Fruit Development and Ripening

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Formation of fruits is a characteristic feature of angiosperms. Fruit types exhibit a great deal of diversity. They form an integral component of diet and provide not only vitamins and minerals but are also important source of antioxidants and fibers. Nutritional quality of the fruit is determined by various factors including those affecting the ripening stage. A fruit must have an optimum degree of ripeness so as to be consumed. Fruits are developed from less attractive immature stages to mature stages which attract seed-dispersing animals as well as human beings. Fruit ripening is a complex process, accomplished through several physiological, biochemical, and molecular mechanisms. These mechanisms also bring about changes in pigmentation due to loss of chlorophylls and a substantial increase in non-photosynthetic pigments, such as anthocyanins and carotenoids. Increase in the activity of cell wall hydrolases leads to fruit softening which is reflected in the texture of the mature fruit. Ripe fruits possess characteristic taste (due to elevated levels of sugars and depletion of organic acids) and aroma (due to biosynthesis of volatile compounds). Postharvest handling of the overripe fruits, however, is very difficult and adversely affects their storage and marketing.

This chapter begins with a brief account of different phases of fruit development. Detailed account of various physiological processes that take place during fruit ripening process, such as change in the color of fruit, change in cell wall architecture, intensification of flavors and fragrances, and synthesis of various bioactive compounds, has subsequently been presented. Both climacteric and non-climacteric patterns of fruit ripening have been dealt with to highlight the differences and common aspects among them. The role of ethylene and other phytohormones in regulating the ripening process has also been discussed. Genes implicated in the biosynthesis of ethylene and oxidative processes, especially those related to reactive oxygen species (ROS) have also been discussed. Fruit ripening is regulated by several genes that play a significant role in accumulation of antioxidants, sugars, and bioactive compounds, release of volatile compounds, and softening of cell wall. The role of transcription factors and genes functioning as

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master switches in fruit ripening has also been explained. Epigenetic studies on *colorless non-ripening (CNR)* mutant of the model system-tomato have been highlighted to discuss the importance of these modifications in affecting fruit phenotypes.

27.1 Stages of Fruit Development and Ripening Stages

Botanically, a fruit is defined as "a seed receptacle developed from an ovary." Many fruits arise from several ovaries, and sometimes associated floral parts are also involved in fruit development after fertilization. Development of fruits is an evolutionary adaptation which helps in seed dispersal through various agencies. Fruit development shows considerable variations and ranges from simple to aggregate and multiple fruits. Simple fruits develop from single ovary, whereas multiple fruits develop from fused ovaries of a single flower (Fig. 27.1). Development of aggregate fruits takes place from several single ovaries of a single flower, whereas multiple fruits develop from ovaries of several flowers in an inflorescence. Simple fruits can be either fleshy (e.g., tomato and apple) or dry, such as that of poppy. Aggregate and multiple fruits are mostly fleshy and sometimes, associated structures, such as a receptacle, become part of the fruit. Berries, such as tomato, possess numerous small seeds within the locule (Fig. 27.2). Drupes (e.g., peach) possess a single large seed which is surrounded by a fleshy mesocarp. In some varieties, such as those of banana, pineapple, and cucumber, the development of fruit takes place without prior fertilization, thereby resulting in seedless fruits (Box 27.1). A wide range of fruit types represent different morphologies which, to an extent, are associated with different metabolic pathways accompanying their maturation. However, many of the biochemical pathways involved are common to almost all fruits. Most of the physiological and biochemical studies on fruit development and ripening have been dealt with fleshy fruits. The pattern of growth curve of fruit development has been reported to be either sigmoidal or double sigmoidal in the majority of flowering plants. The double-sigmoidal curve is obtained sometimes due to the second burst of growth. On the basis of physiological and biochemical activities, fruit development process can be identified in four phases (Table 27.1). It is the physiological and biochemical activities occurring along the course of development which are responsible for the characteristic ripening of fruits. Ripening is the terminal stage of fruit development which is characterized by a change in size, color, and aroma of the fruit. Different ripening stages in model climacteric and non-climacteric fruits, i.e., tomato and strawberry, respectively, have been shown in Boxes 27.2 and 27.3. Table 27.1 highlights the events associated with fruit development and ripening. Phase I of fruit development is characterized by anther dehiscence and subsequent release of pollen grains. Phase I is known as fruit set which decides whether to abort or to go ahead with ovary development to form fruit. Phase II is marked by rapid divisions of cells. Since cessation of cell division takes place in phase III, fruit growth is mostly attributed to cell enlargement. Attainment of final fruit size and shape also occurs

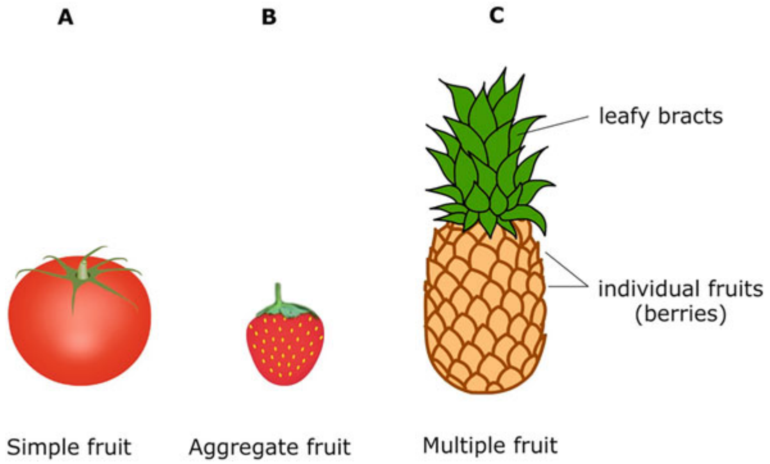


Fig. 27.1 Different types of fruits. (**a**) A simple fruit develops from a single ovary (tomato). (**b**) An aggregate fruit develops from several individual ovaries of a single flower (strawberry). (**c**) A multiple fruit develops from fused ovaries of several flowers (pineapple)

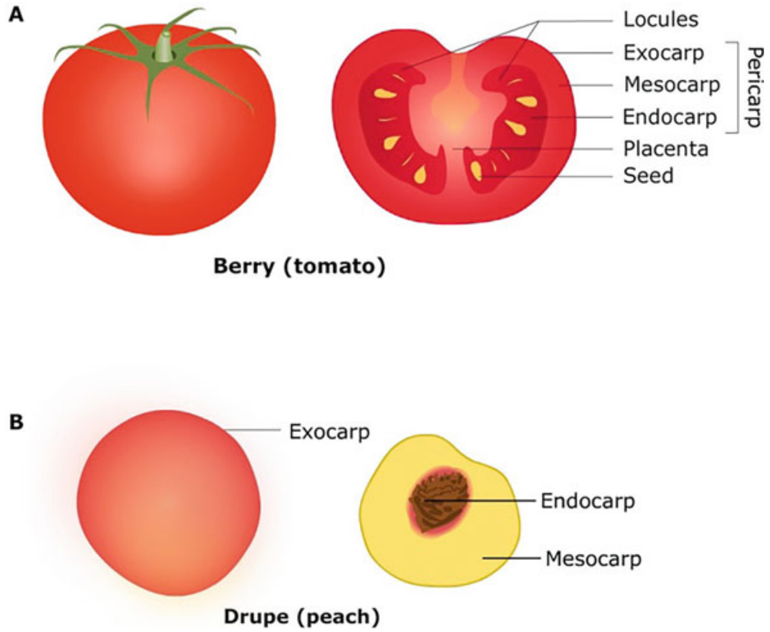
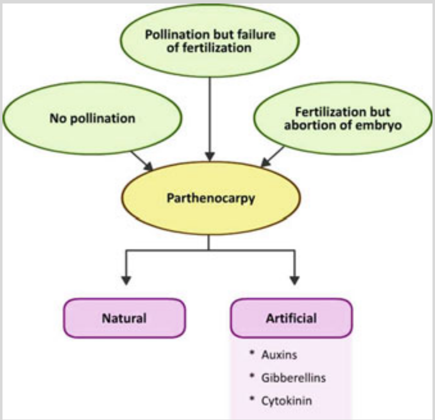


Fig. 27.2 (**a**) A berry has numerous small seeds within the locule (Tomato). (**b**) A drupe possesses a single large seed surrounded by fleshy mesocarp (Peach)

Box 27.1: Parthenocarpic Fruits

The term "Parthenokarpie" was introduced by Noll in 1902 to the seedless condition in fruits. The development of fruit without fertilization is known as parthenocarpy. Parthenocarpic fruits may develop due to (1) the absence of pollination, (2) occurrence of pollination but failure of fertilization, and (3) fertilization followed by abortion of embryo. Parthenocarpy is mainly of two types—natural and artificial parthenocarpy. Naturally parthenocarpy is genetically inherited, and the potential of different cultivars to form parthenocarpic fruits is variable. Parthenocarpy can also be induced artificially using hormones, such as auxins, gibberellins, and cytokinins. However, auxins and gibberellins are well-known to induce parthenocarpy. The treatment of young, unpollinated ovaries with auxins leads to the production of parthenocarpic fruits in selective cultivars of strawberry, orange, grapes, and tomato. Furthermore, GA3 or GA1 treatment to ovaries of unopened and unpollinated flowers during receptive period after anthesis or before anthesis causes development of parthenocarpic fruits in certain cultivars of tomatoes, blueberries, and *Arabidopsis*. The phenomenon of parthenocarpy is popular among horticulturists mainly for crops where seedless fruits are desirable, such as citrus, grapes, and bananas for the preparation of eatables like jam, jellies, sauces, etc. Also, seedless fruit has a longer shelf life than fruits with seeds because seeds produce hormones that trigger senescence. However, parthenocarpy has limited application in agriculture due to pleiotropic effects and occurrence of unfavorable characteristics, such as small fruits.



during this phase. Final phase, i.e., phase IV, is marked by the onset of fruit ripening. Sometimes, there could be a second burst of growth in some species which is responsible for getting double-sigmoidal growth curve.

Table 27.1 Events associated with different phases of fruit development and ripening

Phase of fruit development	Events	Activity
Phase I	Fruit set	Ovary development, anthesis, and, following fertilization, decision whether to abort or to go ahead with fruit development
Phase II	Cell divisions	Embryo growth marked by rapid cell divisions
Phase III	Cell expansion	Cessation of cell division and cell enlargement, leading to attainment of final fruit shape and size
Phase IV	Onset of fruit ripening	Enhanced biosynthesis of carotenoid; changes in color, texture, and taste; second burst of growth in some species

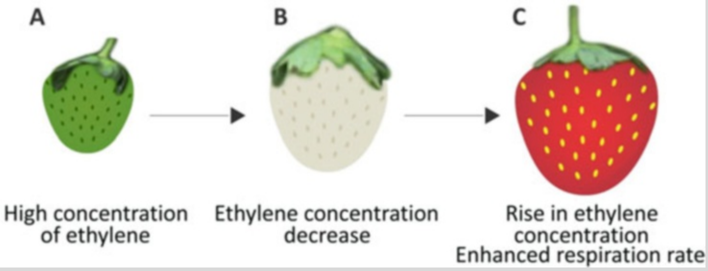
Box 27.2: Tomato as a Model System for Understanding Fruit Development and Ripening

Tomato (*Solanum lycopersicum*), belonging to Solanaceae family, has been considered a favorite model for understanding the physiology of fruit development and subsequent ripening. The pigmentation of tomato fruits is the most important external characteristic feature to assess its degree of ripeness and postharvest life. Red pigment accumulation is the result of chlorophyll degradation accompanied with the synthesis of lycopene and other carotenoids, as chloroplasts are converted into chromoplasts. On the basis of external color, six ripening stages of fruit ripening have been recognized in tomato:

I. Stage 1 or "Green stage" is characterized by 100% green pigmented fruit.
II. Stage 2 or "Breaker stage" is characterized by a noticeable break or change in color with less than 10% of nongreen color.
III. Stage 3 or "Turning stage" brings about 10–30% change in surface pigment to red.
IV. Stage 4 or "Pink stage." At this stage, about 30–60% fruit surface is red (ish).
V. Stage 5 or "Light red stage" is characterized by 60–90% red surface of the fruit.
VI. Stage 6 or "Red stage." During this stage, more than 90% of fruit surface appears red.

Box 27.3: Strawberry: Prime Model for Non-climacteric Fruit Ripening

Studies in ripening of non-climacteric fruits are less in comparison to those done in climacteric fruits. Significant research has been done on some non-climacteric fruits, such as strawberry, citrus, and grapes. Strawberry is considered as the model of choice for understanding ripening in non-climacteric fruits. Some of the changes in strawberry ripening are common in climacteric fruits. This includes loss of chlorophyll and accumulation of anthocyanins, sugars, and volatiles. Biochemical analyses have revealed that ethylene concentration is relatively high at green stage, decreases in white fruits, and increases again at the red ripening stage. Increase in ethylene concentration is accompanied by enhancement in respiration rate similar to that in climacteric fruits before the onset of ripening. Exogenous application of ethylene causes downregulation of several cell wall-related genes for the activity of β-galactosidase, pectin methylesterase or β-xylosidase, and some more. Interestingly, expression of expansin gene FaEXP2 does not change. Transcriptomic and metabolomic studies in transgenic strawberries have shown the requirement of ethylene action during ripening.



Relative differences in the concentration of endogenous ethylene during different developmental stages in strawberry. A. Green stage. B. White stage and C. Mature ripe red stage.

27.2 Physiological Changes During Fruit Ripening

27.2.1 Fruit Texture and Softening

Several metabolic events contribute to changes in the texture of fruits. These changes involve loss of turgor pressure, degradation of starch, physiological changes in the composition of membranes, and modifications in the cell wall architecture (Fig. 27.3). Changes in cell wall composition are considered highly significant during the progression of fruit ripening. Work on transgenic fruits has shown that changes in cell wall responsible for fruit softening and textural changes are complex

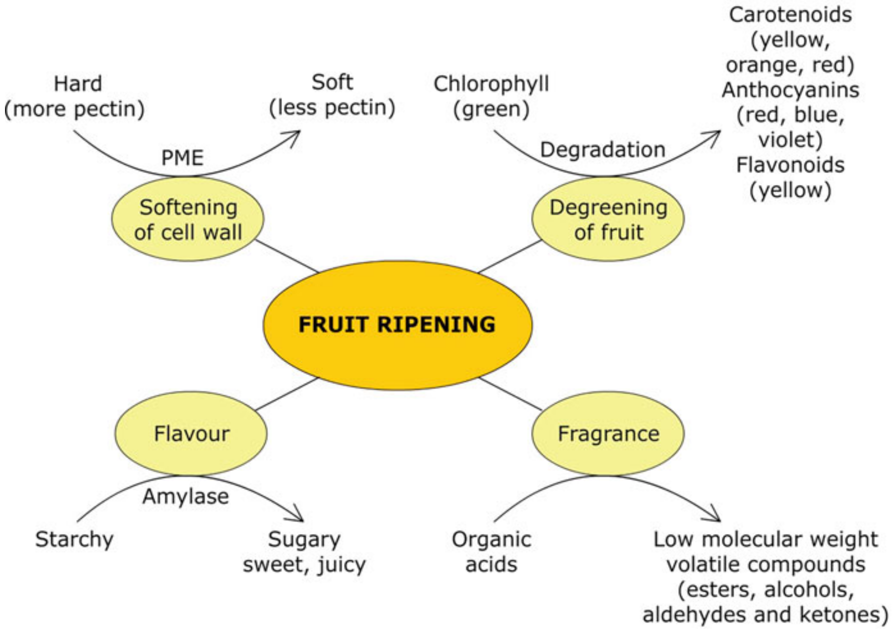


Fig. 27.3 Different physiological changes that take place during fruit ripening process. PME, pectin methyl esterase

(Box 27.4). Coordination and synergistic action of a variety of cell wall-modifying enzymes are responsible for these changes. Structural changes in the cell wall are associated with the dissolution of middle lamella and disruption of the primary cell wall which leads to loss of firmness in mature fruits. These include depolymerization and solubilization of the polysaccharides and pectin components along with rearrangements of their associations. Modifications of polysaccharides may occur depending on the type of fruit. Table 27.2 summarizes the major modifications in the cell wall polysaccharides during ripening in some climacteric and non-climacteric fruits. In tomato, cell wall modifications include both depolymerization and solubilization of polyuronides and hemicelluloses. Cell wall-modifying enzymes are broadly categorized as pectolytic and non-pectolytic, depending on the specific polysaccharide used as substrate. Endo- and exo-polygalacturonases, pectin acetylesterases (PAE), pectate lyases (PL), pectin methylesterases (PME), β-galactosidases, and α- L-arabinofuranosidases are pectolytic enzymes. The activity of these enzymes leads to cleavage or modification of the polysaccharide backbone and removal of neutral sugars from the branched side chains. Removal of methyl ester groups from the fruit cell wall by PME facilitates access of polygalacturonase (PG) to its substrate. PME is expressed before ripening and is downregulated by ethylene during ripening. Repression of PME by antisense resulted in increased fruit viscosity. Non-pectolytic enzymes, such as endo-1, 4-β-glucanases (EGase), endo-1,4-β-xylanases, β-xylanases, xyloglucan endotransglucosylase/hydrolases (XTH),

Box 27.4: Flavr Savr™ Tomato

Vegetable and fruit production suffer from many biotic stresses caused by pathogens, weeds, and insects. A number of transgenic fruits have been deregulated and commercialized in many countries in the recent past. Commercial sale of genetically modified (GM) food began in 1994 when scientists from first GM crop, i.e., Calgene Inc. (USA), developed Flavr Savr which exhibited delayed ripening in tomato. It was engineered to have a longer shelf life by inserting an antisense gene that delayed ripening. To withstand the rigors of shipping, tomatoes must be picked up at "mature green stage" which have already absorbed all the vitamins and nutrients from the plants but have not started producing ethylene that triggers ripening. Calgene, Inc. developed a tomato with a gene that slows down the natural softening process accompanying ripening. Pectin in fruits, which is responsible for their firmness, is degraded by an enzyme called polygalacturonase (PG). As pectin is destroyed, the cell walls of tomatoes breakdown, leading to softening of fruits thus, make them difficult to ship. Reducing the amount of PG in tomatoes shows cell wall breakdown and produces a firmer fruit for a longer time. Calgene's scientists isolated PG gene in tomato plants and converted it into a reverse image of itself, called antisense orientation. The "reversed" tomato gene (the Flavr Savr gene) was reintroduced into the plants. In order to tell if Flavr Savr gene was successfully reintroduced into the plants, Calgene scientists attached a gene that makes a naturally occurring protein that renders plants resistant to the antibiotic kanamycin. By exposing the plants to the antibiotic, Calgene scientists could tell which plant had accepted that Flavr Savr gene. The ones unaffected by kanamycin grow to have designed traits of Flavr Savr. Once in a tomato plant, the Flavr Savr gene attaches itself to the PG gene.

Developing Flavr Savr Tomatoes

With the Flavr Savr gene adhering to it, the PG gene cannot give necessary signals to produce PG enzyme. Thus, these tomatoes retain their flavor. In 1994, US Food and Drug Administration (FDA) announced that Flavr Savr tomatoes are as safe as conventional tomatoes.

Table 27.2 Some climacteric and non-climacteric fruits which undergo polysaccharide depolymerization during ripening

S. No.	Fruit	Climacteric/non-climacteric	Depolymerization of	
			Pectin	Hemicellulose
1.	Tomato (*Solanum lycopersicum*)	Climacteric	Yes	Yes
2.	Apple (*Pyrus malus*)	Climacteric	No	No
3.	Strawberry (*Fragaria vesca*)	Non-climacteric	No	Yes
4.	Grapes (*Vitis vinifera*)	Non-climacteric	Yes	Yes
5.	Kiwifruit (*Actinidia deliciosa*)	Climacteric	Yes	Yes

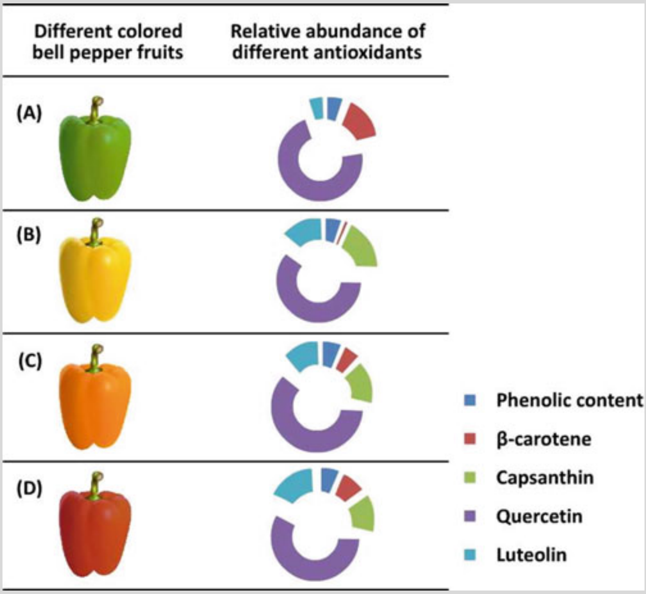
and expansins, are responsible for hemicellulose modifications. In tomato, softening and ripening are accompanied by about 2000-fold increase in PG mRNA. During softening in apples, cell wall polysaccharides do not undergo extensive depolymerization.

27.2.2 Changes in Pigmentation

Along with texture, fruit ripening is often accompanied by changes in flavor and aroma. During ripening, fruits exhibit transition from green to a variety of pigmentations, including red, yellow, orange, purple, and blue. The pigments involved in this process not only increase the visual appeal of the fruit but are also crucial for the taste and aroma. Generally, fruits contain a variety of pigments which includes chlorophyll (green color pigment), carotenoids (yellow, orange, and red colors), anthocyanins (red, blue, and violet colors), and flavonoids (yellow pigment). Degradation of chloroplasts takes place at the onset of ripening which is responsible for degreening of developing fruit. Chloroplasts are converted into chromoplasts because of degradation of chlorophyll and act as the sites for the accumulation of carotenoids. Chromoplasts are nongreen plastids that are responsible for the yellow, orange, and red colors of many fruits. They evolve during fruit ripening by the differentiation of other forms of plastids. In many fruits, such as tomato and bell pepper (*Capsicum annuum*), chromoplasts are derived from chloroplasts with the disintegration of the thylakoid membranes and the formation of new carotenoid-bearing structures. The colors of different kinds of bell pepper are a consequence of selection arrest of chloroplast to chromoplast transition at different stages (Box 27.5). Protein tyrosine nitration is also enhanced during fruit ripening in capsicum (Box 27.6). In other fruits, such as papaya, chromoplasts evolve from leucoplasts or proplastids, as no intermediary amyloplast or chloroplast transitions are encountered. The precursors for carotenoid biosynthesis are synthesized in plastids. The formation of the colorless molecule-phytoene, catalyzed by the enzyme phytoene synthase, is

Box 27.5: Antioxidant Activity of Different Colored Bell Pepper Fruits

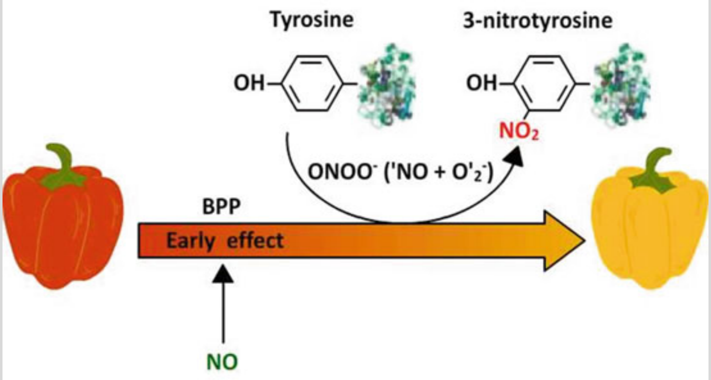
Bell peppers (*Capsicum annuum* L.) are commonly available in green, yellow, orange, and red color. Seed source of all the bell peppers is the same, i.e., *Capsicum annuum*, but they have different characteristics depending upon the special variety of the plant. The two major factors responsible for the difference in a bell pepper's color are its time of harvesting and degree of ripening. A green bell pepper usually matures into a yellow/orange bell pepper and then grows on to gain red color. However, yellow, orange, and red bell peppers are always more ripe than green ones. They are costlier than green bell peppers because they require more time in the ground before they can be harvested. The green color of bell pepper is due to chlorophyll and the carotenoids typical of the chlorophyll. The yellow and orange colors of pepper are generally due to α- and β-carotene, zeaxanthin, lutein and β-cryptoxanthin. The red color is due to the carotenoids, such as capsanthin, capsorubin and capsanthin 5,6-epoxide. Differences exist not only in their colors but also in their nutrient compositions. Bell peppers are good sources of vitamins C and E, provitamin A and carotenoids. Additionally, they contain various antioxidants, such as phenolics and flavonoids.



The figure depicts the relative differences in the abundance of various antioxidants in green, yellow, orange and red colored bell pepper fruits.

Box 27.6: Enhancement of Protein Tyrosine Nitration Characterizes Fruit Ripening In Capsicum

Fruits of *Capsicum annuum* usually ripe within 3–8 days depending on the variety. After the attainment of the breaker point stage (BP stage), the process of ripening proceeds irreversibly in one direction. Exogenous application of NO gas initially delays fruit ripening without promoting protein nitration. However, once the early effect of NO is over as it is a highly reactive radical, it promotes nitration of tyrosine residues of certain proteins. The process of fruit ripening proceeds simultaneously with the conversion of protein tyrosine into 3-nitrotyrosine which involves peroxynitrite through the reaction of NO and superoxide radicals.



Model depicting proposed relationship between reactive nitrogen species (RNS) and the ripening of *Capsicum annuum* fruit.

the first committed step in this process. Phytoene synthase catalyzes a two-step reaction which results in the formation of phytoene (Eqs. 27.1 and 27.2). Through a series of reactions, phytoene is converted into red carotenoid-lycopene (Eqs. 27.3 and 27.4). During ripening in tomato, carotenoid concentration increases about 10–14 times due to the accumulation of lycopene in tomato. In fact, blueberries are known to accumulate more than a dozen different anthocyanins during ripening. These anthocyanins include malvidin-, delphinidin-, petunidin-, cyanidin-, and peonidin-glycosides. The synthesis of anthocyanins takes place through the phenylpropanoid pathway.

$$2\text{Geranylgeranyl diphosphate} \xrightarrow{\text{phytoene synthase}} \text{PP}_{\text{i}} + \text{prephytoene diphosphate} \tag{27.1}$$

$$\text{Prephytoene diphosphate} \xrightarrow{\text{phytoene synthase}} \text{phytoene} + \text{PP}_\text{i} \tag{27.2}$$

$$\text{Phytoene} \xrightarrow{\text{phytoene desaturase}} \zeta - \text{carotene} \tag{27.3}$$

$$\zeta - \text{carotene} \;+ 2\text{PQH}_2 + 2\text{O}_2 \overset{\zeta-\text{carotene desaturase}}{\leftrightarrow} \text{Lycopene} + 4\text{H}_2\text{O} + 2\text{PQ} \tag{27.4}$$

27.2.3 Flavor and Fragrance

Fruit flavor is contributed by several primary and secondary metabolites which include mainly essential amino acids (phenylalanine, leucine, and isoleucine), essential fatty acids (principally linolenic acid), sugars, and carotenoids. **1. Sugars**—Sweetness is one of the important quality parameters in many fruits. It is determined by the total sugar content and the relative ratio of various sugars present during ripening. Accumulation of glucose, fructose, and sucrose has been reported in several fruits, such as watermelon. In developing fruits, such as mango and banana, which act as sink, transported sugar, or sugar alcohols are converted into starch. Sugars are stored as reducing sugars or as sucrose in fruits functioning as storage organs (e.g., tomato and strawberry). Occasionally, as in olive, they may get converted into lipids. **2. Organic acids**—Organic acids are important intermediate metabolites which contribute significantly to fruit flavor. The ratio of organic acid: sugar is crucial in determining the quality parameters at the time of fruit harvest. Primary metabolic pathways, such as glycolysis and the tricarboxylic acid (TCA) cycle, determine the content of organic acids. Citrate and malate are the main organic acids, whereas oxalate, succinate, isocitrate, fumarate, and aconitate are minor ones. **3. Volatile compounds**—Several volatile compounds are produced during ripening process in fruits. Differences in their relative abundance exist. For instance, hexanal is usually present at about few micrograms level per gram fresh weight, whereas β-ionone is present in nanograms per gram. In ripe tomato, approximately 400 volatile compounds have been reported. Some of the volatiles are salicylaldehyde, guaiacol, eugenol, toluene, phenol, styrene, etc. Mostly, important volatile compounds related to flavor are derivatives of essential nutrients, such as phenylalanine, leucine, isoleucine, or linolenic acid.

27.2.4 Antioxidants and Bioactive Compounds

Fruits are rich sources of phytochemical compounds (also called as bioactive compounds) and are considered to be beneficial sources of antioxidants. Numerous bioactive compounds have been shown to play roles in preventing or ameliorating various chronic human diseases. Phytochemicals present in fruits have variable chemical structures and functions, and they are categorized into phenolic

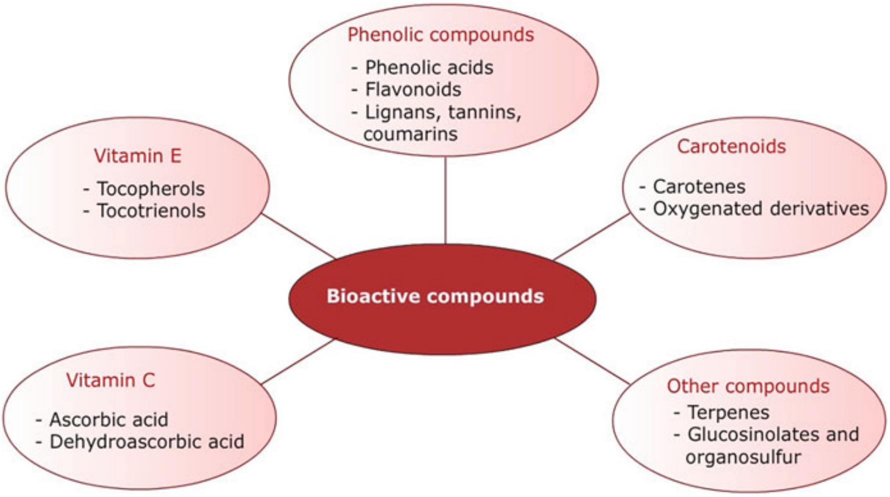


Fig. 27.4 Different categories of bioactive compounds reported during fruit ripening

compounds, carotenoids, and vitamins (C and E) (Fig. 27.4). Major antioxidant and bioactive compounds found in the mature fruits include tocopherols (vitamin E), phenolic acid, flavonoids, tannins, coumarins, carotenoids, ascorbic acid, and terpenes. Noticeable differences in the antioxidant properties of tomato have been reported in different developmental stages. Phenolic compounds encompass a wide diversity of compounds. Mostly, they are derived from phenylalanine and tyrosine and exhibit variable degrees of hydroxylation, methoxylation, and glycosylation. Various factors influence the content of phenolic compounds in fruits. The most common phenolic acids in fruits are caffeic and gallic acid, and thus the total phenol content is usually determined in terms of caffeic or gallic acid. Flavonoids include flavonols, flavones, isoflavones, flavanols, flavanones, proanthocyanidins and anthocyanidins. Rutin, luteolin, and apigenin are the most common flavones. Quercetin, kaempferol, catechin, and epicatechin are typical flavonols present in fruits. Endogenous antioxidant activity is improved by phenolic compounds. They also protect the developing fruit against oxidative damage caused by free radicals. For this reason, fruits containing phenolics are generally associated with health-promoting properties and prevention of several degenerative diseases.

27.3 Climacteric and Non-climacteric Fruit Ripening

Two different types of fruit ripening patterns have been identified on the basis of the presence or absence of a characteristic rise in the rate of respiration associated with ripening. These are referred as climacteric and non-climacteric types, respectively. The term “climacteric” was used to describe the rapid increase in respiration associated with fruit maturation. However, the definition of climacteric has changed

Table 27.3 Some of the climacteric and non-climacteric fruits

Climacteric fruit	Non-climacteric fruit
Apple (*Malus domestica*)	Bell pepper (*Capsicum annuum*)
Apricot (*Prunus armeniaca*)	Grapes (*Vitis vinifera*)
Banana (*Musa balbisiana*)	Lemon (*Citrus chinensis*)
Guava (*Psidium guajava*)	Litchi (*Litchi chinensis*)
Jackfruit (*Artocarpus heterophyllus*)	Orange (*Citrus sinensis*)
Kiwi (*Actinidia deliciosa*)	Pineapple (*Ananas comosus*)
Papaya (*Carica papaya*)	Pomegranate (*Punica granatum*)
Passion fruit (*Passiflora edulis*)	Raspberry (*Rubus idaeus*)
Pear (*Pyrus communis*)	Strawberry (*Fragaria vesca)*
Tomato (*Solanum lycopersicum*)	Cucumber (*Cucumis sativus*)

over time, and it now usually refers not only to an increase in respiration but also to elevated ethylene production at the onset of fruit ripening. Table 27.3 lists some of the climacteric and non-climacteric fruits. Climacteric fruits are characterized by an increase in respiration with a concomitant and rapid production of ethylene with the initiation of ripening. The considerable increase in ethylene production may take place before or just after the respiratory peak. In non-climacteric fruits, no rise in respiration rate is observed during ripening, and the process of ripening proceeds relatively slowly. Furthermore, climacteric fruits ripen even if harvested at an immature stage in contrast to non-climacteric fruits which do not ripe when plucked unripe. Figure 27.5 illustrates the comparative account of climacteric and non-climacteric patterns of fruit ripening processes in terms of ethylene production, CO_2 evolution, and the physical parameters, such as color and firmness of fruits. Tomato is one of the most widely studied model systems for fruit ripening in climacteric fruits (Box 27.2). In climacteric fruits, ethylene plays an important role in the fruit ripening process. In such fruits, initiation of ripening is coupled with a burst of ethylene production which further triggers its autocatalytic production. Subsequently, fruit ripening is accelerated, and dramatic changes in color, texture, and aroma of fruits become evident. Depending on the stage of fruit development, plants producing climacteric fruits exhibit two systems of ethylene production. System 1 operates in immature climacteric fruits and vegetative tissues in which ethylene inhibits its own biosynthesis by negative feedback. Mature climacteric fruits and senescing petals in some species exhibit system 2 for ethylene production in which ethylene stimulates its own biosynthesis. In system 2, the positive feedback for autocatalytic biosynthesis of ethylene ensures ripening of the entire fruit even after the commencement of ripening. Exposure to exogenous ethylene induces a rapid increase in autocatalytic ethylene production in mature climacteric fruits, thereby hastening the onset of the climacteric rise in ethylene and other changes associated with ripening. However, ethylene treatment of fruits at pre-climacteric stage results in gradual increase in the respiration rate due to rise in ethylene concentration, but it does not trigger further autocatalytic endogenous ethylene production or ripening. Treatment of fruits with inhibitors of the ethylene action,

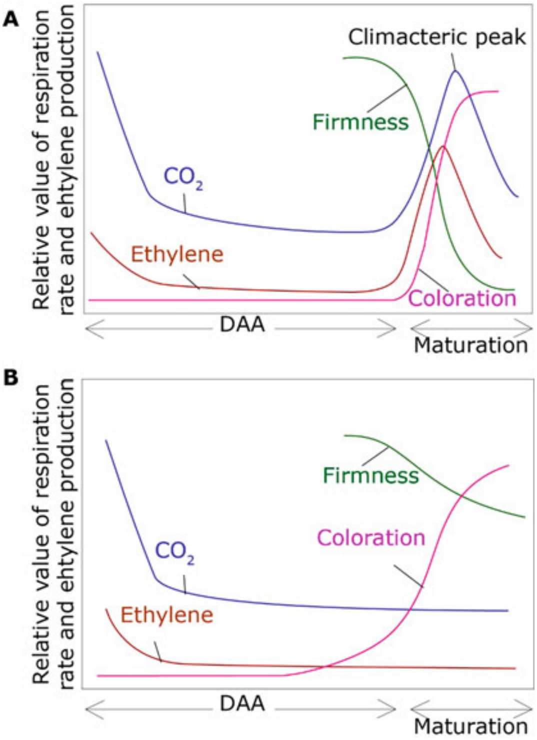


Fig. 27.5 Typical pattern of fruit ripening. (**a**) Climacteric fruits exhibit increased respiration rate concomitant with rapid increase in ethylene production during maturation phase. (**b**) In non-climacteric fruits, increase is neither observed in respiration rate nor in ethylene production. Increase in coloration and loss of firmness are relatively slower as compared to that in climacteric fruits. *DAA* Days after anthesis

such as 1-methylcyclopropane (1-MCP), suppresses or delays fruit maturation. Transgenic tomatoes, in which ethylene production is inhibited by the antisense-induced repression of the 1-aminocyclopropane-1-carboxylic acid (ACC) synthase (ACS) and ACC oxidase (ACO) genes (which encode ethylene biosynthesis enzymes), exhibit delayed fruit maturation. These observations indicate the pivotal role played by ethylene in regulation of ripening process of fruits. Although continuous ethylene action is necessary for adequate ripening, the process in climacteric fruits is not necessarily regulated in an ethylene-dependent manner. In melon fruit, flesh softening and membrane degradation are regulated by both ethylene-dependent and -independent processes. However, some of the processes such as detachment of the abscission zone and color change of the rind, which are associated with chlorophyll degradation and an increase in yellow carotenoids, are regulated by ethylene. In papaya, the degreening of the skin and flesh softening are only partially dependent on ethylene, while production of volatile aromatic compounds is strictly dependent on ethylene. In case of pear, fruit softening is completely suppressed in the absence of ethylene, whereas in melon and papaya, the softening process shows only a partial dependence on ethylene. Thus, ripening of climacteric fruits is under both developmental and ethylene-dependent regulation. The degree of dependence on ethylene varies with respect to individual ripening parameters and the diversity of the species and cultivars.

Table 27.4 Comparison of the physiology of climacteric and non-climacteric fruit ripening

S. No.	Characteristic feature	Climacteric fruit	Non-climacteric fruit
1.	Endogenous ethylene levels prior to start of fruit ripening	Increases	Does not increase
2.	Respiration rate prior to onset of fruit ripening	Increases	Either remains unchanged or there is steady decline until senescence
3.	Effect of exogenous ethylene treatment		
	i. Production of endogenous ethylene	Autocatalytic induction of endogenous ethylene production if treated at mature climacteric fruit stage. However, at pre-climacteric stage, endogenous ethylene production is not triggered	Not triggered
	ii. Respiration rate	Increases	Increases
	iii. Acceleration of fruit ripening	Yes	No. In some fruits, such as citrus, degreening is seen

In non-climacteric fruits, maturation and ripening processes occur without a burst of ethylene production. Increase in the rate of respiration has also not been observed. However, a certain level of endogenous ethylene is produced in non-climacteric fruits. Exogenous ethylene treatment increases respiration rate but does not trigger the production of endogenous ethylene, and fruit ripening is not accelerated in non-climacteric fruits. A comparative account of important events taking place in climacteric and non-climacteric fruits accompanying ripening process is summarized in Table 27.4. Recently, the involvement of ethylene in strawberry fruit ripening has been reported (Box 27.3). Respiration rate in strawberry fruit is stimulated by ethylene in a dose-dependent manner and results in slight acceleration of the cell wall softening process. There is an increase in the expression of ethylene receptor which increases the response to elevated ethylene levels in strawberry, indicating that a low level of ethylene is sufficient to trigger ripening process. Exogenous application of ethylene in citrus accelerates respiration rate and stimulates chlorophyll degradation and carotenoid biosynthesis. Treatment with ethylene antagonists, such as 2, 5-norbornadiene and silver nitrate, prevents degreening process.

27.4 Fruit Ripening: An Oxidative Phenomenon

Fruit ripening is a complex process and involves several oxidative processes, such as peroxidative damage, loss of membrane integrity, increase in ion leakage, and accumulation of lipid peroxides. Accumulation of reactive oxygen species (ROS),

Table 27.5 Variations in the activity of various ROS-scavenging enzymes in ripe fruits

		Antioxidant enzyme				
S. No.	Name of the fruit	SOD	CAT	APX	GR	POX
1.	Tomato (*Solanum lycopersicum*)	↓	↑	↑	----	↓
2.	Guava (*Psidium guajava*)	↓	↑	↑	↓	↓
3.	Papaya (*Carica papaya*)	↔	↑	↑	↑	------
4.	Cucumber (*Cucumis sativus*)	↑	↓	↓	↑	↑
5.	Orange (*Citrus sinensis*)	↓	↓	↓	↓	↓
6.	Ber (*Ziziphus sp.*)	↔	↑	↑	↓	↓
7.	Mango (*Mangifera indica*)	↓	↓	-------	------	------

Abbreviations: *SOD* superoxide dismutase, *CAT* Catalase, *APX* Ascorbate peroxidase, *GR* Glutathione reductase, *POX* Peroxidase (↓ Decrease, ↑ Increase, ↔ Unaffected, ---- Not reported)

such as superoxide radicals ($O^{2\bullet-}$), hydrogen peroxide (H_2O_2), and hydroxyl radicals (OH^-), brings about oxidative damage to the tissue. Accumulation of ROS leads to activation of the antioxidant system. Fruit tissue possesses a natural antioxidant system that includes enzymes, such as catalase (CAT), peroxidase (POX), superoxide dismutase (SOD), ascorbate peroxidase, and glutathione reductase (GR). Components of the antioxidant system act synergistically to scavenge $O^{2\bullet-}$, H_2O_2 and OH^- radicals. Table 27.5 summarizes the variations in the activity of various ROS-scavenging enzymes in ripe fruits. Variations in the pattern of antioxidant enzyme activities could be attributed to the differential activity of various isoenzymes. During early stages of fruit ripening, the antioxidant system protects fruits from the destructive effects of gradual oxidative stress. However, oxidative damage attributed to overproduction and accumulation of ROS takes place at later stages. Inability of the antioxidant system to eliminate ROS is also responsible for oxidative damages. These free radicals initiate the destructive changes during ripening. Fruit ripening is characterized by a gradual increase in oxidative processes, and it is considered a stressful process. Several fruits, such as tomato, mango, peach, and papaya, undergo oxidative stress during ripening. Free radicals induce the process of lipid peroxidation which is responsible for the initiation of ripening-associated deteriorative changes. Increase in lipid peroxidation is often associated with decreased CAT, GR, and SOD activity. Reduced activity of important antioxidant enzymes, such as CAT and SOD, leads to oxidative stress during ripening. However, it has been proposed that this increase in oxidative stress is necessary to facilitate the metabolic changes associated with fruit ripening.

27.4.1 Role of Alternative Oxidase (AOX)

Alternative oxidase (AOX) plays a role in ripening by mediating ethylene signaling as well as scavenging of ROS. AOX levels increase dramatically in ripe tomato fruits. Likewise, increased AOX levels have been reported in mango and apple fruits in which climacteric bursts are associated with an enhanced cyanide-insensitive respiration. The multigene family for the AOX expresses differentially during fruit ripening. Abundance of AOX protein peaks at the ripe stage. Pattern of protein accumulation indicates that an increase in cytochrome chain components plays an important role in facilitating the climacteric burst of respiration and fruit ripening. Transgenic tomato plants with reduced AOX levels, i.e., LeAOX1, show retarded ripening and reduction in carotenoid content, respiration, and ethylene production. Downregulation of ripening-associated genes has also been reported. Treatment of such mutants with 1-methylcyclopropane fails to induce ripening. However, inhibition of the AOX knockout mutant, i.e., 35S-AOX1a, is less effective. Mutant that overexpresses AOX levels, i.e., LeAOX1a, shows increased lycopene content and similar pattern of fruit ripening as reported in wild tomato. Therefore, it has been suggested that both AOX and ethylene are essential for fruit ripening in tomato, and inhibition of both is required to halt ripening process completely.

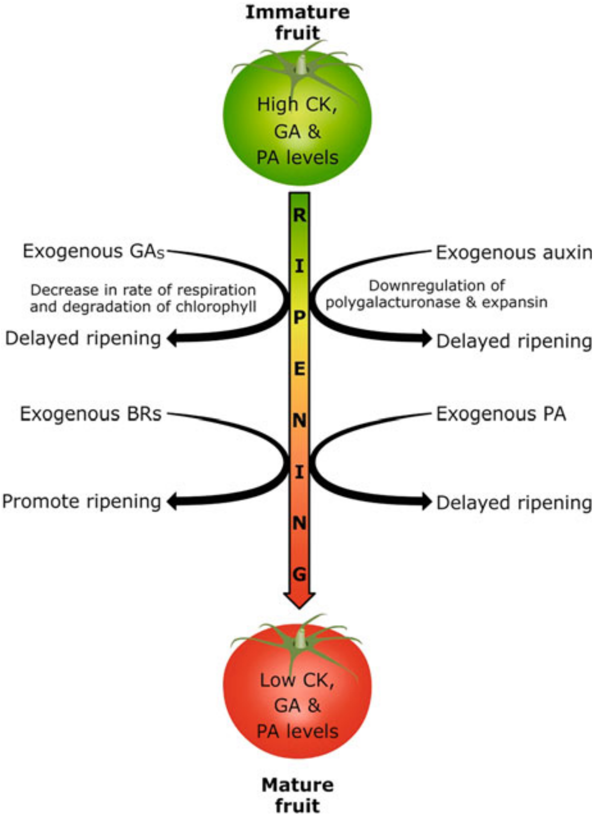
27.5 Role of Phytohormones

Abscisic acid (ABA) regulates the expression of ripening-related genes in strawberry. It has recently been implicated as a ripening factor both in climacteric (e.g., tomato, peach, and grapes) and non-climacteric fruits (e.g., apple, strawberry). In tomato, accumulation of endogenous ABA prior to ethylene burst has been reported, suggesting a possible role of ABA in the modulation of ethylene accumulation. Exogenous application of ABA promotes both ethylene biosynthesis and fruit ripening. However, application of nordihydroguaiaretic acid (NDGA), an inhibitor of ABA biosynthesis, suppresses fruit coloration and softening process. Four different phenomena are brought about by ABA during citrus fruit ripening:

1. Increase in endogenous ABA levels leads to ethylene production. Ethylene treatment, however, does not cause any increase in ABA levels.
2. Suppression of increase in ABA content accompanying fruit ripening by NDGA treatment whereas ethylene production remains unaffected.
3. Inhibition of ethylene production by ABA+1-MCP without any negative effect on ABA content.
4. Inhibition of fruit ripening and softening by treatment with NDGA and 1-MCP.

In tomato, both ABA and ethylene control the expression of genes encoding the cell wall- degrading enzymes. It has been suggested that the presence and perception of both ABA and ethylene are important for normal fruit ripening process. Auxin functions as a negative regulator of fruit ripening in a number of climacteric and non-climacteric fruits. Exogenous auxin delays the onset of ripening in avocado, pear, grape, and strawberry. In strawberry, the expression levels of genes encoding polygalacturonase and expansin (which are related to cell wall degradation) are repressed by auxin. High levels of cytokinins are often found in immature fruits as compared to mature fruits (Fig. 27.6). Exogenous application of cytokinin has been reported to delay fruit ripening in both climacteric and non-climacteric fruits.

Fig. 27.6 Depiction of effect of exogenous application of phytohormones, such as auxin, gibberellins (GAs), brassinosteroids (BRs), and polyamines (PAs) on the process of fruit ripening



Increased concentration of GAs is generally evident during early stages of fruit development. Ripening process is characterized by decline in GA concentration. It seems that high levels of GA might be essential in promoting early fruit development and inhibiting ripening. Exogenous GA application leads to the delay in ripening process. GA application slows down respiratory activity, anthocyanin synthesis, and chlorophyll degradation and delays softening process in strawberry. Negative regulation of ripening process is often associated with the role of cytokinin and GA in increasing fruit size. Concentrations of jasmonates, such as jasmonic acid and methyl jasmonate, attain a peak just prior to ethylene burst in climacteric fruits. In non-climacteric fruits, however, higher levels of jasmonates are observed during early stages of fruit development which decrease during the ripening (later) phase. In grapes, levels of endogenous brassinosteroids (BR) increase with ripening. Exogenous application of BRs promotes ripening process in grapes and tomatoes. However, further work is required to ascertain the role of BRs in fruit ripening process. Polyamine (PA) levels have been reported to be high during early fruit development but decline following commencement of ripening. Putrescine and spermidine are usually the most and least abundant polyamines, respectively, in the developing fruits. Exogenous application of PAs delays ripening process, possibly due to the

Table 27.6 Regulation of ripening process in both climacteric and non-climacteric fruits by various phytohormones other than ethylene

S. No.	Hormone	Climacteric fruits	Non-climacteric fruits
1.	Auxin	Negative regulator	Negative regulator. Downregulation of genes related to cell wall degradation, such as polygalacturonase and expansins
2.	GA	Inhibition of ripening	Delay of fruit softening
3.	Cytokinin	Delay in fruit ripening	Delay in fruit ripening
4.	ABA	Positive regulator	Positive regulator
5.	Jasmonates	Enhancement/delay of ripening, e.g., peach	Enhancement of ripening, e.g., blackberries
6.	Brassinosteroids	Increase in ripening-related parameters, such as an increase in sugar levels, reduction in chlorophyll levels, increased lycopene accumulation, and a decrease in ascorbic acid levels, e.g. tomato	Enhancement of ripening, e.g., grape
7.	Polyamines (PA)	Delay in some ripening-related parameters, e.g., tomato	Expression of PA biosynthetic gene increases, e.g., grape

antagonistic interaction between PAs and ethylene. In climacteric fruits, the role of ethylene is considered as the most crucial factor for ripening process; however, fruit ripening seems to be regulated by the coordinated action of phytohormones irrespective of climacteric and non-climacteric pattern of fruit ripening. Table 27.6 summarizes the regulation of ripening process by phytohormones other than ethylene in both climacteric and non-climacteric fruits.

27.6 Nitric Oxide and Ethylene CrossTalk

In many fruits, especially tropical ones, the increase in ethylene production during ripening leads to induction of senescence which adversely affects their post-climacteric storage. As a result, the quality of these fruits in terms of nutrients, color, texture, and flavor is drastically reduced. There are many signals that regulate ethylene production and its perception in different parts of plants. Nitric oxide (NO) directly interferes with ethylene production during fruit ripening either by suppression of the activity of various enzymes implicated in ethylene biosynthesis or through direct stoichiometric inhibition. In ethylene biosynthesis pathway, the autocatalytic activity of ethylene results in the formation of S-adenosyl methionine (SAM) from methionine (MT) which is catalyzed by SAM synthetase. 1-aminocyclopropane-1-carboxylic acid synthase (ACC synthase) converts SAM into ACC which subsequently get oxidized to ethylene by the action of ACC

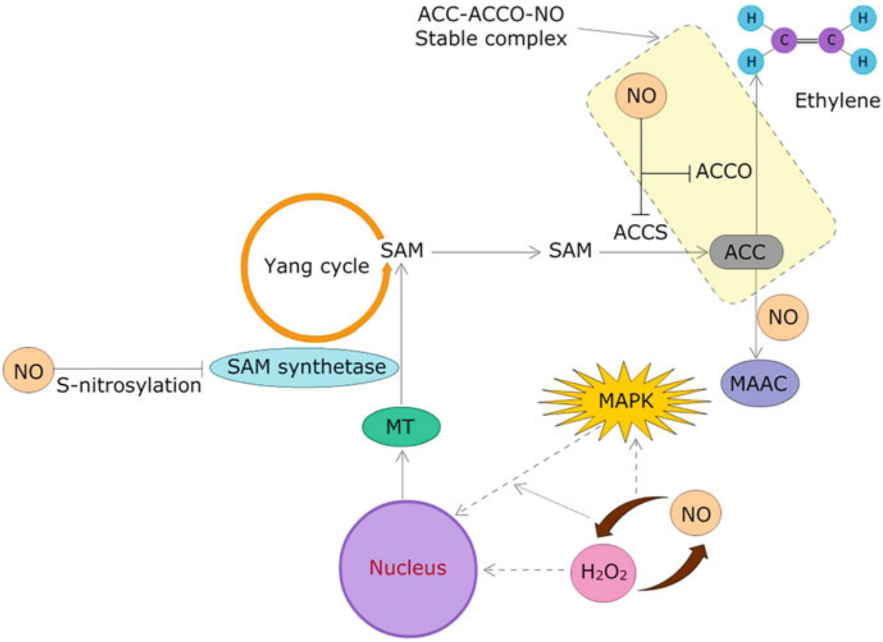


Fig. 27.7 Schematic model depicting effect of NO on ethylene biosynthesis during fruit ripening. *NO* Nitric oxide, *MT* Methionine, *SAM* S-adenosyl methionine, *ACCS* 1-aminocyclopropane-1-carboxylic acid synthase, *ACC* 1-aminocyclopropane-1-carboxylic acid, *MAAC* 1-melonyl aminocyclopropane 1-carboxylic acid, *ACCO* 1-aminocyclopropane-1-carboxylic acid oxidase, H_2O_2 Hydrogen peroxide. H_2O_2 and NO reciprocally stimulate production of each other

oxidase. NO has been known to adversely affect ethylene biosynthesis due to S-nitrosylation of methionine adenosyltransferase (MAT), resulting in inhibition of SAM production. NO binds to ACC oxidase thereby inhibiting the autocatalytic biosynthesis of ethylene. This binding causes formation of ACCO-NO complex. Eventually, ACCO-NO complex forms a stable ternary complex, ACC-ACCO-NO thereby inhibiting ACO activity and consequent ethylene production (Fig. 27.7). NO also regulates ethylene production through interaction with hydrogen peroxide (H_2O_2). H_2O_2 effectively induce the biosynthesis of ethylene via gene transcription. Reciprocal interaction of NO and H_2O_2 also affects mitogen-activated protein kinase (MAPK)-mediated downstream components of ethylene biosynthesis. Being highly reactive, NO has the potential to directly trigger redox processes related to ROS production. Through post-translational modifications, such as S-nitrosylation and tyrosine nitration, NO targets transition metals, such as Cu, Zn, and Fe, which are components of various signaling molecules, receptors, enzymes, transcription factors, and proteins. NO reduces the production of ethylene leading to the delay in the expression of enzymes which are responsible for cell wall degradation, lignification, and pigmentation of fruits, thereby significantly extending the shelf life of fruits. Postharvest application of exogenous NO causes delayed ripening in many fruits, such as strawberry, avocado, and mango. NO treatment reduces disintegration of the cell membrane with reduced electrolyte leakage leading to facilitate better retention of the cellular components. Furthermore, exogenous application of NO has also been reported to be beneficial for the protection of fruits from a variety of microbes, such as *Aspergillus niger*, *Monilinia fructicola*, *Penicillium italicum*, and *Rhizopus nigricans*.

27.7 Transcriptional Regulation

Characterization of certain tomato mutants, such as non-ripening (*NOR*), ripening inhibitor (*RIN*), green-ripe (*GR*), colorless non-ripening *(CNR*), and never-ripe (*NR)*, has contributed significantly toward understanding the mechanisms that regulate ripening (Fig. 27.8). The *RIN* mutant encodes a partially deleted MADS-box protein of the SEPALLATA clade (*SEP4*). *CNR* expression is due to an epigenetic change which alters methylation of SQUAMOSA promoter-binding (SPB) protein. *NOR* is a member of the NAC-domain (NAM, ATAF1/ATAF2, and CUC2) transcription factor family. *GR* mutant has a mutation in a gene which encodes a conserved protein that inhibits ethylene signaling. Recently, transcriptome and proteome analyses have shown that *NOR* and *RIN* act synergistically to control ripening. *NOR* has been suggested to operate upstream of *RIN* because *NOR* has more profound effect on ripening-related gene expression. Chromatin immunoprecipitation (ChIP) and transcriptome analyses have revealed the interaction of *RIN* locus

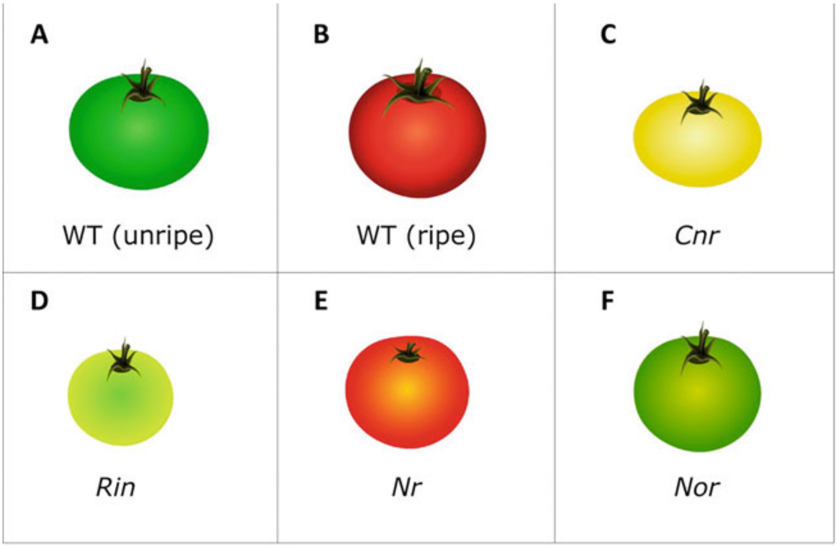


Fig. 27.8 Wild type and different types of tomato mutants. (**a**) Wild-type unripe tomato. (**b**) Wild-type ripe tomato. (**c**) *Colorless non- ripening (Cnr)* mutant. (**d**) *Ripening inhibitor (Rin)* mutant. (**e**) *Never ripe (Nr)* mutant. (**f**) *Non- ripening (Nor)* mutant

Table 27.7 Different mutants of tomato and their respective modification exploited in fruit ripening studies

S. No.	Name of the mutant	Modification
1.	Non-ripening (NOR)	Member of the NAC-domain (NAM, ATAF1/ATAF2, and CUC2) transcription factor family
2.	Ripening inhibitor (RIN)	Encodes a partially deleted MADS-box protein of the SEPALLATA clade (*SEP4*)
3.	Green ripe (GR)	Mutation in a gene which encodes a conserved protein that inhibits ethylene signaling
4.	Colorless non-ripening (CNR)	Epigenetic change which alters the promoter methylation of SQUAMOSA promoter-binding (SPB) protein

with over 200 genes. This interaction modulates the expression of *RIN* target genes by either activation or repression. *CNR* and *NOR* are the *RIN* target genes, which are the major regulators of ripening process. *RIN* target genes are also implicated in pathways active during the transition from green unripe fruits to ripe fruits, such as carotenoid accumulation, chlorophyll breakdown, and ethylene biosynthesis. Table 27.7 shows the modification that has given rise to different tomato mutants.

MADS-box genes are involved in the regulation of fruit ripening in a variety of fruits, such as banana, tomato, and strawberry. Numerous fruit development and ripening-related genes have been isolated and characterized using differential gene expression patterns and biochemical functions, including the regulatory genes involved in ethylene biosynthesis. *RIN* and *CNR* encode certain transcription factors, which play important roles in the regulation of the ethylene biosynthesis pathway. A MADS-box transcription factor *MADS-RIN*, encoded by *RIN* locus, is induced at the onset of fruit ripening. In transgenic tomatoes, the suppression of *MADS-RIN* gene expression results in the formation of non-ripening. *RIN* mutants were rescued after complementation with MADS-RIN gene, thus demonstrating that *MADS-RIN* gene is essential for normal fruit ripening. *MADS-RIN* gene interacts with the promoter for *ACC synthase* gene suggesting its role in the regulation of ethylene biosynthesis. The expression of several ripening-related genes, such as *polygalacturonase* (*PG)*, *expansins (EXP)*, *lipoxygenase (LOXC)*, and *phytoene synthase (PSY1)*, is also controlled by *MADS-RIN* gene. These genes encode proteins involved in the metabolism of the cell wall and carotenoid biosynthesis, such as *PSY1*. Expression of some genes, such as *LOXC* and alcohol dehydrogenase, is also regulated by *MADS-RIN*. Strawberry, a non-climacteric fruit, has a fruit-specific *LeMADS-RIN* orthologue, which indicates the presence of ethylene-independent regulatory pathway involving MADS-box genes both in climacteric and non-climacteric fruit ripening. Furthermore, it has been suggested that *MADS-RIN* and ethylene signaling act synergistically for normal fruit ripening. Figure 27.9 summarizes the regulation of climacteric and non-climacteric patterns of fruit ripening.

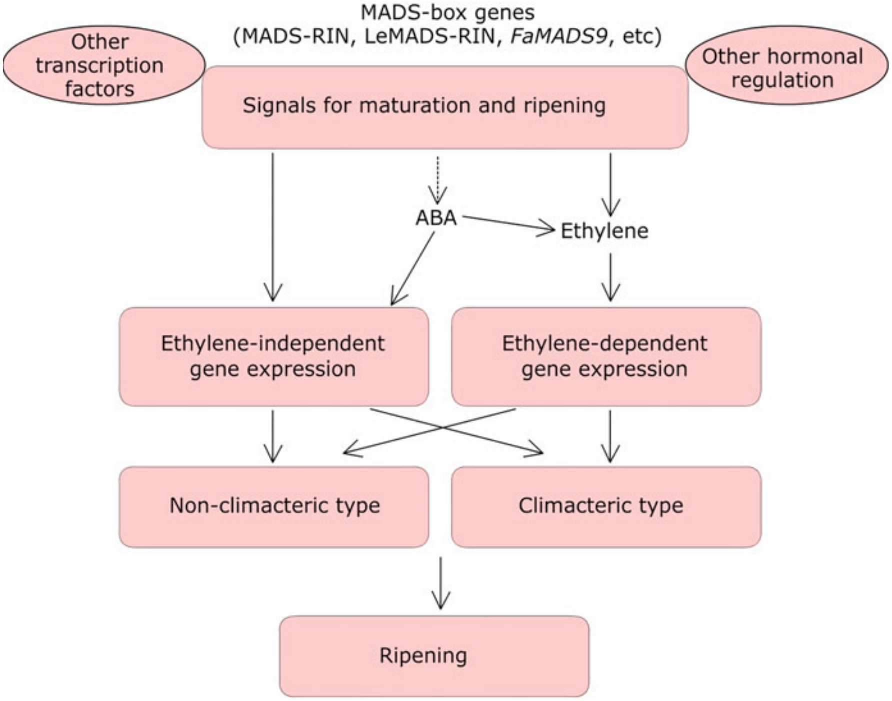


Fig. 27.9 Schematic diagram illustrating the involvement of various genes, transcription factors, and hormones in regulation of climacteric and non-climacteric type of fruit ripening via ethylene-dependent and ethylene-independent pathways. *MADS-RIN* MADS-box transcription factor, *LeMADS-RIN* MADS-RIN in tomato, *FaMADS9* SEP1-/SEP2-like gene in strawberry, and *ABA* Abscisic acid

27.8 Epigenetic Regulation of Gene Expression

Epigenetic variations generally affect gene expression at the level of chromatin organization, mainly through DNA methylation, cytosine methylation, and histone modifications without affecting primary DNA sequence. Molecular nature of *CNR* mutant phenotype has effectively explained the impact of cytosine methylation on fruit ripening in tomato. *NR* phenotype is due to the heritable cytosine hypermethylation, including that in the *CNR* gene promoter region. During fruit development, in wild tomato, demethylation of *CNR* promoter takes place in specific region just before the onset of ripening. Thus, it has been hypothesized that DNA methylation seems to be crucial in the regulation of fruit ripening. Epigenomic studies have highlighted mainly three important aspects in the ripening process. 1. The degree of methylation of regions upstream the transcription start sites (TSS) decreases gradually along with fruit development. The decline in the degree of methylation has not been observed for *CNR* and *RIN*, whose CG methylation levels were constantly higher at TSS. 2. Promoters of typical ripening-related genes are gradually demethylated during fruit development. Analysis of methylation status of TSS has demonstrated their progressive demethylation during the transition from green unripe to red ripe fruits. 3. Analysis of epigenetic variation in *Arabidopsis* revealed that at least one-third of expressed genes are methylated in their coding region and only 5% of genes are methylated within promoter regions. However, the promoter-methylated genes have a higher degree of tissue-specific expression. It has been suggested that progressive demethylation of ripening-related gene promoters may be necessary for the binding of transcriptional regulators triggering the accumulation of ripening-related genes.

Summary

- Fruit development involves four different phases characterized by fruit set, rapid division of cells, cell enlargement, and onset of ripening. Different ripening stages have distinct physicochemical markers in different model systems.
- Fruits are generally categorized into climacteric and non-climacteric types on the basis of respiratory activity and are associated with ethylene biosynthesis during ripening process. Climacteric fruits are characterized by an increase in respiration rate and ethylene production at the onset of ripening, whereas non-climacteric fruits are characterized by the absence of ethylene production burst and respiratory rise. However, they have the capacity to produce ethylene endogenously. Climacteric fruits possess two systems for ethylene production depending on development stage of fruit. Initial stages of ripening have been characterized by the auto inhibition of ethylene biosynthesis by negative feedback (System 1), whereas at later stages, ethylene autocatalyzes its biosynthesis by positive feedback loop (System 2). Involvement of other phytohormones such as auxins, ABA, GA, jasmonates, and BRs in the regulation of the fruit ripening has also been reported.
- Cell wall architecture is progressively modified during fruit ripening, due to metabolic changes leading to softening (textural change) of the fruit due to the activity of various cell wall-degrading enzymes. Transition from green to a variety of colors due to development of different pigments, such as carotenoids, anthocyanins is responsible for the taste and aroma of the fruits. Sugars, organic acids, and volatiles are essential for intensification of flavors and fragrances.
- Oxidative processes during ripening involve an increase in ion leakage due to loss of membrane integrity and decrease in membrane fluidity. Antioxidant system becomes activated due to rise in the level of reactive oxygen species. NO inhibits ethylene biosynthesis via binding to ACC oxidase (ACCO). ACCS and ACCO genes are downregulated by NO leading to inhibition of ethylene.
- MADS-box genes, such as *MADS-RIN*, have been implicated in the regulation of fruit ripening process. Synergistic and coordinated actions of transcription factors and ethylene along with other phytohormones regulate the normal process of fruit ripening.

Multiple-Choice Questions

1. A fruit is best defined as:
    (a) Ripened ovule enclosing one or more seeds
    (b) Ripened ovary enclosing one or more seeds
    (c) Ripened endosperm enclosing one or more seeds
    (d) Ripened nucellus enclosing one or more seeds
2. Which of the following statements is incorrect in the context of fruit development?
    (a) Phase I is characterized by anthesis and successful fertilization.
    (b) Phase II is characterized by rapid cell divisions during embryo development.
    (c) Phase III is characterized by cessation of cell divisions and cell enlargement.
    (d) Phase IV is characterized by enhanced carotenoid biosynthesis.
3. Which of the following is a non-climacteric fruit?
    (a) *Solanum lycopersicum*
    (b) *Musa balbisiana*
    (c) *Fragaria vesca*
    (d) *Artocarpus heterophyllus*
4. In climacteric fruits, exogenous application of ethylene results in:
    (a) Autocatalytic induction of endogenous ethylene production if treated at pre-climacteric stage
    (b) Autocatalytic induction of endogenous ethylene production if treated at mature climacteric fruit stage
    (c) Increase in respiration rate
    (d) Acceleration of fruit ripening
5. Which of the following statement is correct?
    (a) Exogenous application of gibberellic acid (GA) leads to decrease in rate of respiration and degradation of chlorophyll.
    (b) Exogenous application of auxin causes upregulation of polygalacturonase and expansin.
    (c) Exogenous application of cytokinin enhances fruit ripening.
    (d) Abscisic acid (ABA) is a positive regulator of fruit ripening.
6. Development of fruit without fertilization is called:
    (a) Apospory
    (b) Parthenogenesis
    (c) Polygamy
    (d) Parthenocarpy

Answers

1.b 2.a 3.c 4.a 5.b 6.d

Suggested Further Readings

Gapper NE, Giovannoni JJ, Watkins CB (2014) Understanding development and ripening of fruit crops in an 'omics' era. Horticl Res 23:14034–14044

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