United States Department of Agriculture

Agricultural Research Service

Agriculture Handbook Number 66

The Commercial Storage of Fruits, Vegetables, and Florist and Nursery Stocks

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Abstract

Gross, Kenneth C., Chien Yi Wang, and Mikal Saltveit, eds. 2016. The Commercial Storage of Fruits, Vegetables, and Florist and Nursery Stocks. Agriculture Handbook 66, U.S. Department of Agriculture, Agricultural Research Service, Washington, DC.

Agriculture Handbook 66 (AH-66) represents a complete revision and major expansion of the 1986 edition. It has been reorganized and now includes 17 Chapters and 138 Commodity Summaries written by nearly a hundred experts in plant science and postharvest technology. This version, like the previous editions of AH-66 in 1954, 1968, 1977, and 1986, presents summaries of current storage requirements of fresh fruits, vegetables, cut flowers, and other horticultural crops. However, this highly expanded version also includes information on quality characteristics, maturity indices, grading, packaging, precooling, retail display, chilling sensitivity, ethylene production and sensitivity, respiration rates, physiological disorders, postharvest pathology, quarantine issues, and suitability as fresh-cut product. A large number of fruits and vegetables were added, as well as sections on food safety, nutritional quality, texture, and fresh-cut produce. The purpose of storing plant material is to lengthen the time it can be consumed or utilized. In doing so, it is critical to provide an environment that minimizes deterioration, maintains microbial safety, and retains other quality attributes. AH-66 provides guidelines and other important information for storing and handling horticultural commodities to accomplish this.

Keywords: carbon dioxide, chilling injury, cold storage, controlled atmosphere storage, cut flowers, ethylene, flavor, food safety, fresh-cut, fresh produce, fruit softening, heat load, 1-methylcyclopropene, microbial safety, minimally processed, modified-atmosphere packaging, potted plants, nutritional quality, nuts, orchids, packaging film, perishable, postharvest biology, precooling, respiration, sensory evaluation, shelf-life, texture.

The information contained in AH-66 has been assembled from material prepared by nearly a hundred authors from around the world. All of the information contained herein was peer reviewed and edited for scientific content. Every effort was made to provide the most accurate and current information available.

The contributors' professional affiliations and addresses were up-to-date at the time of submission of their chapters, and the editors made all reasonable efforts to update any changes received during the review and publishing process. However, due to the large number of contributors and countries represented, it is not inconceivable that some of the contributors may have changed organizations in the interim and thus are no longer at the addresses given in this handbook. In cases where the editors received specific address changes or death notices, all such updates are reflected in this volume.

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While supplies last, printed copies of this publication may be obtained at no cost from the USDA-ARS Food Quality Laboratory, Building 002, Room 117, 10300 Baltimore Avenue, Beltsville, MD 20705-2350.

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Ethylene Effects

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Introduction

Ethylene (C_2H_4) is a simple, naturally occurring organic molecule that is a colorless gas at biological temperatures. The following is a list of biological attributes of ethylene:

- It is a colorless gas at biological temperatures.
- It is a naturally occurring organic compound.
- It readily diffuses from tissue.
- It is produced from methionine via aminocyclopropane carboxylate (ACC) by a highly regulated metabolic pathway.
- Key enzymes are ACC synthase and ACC oxidase.
- C_2H_4 synthesis is inhibited by C_2H_4 in vegetative and immature reproductive tissue.
- C_2H_4 synthesis is promoted (autocatalytic) by C_2H_4 in mature reproductive climacteric tissue.
- It is effective at ppm and ppb concentrations (1 ppm = 6.5×10^{-9} M at 25 °C).
- It requires O_2 to be synthesized, and O_2 and low levels of CO_2 to be active.

Many biotic and abiotic sources contribute to the presence of C_2H_4 in the postharvest environment. Ripening and diseased plant tissues are a significant source of C_2H_4, as are industrial sources, the most prominent ones being internal combustion engines and fires.

Ethylene is biologically active at very low concentrations measured in the ppm and ppb range. Most plants synthesize small amounts of C_2H_4 that appear to coordinate growth and development. Because it is a gas, C_2H_4 readily diffuses from sites of production, and continuous synthesis is needed to maintain biologically active levels in the tissues. Barriers to diffusive loss include not only the commodity's epidermis but also postharvest coatings and packaging. Under biotic or abiotic stress or during climacteric ripening, C_2H_4 production can increase dramatically, and emanations from stimulated tissue can accumulate in packages or storerooms and produce unwanted effects in adjacent tissue. Other molecules with specific configurations can mimic C_2H_4 but are less effective. For example, C_2H_4 analogs propylene (C_3H_6) and acetylene (C_2H_2) require 100- and 2,700-fold, respectively, the concentration of C_2H_4 to elicit the same effect.

Plants produce C_2H_4 through an actively regulated biosynthetic pathway in which the amino acid methionine is converted to ACC (1-aminocyclopropane-1-carboxylic acid) and then to C_2H_4 through a series of biochemical reactions. O_2 is required for the synthesis of C_2H_4 and both O_2 and CO_2 are required for its biological activity. Each reaction in the synthesis and action of C_2H_4 involves a biological catalyst, an enzyme that focuses the reaction into producing the next specific chemical for that pathway. Enzyme activity is regulated either through its synthesis and/or destruction, or by interactions with substrates and products. These interactions can create a positive or a negative feedback of C_2H_4 on its synthesis (figure 1).

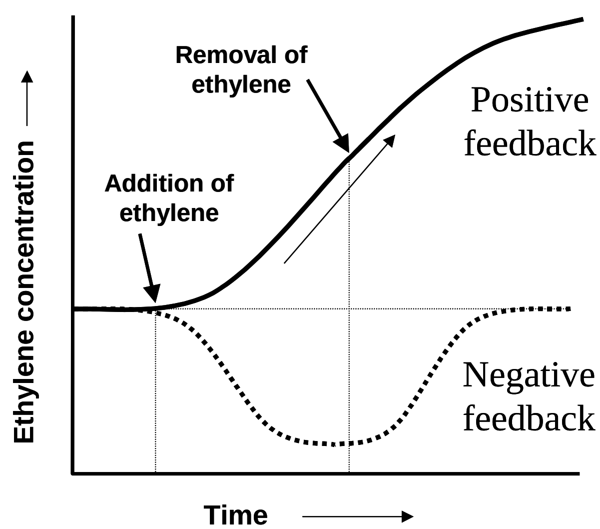

Figure 1. Effect of adding and removing ethylene from the atmosphere surrounding tissues that respond with a positive (ethylene promotes its own synthesis) or negative (ethylene inhibits its own synthesis) feedback. Modified from Saltveit (1999).

In vegetative tissue and in nonclimacteric and immature climacteric fruit tissue, C_2H_4 suppresses its own synthesis, and in ripening climacteric fruit C_2H_4 enhances its own synthesis. This positive feedback of C_2H_4 on C_2H_4 synthesis is called autocatalytic C_2H_4 production. Plants respond to C_2H_4 in a number of ways.

Ethylene stimulates the following:

- Synthesis of C_2H_4 in ripening climacteric fruit
- Ripening of climacteric fruit and some nonclimacteric fruit
- Anthocyanin synthesis in ripening fruit
- Chlorophyll destruction and yellowing (for example, degreening of citrus)
- Seed germination
- Adventitious root formation
- Respiration and phenylpropanoid metabolism
- Flower initiation in bromeliads (for example, pineapple)
- Abscission and senescence

Ethylene inhibits the following:

- Ethylene synthesis in vegetative tissue and nonclimacteric fruit
- Flowering and flower development in most plants
- Auxin transport
- Shoot and root elongation; that is, growth

Depending on a number of variables, C_2H_4 has both beneficial and deleterious effects on harvested fruits, vegetables, and ornamentals.

Beneficial effects:

- Promotes color development in fruit
- Stimulates ripening of climacteric fruit
- Promotes degreening of citrus
- Stimulates dehiscence in nuts
- Alters sex expression (Cucurbitaceae)
- Promotes flowering (for example, in pineapple)
- Reduces lodging of cereals

Detrimental effects:

- Accelerates senescence
- Enhances excessive softening of fruits
- Stimulates chlorophyll loss (for example, yellowing)
- Stimulates sprouting of potato
- Promotes discoloration (for example, browning)
- Promotes abscission of leaves and flowers
- Stimulates phenylpropanoid metabolism

Often an C_2H_4-induced change in one commodity is viewed as beneficial, while the same change in another commodity is viewed as detrimental. For example, C_2H_4 is used to promote ripening of bananas, melons, and tomatoes; degreening of oranges; and synthesis of pigments in apples. Yet the same changes are unwanted when C_2H_4 promotes over-ripening of fruit, yellowing of broccoli, development of brown russet spot lesions in lettuce, and senescence of flowers. Because of these diverse and often opposite effects of C_2H_4, controlling its action in plants is of great economic importance to producers, wholesalers, retailers, and consumers of fresh fruits, vegetables, and ornamentals.

In most vegetative tissues, C_2H_4 is only produced in biologically active amounts during early stages of development or in response to biotic or abiotic stress. Mutant plants that do not respond to C_2H_4 often grow normally, with only a few insignificant alterations in development. Most of the effects of C_2H_4 on vegetative tissue are therefore the result of the tissue's response to a stress or to the intentional or unintentional exposure of tissue to active levels of C_2H_4.

In contrast to its effects on vegetative tissue, biologically produced C_2H_4 plays a crucial role in the development of reproductive tissues and in the ripening of certain climacteric fruit. The rates of C_2H_4 production and its internal concentration often vary by orders of magnitude during early stages of development and during the initiation and development of reproductive structures. Increased rates of C_2H_4 production are especially pronounced during the ripening of climacteric fruit such as apples, avocados, bananas, melons, pears, and tomatoes. In these fruit, the autocatalytic production of C_2H_4 heralds the onset of ripening and is required for many of the reactions associated with ripening to continue. See section "Summary of Respiration and Ethylene Production Rates" in the Introduction of this handbook.

Once internal C_2H_4 exceeds a level characteristic for the species, tissue, and developmental stage, the further production of C_2H_4 is stimulated by presence of previously produced C_2H_4. In this way, autocatalytic positive feedback can increase rates of C_2H_4 production and internal concentration of C_2H_4 by 1,000-fold during ripening. External application of C_2H_4 can promote the ripening of climacteric fruit—for example, avocado, banana, honeydew, and tomato—and beneficial quality changes in nonclimacteric fruit; for example, degreening of lemon and orange. Once autocatalytic C_2H_4 production has started in climacteric fruit, lowering its external concentration has an insignificant effect on its internal levels, rates of production, or action.

Ethylene is an important plant growth regulator that has pronounced effects on many aspects of plant growth and development. Regulating its effectiveness is commercially important for many crops. Controlling its effectiveness can mean either increasing its beneficial effects or decreasing its detrimental effects. There are a number of ways to accomplish either objective.

Reducing Effectiveness of Ethylene
Use C_2H_4-tolerant cultivars
Keep atmosphere free of C_2H_4
Maintain at coldest possible temperature
Store under CA or MA or in MAP
Minimize time between exposure and use

Increasing Effectiveness of Ethylene
Use C_2H_4-sensitive cultivars
Keep an active level of C_2H_4 in the air
Maintain at optimum temperature
Store under adequate levels of O_2 and CO_2
Allow sufficient time for plant response

Ethylene Interactions in Plants

There are some significant interactions between the plant and its environment that are important in understanding how to control biological activity of C_2H_4 in plants (figure 2).

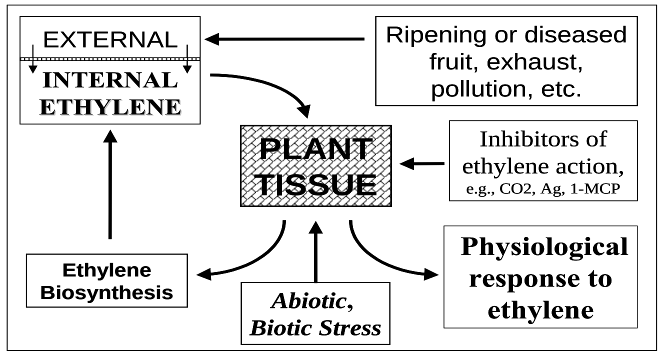

Figure 2. Interactions among a plant and ethylene in its environment (Saltveit 1999).

Ethylene in the atmosphere can have a direct effect on plant tissue by raising the internal concentration to an active level. Sources of atmospheric C_2H_4 include exhaust from trucks and forklifts, pollution from industrial activity and from the burning of fuels, and biosynthesis by diseased plants or ripening fruit. In some cases, C_2H_4, whether applied as a gas or as an C_2H_4-releasing compound such as ethephon, is intentionally added to the plant's environment to stimulate desirable changes. The changes can include promotion of flowering in pineapple; ripening of avocado, banana, melon, and tomato fruit; degreening of citrus; altering sex expression in cucurbits; defoliation; and promotion of latex secretion by rubber trees.

The activity of C_2H_4 inside plants is regulated not only by the absolute level of C_2H_4 but also by the responsiveness of tissues and the presence of CO_2, the natural antagonist of C_2H_4 action. The response of plants to C_2H_4, therefore, depends on a number of factors, only one of which is the rate of C_2H_4 production by the plant. Tissue sensitivity depends on species, cultivar, cultural practices, and stage of development.

Prior and current stresses have a significant effect on modulating the effect of C_2H_4. For example, wounding stimulates C_2H_4 production, as well as a host of plant defense responses such as increased phenylpropanoid metabolism. Some of these responses involve C_2H_4, while others do not. Increased phenolic metabolism greatly increases the susceptibility of some crops like lettuce to develop browning—for example, russet spotting—when exposed to C_2H_4 and/or mechanical injury.

The effect of tissue susceptibility is most clearly seen in fruit tissue. Immature climacteric fruit respond to C_2H_4 with increased respiration and reduced C_2H_4 production. Once the tissue has reached a certain stage of maturity, however, C_2H_4 not only promotes increased respiration but also increased C_2H_4 synthesis.

Controlling the effectiveness of C_2H_4 does not always involve a reduction in its activity. There are many beneficial effects of C_2H_4 that can be enhanced (see above). The techniques used to increase the effectiveness of C_2H_4 are almost the mirror image of techniques used to reduce its effectiveness.

Ethylene action can be enhanced by using cultivars that are sensitive and respond uniformly to C_2H_4 rather than cultivars that are C_2H_4 insensitive. An effective concentration of C_2H_4 should be maintained around the tissue for a sufficient time to elicit the full response. However, since the response to C_2H_4 is log-linear (a log increase in C_2H_4 concentration results in a linear increase in the response), there is an extremely large range over which the concentrations are effective. The application of C_2H_4 must be at the proper stage of development and at the proper temperature for the desired effects to be induced. Ethephon and similar C_2H_4-releasing chemicals permit the commercial application of C_2H_4 in the field. After harvest, C_2H_4 gas, either from compressed gas cylinders or catalytically generated from alcohol, can be used in enclosed storage rooms.

Controlling Ethylene Action

There are roughly three ways to control the action of C_2H_4 in plants. The first is to prevent the plant from being exposed to biologically active levels of C_2H_4. The second is to prevent the plant tissue from perceiving the C_2H_4 that is in its surrounding atmosphere or that is being produced by the tissue. The third is to prevent the plant from responding to the perceived C_2H_4 by controlling exposure to C_2H_4.

Preventing Exposure to Ethylene

The following should be done to prevent exposure to C_2H_4:

- Keep the air around the commodity C_2H_4 free.
- Use fresh, C_2H_4-free air from outside.
- Scrub C_2H_4 from the storage atmosphere.
- Use sachets of C_2H_4 absorbers inside packages to reduce levels.
- Segregate C_2H_4-producing commodities from C_2H_4-sensitive ones.
- Keep exposure to a minimum (in terms of both duration and level).
- Inhibit C_2H_4 synthesis (AVG, ACC synthase, low O_2, ACC oxidase).

Risk of exposure to C_2H_4 is usually not much of a problem in the field because the levels of C_2H_4 found even in polluted air rarely reach biologically active levels. However, in greenhouses, cold-storage rooms, and transportation vehicles, C_2H_4 can frequently accumulate to reach biologically active levels. Ethylene found in these enclosed spaces comes from varied sources, and the two most prominent sources are diseased, stressed, or ripening plant tissue and the incomplete combustion of organic fuels.

With proper ventilation of enclosed spaces and with persistent attention to the condition of adjacent plants and the operation of heaters and gas-powered forklifts, C_2H_4 can be kept below biologically active levels. Sometimes the C_2H_4 that we are concerned with comes from the plant itself. Application of inhibitors of C_2H_4 biosynthesis, such as AVG and AOA, to the tissue before or after harvest can significantly reduce this source of C_2H_4 exposure. For example, tissue can be prevented from making either stress or autocatalytic C_2H_4 by blocking the biosynthetic pathway for C_2H_4 synthesis. If exposure cannot be prevented or has already occurred, then both the duration of exposure and the level of C_2H_4 in the atmosphere should be kept as low as possible.

Preventing Perception of Ethylene

If significant amounts of C_2H_4 are in the immediate environment, certain methods can be used to block the perception of C_2H_4 by the plant. Here are some possible methods:

- Store at the coldest possible temperature.
- Use inhibitor of C_2H_4 perception: CO_2, silver (for example, silver thiosulfate), and 1-methyl cyclopropene (1-MCP).
- Use C_2H_4-insensitive cultivars.
- Interrupt the C_2H_4-induced signal.

Since perception is a metabolic process, holding the tissue at the lowest possible temperature will effectively reduce perception. Specific chemical inhibitors can also be used that directly interfere with the perception event.

A gaseous inhibitor like CO_2 or 1-MCP can be introduced into the atmosphere. The tissue can be dipped or fed a nonvolatile inhibitor such as silver thiosulfate, but this treatment is limited to nonfood crops. Ethylene-resistant cultivars can be selected or the tissue genetically engineered to lack the necessary biochemical receptors for ethylene or the signal pathway necessary to transduce the signal into a physiological event.

Even after the molecular perception event has occurred, blocking the transduced signal will effectively prevent perception. However, effective methods to do this will require a far greater understanding of the signal pathway than is currently available.

Preventing Response by the Plant

The third way to control C_2H_4 is to prevent the plant from responding to the perceived C_2H_4. This can be done by interfering with the metabolic machinery that is induced by exposure to C_2H_4, by methods such as the following:

- Store at coldest possible temperature.
- Store under CA or MA or in MAP.
- Inhibit or reduce specific enzyme activities using chemical inhibitors (for example, AIP) or genetic engineering (for example, antisense or other gene knockout techniques).
- Divert protein synthesis—by heat-shock, for example.
- Minimize time before use (for example, consumption).

Since all the effects of C_2H_4 on plants that we are interested in involve metabolic changes, reducing the rate of metabolism by lowering the temperature, withholding a vital reactant (for example, O_2), or by inhibiting a specific enzyme (for example, with a chemical or through genetic engineering) will prevent a response to C_2H_4. For example, ripening promoted by C_2H_4 often entails tissue softening that significantly reduces shelf-life. Using antisense technology to reduce the activity of enzymes involved in tissue softening has produced fruit that remain firmer longer. Ethylene also promotes phenylpropanoid metabolism in many tissues that use stress-produced C_2H_4 as a signal to induce defense mechanisms. Interfering with synthesis or activity of phenylalanine ammonia lyase (PAL, the first enzyme in phenolic metabolism) with chemical inhibitors or heat treatment eliminates tissue response to C_2H_4, preventing development of postharvest disorders.

Application of Ethylene

The quality of some fruits is increased when they are harvested at a mature but unripe stage that can withstand the rigors and duration of transport and then treated with C_2H_4 to promote ripening before sale. These fruit include avocados, bananas, honeydew melons, lemons, oranges, and tomatoes.

An effective atmosphere of 100 to 150 $\mu L\ L^{-1}$ C_2H_4 in air can be produced by a number of methods. The "shot" method introduces a relative large amount of gaseous C_2H_4 into a ripening room by metering C_2H_4 from compressed gas cylinders. Ethylene in air mixtures between 3.1% and 32% are explosive. While these concentrations are more than 200-fold higher than recommended, they have been reached when metering equipment has malfunctioned. Use of compressed gas containing around 3.1% C_2H_4 in N_2 ("banana gas"), eliminates this problem.

Catalytic converters are instruments that use a heated metal catalyst to convert alcohol into C_2H_4. They deliver a continuous flow of low C_2H_4 into the storage room. Ethylene can also be applied in aqueous form from decomposition of compounds such as Ethrel. While stable at acidic pH, Ethrel quickly breaks down to C_2H_4 as temperature and pH increase. Field application is approved for many food crops, but postharvest application is not approved.

Treatment with C_2H_4 stimulates many metabolic pathways, including respiration. Oxygen use is increased, as is the production of CO_2 and heat. Rooms designed to hold produce being exposed to C_2H_4 must be designed with extra air-moving capacity to ensure that an optimal ripening environment is maintained around the crop. Exposure to C_2H_4 must be uniform throughout the room and within packages. Heat of respiration and excessive CO_2 must be removed to maintain a proper environment. Loss of water by the crop will be increased by the rise in respiratory heat production. Maintaining a high RH can lessen water loss, but too much water vapor can decrease the strength of cardboard boxes and promote pathogen growth. Judicious maintenance of proper ripening environments will ensure production of high-quality fruit. Care must be exercised in venting and opening ripening rooms to prevent release of sufficient amounts of C_2H_4 to adversely affect other commodities stored in the same warehouse.

Conclusion

Ethylene can be both beneficial and detrimental to horticultural crops in storage. Practical uses for C_2H_4 and treatments to minimize its adverse effects have slowly accumulated over almost a century of study. The three general methods used to modulate C_2H_4 activity involve controlling exposure, altering perception, and varying the response of the tissue. An understanding of ethylene's synthetic pathway and mode of action has greatly improved the ability of postharvest physiologists to devise treatments and storage conditions to control C_2H_4 during the commercial storage and handling of horticultural crops. Simple methods like ventilation and temperature management can be combined with more sophisticated treatments like MAP and inhibitors of specific induced enzymes to provide conditions that optimize both storage life and product quality.

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Further Reading

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