# Physiological Activities of Partially Processed Fruits and Vegetables

***Injuries and wounds caused by the physical actions of partial processing affect physiological activities and subsequently the quality of produce***

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□ FRESH FRUITS AND VEGETABLES must be of excellent quality and condition at the time of harvest and after packing for the produce to be of top quality for consumers. One of the essential requirements of condition is that the commodity be free of skin breaks, bruises, injuries, and other mechanical damages. Any opening of tissue generally leads to changes in physiological activities and/or infection by pathogens and subsequently results in deterioration of the commodity. In the partial processing of fresh fruits and vegetables, trimming, peeling, cutting, slicing, and other physical actions cause injury and damage to tissues. These conditions are not acceptable and should be discouraged when handling fresh intact produce. However, these physical actions are required and necessary when preparing fresh product for direct use without any additional preparation, by restaurants, dining commons, and fast-food services, which places special importance in the handling procedures and processing sites (Huxsoll and Bolin, 1989).

## Changes at the Harvest Stage

Fruits and vegetables at harvest are at a stage of plant development when they are more susceptible to undesirable responses and consequences from injuries and wounds than at the earlier stages of development. At harvest stage, plant tissues are beginning to senesce and the balance in the dynamic processes of cellular structures and organelles shifts with the total degradative reactions becoming greater than the total biosynthetic reactions. This shift leads to such changes as general lipid breakdown, disorganization of membranes in cells or organelles, and quick deacylation of glycolipids in chloroplast thylakoids (Mazliak, 1983, Rolle and Chism, 1987). As the senescing process continues and cellular structures and membrane integrity are weakened, the mature tissues become increasingly susceptible to deterioration processes induced by stress and injuries caused by the physical actions of partial processing.

Deacylation of membrane glycolipids, phospholipids, and galactolipids results in liberation and accumulation of free fatty acids, which are toxic to many cellular processes. Free fatty acids are capable of disrupting biological systems causing lysis of organelles and binding to and inactivating proteins (Galliard, 1979). The free polyunsaturated fatty acids can also be degraded to hydroperoxides enzymatically by lipoxidases or lipoxygenases (Mazliak, 1983). The hydroperoxides are unstable and are cytotoxic particularly affecting proteins and membranes. Due to their instability, the hydroperoxide isomers lead to free radicals, and to volatile end products which are responsible for food spoilage.

Free radicals react at random with other compounds through hydrogen removal and a variety of additional reactions. Through these reactions, membrane lipid and protein components would be damaged chemically. Damages to the membrane can result in breaking of the diffusion barrier, which would allow cell contents to leak out and wound adjacent cells (Omarkhayyam, 1986).

A series of side reactions occur concurrent with membrane damage to facilitate continuation of respiratory processes (Laties, 1978). Steps of the electron transport chain which are controlled in intact tissues are unblocked. Thus, throughout wound healing, induced and accelerated respiration progresses through changes brought about by damage to the membranes.

Degradation or wounding of the membrane results in increased permeability, which has sequential effects. In particular, the reduction in cellular compartmentation is thought to cause mixing of previously sequestered metabolites of the ethylene-generating system, thereby stimulating ethylene evolution (Mazliak, 1983), which is identified as 'wound ethylene'. Wounding increases the activity of 1-aminocyclopropane-1-carboxylic acid (ACC) synthase and results in the accumulation of ACC, which is then oxidized to ethylene (Boller et al., 1979). The biosynthetic pathway for the production of 'wound ethylene' is the same as that for ethylene synthesis in ripening fruit (Mattoo and Anderson, 1984).

## Effects of Partial Processing

The degradative changes that occur during senescence are induced or enhanced by the physical action of processing of fruits and vegetables. This response is noted particularly with cells or tissues adjacent to those that are damaged by the cutting action and when acids and hydrolyzing enzymes of the vacuole are released. 'Wound ethylene' produced under these conditions can

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increase the permeability of membranes and perhaps reduce phospholipid biosynthesis, which can upset the dynamic processes of cellular structures and membrane integrity. This can contribute to the production of volatile long chain aldehydes, which are responsible for the 'wound respiration' (Laties and Hoelee, 1967), and would rapidly utilize the reserve substrate. However, some of the volatiles are associated with characteristic aroma of vegetables, which would be acceptable (Mazliak, 1983).

Increased respiration and ethylene production have been noted by slicing of muskmelon fruit (McGlasson and Pratt, 1964). Higher rates of respiration and ethylene production are thought to cause the onset of climacteric and result in different physiological age between intact and sliced tissue. Cutting of tomato into small disks causes the ethylene production to be about 20-fold higher than that of whole fruit and the increase was noted 15-20 min after cutting (Lee et al., 1970).

The effects of slicing on rate of respiration and ethylene production differs between climacteric and nonclimacteric fruit and with physiological age of climacteric fruit (Rosen and Kader, 1989). Slicing increased respiration of strawberries by 50% at 2.5°C, but had no effect on ethylene production at the low temperature, which is typical of nonclimacteric fruit. With partially ripe pears, a climacteric fruit, slicing increased respiration by 30% and decreased ethylene production at 2.5°C. Wounding induces ethylene production, particularly with climacteric fruit at the preclimacteric stage, but not with those at the postclimacteric stage as noted with the partially ripe pears.

In green bananas, ethylene production is extremely low, less than 50 nl $kg^{-1}$ $hr^{-1}$, but slicing causes the rate of production to be several times higher than the rate in intact fruit (McGlasson, 1969). A rise was noted within 2-hr after cutting and a second rise in the rate was detected 4-hr after cutting, and the maximum rate was reached within 6–8 hr. Rate of increase was dependent on the thickness of slices, with the rate of 2-mm slices being substantially greater than that of 4- or 6-mm slices. Differences in respiration rates between thin slices and intact fruit was noted also with bananas cut at the climacteric stage (Ku et al., 1965).

On the other hand, we found with ripe bananas and kiwifruit that were sliced and prepared for a combined package, respiration of only kiwifruit was affected by slicing (Fig. 1). Respiration rate of intact ripe bananas was about 100 mg $kg^{-1}$ $hr^{-1}$ and slicing to 4-cm thick sections with peel intact had minimal or no effect on the respiration rate. The bananas were completely yellow and beyond the climacteric stage.

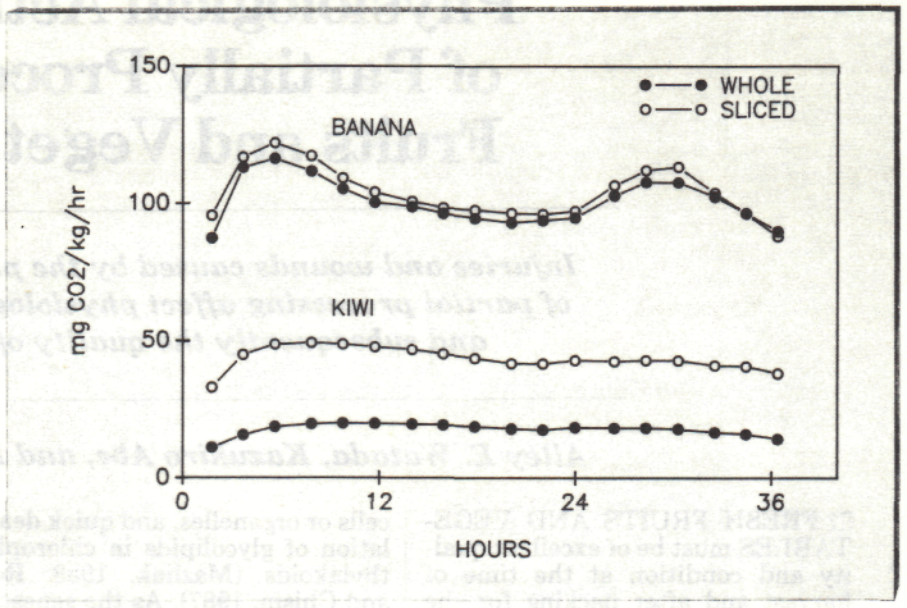


*Fig. 1*—**Respiration rates** *of sliced and whole banana and kiwifruit held at 20°C. Banana sliced to 4-cm length sections and kiwifruit to 1-cm thick slices.*

Respiration rate of intact kiwifruit was about 25 mg $kg^{-1}$ $hr^{-1}$ and peeling and slicing to 1 cm thickness caused the rate to double to about 50 mg $kg^{-1}$ $hr^{-1}$ at 20° C. The elevated rate was sustained for the 36-hr period.

Ethylene production by the kiwifruit was very low, about 5 nl $kg^{-1}$ $hr^{-1}$, and slicing caused the rate to be about 40 nl after 2-hr and about 80 nl $kg^{-1}$ $hr^{-1}$ after 4-hr at 20° C. After 24-hr, the rate had increased to an average of 1.3 $\mu$l $kg^{-1}$ $hr^{-1}$. The continual increase in rate probably was due to stimulation of ethylene production by the endogenous ethylene as well as the slicing action. Ethylene production by intact fruit remained unchanged during the 24-hr period.

Ripe banana cut to 4-cm length sections had a constant ethylene production of about 24 nl $kg^{-1}$ $hr^{-1}$ over a 4-hr period at 20°C, which was about the rate exhibited by intact fruit. Although the bananas used were in the post-climacteric stage of maturity, some response in ethylene production would be expected due to wounding from the slicing action. Reason for the lack of response is unknown.

## Textural Changes

As senescence is hastened by wound ethylene, associated quality changes are noted. One of these changes occurs with the texture, where firmness is desired for storage and transit of the produce, but softening is essential for sensory acceptance. The mechanism by which softening is regulated is not understood. In climacteric fruit, such as tomatoes, ethylene induces the ripening process. The softening noted with ripening is a phenomena that is already in progress and is accelerated with conditions that hasten ripening (Marshall et al. 1989).

Watermelon has been shown to deteriorate rapidly and lose firmness in 3 days when held in ethylene atmosphere at 18°C (Risse and Hatton 1982). Maceration of watermelon tissue appears to be due to increased activities of pectinase, cellulase, esterase, polyphenol oxidase, and peroxidase induced by ethylene (Shimokawa, 1973).

Softening rate of kiwifruit is dependent on ethylene and temperature (Arpaia et al., 1986). An ethylene level of 0.05–5 ppm accelerated softening and induced white core inclusion of kiwifruit. Acceleration increased with time and the ethylene level. Softening was accelerated by 0.5 ppm ethylene at 0°C and the rate increased with temperature (Arpaia et al., 1985). At 0°C, firmness of control fruit decreased from 70 to 60 newtons in 4 weeks; whereas, those treated with 0.5 ppm ethylene firmness decreased down to 20 newtons.

We found softening of ripe, sliced

kiwifruit that were packed together with banana sections was also accelerated by ethylene. Average firmness of 1 cm thick slices, which initially were about 4 newtons, decreased by about 25% after 24-hr and 40% after 48-hr at 20° C (Table 1). Exposure of slices to 2 or 20 ppm ethylene accelerated the loss of firmness down to about 2 newtons by the end of a 24-hr period. After 48-hr, the firmness of all samples was similar, probably because the values were at the minimum level of about 2 newtons.

Firmness of banana fruit were affected similarly as with kiwifruit by ethylene (Table 1). Average firmness of sliced bananas, which initially was about 5 newtons, decreased to 4 and 3.5 newtons with 2 and 20 ppm ethylene respectively after 48-hr. It is questionable whether taste panels can recognize these small changes, particularly with both fruit being relatively soft. However, these changes do indicate that ethylene accelerates the senescing and deteriorating processes.

*Table 1*—**Average Firmness (newtons) of Sliced Kiwifruit and Bananas** *held at 20°C in atmosphere of 0, 2, or 20 ppm $C_2H_4$ in air*[a]

| Fruit | $C_2H_4$ (ppm) | Hours 0 | 24 | 48 |
|---|---|---|---|---|
| Kiwifruit | 0 | 3.67 ± 0.21 | 2.78 ± 0.11 | 2.21 ± 0.06 |
| | 2 | 3.67 ± 0.21 | 2.28 ± 0.07 | 1.96 ± 0.05 |
| | 20 | 3.67 ± 0.21 | 2.0 ± 0.07 | 1.89 ± 0.07 |
| Banana | 0 | 4.83 ± 0.12 | 4.61 ± 0.09 | 4.86 ± 0.10 |
| | 2 | 4.83 ± 0.12 | 4.24 ± 0.07 | 4.04 ± 0.08 |
| | 20 | 4.83 ± 0.12 | 4.03 ± 0.09 | 3.53 ± 0.05 |

[a]Mean ± standard deviation, n = 6

*Table 2*—**Average Ethylene (ppm) and Carbon Dioxide (%) Content** *in enclosed containers of sliced kiwifruit and bananas held at 20°C with and without charcoal (ethylene absorbent)*[a]

| Volatile | Ethylene Absorbent | Hours 4 | 24 | 48 | 72 |
|---|---|---|---|---|---|
| $C_2H_4$ (ppm) | − | 1.27 ± 0.05 | 4.07 ± 0.15 | 4.69 ± 0.51 | 4.37 ± 0.47 |
| | + | 0.08 ± 0.01 | 0.08 ± 0.01 | 0.12 ± 0.02 | 0.22 ± 0.02 |
| $CO_2$ (%) | − | 4.14 ± 0.30 | 16.25 ± 0.46 | 20.19 ± 0.84 | 21.83 ± 0.73 |
| | + | 2.98 ± 0.20 | 13.11 ± 0.46 | 17.29 ± 0.92 | 17.45 ± 0.94 |

[a]Mean ± standard deviation, n = 6

*Table 3*—**Average Firmness (newtons) of Kiwifruit and Bananas** *placed together in an enclosed container held at 20°C with and without charcoal (ethylene absorbent)*[a]

| Fruit | Ethylene absorbent | Hours 0 | 24 | 48 | 72 |
|---|---|---|---|---|---|
| Kiwifruit | − | 4.30 ± 0.11 | 2.38 ± 0.09 | 2.14 ± 0.08 | 1.92 ± 0.06 |
| | + | 4.30 ± 0.11 | 2.91 ± 0.10 | 2.46 ± 0.07 | 2.30 ± 0.07 |
| Banana | − | 3.90 ± 0.06 | 3.29 ± 0.05 | 3.26 ± 0.06 | 3.21 ± 0.09 |
| | + | 3.90 ± 0.06 | 3.85 ± 0.08 | 3.69 ± 0.07 | 3.50 ± 0.07 |

[a]Mean ± standard deviation, n = 6

## Absorbing the Ethylene

With ethylene having the capability of accelerating the senescence and deterioration processes within a few hours, use of compounds to absorb ethylene needs to be considered in packages containing partially processed fruits. Packets containing charcoal with palladium chloride are effective in absorbing ethylene and can be placed conveniently in containers without contaminating the produce.

In an enclosed package containing four 4-cm length sections of banana and six 1-cm thick slices of kiwifruit, the charcoal with palladium chloride packet was effective in absorbing ethylene for 72-hr at 20° C. (Table 2). Without the absorbent, ethylene accumulated to 1.3 ppm in 4-hr and reached a plateau at about 4 ppm by the 24th hour.

Benefit of ethylene absorbent was noted in reducing both respiration rate and loss of firmness. Carbon dioxide in enclosed packages containing sliced bananas and kiwifruit without the ethylene-absorbent accumulated to 16% in 24-hr and continued to accumulate to 21.9% in 72-hr (Table 2). With the absorbent, carbon dioxide accumulated to about 80% of that in containers without the absorbent. Reduction in loss of firmness by the absorbent was apparent with both fruits (Table 3), but the degree of reduction was small, probably because they were already quite soft, possibly at the minimum level of firmness and/or possibly elevated carbon dioxide had an inhibitory effect on ethylene-induced softening (Arpaia et al., 1985, Palmer, 1971, Rosen and Kader, 1989). Nevertheless, if firmness was used as a criterion for determining shelf life, use of ethylene absorbent would allow marketing of packaged fruits another day, which is an additional 30% extension in shelf life.

## Loss of Pigment

Green color of vegetative tissue is another quality attribute that is affected deleteriously by ethylene. An ethylene level of 5 ppm can cause the green color of cabbage to fade significantly after 1 month of storage, and a low level of 1 ppm can have a similar effect on cabbage stored for 5 months (Hicks et al., 1982). Yellowing induced by 4 ppm ethylene has also been reported for Brussels sprouts, broccoli, cauliflower, and cabbage (Toivonen et al., 1982). Destruction of chlorophyll by ethylene has been reported to be due to increased chlorophyllase activity (Amir-Shapira et al., 1987, Shimokawa et al., 1978). The chlorophyllase activity of calmondin fruit increased by 50% after a 12-hr treatment with 10 ppm ethylene, and by 137% after a 24-hr treatment. At the end of 14-hr, calmondin fruit lost 11% of the chlorophyll content, as estimated with a color difference meter. These chlorophyll changes probably result from loss of membrane integrity that occurs with senescence, hastened by ethylene (Rolle and Chism, 1987).

The mechanism or pathway of chlorophyll degradation under normal senescence or that induced by ethylene in vegetative tissue is not clear. Chlorophyllase activity has been noted to increase with yellowing of barley and oat leaves (Rodriquez et al., 1987) and to decrease with senescence of tobacco and radish leaves (Phillips et al., 1969; Shimizu and Tamaki, 1963). These results imply that chlorophyllase is involved in both the degradative and biosynthetic processes of chlorophyll, and is not always required in the degradative processes. Chlorophyll oxidase-linoleic acid system located in the chloroplast thylakoids is involved in the oxidation of chlorophyll and the degradative product is chlorophyll a-1. Pathways requiring lipolytic acyl hydrolase in the choroplast of spinach leaves (Yamauchi et al., 1987) and peroxidase-hydrogen perioxide system in parsley (Yamauchi and Minamide, 1985) have been shown to be involved in degradation of chlorophyll. These results indicate that chlorophyll degradation pathway probably differs among plant species and it is unknown if ethylene activates other pathways.

In determining the chlorophyll degradative pathway in packaged spinach stored at 25° C, we found chlorophyll a and b to decrease with 'a' decreasing more rapidly than 'b' (Table 4). Of the degradative products of chlorophyll, chlorophyll a-1, which was very low initially decreased slightly rather than the expected increase and chlorophyllide a increased slightly. Pheophytin, also present in very low amounts, did not change with time. Of the enzymes catalyzing degradative reactions, chlorophyllase activity increased by about 2-fold the first 3 days and then decreased sharply between day 3 and 4 down to the initial level in spinach held at 25° C. Peroxidase activity based on a unit protein weight increased by about 30% during the 4 day period (unpublished data).

With the increased chlorophyllase activity, an increase in chlorophyllide would be expected since the enzyme catalyzes the removal of the phytol chain. Reason for the lack of relationship is unknown. Lipid peroxidation has been implied to be involved in the degradation of chlorophyll (Orthoefer and Dugan, 1973, Imamura and Shimizu, 1974), but it is questionable whether this reaction is strongly involved with the spinach leaves due to the decreasing content of chlorophyll a-1. It is also questionable that the degradative pathway catalyzed by chlorophyll oxidase was active in the spinach because the product of this pathway is chlorophyll a-1 (Schoch et al., 1984). The peroxidase-hydrogen peroxide system appears to be the major chlorophyll degradative pathway in spinach, based on the high activity that we had noted and presence of a phenolic compound, apigenin, that is necessary for degradative reaction to proceed (Yamauchi and Minamide, 1985).

In spinach that was exposed to 10 ppm ethylene, degradation of chlorophyll a was hastened (Table 4). Ethylene hastened the slight decrease of chlorophyll a-1, and had no effect on the changes in chlorophyllide or pheophytin. Activities of neither chlorophyllase or peroxidase were affected by the volatile (unpublished data). These results indicate that ethylene hastened the normal pathway of chlorophyll degra-

dation and did not appear to initiate other pathways.

## Additional Research Required

Ethylene, which can be produced readily and in substantial quantity by the physical action of partial processing, has the potential of having a significant effect on reactions associated with quality. Firmness and color are only two of the many quality attributes that are affected by ethylene, but probably the most significant ones that must be regulated to provide the consumer with a desired product. Additional research is required to understand the mechanism of how quality attributes are regulated and affected by ethylene.

*Table 4*—**Average Contents (mg/100g fresh weight) of Chlorophyll a and b** *in spinach held at 25°C in atmosphere with and without 10 ppm ethylene in air*

| | | Days | | | | | Significance | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Chlorophyll | Ethylene | 0 | 1 | 2 | 3 | 4 | Time linear | Time quadratic | Treatment | Interaction |
| a | − | 125 | 129 | 125 | 111 | 89 | | | | |
| | | | | | | | xxx[c] | xx[b] | xxx | xxx |
| a | + | 125 | 131 | 100 | 76 | 48 | | | | |
| b | − | 49 | 50 | 51 | 45 | 37 | | | | |
| | | | | | | | xx | xx | xxx | x[a] |
| b | + | 49 | 51 | 42 | 31 | 18 | | | | |

[a]Significant at P = 0.05
[b]Significant at P = 0.01
[c]Significant at P = 0.001

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