Chapter 3. Ripening and changes occurring during ripening

Ripening is the process by which fruits attain their desirable flavour, quality, colour, palatable nature and other textural properties. Ripening is associated with change in composition i.e. conversion of starch to sugar. On the basis of ripening behavior, fruits are classified as climacteric and non-climacteric fruits.

Climacteric fruits: Climacteric fruits are defined as fruits that enter ‘climacteric phase’ after harvest i.e. they continue to ripen. During the ripening process the fruits emit ethylene along with increased rate of respiration. Ripe fruits are soft and delicate and generally cannot withstand rigours of transport and repeated handling. These fruits are harvested hard and green, but fully mature and are ripened near consumption areas. Small dose of ethylene is used to induce ripening process under controlled conditions of temperature and humidity.

Climacteric fruits are:

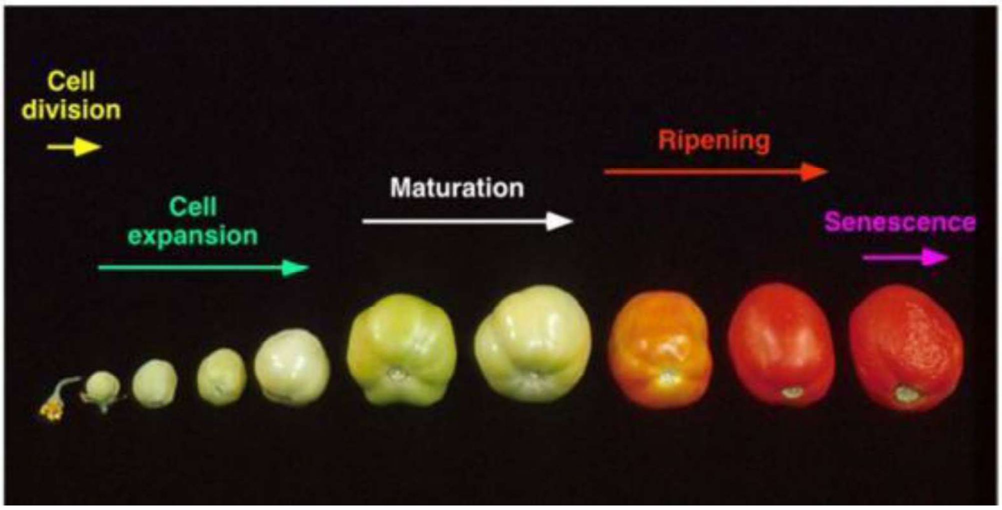
*Mango *Banana *Papaya *Guava *Sapota *Kiwi *Fig *Apple *Passion fruit *Apricot *Plum *Pear

These fruit in fully ripe state are too delicate to withstand transportation over long distances and should preferably be ripened near the consumption area.

Non-Climacteric fruits: Non-climacteric fruits once harvested do not ripen further. Nonclimacteric fruits produce very small amount of ethylene and do not respond to ethylene treatment. There is no characteristic increased rate of respiration or production of carbon dioxide. In order to improve external skin colour and market acceptance, citrus like orange, lemon, mousambi and kinnow can be treated with ethylene, as a de-greening agent. Ethylene treatment breaks down the green chlorophyll pigment in the exterior part of the peel and allows the yellow or orange carotenoid pigments to be expressed.

Non-Climacteric fruits are:

*Orange *Mousambi *Kinnow *Grapefruit *Grapes *Pomegranate *Litchi *Watermelon *Cherry *Raspberry *Blackberry *Strawberry * Carambola *Rambutan * Cashew



Several processes take place as fruit ripen and become edible, and then senesce. These changes may take place while fruit are still attached to the plant or after harvest. Tomato, banana and avocado are examples of fruit that at harvest can be at a mature green but unripe stage and are inedible until subsequent ripening processes have occurred. In contrast, strawberry, orange, boysenberry and grape are examples of fruit that need to stay on the tree or vine until ready to eat in order to have their desired eating characteristics.

The maturity has been divided into two categories i.e. physiological maturity and horticultural maturity.

1. **Physiological maturity:** It is the stage at which a plant or plant part continues ontogeny (complete developmental history of an organism from egg/spore/ bud etc. to an adult individual) even if detached from the parent plant or the point of origin. It can also be defined as the stage at which a plant or plant part is capable of further development or ripening when it is harvested i.e. ready for eating or processing. Ex. A French bean pod or okra pod is at its physiological maturity when the seeds are fully developed and the pod is lignified which will dehisce with little pressure.
2. **Horticultural maturity / Harvest maturity**: It may be defined as the stage at which a plant or plant part possesses all the prerequisites for use by consumers for a particular purpose, i.e. local, distant, export market **(shipping** maturity) or exhibition or processing (processing maturity), culinary maturity, desert maturity etc. Ex. A pod vegetable is matured when it is tender with maximum size. Horticultural maturity stage of tomato if harvested for long distance transportation would be the "turning stage of skin from green to red", while the optimum stage of harvesting of the same crop for home use or local markets would be "when the fruits have attained full red colour".

Several major changes take place as fruits ripen, and taken collectively they characterise ripening processes.

These changes make the ripe fruit attractive to animals, which in eating the fruit will disperse the seeds and enlarge the range and improve the survival chances of the next generation of the plant. Lignified pits and seeds encased in a fibrous core might be discarded after eating the flesh, whereas smaller seeds might pass through the animal's digestive system and be deposited with the animal's excrement.

1. **Change in carbohydrate composition -** The ripening induces the breakdown of carbohydrate polymers, by various carbohydrases and leads to near total conversion of starch to sugars. This has the dual effect of altering the taste and texture of the produce. The increase in sugar renders the fruit much sweeter and therefore more acceptable.

2. **Change in color -** Pigments are essential for the attractiveness of fruits and accumulate most often in the skin during the ripening process. Color is often the major criteria used by consumers to determine whether the fruit is ripe or unripe. As fruit matures and ripen, green colour decline and develops yellow, red or other colours due to the presence of accessory pigments, which are characteristic of the various cultivars.

➢ **Formation of pigments:** During ripening there is formation of pigments mainly carotenoids and anthocyanins. Carotenoids include β-carotene, xanthophyll esters, xanthophylls and lycopene while anthocyanins include cyaniding, pelargonidin, petunidin etc

➢ **Degradation of pigments:** Climacteric fruits show rapid loss of green colour with attainment of optimum eating quality. Some non-climacteric fruits also exhibit a marked loss of green colour with attainment of optimum quality. The green colour loss is due to the degradation of chlorophyll structure. The main factors responsible for chlorophyll degradation are: pH changes, oxidative systems and enzyme chlorophyllase.

3. **Flesh softening and textural changes -** Textural change is the major event in fruit softening, and is the integral part of ripening, which is the result of enzymatic degradation of structural as well as storage polysaccharides. Cell walls of fruit undergo a natural degradation during fruit ripening, reducing cell wall firmness and intercellular adhesion. This leads firstly to the attainment of a desirable eating texture and then, as senescence begins, to a loss of this desirable texture. Enzymatic degradation of structural as well as storage polysaccharides occurs - pectin, cellulose, and hemicelluloses.

4. **Taste:** Taste depends on the proper proportion of sugars and acids. So, it is convenient to measure taste as sugar-acid ratio (Brix-acid ratio). Acidity and astringency gradually disappear, while sweetness increases due to conversion of starch to sugars during the course of fruit ripening. Starch content of banana decreases from initial 21% to about 15% in ripened fruit. This is accompanied by accumulation of sugars mainly sucrose to the extent up to 20% by fresh weight.

5. **Flavour changes -** The increase in flavor and aroma during fruit ripening is attributed to the production of a complex mixture of volatile compounds and degradation of bitter principles, flavanoids, tannins, and related compounds.

6. **Aroma:** Aroma plays an important part in the development of optimal eating quality in most fruit. It is due to synthesis of many volatile organic compounds (often known merely as volatiles) during ripening phase. Together with taste, it constitutes flavour. Aroma usually develops during ripening but occasionally in storage also. During ripening enzymes break down large organic molecules into smaller one that can be volatile (evaporate into the air) and can be detected as an aroma. The flavouring compounds are found to be different in different types of fruit but all of them are volatile. The aroma of fruit is not due to a single chemical compound but it is a mixture of no. of chemicals, which may be derived from aliphatic compounds, alcohols, acetates, ketones or esters and terpinoides.

7. **Change in organic acids -** In oranges and grapefruits the acid content drops during ripening, while in lemons, there is an increase in acids. Synthesis of ascorbic acid also occurs in many fruits during ripening. Generally, the acidity decreases during ripening as organic acid are utilized in respiration of fruits.

8. **Ethylene and respiration –** Climacteric peak (increase in ethylene and respiration rate) is observed during ripening in climacteric fruits.Respiration is essential for ripening as it provides the energy required to drive many of the reactions and changes. If respiration is inhibited, ripening is also inhibited.

9. **Abscission:** During ripening the pectinase enzyme also unglue the cells of the abscission zone (the layer of cells in the pedicels often called abscission zone). So, the cells in this zone become weak and the weight of the fruit will cause it to all from the plant.
10. **Development of surface wax:** The delicate waxy or powdery substance develops on the surface of certain fruits like grape and berries.

Chapter 4. Respiration

Metabolic activity in fresh fruits and vegetables continues for a short period after harvest. The energy required to sustain this activity comes from the respiration process (Mannapperuma, 1991). Respiration involves the oxidation of sugars to produce carbon dioxide, water and heat. The storage life of a commodity is influenced by its respiratory activity. By storing a commodity at low temperature, respiration is reduced and senescence is delayed, thus extending storage life (Halachmy and Mannheim, 1991). Proper control of the oxygen and carbon dioxide concentrations surrounding a commodity is also effective in reducing the rate of respiration.

There are mainly two types of respiration: aerobic respiration and anaerobic respiration, depending upon availability of oxygen. In aerobic respiration, oxygen (O2) is the final electron acceptor while in anaerobic respiration, or fermentation, some other compound is the final electron acceptor.

Aerobic respiration

Aerobic respiration is the major biochemical process supplying energy. It involves the oxidation of certain organic compounds i.e. glucose, maleic acid, stored in the tissues. The compounds that are oxidised during this process are known as respiratory substrates. Aerobic respiration occurs in three phases: glycolysis or Embden-Meyerhoff-Parnas (EMP) pathway, the Krebs or Tri-carboxylic acid cycle and the electron transport chain.

In glycolysis, a molecule of the six-carbon sugar glucose is oxidized to two molecules of the three-carbon pyruvate. The Krebs cycle completes the oxidation of pyruvate to produce carbon dioxide (CO2) and reduced electron carriers. In the electron transport chain, a proton (H+) gradient drives the production of even more ATP and is coupled with the transfer of electrons to oxygen (O2), producing water (H2O). After the entire process of respiration is complete, much of the energy released from the glucose is recaptured in the production of ATP.

Glucose is the most favoured substrate for respiration. All carbohydrates other than glucose are converted into glucose first before they are used for respiration. Fats are metabolized into glycerol and fatty acids first and then to acetyl CoA and glyceraldehyde-3- phosphate, respectively. Proteins are degraded individual amino acids (after deamination) and then enter the respiratory pathway. The respiration is exothermic in nature and theoretically, 60% of the bond energy is lost as heat. However, calorimetry studies have shown that respiration in postharvest tissues often results in even more dissipation of energy as heat loss (90% or more) and less ATP synthesis. This heat contributes to an increase in the temperature of the commodity and is known as **vital heat or heat of respiration**. Heat of respiration is a primary consideration in designing the storage for horticultural crops.

Anaerobic respiration

Aerobic respiration is preferred energy producing pathway in fruits and vegetables. But under the limiting O2 conditions, fermentation becomes increasingly important. During this process, pyruvic acid produced during the glycolysis is converted to lactic acid, ethanol or acetaldehyde. Increase in fermentation helps the cell meet its ATP requirement under anaerobic conditions. Anaerobic respiration produces much less energy than aerobic pathway and elevated CO2 concentrations have deteriorative effect on the product quality. Anoxia results in injuries to tissue. High concentrations of fermentative metabolites are also associated with various physiological disorders like necrosis, discoloured tissues, offflavours, off-odours etc. The oxygen concentration at which anaerobic respiration starts is called extinction point. It varies between the tissue types and also species, cultivar, development stage, maturity etc.

SIGNIFICANCE OF RESPIRATION

Respiration plays a major role in the postharvest life of fresh fruits and vegetables. Respiration continues even after harvest. After harvest the produce is dependent entirely upon its own food reserves as no replenishment is there. Therefore, losses of respiratory substrates and moisture are not made up and deterioration has started. Produce' possessing a high respiration rate can be stored for longer time duration. An enhanced respiration rate is associated with perishability of food. The significant effects of respiration are:

- **Loss of substrates:** The process of respiration utilizes various substrates and thus results in loss of food reserves in the tissue, loss of taste quality and food value. During the extended storage the loss in weight can be highly significant.
- **Oxygen consumption:** For aerobic respiration, presence of oxygen is must. Reduction of oxygen concentration is a useful tool for controlling respiration rate and slowing down the senescence.
- **Carbon Dioxide production:** Accumulation of CO2 produced due to respiratory metabolism can be beneficial or harmful. Carbon dioxide concentration is also used as an effective measure for delaying senescence.
- **Release of Heat:** As respiration is exothermic in nature, heat generated raises the temperature of produce. The heat generated is one of the prime consideration for designing the packaging/ storage of horticultural crops.
- **Shelf life indicator:** Respiratory process indicates the metabolic activity of living produce and determines the post-harvest physiology and deteriorative ability of plant produce. Respiration rate is well correlated with rate of deterioration, and thus is a good measure of the storage potential of the plant produce. Generally, a higher respiration rate indicates shorter shelf life.
- **Change in quality:** The physiological processes leading to enhanced quality (e.g., color development, softening, astringency loss, and aroma production) are deeply influenced by respiration. Extremes in respiration rate results in the development of specific physiological disorders, resulting in loss of quality. Respiration is beneficial in providing carbon skeleton intermediates for pigment synthesis, flavor development, formation of ripening enzymes, fats, sterols etc.
- Classification: Respiration rate is an important criterion to compare perishability of fruits and vegetables. Depending upon respiratory rate Kader and Barrett classified fruits and vegetables into five different classes, rate as shown below:

Classification of Fruits and Vegetables Based on Respiration Rate

Class	Respiration Rate (mg carbon dioxide/kg h) 10°C	20°C	Examples
I. Very low	<10	<40	Nuts, dates, dried fruits
II. Low	10	40	Potatoes, onions, cucumbers, apple, pear, kiwi fruit, pomegranate, Chinese date
III. Moderate	10–20	40–80	Peppers, carrots, tomatoes, eggplant, citrus fruits, banana
IV. High	20–40	80–120	Peas, radish, apricot, fig, ripe avocado, cherimoya, papaya
V. Very high	>40	>120	Mushrooms, green onions, cauliflower, dill, parsley, melons, okra, strawberry, blackberry, raspberry

Source: Data from I. Burzo. *Acta Hortic.* 116:61, 1980; A. A. Kader and D. M. Barrett. In *Processing Fruits: Science and Technology, Vol. 1, Biology, Principles, and Applications.* (L. P. Somogyi, H. S. Ramaswamy, and Y. H. Hui, Eds), Technomic Publishing Co., Pennsylvania, 2003, p. 1.

Respiratory Quotient The ratio of moles of CO2 produced per mole of O2 consumed is called the respiratory quotient. RQ is as an indication of which substrates are being used in the respiratory pathway. RQ is 1 for glucose (carbohydrate) catabolism. When substrates other than glucose are respired, the RQ is different than 1. The complete oxidation of malate by the TCA cycle results in a RQ of 1.6 (oxidation of respiratory malic acid, leads to the production of additional CO2). The RQ can also exceed 1 when O2 is not involved, such as in fermentation. RQ values below 1 are expected when lipids or proteins, molecules often containing less oxygen than carbohydrates, are respired.

Respiratory Substrate – carbohydrates, lipids, and organic acids.

$$\text{Respiratory quotient (RQ)} = \frac{\text{CO2 evolved}}{\text{O2 consumed}}$$

• RQ range from 0.7 to 1.3 for aerobic (with O2) respiration.

• RQ is much greater if tissue goes into anaerobic (without O2) respiration.

• Carbohydrates: RQ = 1
• Lipids: RQ < 1
• Organic Acids: RQ > 1

Factors affecting Respiration

1. **Temperature:** Typically, for every 10 °C increase, respiration increases between 2 and 3 fold (Van't Hoff Rule).The temperature dependence of respiratory rate varies among and within commodities. Generally, the respiration increases significantly as the storage temperature increases. Within the physiological range of temperature (0°C–30°C), the rate of respiration increases exponentially, and a large amount of heat is produced as heat of respiration.

Heat Stress: If the temperature rises beyond the physiological range, respiration rate falls. When tissue reaches its thermal death point, metabolism is disordered as enzyme proteins are denatured. Continued exposure to high temperatures causes phytotoxic symptoms and tissue collapse.
Chilling stress: Although respiration is normally reduced at low, but non-freezing temperatures, certain commodities particularly those originating in the tropics and subtropics, exhibit abnormal respiration when their temperature falls below 10 to 12 °C. Respiration may increase dramatically at the chilling temperatures or when the commodity is returned to nonchilling temperatures. Enhanced respiration rate is cells' effort to detoxify metabolic intermediates that accumulated during chilling, as well as to repair damage to membranes and other sub-cellular structures. Enhanced respiration is one of the symptoms that signal the onset of chilling injury.

2. **Oxygen and carbon dioxide:** Low O_2 concentrations reduce respiration. High CO_2 also reduced respiration.Reduction of O2 concentration below 2%–3% gives beneficial reduction in rates of respiration and other metabolic processes for most produce. However, complete removal of O2 is not recommended as anaerobic environment is detrimental to the quality of the produce as it leads to fermentation, decay and development of off flavor, and change in color and texture. Increasing the CO2 level around some commodities reduces respiration, delays senescence and retards fungal growth. Different commodities vary widely to their ability to tolerate high CO2

3. **Ethylene:**Climacteric & Non-Climacteric fruits differ in their response to ethylene in the environment. Exposure of climacteric tissues during their preclimacteric stage to ethylene (C2H4) shortens the time to the start of the climacteric rise in respiration. Once the respiratory rise has begun, the tissue's endogenous rate of C2H4production increases and the internal C2H4 concentration also increases, reaching levels that saturate its biological activity. In contrast, C2H4 treatment of non climacteric tissues, in which endogenous C2H4 levels are very low, induces a climacteric-like rise in respiration that is proportional to C2H4 concentrations. However, unlike the case in climacteric tissues, endogenous C2H4 production remains unaffected. Removal of C2H4 results in a return of the respiration rate to its pretreatment level. The respiratory response of nonclimacteric tissues to C2H4 can be repeatedly induced throughout their postharvest life

4. **Stage of development:** Respiration rates vary due to stage of the development. Storage organs such as nuts and tubers have low respiration rates. Tissues with vegetative or floral meristems such as asparagus and broccoli have very high respiration rates. The commodities harvested during active growth, such as many vegetables and immature fruits, have high respiration rates. Mature fruits, dormant buds and storage organs have relatively low rates.After harvest, the respiration rate typically declines; slowly in non-climacteric fruits and storage organs, rapidly in vegetative tissues and immature fruits. The rapid decline presumably reflects depletion of respirable substrates that are typically low in such tissues.

5. **Stress/ Injury:**Physical stress during cultivation, harvesting, and postharvest handling influences respiratory behaviour significantly. Tissue injury increases the rate of respiration and induces ethylene production, which may further catalyse an increase in respiration with consequent loss of quality. The extent of increase in respiration rate is usually proportional to the severity of bruising. Water stress which is induced by lower than optimal relative humidity in air surrounding the commodity can increase its respiration rate. Biological stress like disease also increases the

respiration rate. Other stresses that stimulate the respiration rate of vegetables including exposure to ionizing radiation and to various chemicals such as methyl bromide (fumigant) etc.

6. **Surface of tissue:** Thickness of surface dermal system, wax composition and arrangement, number and distribution of stomata on the tissue influences the respiration rate. More the thickness of coating, less is the respiration rate.

7. **Surface to volume ratio:** Smaller fruits have large surface area leading to high respiration rate.

8. **Growing conditions:** Cultural practices, irrigation, fertilizer also influence the respiration rate. High nitrogen fertilizer increases respiration while high calcium fertilizer decreases respiration.

9. **Application of chemicals:** Certain chemicals like malic hydrazide (MH), methyl cyclo- propene (MCP), polyamines like putrescine, spermidine and spermine slows down the rate of the ethylene production and respiration while application of ethylene, acetylene, propylene, ethephon has a positive effect on respiration rate due to positive impact on ethylene (ripening hormone) generation.