

Fresh-Cut Vegetables

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I. INTRODUCTION

The term *fresh-cut* (i.e., lightly processed, minimally processed) identifies fresh vegetables that have been cut into small serving-size portions and are ready to eat [e.g., broccoli (*Brassica oleracea* L. Botrytis group), carrots (*Daucus carota* L.), lettuce (*Lactuca sativa* L.), spinach (*Spinacia oleracea* L.)] or to cook [e.g., artichokes (*Cynara scolymus* L.), broccoli, sweetcorn (*Zea mays* L. var. *rugosa* Bonaf.), peeled potatoes (*Solanum tuberosum* L.), etc.] (Saltveit, 1997; Schlimme, 1995; Shewfelt, 1987, Stanley, 1998). Consumption of fresh-cut vegetables is rapidly increasing and new products are continually being developed. Demographic changes over the next few decades promise to further increase the demand for fresh-cut vegetables. Consumers are becoming increasingly concerned about the therapeutic and nutritive quality of their diet. The minimal application of chemicals to fresh-cut vegetables and their fresh appearance satisfy many of these concerns (Bruhn, 1995). The ease of use of fresh-cut should also significantly improve the diet through promotion of the consumption of vegetables.

A plant cell contains many compounds that are kept in separate compartments by semipermeable membranes. The cell membrane that surrounds the living cytoplasm of the cell establishes a boundary between it and its external environment. The membrane surrounding the largest compartment of a mature cell, the vacuole, separates the cytoplasm, with its many enzymes, from stored organic acids and phenolic compounds. Wounding not only physically damages the membranes in the injured cells but also disrupts membrane function in adjacent cells, so that incompatible compounds mix and produce unwanted and uncontrolled reactions. For example, phenolic compounds from the vacuole mix with enzymes (e.g., polyphenol oxidase, PPO) in the cytoplasm to produce a brown compound that can discolor the tissue. Wounding changes membrane lipid metabolism and cell permeability in fresh-cut carrots (Picchioni et al., 1994).

During respiration, plant cells use O_2 from the atmosphere and food in the cell to produce energy and CO_2 . If the O_2 concentration within the tissue falls below about 2% or if the CO_2 concentration rises about 5%, the predominant respiratory reactions within the tissue could change from aerobic to anaerobic (see Chaps. 2 and 9). The tissue will then undergo fermentation with the production of compounds that give the product an undesirable flavor and aroma.

Oxygen diffuses into the tissue and CO_2 and heat diffuse out. The rate of O_2 and CO_2 diffusion is governed by the concentration gradient and by the resistance of the surface (epidermis) and the internal tissues to gas movement. The resistance of the surface usually has a pronounced effect on the rate of diffusion (Cameron et al., 1995). Temperature affects both the rate of respiration and the rate of gas diffusion (see Chap. 2). However, the effect of temperature on respiration is much more pronounced (i.e., Q_{10} is often greater than 2) than its effect on gas diffusion (i.e., changes are proportional to the changes in the absolute temperature). Although diffusion of O_2 and CO_2 increases at elevated temperatures, the increase in respiration (i.e., consumption of O_2 and production of CO_2) so predominates that the concentration of O_2 in the tissue actually decreases while the concentration of CO_2 increases. This relationship also holds for packages where the diffusion of O_2 and CO_2 through a plastic film can lag behind respiration at elevated temperatures and produce an unwanted atmosphere low in O_2 and high in CO_2 in the package.

The injury and trauma associated with the preparation of fresh-cut vegetables (e.g., abrading, chopping, cutting, dicing, mincing, shredding, and slicing) mimic some of the naturally occurring stresses to which the plant has evolved elaborate defense responses. In some postharvest situations, these defense responses are encouraged. For example, the curing of potatoes promotes healing of harvest-related injuries through the development of wound periderm and suberization of tissue adjacent to the wound. But in fresh-cut vegetables, these responses are usually detrimental to the overall quality of the product. For example, wound-stimulated phenylpropanoid metabolism promotes the synthesis and accumulation of phenolic compounds that promotes the browning of fresh-cut lettuce.

Development of effective postharvest measures to maintain quality and extend the post-cutting life of fresh-cut vegetables requires an understanding of both the basic physiological processes of the commodity (these are covered in many chapters of this book) and how these processes are altered by the stresses of processing. Reviews on fresh-cut vegetables include an entire issue of five articles in the *Journal of Food Quality* [1987, Vol. 10(3):143–217; Barmore; Brackett; Klein; Rolle and Chism; and Shewfelt], a book chapter (Cantwell, 1992), a book (Wiley, 1994), the proceedings of a colloquium of seven articles on lightly processed fruit and vegetables in *HortScience* (1995, Vol. 30(1):14–40; Baldwin, et al.; Brecht; Burns; Cameron et al.; Hurst; Romig; Schlimme), an entire issue of 11 articles in *Postharvest Biology and Technology* [1996, Vol. 9(2):115–245; Babic and Watada; Barth and Zhuang; Bennik et al.; Baldwin et al.; Blanchard et al.; Guerzoni et al.; Izumi et al.; López-Gálvez et al.; Picchioni et al.; Varoquaux et al.; Watada et al.], one volume of the Proceedings of the Controlled Atmosphere Research Conference held at UC Davis (Gorny, 1997), and parts of the Proceedings of the USDA Beltsville Symposium on Quality that was published in *Postharvest Biology and Technology* [1999, Vol. 15(3):195–340; Cavalieri; Saltveit; Watada and Qi-Ling; Zagory]. Other individual reviews include Huxsoll et al. (1989), King and Bolin (1989), Watada, et al. (1990), Ahvenainen (1996), and Saltveit (1997). A review by Laurila et al. (1998) that is pertinent to

this topic focuses on methods to inhibit enzymatic browning in fresh-cut fruits and vegetables, while one by Sapers (1993) also includes a general discussion of enzymatic and nonenzymatic browning reactions in foods. This chapter focuses on the general response of fresh-cut vegetables and to representative responses of some major fresh-cut vegetables. Specific recommendations are rapidly changing as new treatment methods and products are developed. However, the basic strategies for minimizing physical damage, maintaining optimum ranges of temperature and relative humidities, and avoiding microbial contamination remain unchanged.

II. PHYSICAL RESPONSES TO WOUNDING

The preparation of fresh-cut vegetables entails physical wounding of the tissue; e.g., carrots are peeled and cut, cucumbers (*Cucumis sativus* L.) are sliced, and lettuce and cabbage (*Brassica oleracea* L. Capitata group) are shredded. These unavoidable physical injuries cause both an immediate and a subsequent physical and physiological response in the tissue. The immediate physical effects of fresh-cut processing are to cause mechanical shocks to the tissue, to remove the protective epidermal layer, to accumulate surface moisture, and to expose tissue to contaminants. Later, as the surface water evaporates and the tissue starts to respond physiologically, there is a further alteration in gas diffusion and surface appearance. Some of the immediate physical effects of wounding are listed in Table 1.

Accelerated water loss, altered surface appearance, and entry of pathogens are the three major physical problems with fresh-cut vegetables. Removal of a significant portion of the epidermis during preparation exposes hydrated tissue that now has no physical barrier to impede the evaporation of water. Maintaining a high relative humidity (RH) around the fresh-cut product will reduce the vapor-pressure deficit and minimize water loss. This is most easily accomplished by packaging in barrier films. However, some water

Table 1 Immediate Physical Effects Caused by the Preparation of Fresh-Cut Vegetables

Mechanical shock to tissue
Bruises, cracks, fractures, tears
Hydraulic shocks are dispersed or focused by reflective and refractive properties of nonhomogenous tissues within the commodity
Removal of protective epidermal layer
Alters gas diffusion
Water vapor, O ₂ , CO ₂ , C ₂ H ₄
Provides entry for contaminants
Chemicals, micro-organisms
Liquid on cut surface
Reduces gas diffusion
Elevates CO ₂ , C ₂ H ₄
Reduces O ₂
Accelerates water loss
Provides substrate for microbes
Liquid in tissue
Water in intracellular spaces causes translucent tissue
Changes density of the commodity

loss is unavoidable, since removal of the heat of respiration by external cooling creates a gradient of water potential that drives water from the product to the cooling surface. In a package, the cooling surface is the inner surface of the bag. Condensation of water evaporated from the product on the inner surface of the package shows that this concomitant movement of heat and water vapor is ubiquitous. Solutes in the water on the surface of the commodity lower its vapor pressure, but the result is slight in the dilute solutions associated with fresh vegetables and has only marginal effects on evaporation.

Surface coatings to minimize water loss are slightly effective, but they may interfere with the diffusion of other gases and produce unwanted anaerobic conditions within the commodity. This is especially true immediately after processing, when respiration is temporarily stimulated by wounding. It is also difficult to formulate coatings that will adhere to the wet, unstable surface of cut vegetables yet act as a barrier to water loss (Avena-Bustillos et al., 1994, 1997).

Edible coating have been formulated to prolong the shelf-life and maintain quality of fresh-cut vegetables (Baldwin et al., 1996; Li and Barth, 1998). White blush on peeled carrots was reduced by edible coatings that increased water vapor resistance (Avena-Bustillos et al., 1993). An edible sodium caseinate/stearic acid emulsion also controlled white blush and, in addition, reduced respiration by about 20% when compared to the uncoated control (Avena-Bustillos et al., 1994). However, white blush was also controlled by treatments that modified the hygroscopic properties of the surface and did not leave a proteinaceous residue (Cisneros-Zevallos et al., 1997; Krochta et al., 1996). Inclusion of potentially allergenic compounds in edible coatings and the necessity of labeling the product as containing "artificial chemical compounds" may detract from their use on "natural" fresh-cut vegetables.

Liquids coming from the wounded tissue or water remaining on the surface after washing constitute a formidable barrier to gas diffusion (see Chap. 9). Gases diffuse through liquid water 10,000 times slower than through an equal thickness of air. A thin 0.1-mm film of water has the same diffusive resistance as 1 m of air. The combined effect of wound-stimulated respiration and reduced gas diffusivity in wet, fresh-cut vegetables can significantly alter the concentration of internal gases. The balanced concentrations of internal gases (e.g., O₂, CO₂, and C₂H₄) has a pronounced effect on tissue maturation and development. Changes in gas concentrations can accelerate ripening, alter phenolic metabolism, and promote senescence of the tissue. Disrupting this balance could influence some of the postharvest problems associated with fresh-cut vegetables. Later, as the rate of respiration and the diffusive resistance of the surface decreases because the surface moisture is either absorbed by the adjacent cells or evaporates, the internal concentrations of these metabolically active gases will again change. At that time, the lack of any resistive barrier on the surface permits rapid diffusion of all internal gases, including water vapor. A coating that mimicked the diffusive properties of the epidermis would be very effective in protecting the commodity against excessive water loss. Some of the subsequent physical effects of wounding are listed in Table 2.

The shock of cutting affects not only those cells cut and those adjacent to the cut but also cells far removed for the actual site of injury. The incompressible nature of water and the reflective and refractive properties of the tissues in a commodity to hydraulic pressure waves can channel and focus the force imparted to the tissue by cutting to cells often far removed. This may account for some of the injury and browning that is seen in tissue apparently uninjured in the initial preparation of fresh-cut vegetables. Sharper knives and slower processing speeds would lessen this source of damage.

Table 2 Subsequent Physical Effects Caused by the Preparation of Fresh-Cut Vegetables

Elimination of natural barriers
Enhanced gas diffusion
Reduced CO ₂ , C ₂ H ₄
Elevated O ₂
Accelerated water loss
Entry of contamination
Changes in appearance
White blush formation because of surface debris
Uneven surface resulting from uneven water loss by tissues
Splitting or fracturing resulting from differential changes in turgor
Intrusion of water into intracellular spaces causing translucent tissue

III. PHYSIOLOGICAL RESPONSES TO WOUNDING

There are immediate and subsequent effects of wounding on a wide range of physiological and biochemical processes. These responses to wounding are elicited in both adjacent and distant tissues by a wound signal that propagates through the tissue and induces a myriad of responses. Many of these induced responses are detrimental to the quality of fresh-cut produce. A few of these changes happen very quickly after wounding, while other can take many days to complete. Some of the immediate physiological effects of wounding are listed in Table 3.

A. The Wound Signal

The cutting and abrasion of tissue rapidly produces a wound signal that is thought to be responsible for the induction of many physiological responses, including increased respiration, increased production of C₂H₄ and phenolic compounds, and the induction of

Table 3 Immediate Physiological Effects Caused by the Preparation of Fresh-Cut Vegetables

Wound signal (nature, speed)
Plant growth regulators (e.g., ABA, ethylene, jasmonic acid, salicylic acid, systemin, traumatin, etc.)
Wall fragment
Hydraulic
Bioelectrical wave
Membrane depolarization
Increased permeability
Mixing of cellular compounds
Calcium and signal transduction
Vacuole contents
Membrane disorganized
Lipids oxidized
Free fatty acids produced
Loss of protoplasmic streaming

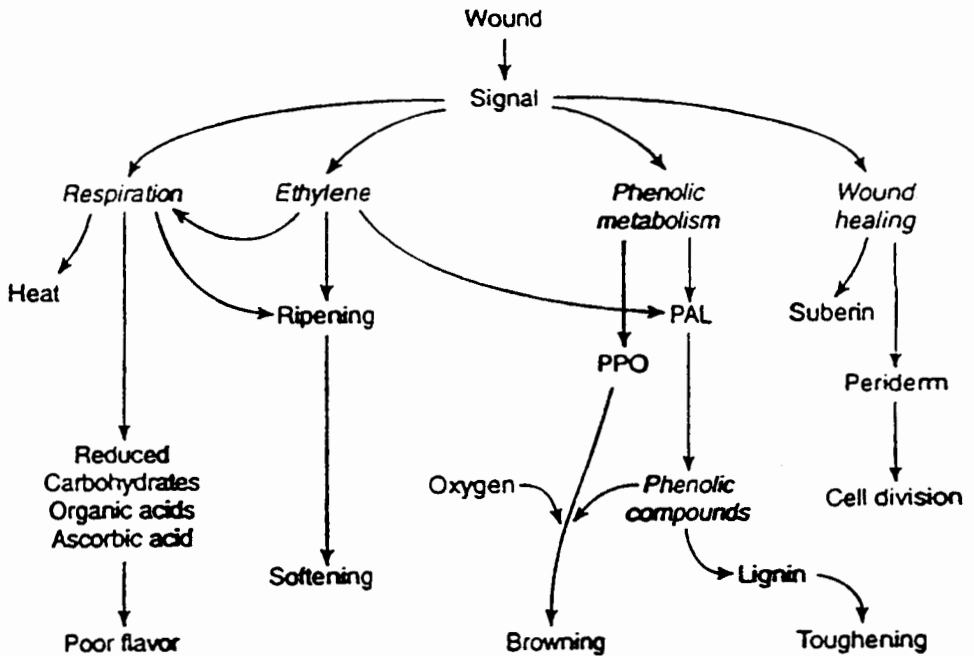


Figure 1 The interrelationship among the many effects of wounding on physiological processes in fresh-cut vegetables. (From Saltveit, 1997.)

wound healing (Fig. 1). The nature of the wound signal and its method of abscisic acid (ABA) propagation from the site of injury are unknown. Products of the lipid metabolism and oxidation, e.g., jasmonic acid, are thought to be constituents of the plethora of possible wound signals in plants. Other candidates for the wound signal include chemical compounds such as ethylene, systemin, ABA, salicylic acid, elicitors; and physical changes such as electric and hydraulic waves (Peña-Cortes and Willmitzer, 1995). Although many of these candidates are very potent wound signals in specific plants and tissues, they are inactive in others. Whatever the signal may be, it is becoming increasingly obvious that a better understanding of the wound signal is necessary to reduce its impact on the quality of the processed product. In field crops, enhancement of protective responses induced by a wound signal would be beneficial to the growth of the crop in a stressful environment.

The wound signal appears to migrate from the site of injury into adjacent tissue. This progressive movement can be seen by measuring specific responses of the tissue to injury. Wounding lettuce tissue stimulates phenolic metabolism, and one of the most important enzymes in phenolic metabolism is phenylalanine ammonia-lyase (PAL). The activity of PAL in tissue within 5-mm of a cut increases within 4 h, while it takes 6 and 8 h to increase in tissue 1 and 2 cm away (Fig. 2). This suggests that the wound signal is moving at a speed of about 0.5 cm/h. The signal seems to persist for some time (e.g., around 24 h), since inhibiting its movement into adjacent tissue with cold or anaerobic treatments does not greatly curtail its affect once these inhibitors are removed (Saltveit and Dille, 1978). If the wound signal is a chemical, it does not appear to be water-soluble, since the cutting of tissue immersed in a variety of aqueous solutions does not greatly perturb the response.

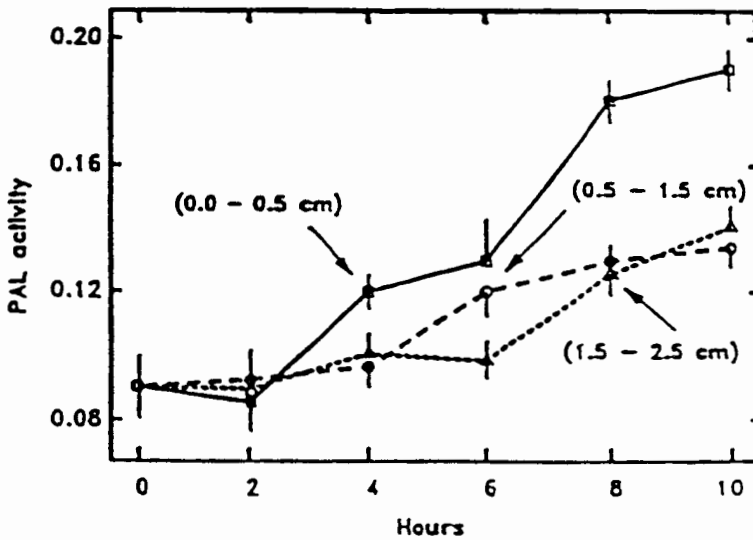


Figure 2 Induction of PAL activity ($\mu\text{mol g}^{-1} \text{h}^{-1}$) as affected by the distance from the wound and the time after wounding (Ke and Saltveit, 1989). PAL activity was measured after incubation at 5°C . The numbers in parentheses represent the distance from the cut surface. The arrows represent the time when PAL activity started to increase. The vertical bars represent the standard error of the mean.

B. Severity of the Injury

The response of tissue to wounding usually increases as the severity of the injury increases; i.e., as more tissue is injured, there is more of a response. For example, increasing the number of puncture wounds in lettuce increases the response of the tissue; e.g., the level of PAL activity (Ke and Saltveit 1989). However, after a certain level of injury has been reached, additional injuries cause less and less of a response (Fig. 3). This probably happens because the expanding areas of induced tissue start to overlap each other. Since the level of response from individual cells appears to be quickly saturated by a wound signal, induction by wound signals from two or more sites of injury would have progressively less of an effect than the initial induction from one.

These overlapping areas of induced tissue can be thought of as expanding circles. Cells near the site of injury experience the greatest induction for the longest period of time. As the wound signal moves outward, it appears to dissipate in strength, so cells some distances away from the site of injury are induced less and for a shorter period of time. When wound signals converge from two directions there is a double induction; but because of the distances and limitations of the tissue response, there is only a slightly increased physiological effect (Fig. 4). Following the propagation of the wound signal through the tissue, there is a subsequent response that entails many physiological, biochemical, and morphological changes. Some of the subsequent physiological effects of wounding are listed in Table 4.

Commodities and tissues are also made up of cells that have different capacities and modes of responding to injury. For example, the pith of a celery [*Apium graveolens* L. var. *dulce* (Mill.) Pers.] stalk has lower rates of metabolism than cells associated with the phloem in the vascular tissue. A few hours after wounding, tissue at the blossom end

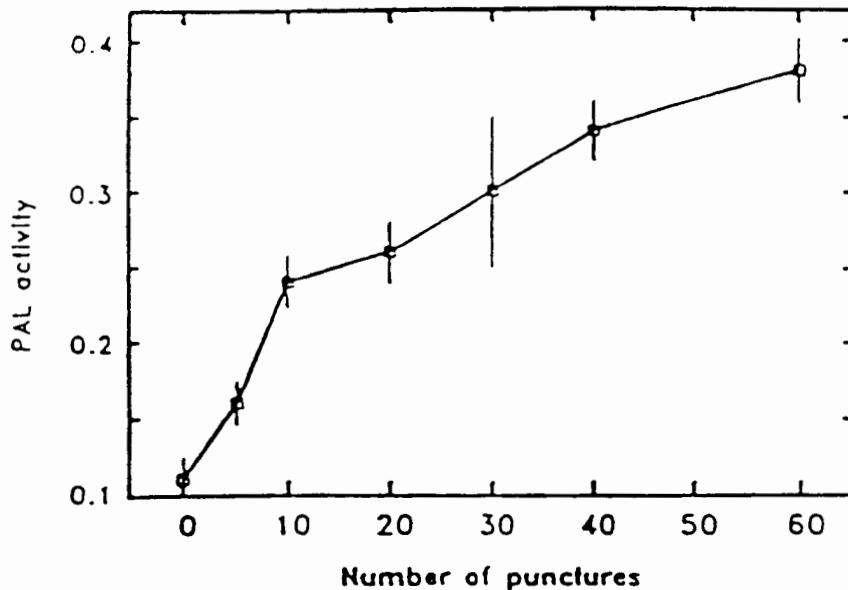


Figure 3 Increase in PAL activity ($\mu\text{mol g}^{-1} \text{h}^{-1}$) with increased wounding of lettuce leaf tissue (Ke and Saltveit, 1989). Wounding was done by puncturing an 8-cm square of midrib tissue with a sterile 26-gauge hypodermic needle. All measurements were done 48 h after incubation at 5°C. The vertical bars represent the standard error of the mean.

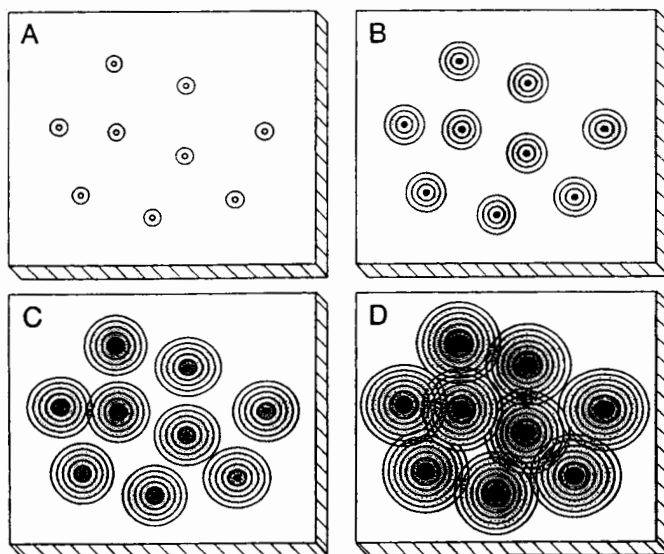


Figure 4 Expanding areas of wound-induced tissue as the wound signal moves out from the initial site of injury (A) to adjacent tissue (B) over time. The induced zones can overlap (C and D) to form zones of multiple induction. (From Saltveit, 1997.)

Table 4 Subsequent Physiological Effects Caused by the Preparation of Fresh-Cut Vegetables

Elevated C ₂ H ₄ production
ACC pathway
Wound pathway? (ethane)
Elevated respiration
Carbon dioxide production
Oxygen consumption
Heat production
Anaerobic respiration
Oxidative reactions
Nonrespiratory O ₂ consumption
Browning
Induction of enzymes
ACC synthase and ACC oxidase
PAL, PIF, etc.
Altered phenolic metabolism
Phytoalexins
Browning substrates
Induction of wound healing
Lignin and suberin synthesis
Cell division
Altered protein synthesis
Compositional changes
Ascorbic acid
Organic acids
Carbohydrates
Sugar to acid balance
Toughening
Softening
Loss of flavor

of a mature-green tomato (*Lycopersicon esculentum* Mill.) fruit produces wound-induced C₂H₄ at over twice the rate as tissue excised from the equator or stem end (Brecht, 1995). When incubated at 20°C, the maximum rate of C₂H₄ production from wounded tomato tissue is similar, but tissue from the blossom end produced its maximum about 2 days before tissue from the equator, and tissue from the stem end reached its maximum about 2 days later. Inclusion of such differently responding tissues in the same consumer package may result in produce of uneven quality.

C. Interaction of Stresses

The response of the tissue to one stress often modifies its response to another stress. For example, lettuce or celery plants stressed in the field by drought or pathogens are more susceptible to tissue browning and pithiness after harvest. Tomato fruit marginally chilled in the field is more susceptible to postharvest decay. In some cases, the prior stress can actually protect the plant from subsequent stresses. A heat shock can protect plants from subsequent temperatures that would have been lethal. There also appears to be a hierarchical order to the importance of abiotic stresses. When confronted with two stresses, such

as wounding and heat shock, the plant can preferentially respond to one over the other (see Chap. 18). In the case of lettuce, heat shock appears to redirect protein synthesis from the production of wound-induced enzymes to heat-shock proteins. This redirection of protein synthesis away from the synthesis of enzymes of phenylpropanoid metabolism to innocuous heat-shock proteins can be used to reduce the browning of fresh-cut lettuce (Loaiza-Velarde et al., 1997).

Browning is a severe problem, and the browning potential of many tissues is affected by their prior treatment (Lopez-Galvez et al., 1996a; 1996b). Stresses (e.g., temperature, physical injury, and disease) tend to increase the production of many phenolic compounds that brown easily upon injury. Being able to predict the browning potential of tissue before processing would help in making marketing decisions and in deciding which treatments, packaging, and storage conditions would be needed to maintain maximal quality and shelf-life. Many tissues initially low in the activity of enzymes of phenylpropanoid metabolism and in phenolic content (e.g., celery, lettuce) however, are predisposed by previous stresses to rapidly mobilize the phenylpropanoid pathway and accumulate significant quantities of brownable phenolic compounds (Lopez-Galvez et al., 1996b). Measuring the initial levels of enzymes of phenylpropanoid metabolism (e.g., PAL, PPO) or the level of phenolic compounds in these tissues does not give a good indication of their shelf life (Couture et al., 1993). In contrast, these factors measured a few days after wounding are highly correlated with subsequent shelf life. Attempts have been made to devise models for the shelf life of fresh-cut vegetables (Guerzoni et al., 1996). A rapid test that measured inducibility before processing would certainly be preferred over one that measured the induced products a few days after processing.

The method of preparation can significantly affect the storage life of fresh-cut vegetables (Bolin and Huxsoll, 1991). Cutting a whole commodity into smaller portions usually shortens its storage life. Sharp knives cause less damage than dull knives, while tearing along sutures causes the least damage. Shredding a commodity and abrading its surface causes the most damage. Even the direction of the cut (i.e., transverse, longitudinal, or diagonal) can influence the response of the commodity (Brecht, 1995). However, whether this is due to the directionality of the cut or to the inclusion of tissues in the severed portions that have different metabolic activities is unanswered. Cutting a tomato along the equator would produce halves with very different rates of respiration, while a longitudinal cut would produce halves with similar rates of respiration.

D. Wound Respiration and C₂H₄ Production

A rapid and important response to wounding is the increase in respiration and C₂H₄ production rates. These increases may occur through the uncontrolled mixing of cellular components (e.g., disruption of membrane semipermeability) or through controlled cellular repair mechanisms. Both processes produce heat that may increase the tissue's temperature and accompanying rate of water loss. Wounding may thereby increase both the basal rate of heat production and the amount of heat produced, because the tissue is metabolizing faster at an elevated temperature. Although often transitory in nature, this increase in heat production should be taken into account in designing packages and storage conditions for fresh-cut produce so as to prevent prolonged exposure to elevated temperatures that will shorten the shelf life of the processed commodity.

The rates of O₂ consumption and CO₂ production increase in wounded plant tissue (Fig. 5). In some tissues it is minor and transitory (e.g., lettuce), while in others it is major

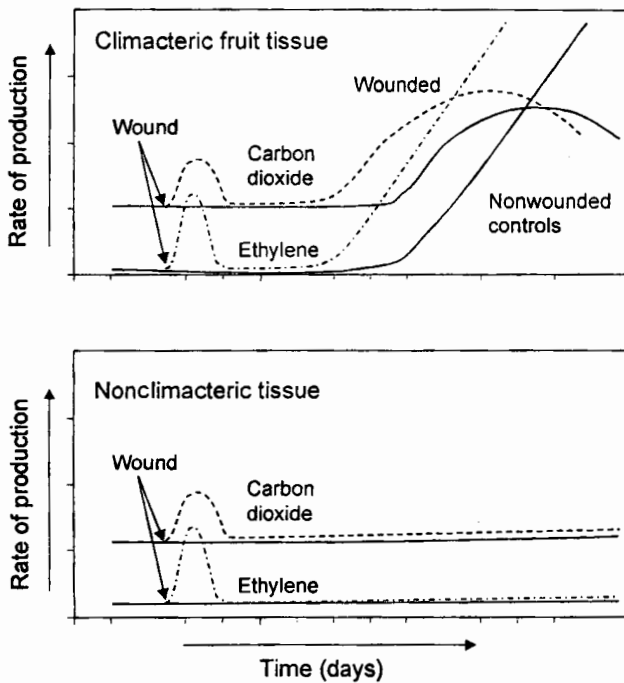


Figure 5 Effect of wounding on the rate of CO_2 and C_2H_4 production by nonclimacteric vegetative and climacteric fruit tissue.

and persistent (e.g., potato). Depletion of carbohydrate reserves as a result of stimulated respiration rate can lower the organoleptic quality of some commodities like melons (*Cucumis* spp.), whose quality is highly dependent on sugar content, or those with naturally low levels of sugars and reserve carbohydrates (e.g., starch). In contrast to apple and banana fruits, which have large amounts of starch to convert to sugars, melon and tomato fruits have very limited stores of starch to replenish any sugars lost to accelerated respiration during storage or ripening.

Not all the O_2 consumed by the tissue is used in respiration. The uncontrolled increase in O_2 consumption by fresh-cut tissue is often an indication of oxidative browning (Laurila et al., 1998). The oxidation of phenolic compounds to colored pigments can be enzymatic or nonenzymatic and may or may not be accompanied by the increased production of CO_2 (Sapers, 1993).

Carbon dioxide production increases in tissue undergoing wound repair as respiration is stimulated to furnish not only energy but also to synthesize the molecules needed for repair. The substrates used in these reactions are often the very compounds that are prized components of quality—e.g., sugars and organic acids. The reductions and interconversion of these compounds during metabolism can significantly reduce quality. For example, the preferential respiration of organic acids can alter the sugar-to-acid ratio, making the commodity insipid-tasting. Other respiratory reactions accelerate softening of some tissue and the toughening of others. The breakdown of cell-wall components produces soft tissues (e.g., tomato), while the synthesis of lignin strengthens the cell walls of fibers, making the tissue tough and stringy (e.g., asparagus).

Wounding also activates the C_2H_4 biosynthetic pathway in many commodities (Abeles et al., 1992). Large quantities of C_2H_4 can be produced by some injured tissues. However, not all tissues respond to wounding with increased C_2H_4 production. Many vegetative or immature tissues (e.g., broccoli, cabbage, celery, and lettuce) normally produce small amounts of C_2H_4 , and wounding causes only a small and transitory increase in production rate. Although exposure to C_2H_4 adversely affects most fresh-cut commodities, wound-induced C_2H_4 production is probably not a major problem because of its low level of production and its transient induction by wounding (Saltveit, 1999). Wound-induced C_2H_4 could become a problem if the tissue were exposed to high levels over an extended period because the wounded tissue was confined in a small, unventilated container. Ventilation of the container or inclusion of an C_2H_4 scrubber are simple remedies to that problem (Abe and Watada, 1991; Saltveit, 1998).

While C_2H_4 production in climacteric fruit is promoted by the endogenous concentration of C_2H_4 through positive feedback and increases dramatically during ripening, the feedback of C_2H_4 on C_2H_4 production in vegetative and nonclimacteric fruit tissue is negative (Saltveit, 1999). In these tissues, C_2H_4 actually inhibits C_2H_4 production. Apart from some transient increases in C_2H_4 production associated with the traumas of harvest, endogenous C_2H_4 levels are maintained at low levels by this negative feedback, and endogenously produced C_2H_4 probably has minimal effect on the postharvest quality of vegetables and nonclimacteric fruit. For example, C_2H_4 production from fresh-cut lettuce (Ke and Saltveit, 1989) is much smaller than that from wounded mature-green tomato fruit tissue (Brecht, 1995). The maximum rate of C_2H_4 production from wounded lettuce and tomato was 0.6 and 8.0 $\eta L g^{-1} h^{-1}$, respectively, and elevated C_2H_4 production lasted for less than a day for lettuce (Fig. 6), while it was still elevated after 2 weeks for tomato.

Both wounding and C_2H_4 stimulate phenolic metabolism in plant tissue. Wounding may stimulate phenolic metabolism either through the induction of C_2H_4 production or other plant growth regulators or through an unidentified wound signal. If wounding did

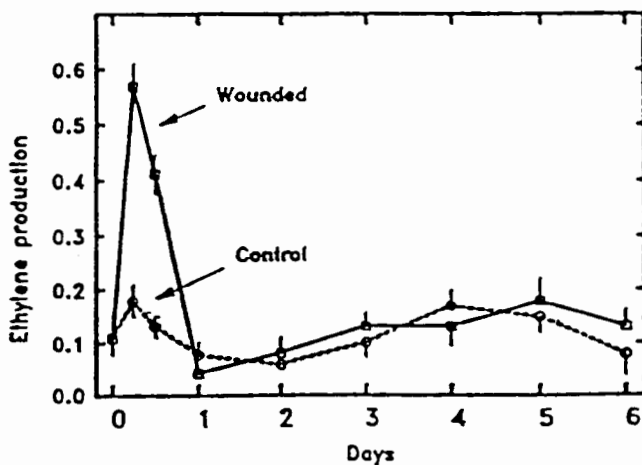


Figure 6 Ethylene production ($\eta L g^{-1} h^{-1}$) by wounded lettuce tissue over time, showing the low levels of production and the transitory nature of wound-induced C_2H_4 production (Ke and Saltveit, 1989). All measurements were done after incubation at 5°C. The vertical bars represent the standard error of the mean.

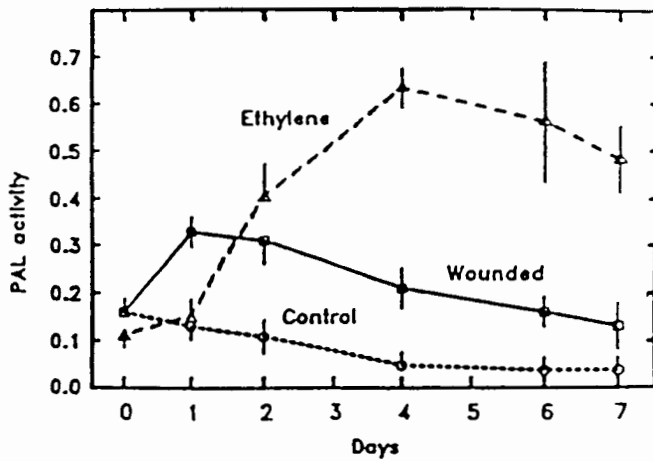


Figure 7 Increase in PAL activity ($\mu\text{mol g}^{-1} \text{h}^{-1}$) over time in lettuce tissue exposed to C_2H_4 ($3 \mu\text{L L}^{-1}$) or wounded (Ke and Saltveit, 1989). The vertical bars represent the standard error of the mean.

stimulate phenolic metabolism through the production of wound-induced C_2H_4 , then increases in phenolic metabolism, which are at the basis of many unwanted wound responses in fresh-cut produce, could be eliminated by simply inhibiting the synthesis or action of C_2H_4 . However, it appears that C_2H_4 is not the wound signal for the induction of phenolic metabolism in many vegetative tissues.

For example, the induction of PAL activity in lettuce tissue is more rapid in wounded tissue than in C_2H_4 -treated tissue (Fig. 7). If wounding acted through the induction of C_2H_4 , then the level of PAL in lettuce tissue exposed to C_2H_4 should have been higher than in wounded tissue, since the step in which wounding induced C_2H_4 production was bypassed (Ke and Saltveit, 1989). In the case of lettuce, elimination of wound-induced C_2H_4 production would therefore have no effect on the induction of PAL.

There are other crops, however, in which wound-induced C_2H_4 production could induce significant changes. Many fruit are climacterio—e.g., apple [*Malus sylvestris* (L.) Mill. var *domestica* (Borkh.) Mansf.], avocado (*Persea americana* Mill.), banana (*Musa acuminata* Colla.), melon, and tomato—and experience increased respiration and C_2H_4 production during ripening. Exposure of mature, preclimacteric fruit tissue to C_2H_4 stimulates C_2H_4 production and ripening. The self-stimulation of C_2H_4 production by C_2H_4 means that a short exposure to C_2H_4 as the result of injection of C_2H_4 into the storage atmosphere (as is done to ripen bananas and tomatoes, or as the result of wounding) could trigger additional C_2H_4 production and possible unwanted physiological activity—e.g., ripening, softening, etc.

IV. FRESH-CUT VEGETABLES

The fresh-cut vegetables that are available commercially include broccoli, cabbage, carrots, cauliflower (*Brassica oleracea* L. Botrytis group), lettuce, potatoes, and spinach. To a lesser extent, artichokes, cucumbers, onions (*Allium cepa* L.), and tomatoes are also sold as fresh-cut.

The two major fresh-cut vegetable crops are lettuce and carrots. Their methods of preparation are radically different, as are their responses to wounding. Quality is decreased in both commodities by changes in their visual appearance: carrots turn white and lettuce turns brown. However, the whitening of peeled carrot segments results from a purely physical phenomenon (Cisneros-Zevallos et al., 1995), while the browning of cut lettuce is predominately physiological in nature (Peiser et al., 1998). Methods to control whitening in carrots therefore concentrate on modifications to the surface of the peeled root segment (Cisneros-Zevallos et al., 1997; Krochta et al., 1996), while browning is controlled in lettuce by modifying the physiological processes induced by wounding (Ke and Saltveit, 1989; Peiser et al., 1998).

The surface debris left on the carrot after peeling is held appressed to the tissue and rendered transparent by a thin film of water (Cisneros-Zevallos et al., 1995). As the water evaporates, portions of the abraded cell walls that are still attached to the tissue are released from the surface tension of the water film and form a "fluffy" layer (Fig. 8). This layer

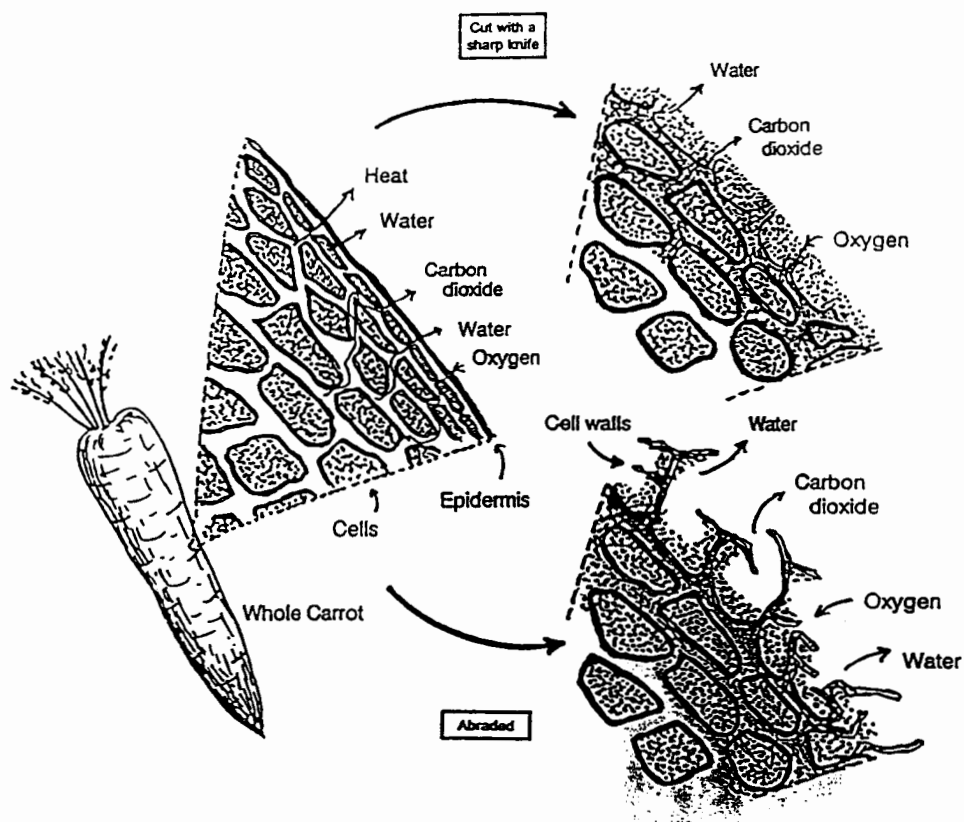


Figure 8 Representation of how cutting and abrading can alter the surface of minimally processed vegetables. A. The cellular structures near the surface of a whole, uninjured carrot root. B. The immediate effect of cutting the epidermal layer from the carrot root with a sharp knife. Water on the cut surface and in the intercellular spaces between the first few layers of cells is represented by stipples. C. The abraded surface of the carrot, showing the surface debris that is responsible for the whitish discoloration upon slight dehydration.

of material that is oriented perpendicular to the intact tissue produces a whitish “bloom” on the surface. Treatments that increase the water-holding capacity of the surface or that in any other way “glue” the abraded debris to the surface would be effective in preventing whitening (Cisneros-Zevallos et al., 1997; Avena-Bustillos et al., 1994). Maintaining high RH around the commodity and storage at cold temperatures are effective control measures, but these measures have no residual effectiveness when the consumer sets out the peeled carrots in a bowl at room temperature (Cisneros-Zevallos et al., 1995). A peeling process that did not leave surface debris that could form the whitish layer would effectively eliminate the problem. Physically polishing the peeled carrot sections or enzymatically digesting away the loose cellular debris would produce a smoother surface.

Extensive research with fresh-cut lettuce has produced a detailed picture of how the many wound-related responses affect one another and contribute to browning (Fig. 9). An examination of the diagram in Figure 9 shows how browning could be reduced at many different steps. Browning of fresh-cut lettuce can be partially controlled by rapid cooling and storage at 0°C to minimize wound-induced changes in phenylpropanoid metabolism (Couture et al., 1993; Lopez-Galvez et al., 1996b). Storage under low O₂ and high CO₂ controlled or modified atmospheres is also effective in reducing the synthesis, accumulation, and browning of phenolic compounds (Lopez-Galvez et al., 1996a; Smyth et al., 1998). However, just as in the case of peeled carrots, these preventive measures have little if any residual effects. Application of chemicals that denature proteins or act as antioxidants has a residual effect on browning as long as the chemicals remain active. For example, application of some organic acids to the cut ends of harvested lettuce effectively

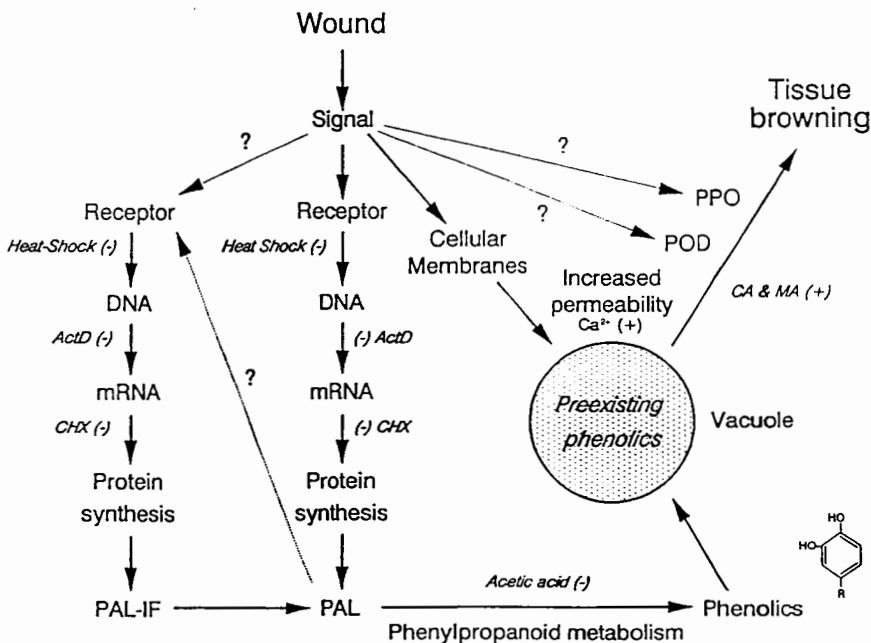


Figure 9 Diagram showing the interrelationships between a wound and the subsequent induced changes in phenolic metabolism that leads to tissue browning. Many control points for postharvest modification of the wound-induced browning processes are evident. The question marks indicate relationships that are still under investigation.

reduces stem-end browning (Tomás-Barberán et al., 1997a). Low-dosage irradiation in combination with modified atmospheres is also effective in extending the shelf life of fresh-cut lettuce, mainly through a reduction in microbial populations (Hagenmaier and Baker, 1997). Disrupting the formation and spread of the wound signal, its perception, or its implementation would effectively prevent adjacent cells from sensing that the tissue had been wounded and would work long after the initial treatment was applied.

Induction of other metabolic pathways that are naturally antagonistic to wound responses could also have a protracted effect on browning (Fig. 9). For example, a PAL-inactivating factor (Gupta and Creasy, 1991) is produced in lettuce tissues in which increased PAL activity was induced by wounding (Ritenour and Saltveit, 1996). Stabilization of cellular membranes, possibly through the addition of calcium salts to the wash water, appears to prevent some of the uncontrolled mixing within the cell after wounding and helps control browning reactions.

While many modifications of the wound response could be implemented through genetic engineering, the vigor of the resultant plant may be diminished because of its inability to respond to the many natural stresses encountered during growth. For example, unless a reduction in PAL activity were site- and stage-dependent, it could result in reduced phenolic substrates available for the synthesis of aromatic compounds such as the precursors of lignin. The current trend in plant breeding is to produce plants that are more resistant to biotic and abiotic stresses. To actively pursue the development of a plant less resistant to environmental stresses would seem to be counter to this trend. However, in other plants, different isoenzymes of PAL are produced in response to different stimuli. If a constitutively expressed antisense mRNA could be developed for the unique isoenzyme of PAL induced by wounding, then wound-induced phenolic production and browning could be selectively reduced. A synthetic antisense mRNA could be constructed for the wound-induced PAL isoenzyme that recognized a unique region of the mRNA and not the conserved portion common to all PAL mRNA. Coupling the antisense PAL mRNA to a wound promoter is also a possibility, but isolation of such a specific promoter has not yet been achieved.

Commodities with constitutive high levels of phenolic compounds, like artichoke and potato, brown easily when wounded tissue is exposed to the O₂ in air. The exclusion of O₂ or the application of antioxidants controls browning in these commodities. In contrast, other commodities, like lettuce and celery, have low levels of naturally occurring phenolic compounds. Wounding stimulates phenylpropanoid metabolism and the accumulation of phenolic compounds leads to browning in these commodities (Tomás-Barberán et al. 1997b) (Fig. 9). After the phenolic compounds have accumulated in wounded lettuce or celery, the techniques used on artichokes and potatoes need to be employed to control browning of these tissues. However, interfering with the wound signal, the synthesis of enzymes of phenolic metabolism, or the synthesis of the phenolic compounds themselves will prevent the accumulation of deleterious levels of phenolic compounds and eliminate browning.

The synthesis of wound-induced enzymes of phenylpropanoid metabolism (e.g., PAL) can be prevented by giving the lettuce tissue a brief heat shock (e.g., immersion in 45°C water for 90 s) after processing (Fig. 10). The heat-shocked tissue synthesizes innocuous heat shock proteins in preference to enzymes of phenolic metabolism. By the time the tissue has recovered from the heat shock, the wound signal has dissipated and there is no further induction of enzymes of phenolic metabolism. While this technique is very effective at preventing browning in plant tissue with constitutively low levels of phenolic

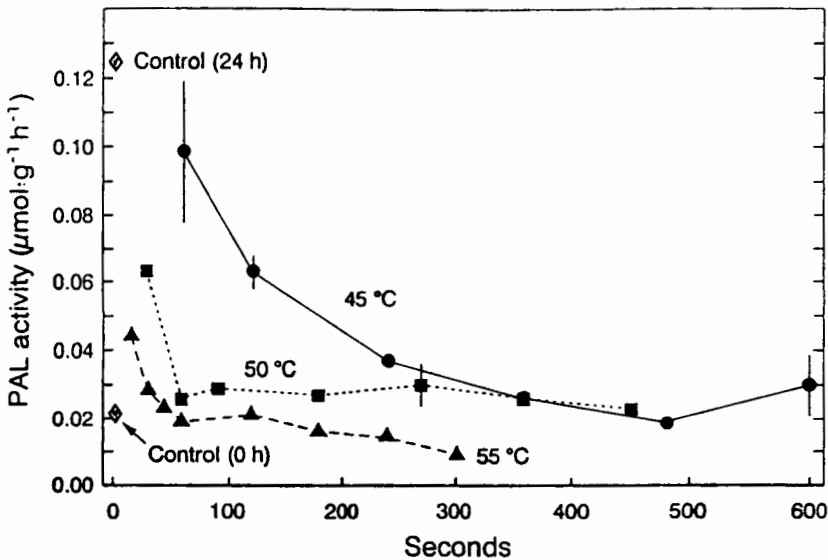


Figure 10 Effects of duration of exposure to selected temperatures on the subsequent development of PAL activity ($\mu\text{mol g}^{-1} \text{h}^{-1}$) in excised iceberg lettuce midrib tissue segments (Loaiza-Velarde et al., 1997). Control midrib segments were assayed before treatment (0 h) and after 24 h at 10°C (24 h). Standard deviation bars are present only when larger than the symbol.

compounds (e.g., celery, lettuce), it is ineffective in tissue with constitutively high levels of phenolic compounds (e.g., artichokes, potatoes).

The postharvest application of aqueous solutions of calcium salts as dips or sprays has long been used to control postharvest disorders in storage and maintain tissue firmness of fresh fruits and vegetables. Application of calcium as the chloride and lactate salts to fresh-cut cantaloupe (*Cucumis melo* L. Reticulatus group), honeydew (*Cucumis melo* L. Inodorus group), and watermelon [*Citrullus lanatus* (Thunb.) Matsum and Nak.] pieces helps maintain firmness (Luna-Guzmán et al., 1999). The quality of zucchini (*Cucurbita pepo* L.) slices was also improved by the application of calcium salts (Izumi and Watada, 1995).

V. SPECIAL PROBLEMS ASSOCIATED WITH FRESH-CUT

Limiting water loss, reducing unwanted metabolic reactions, and protecting against microbial contamination are three problems of crucial importance with fresh-cut vegetables. The removal of the natural barrier to water loss and microbial entry during processing and the exposure of hydrated tissue rich in available nutrients requires that processing of fresh-cut vegetables occurs under sterile or at least aseptic conditions. Because of the delicate nature of many fresh-cut vegetables and their enhanced responsiveness to additional stresses, temperature abuses that whole vegetables could endure with little loss of quality may severely affect the quality of fresh-cut produce.

The common use of barrier bags to produce and maintain modified atmospheres, the differential response of gas diffusion through plastic films, and the rate of respiration by the commodity combine to make temperature management extremely important. Elevated

temperatures would not only cause a more rapid senescence of the commodity, but changes to the modified atmosphere could result in the production of fermentative "off" odors (Kato-Noguchi and Watada, 1997) and the growth of anaerobic microorganisms (Brackett, 1987).

Microbial contamination and growth on fresh-cut vegetables has generated a great deal of concern, and much research has focused on this problem (Hurst, 1995; Nguyen-the and Carlin, 1994). Wounding removes the natural protective barrier to microbial attack and produces a moist surface rich in nutrients that is a superb medium for growth of micro-organisms. Rigorous sanitation of preparation areas reduces the level of contamination, while chemical treatments and low temperatures restrict growth during storage and marketing. Plant growth regulators may be able to induce protective reactions by the tissue. For example, treatment with methyl jasmonate extends the shelf life and reduces the microbial load on fresh-cut celery and peppers (Buta et al., 1998). Some modified atmospheres have biostatic effects on a number of microorganisms and inhibit the growth of others. However, a number of studies have shown that fresh-cut vegetable products are inedible by the time microbial populations have risen to dangerous levels. For example, samples of fresh-cut romaine lettuce (*Lactuca sativa* L. var. *longifolia* Lam.) and shredded cabbage inoculated with *Clostridium botulinum* spores were judged to be inedible by the time they became toxin-positive (Petran et al., 1995).

Another serious problem can arise when fresh-cut vegetables respond in uncharacteristic ways. For example, vivipary (i.e., the precocious germination of seeds in the mature fruit) has been eliminated through selective breeding in most vegetable crops (Marrush et al., 1998). However, when slices from mature-green tomato fruit were ripened, seed germination became the predominant quality defect (Mencarelli and Saltveit, 1988). Exposure to endogenous C_2H_4 solved the problem by hastening ripening and at the same time inhibiting seed germination and radicle elongation. Such unexpected responses from the tissue can make the development of new fresh-cut products a challenging endeavor.

Selecting the proper cultivar is always an important criterion in growing and marketing vegetable crops. However, because of the differential responses of individual cultivars to fresh-cut processing, selecting a cultivar with minimal unwanted wound responses is especially critical with vegetables destined for the fresh-cut market (Romig, 1995). Before fresh-cut attained its present importance, generic product and culls were often used. Today, special cultivars of lettuce and carrots are grown especially for the fresh-cut market. Identification of cultivars suitable for fresh-cut would be very useful for other vegetables.

Some selection can be done to improve the quality and storability of unspecific cultivars after harvest. For example, celery petioles segments that are less likely to develop pithiness can be selected by sorting by density (Saltveit and Mangrich, 1996). The higher-density segments develop pithiness slower than lower density petioles.

VI. PACKAGING FOR FRESH-CUT

Packages usually provide a number of features, including unitization of a number of small commodities into a reasonably sized consumer package, protection against contamination with foreign matter, protection against physical injury, and identification of the product (Barmore, 1987). Along with these qualities, a package for fresh-cut vegetables must also protect the commodity against water loss and contamination by microorganisms. Many packages are also designed to create and maintain a modified atmosphere around the commodity to reduce unwanted metabolic reactions. Because of these additional

needs of fresh-cut vegetables, their packages are often considerably more expensive (e.g., barrier film bags) than packages for whole vegetables (e.g., perforated low-density polyethylene).

Package design, through its effect on rate of cooling and atmospheric modification, can have an effect on the quality of fresh-cut vegetables. For example, the retention of antioxidant vitamins in fresh-cut broccoli was significantly affected by package design (Barth and Zhuang, 1996). Designs that allow rapid cooling are especially important in dealing with the higher respiration rates frequently found in fresh-cut vegetables. Because of the unprotected and delicate nature of the cut surface of many fresh-cut commodities, the package should also stabilize the portions of cut product to prevent additional damage. Containers that can be resealed seem to be an attractive design for packages of fresh-cut vegetables. However, the modified atmosphere established at the processing facility and maintained during marketing by proper temperature management cannot readily be reestablished under conditions usually present in the consumer's environment. A package designed to reduce water loss and allow adequate gas exchange should be preferred over one designed to reestablish a modified atmosphere.

VII. THE FUTURE OF FRESH-CUT

Demographic changes in family structure during the coming decades, the demand for easier-to-prepare fresh vegetables, the recognition of the nutritive importance of fresh vegetables to a healthy diet, and the consumption of more meals away from home all combine to make the demand for fresh-cut vegetables likely to increase for the foreseeable future.

This chapter has only touched on a few of the many responses and interactions that occur during and after processing of fresh-cut products. Wounding associated with the preparation of fresh-cut produce induces many physical and physiological responses. Many of these changes are unwanted and some actually decrease product quality. The best way to control these changes is to maintain low temperatures and high humidity during storage and marketing. Selecting high-quality starting material is another way to ensure a good-quality fresh-cut product. Postprocessing treatments may be necessary to control problems such as browning, white blush, diseases, and textural changes. When possible, these types of treatments should be kept to a minimum, since they add cost and complexity to an already complex system.

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