchy and influences the expression of numerous ripening genes. The **RIN** transcription factor directly regulates a wide range of downstream targets by binding to promoters of genes involved in ethylene biosynthesis, carotenoid accumulation, and chlorophyll degradation. Together, these transcription factors form a regulatory cascade that coordinates the initiation and progression of ripening (Osorio, 2013).

Role of Other Hormones in Ripening

Although ethylene plays a dominant role in climacteric fruits, several other plant hormones also contribute to the regulation of ripening.

Auxin (IAA) levels typically decline before the onset of ripening, while conjugated forms increase. Elevated auxin levels can delay ripening by suppressing the expression of ripening-specific genes, particularly in non-climacteric fruits.

Gibberellins (GA) also influence ripening dynamics. Exogenous application of GA can delay color development and modify the expression of genes associated with cell expansion and cell wall remodeling.

In contrast, **abscisic acid (ABA)** acts as a positive regulator of ripening. ABA

promotes sugar accumulation and activates ripening-related gene expression. Reduced ABA biosynthesis often results in firmer fruits and delayed ripening. In fruits such as strawberry and grape, ABA functions as a major ripening signal.

Genetic and Molecular Basis of Ripening

The physiological changes associated with fruit ripening are controlled by coordinated expression of genes involved in several functional processes:

⇒ **Ethylene biosynthesis and signaling genes** such as *ACS*, *ACO*, *CTR1*, and *EIN* regulate hormone production and signal transduction.

⇒ **Cell wall modification genes**, including *polygalacturonase (PG)*, *pectin methylesterase (PME)*, *expansin (EXP)*, and *xyloglucan endotransglycosylase (XET)*, are responsible for fruit softening.

⇒ **Carbohydrate metabolism genes** such as *fructofuranosidase (FFT)*, α -*amylase*, and *phosphorylase* control starch degradation and sugar accumulation.

⇒ **Pigment biosynthesis genes**, including *chalcone synthase (CHS)* and *lycopene cyclase*, contribute to color development.

⇒ **Aroma biosynthesis genes** such as *alcohol dehydrogenase (ADH)*,

lipoxygenase (LOX), and *cytochrome P450 enzymes* participate in the production of volatile flavor compounds.

Species-specific differences exist despite shared regulatory mechanisms. For example, banana ripening is characterized by strong induction of **α -amylase**, leading to rapid starch degradation, whereas mango ripening involves extensive **pectin degradation** and activation of pigment biosynthesis pathways (Zhong *et al*, 2013).

Epigenetic Regulation of Ripening

Epigenetic mechanisms also contribute significantly to the regulation of fruit ripening. **DNA methylation** plays a key role in controlling the accessibility of ripening genes to transcription factors.

The phenotype of the **Cnr mutant** results from promoter hypermethylation rather than a change in gene sequence. During normal ripening, progressive **demethylation of ripening gene promoters** allows transcription factors such as **RIN** to bind and activate ripening-associated genes, thereby enabling the initiation of ripening programs (Osorio, 2013).

Applications and Future Perspectives

Advances in the molecular understanding of fruit ripening have opened new possibilities for improving fruit quality, extending shelf life, and reducing post-harvest

losses. Modern approaches such as **genetic engineering, genome editing using CRISPR/Cas systems, and advanced post-harvest technologies** enable targeted manipulation of ripening-related genes.

These strategies can delay fruit softening, enhance sweetness, improve nutritional quality, and increase resistance to post-harvest deterioration. Practical applications already include the use of **ethylene inhibitors such as 1-methylcyclopropene (1-MCP)**, improved storage technologies, and breeding programs aimed at enhancing antioxidant and vitamin content. Understanding conserved ripening regulators across different fruit species will further support sustainable crop improvement and horticultural production.

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