Identification

Common Name	Ethylene	**Class**	Processing
Other Names	banana gas	**Code #: CAS**	74-85-1
		Code #: Other	DOT #: UN 1962/UN 1938
Chemical Name	Ethene	**MSDS**	yes

Characterization

Composition C_2H_4

Properties A colorless, flammable gas with a slightly sweet odor, soluble in water.

How Made Made from hydrocarbon feedstocks, such as natural gas liquids or crude oil. Produced almost exclusively from the pyrolysis of hydrocarbons in tubular reactor coils installed in externally fired heaters. These heaters are operated at high temperatures (750-900°C), short residence times (0.1-0.6s), and low hydrocarbon partial pressure. Steam is added.

Ethylene may also be produced from ethanol in fixed or fluid-bed reaction systems. This means it could be made from biomass fermentation, but that is not currently done in the U.S. The amount of heat necessary for this process would still result in what may be considered a "synthetic" material.

Ethylene is given off naturally by ripening fruit and by some micro-organisms. These sources of ethvlene have never been harnessed commerciallv.

Specific Use(s) Ripening and coloring fruit, including bananas, pears, mangoes, tomatoes and citrus. Induces flowering in pineapples when applied in the field. Can be used to improve growth and appearance of bean sprouts.

Action Acts like the natural analog plant growth regulator by accelerating the ripening process through an exact mechanism that is not fully understood. Many volumes of technical information have been generated in books and journals about ethylene's effects on plants and theories about the mode of action. In general the gas is produced in fruit when physiological maturity is reached and the gas triggers the chemical changes which take place at ripening. In pineapple, the gas is generated at vegetative maturity of the plant which triggers the flowering and fruiting cycle.

Combinations One of the more commonly used forms is the ethylene generating chemical, (2-chloroethyl) phosphonic acid, known as ethephon. This is mostly used for pre-harvest applications with the only post-harvest one being the degreening of lemons in Florida (Sherman, 1985). "Banana gas" is pure ethylene gas in a compressed cylinder which is diluted with an inert gas.

Status

OFPA In processing this would be considered a processing aid. For crop use it could be considered a production aid but is not specifically mentioned in any of the exempt categories in 6517(1)(B) (i).

Regulatory Status Ethylene is regarded legally as a pesticide for regulatory purposes. It must be registered with the EPA and appropriate state agencies.

Historic Use Allowed for use on bananas by many organic certifiers and state programs. Some certifiers have certified tropical fruit drying facilities that use ethylene for mango ripening.

International IFOAM - not mentioned. CODEX - not mentioned specifically.

Existing Restrictions Allowed for use on bananas only. (NOSB recommendation, OMRI list)

Proposed Annotation

OFPA Criteria for Processing

2119(m)1: chemical interactions Not Applicable **2119(m)2: toxicity & persistence** Not Applicable

NOSB Processing Criteria #2 2119(m)3: manufacture & disposal consequences

Ethylene or olefin plants require extensive support facilities to comply with environmental regulations. These include boiler feed water yreyaration, treatment of noxious effluents, and steam and electric generation.

NOSB Processing Criteria #3 - 2119(m)4: effect on human health
Also "Nutritional quality of the food is maintained."

Highly flammable and explosive. See discussion for more on explosiveness. Exposure to gas causes dizziness and could cause suffocation from decreasing the amount of oxygen. See discussion for more.

2119(m)5: agroecosystem biology Not Applicable

NOSB Processing Criteria #1 2119(m)6: alternatives to substance

Natural ripening alternatives: temperature control, mix with apples for natural ethylene generation. The alternative for induction of flowering in pineapples is calcium carbide used to form acetylene. See discussion for details. The alternative for sprout production is to do without ethylene and get inferior production, or to use ethylene generated by a different means if possible to develoy a natural source. See discussion for more.

NOSB Processing Criteria #6 2119(m)7: Is it compatible?

NOSB Criteria for Processing

NOSB Proc Criteria #4: Preservative?

It is not a preservative, nor is it used to replace anything lost in processing.

NOSB Proc Criteria #5: GRAS & residues?

It is not on the GRAS list because it is regulated by the EPA instead of the FDA. It does not have any residues because it goes off as a gas into the air.

NOSB Proc Criteria #7: Essential & Minimum?

Fruit will ripen without using ethylene gas. It does not ripen evenly and this necessitates more handling in the form of sorting. It is used at the minimum concentration necessary to get a consistent reponse.

Discussion

See attached.

Ethylene Discussion

History

The first use of natural ethylene in fruit ripening was described in the Bible. The prophet Amos was described as a "gasher and gatherer" of figs. Gashing figs was known to promote stress ethylene production mimicking the action of the wasps when they exit pollinated fruits, and this triggered ripening. Also in ancient times the Chinese placed weighted lids on growing bean sprouts to promote hypocotyl thickening and crispness (Abeles, 1992). Ethylene was used unknowingly to ripen bananas in both East Africa and Samoa by burying them in fire-warmed pits, thus using residual ethylene from the smoke of the fire as the ripening agent.

In more recent times it was discovered that warming citrus fruit with kerosene heaters in closed spaces caused a degreening effect that wasn't just due to the heat (Denny, 1924). In the 1920's it was shown that the cause of this ripening effect was ethylene gas, which fortunately was already being produced commercially for other purposes (Chace, 1934). In the 1930's it was verified that ethylene was indeed produced by plants and that this gas was the same composition as that given off by the kerosene heaters (Abeles, 1992). In the years following this, researchers determined that ethylene produced a variety of effects in plants and it could be classified as a plant hormone, or more correctly, a plant growth regulator.

Concern was voiced early on in the commercial exploitation of ethyiene about the residual effects of ethylene treatment; that it would be used to allow inferior fruit to be sold at a higher price. Researchers then showed that the ethylene treated fruit were equivalent in quality and "healthfulness" to naturally ripened fruit (Chace, 1934). This is largely because the fruit must have reached its physiological green maturity stage in order to respond to external ethylene, and then the ripening changes triggered by ethylene are essentially the same between the treated and naturally ripened fruit. These changes include starch and sugar content, acidity and concentration of pectic substances (Clendennen, 1997).

Environmental Concerns

The main safety concern in relation to ethylene use has been due to the explosive nature of the gas in the air. This is of primary concern in design and operation of ethylene application facilities. Both the EPA, local fire marshal rules, and insurance companies have very specific labelling and registration requirements for the ethylene itself and the process used to apply it, down to the electirical wiring and piping used in ripening rooms. Note that the gas is explosive in air at concentrations from 3.1% to 32% (31,000 to 320,000 ppm). The minimum explosive concentration (3.1%) exceeds the suggested ethylene concentrations for tomato ripening and citrus degreening respectively by 200 and 6200 times (Sherman, 1985). The "banana gas" and catalytic generator sources of ethylene are considered the safest because they are more easily monitored, but explosive accidents have happened in the past and operaors should be well trained and prepared (Sherman, 1985).

Another concern with ethylene is the issue of air pollution. The amount of ethylene given off from either manufacturing or ethylene treatment facilities is miniscule compared to the ethylene released into the air from hydrocarbon emissions from auto exhaust, petrochemical plants or even fires. There are no national air quality standards for ethylene levels, but there are some from the American Industrial Hygiene Association (Abeles, 1992). Ethylene is degraded in the atmosphere by UV light present in sunlight. Ethylene air pollution can reduce ozone pollution. It can however be present in strong enough concentration to produce phytotoxic effects.

Nutrition Effects

While the effects of ethylene on the ripening process have been studied well for both bananas and pineapple, there is no clear and consistent evidence that artificially ripened or induced fruit has any more or less nutritive value than naturally ripened fruit (Chace, 1934; Abeles, 1992; Clendennen, 1997). If anything, pineapple treated with ethyiene had increased sugars, proteins, fruit acid and soluble solids, but lower fruit weight (Mwaule, 1985; Ahmed, 1987)

Specific Uses: **Mango**
The situation with mangoes is similar to that with bananas in several ways (Sarnataro, 1999). Mango trees ripen their fruit unevenly on the tree so **generally** whole trees are picked by hand at an average maturity time, resulting in a mixture of maturities of fruit. **Ethylene** treatment results in even ripening which cuts down the extensive h i t handling and sorting needed for untreated fruit when they are being cut for drying **(Sarnataro,** 1999). Fruits harvested at their physiologically mature and half-mature stages develop good quality **characteristcs** when treated. Immature fruit **will** not respond properly to ethylene and will not be useful for drying **(Abeles,** 1992).

The tropical fruit drying operations typically process bananas, mangos **and/or** papayas in the same facility and are currently allowed to use ethylene on the bananas but not the other fruits. From a certification point of view it has been very **difficult** to enforce the use of the gas for bananas only in these situations with just one annual inspection, and several certifiers have gone ahead and certified the use of ethylene on mangoes and papayas.

Specific Uses: **Pineapple** Flowering
While the cultural techniques for pineapple may not be well known here in the US, pineapple is a large economic crop on several continents and in many countries. The market for organic pineapple has been quite small so far but it could expand dramatically with the demand for processing pineapple on the upswing. The issue limiting pineapple production at this time is the use of a material, either ethylene or calcium carbide, to induce flowering (see petitions from Made in Nature, **TropOrganics** S.A., Pina **Perfecta** S.A., and Eco-LOGICA). The literature does support the petitioners in that a crop of **sufficient** size, uniformity, and ripening period cannot be produced without something to induce flowering **(Abeles 1992; Bose,** 1983; Williams, 1987; **Lacoeuilhe,** 1983)

flowering in pineapple occurs when plants reach a certain size. While some research shows an enhancement in flowering from short **days,** low night temperatures and water stress, the flowering can happen in one field at quite different times because the plants have been growing for at least 20 months before they start to flower **(Reinhardt,** et al, **1986).** Different pineapple regions also report quite different results about what triggers natural flowering. All recommendations for pineapple culture **suggest** using a material for flower induction to acheive even flowering and a uniform harvesting period. **While** the induction can result in producing a crop out of season, it is also necessary to produce a uniform crop in season. **This** is considered important for processing as well as for **predicatable** marketing of the crop, **as** most pineapple is grown for export and a shipping container must be filled for each harvest.

Numerous chemicals have been studied for their usefulness in flower induction, including NAA (Naphthaleneacetic acid), Urea, Ethylene (mostly Ethephon) (with or without Activated Charcoal), CCC (chlormequat), GA3 (gibberellic acid) and Calcium **Carbide/Acetylene.** The petitions received by the NOSB are for Ethylene and Calcium **Carbide/Acetylene.** These two systems both need to be discussed, for either or none to be selected.

1. Ethylene – **Normally** applied to the plants at 12 to 24 months in the form of Ethephon at a rate of 25 to 100 ppm. Can be mixed with either urea or activated charcoal as carrier, or applied alone. flower induction is triggered in 25 to 45 days, depending on time of year, size of plant, and temperature. The ethylene breaks down either in the plant cells or is volatilized into the air very quickly. Activated charcoal is made from steam treatment of natural coal or carbonized wood materials and would be considered to be natural.

2. Calcium **carbide/Acetylene** – Calcium carbide is a synthetic material made from limestone or quicklime mixed with crushed coke at high temperatures. Its chemical formula is CaC_2 and CAS number is 75-20-7. When sprayed on the plants in water it forms acetylene gas which is a precursor to ethylene. The acetylene enters the plant and is transformed into ethylene in the cells, thus triggering flowering. A study of the subject by Oregon Tilth (Coody, **1996)** determined that the calcium carbide and acetylene are considered synthetic under **OFPA,** while the ethylene made by the plants, being made from a "**naturally** occuring biological process" **(OFPA, 1990),** would be considered natural. Besides acetylene created from the reaction of calcium carbide and water, there is calcium hydroxide produced. This is a prohibited synthetic material for organics **as** a fertilizer. Due to lack of information about the nature of calcium carbide, it was used by several of the petitioners and other growers under the

mistaken impression that it was derived from limestone and therefore natural. It is also perceived (but there is no direct evidence behind this) to be more desireable from an organic point of view than ethylene use.

As far as which of the two materials works better, there seems to be considerable disagreement from region to region. Both work better than the control treatments, and in general better than the NAA or other chemicals tested (Bose, 1983; Prasad, 1987). Both materials as externally applied would be considered to be synthetic under OFPA and no testing was found in the literature of any material that would be considered natural. Because these materials are so simple chemically they possibly could be able to be formed naturally. Charcoal and limestone for instance, may be able to create the same effect as calcium carbide. Ethylene has possibilities of being generated naturally, either from micro-organisms, natural ethanol, or captured from ripening fruit. The production of organic pineapple is way too small at this time for these alternatives to be realistically explored.

Specific Uses: Bean Sprouts

About half of the bean sprouts commercially produced in California today are gassed with ethylene (Abeles, 1992). Both the growth and the quality of the sprouts are improved by this practice. In natural germination in soil a bean sprout is exposed to a higher concentration of ethylene that in the air and this improves the trasfer of nutrients from the cotyiedon to the hypocotyl (future shoot to future root). When grown in large quantities out of the soil environment, any ambient ethyiene is soon used up, and the addition of ethylene to sprout rooms at low concentrations mimics the atmosphere present in soil. Ethylene stimulates the physiological aspects of root growth (Abeles, 1992).

One organic sprout grower who has operations in Japan and California has developed a unique and proprietary technique for creating ethylene gas for sprout rooms from food grade ethyl alcohol. Although the ethanol is "natural", the process used involves heating it to 360°F and therefore needs a determination from the TAP reviewers as to whether this would be considered synthetically produced ethylene. Whether synthetic or natural, it also needs to be decided whether this use of ethylene is allowed for organic production. (see attached memo from CCOF to OMRI). The concentration used in the sprout rooms is about 0.1ppm, compared to the 10 ppm or so used for ripening bananas.

Conclusion

This is one of the more difficult subjects facing the NOSB in the National List process. Ethylene as used today is a synthetic analog of a natural gas produced by plants. There is precedent in the previous NOSB recommendations for the approval of analogs of natural materials such as magnesium sulfate, copper sulfate, hydrogen peroxide and ethylene for bananas. There is also precedent for materials being used as plant growth regulators being approved by the NOSB, such as natural gibberelic acid and possibly amino acids.

The basic argument in force here is that agriculture is inherently not the same as a natural system, but organic agriculture can be thought of as an augmented natural system. The augmentation takes the form of materials and practices designed to acheive agricultural production of crops in sufficient quality and quantity for human consumption while maintaining the ecosystem without adding chemicals that have a lasting degradative impact. Ethylene fits this argument in most respects.

On the other hand, this material is being used out of its strictly natural context and is being used as a plant growth regulator to potentially "trick" plants into doing something they may not be ready to do naturally. It is formed from a synthetic process which could have negative environmental impacts from its manufacture. It may be able to be made from a natural starting point at some time in the future if the economic pressure is applied for that to happen.

The Technical Advisory Panel reviewers, and then the NOSB members are being asked to decide on each use of ethylene for organic crop production and post-harvest handling. Please weigh all the factors carefully and make your best recommendation.

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* Articles and documents that are attached to this file

To: OMRI materials committee
From: Ryan Sarnataro, Committee for Sustainable Agriculture
RE: Materials classification - Ethylene
Use: Ripening mango (and papaya) for processing dreied fruit.
1/20/99

Background.

Ethylene gas is currently an approved material for use on bananas for the fresh market. Its approval is based on the specific requirements of banana production and marketing as well as the **fact** that ethylene is naturally occurring byproduct of ripening fruits.

Since ethylene is approved by the NOSB and the European community for certain applications I am presenting the case for extending it's use in another specific instance rather than information on the material itself. Specifically, I would like to see the gas used to facilitate the production of high quality dried fruit. Without the use of a ripening agent the cost of producing organic dried mango will be significantly higher than conventional and the quality will be lower.

Some certification agencies allow the use of ethylene in tropical dried fruit processing. Others do not specifically disallow it. Tropical dehydration facilities that process both banana and mango (or papaya and other fruits) can **use** ethylene for one item and not **another.** There is no logical basis for this. **A** ruling from OMRI will take the current ambiguity and inconsistency out of this area.

Use:
Ripening for preparation for drying is done in well sealed rooms typically on the scale of 3000 **Cu.** Ft. Ethylene gas is introduced to manufacturer specified concentrations for a period of 16 to 24 hours. No temperature controls are used in the rooms. Typically 16 hours is sufficient and the rooms are vented at this time as the fruit begins to produce ethylene gas by itself and will continue ripening without help.

Justification

While the fresh mango season can extend from March to September the season for each individual variety of mango only lasts for a few weeks or at most two months. This means that most mangos ripen at one time on a tree. Consequently mango harvesting (or cutting) is usually done on a tree by tree basis with a mixture of levels of ripeness in a cut.

Tropical mango harvesting is often done by whole families climbing trees without any safety equipment. Picking only ripe fruit means more trips up each tree for the same quantity of fruit. This is both less efficient and more dangerous for the cutters.

To get the optimal flavor and nutritional content in a dehydrated mango it is important to start with an unbruised fully ripened fruit. When fruit is sorted for drying only a small percentage is ripe enough for initial processing. Left on its own mango can take two weeks or longer to ripen sufficiently for processing. Sorting and reselecting mango for this period will cause a variety problems and increase costs and spoilage for any medium or large size processing facility.

The first problem is that a very large amount of storage space will need to be allocated for ripening mangos. Approximately 15 pounds of fresh fruit are required for each pound of dried fruit. A dryer producing 1000 Lbs a day in season would need to have over 200,000 lbs of fresh mango on hand at a time. Both the cost of storage space and the cost of field boxes to store the mango in are quite high. Using a ripening gas reduces the storage requirement to 3 days supply or 75% less mango.

Each time the mango is handled bruising increases and quality decreases. The extra handling, of course, also increases costs.

Mango left to ripen naturally will mature at an unpredictable rate. This means that each sort will uncover overripe mango cannot be processed. It also means that more ripe mango will be "discovered" on some days than the plant can process while less will be available on others. The combination of waste and inefficiency is another unnecessary cost. Ripening mango with ethylene can assure a plant of a much more predictable supply of fruit to work with. Using ethylene is not a substitute for quality control and sorting. However, the predictability it introduces is extremely valuable.

I think it is important that organic producers be permitted to use natural materials to achieve high quality and productivity. In the case of ethylene gas for dehydration we are talking about a processing aide that simply increases the concentration of what is naturally in the fruit and is not detectable in the finished product.

Bananas are a much larger crop than mangos. Consequently bananas have a much stronger advocacy base than mangos and this advocacy has created a different set of rules for bananas and other tropical fruit crops. Allowing ethylene for dried mango production will help level the playing field between different tropical crops.

Residues:

Ethylene is a natural byproduct/ cause of fruit ripening and is therefore not detectable as a separate additive. The process of dehydration involves peeling and cutting the fruit and laying the cut fruit out exposed to air for a period of 3 to 48 hours depending on the temperature used. The cut fruit will lose over 80% of it's mass in this process. This process vents most volatile compounds from the fruit.

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Control of Ethylene in the Postharvest Environment

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The beneficial and harmful effects of ethylene (C_2H_4) on harvested horticultural products are well documented. The purpose of this presentation is to outline the technologies currently available in commercial horticulture for both the application of C_2H_4 to harvested horticultural crops. and the protection of these crops from the undesirable effects of C_2H_4. The research base for this topic cuts across many areas of considerable postharvest research. including temperature effects. atmospheric modification. and regulation of C_2H_4 synthesis. A complete review of any of these topics is beyond the scope of the presentation. and. in most instances. the reader is referred to recent research reports or comprehensive reviews of the specific subject.

Postharvest application of ethylene

History and economic importance. The use of practices to hasten fruit ripening dates to antiquity (35). although it was not known until relatively recently that C_2H_4 was the causal agent. The earliest practices employed the emanations from ripe fruit or the smoke generated from burning combustible products. The use of kerosene stoves for "sweating" or "forced curing" of citrus was a well-established commercial practice by the late 1800s. It was believed that the high temperatures and high humidities in the "sweating" rooms resulted in degreening. Sievers and True (43) demonstrated that degreening was due to some unknown gaseous product from the incomplete combustion of kerosene. Denny (15) provided convincing evidence in 1924, that C_2H_4 was the effective constituent in "stove gas", and described a method for commercially degreening lemons using cylinders of compressed C_2H_4 gas.

Denny's work (IS) marked the beginning of the knowledgeable use of C_2H_4 gas in commercial degreening and fruit ripening practices. Harvey (27) published the 1st comprehensive bulletin describing the commercial application of C_2H_4 for ripening bananas. pineapples, dates, Japanese persimmons, tomatoes, hard pears, apples, and muskmelons. Interestingly, one early commercial use for C_2H_4 was for blanching celery (26). a practice which has long since been discontinued.

Rosa (39) predicted that for tomatoes "the acceleration of the coloring process by low concentrations of C_2H_4 is sufficiently great to make it commercially valuable. . .". These words seem especially prophetic when considering the economic importance of C_2H_4 applications in postharvest horticulture today. Although it is not possible to assign an exact monetan value for C_2H_4 use. it is accurate to state that C_2H_4 applications play an important role in the orderly marketing of fresh citrus (excluding limes). tomatoes. bananas. mangoes. and honeydew melons. Recent reports (20. 47. 48) indicate that these products have a value of over 1.9 billion dollars in the United States alone.

Sources of ethylene. There are 3 potential sources of C_2H_4 for commercial use: liquids, gases, and ripening fruit. Liquid sources are C_2H_4-releasing chemicals such as (2-chloroethyl) phosphonic acid, commonly known as ethephon. This chemical is registered and widely used for preharvest applications to concentrate maturity or otherwise facilitate harvest of several horticultural crops. According to the label. the only presently registered use for postharvest application is for the degreening of lemons in Florida (49). Therefore. further discussion will be limited to the other sources of C_2H_4 for postharvest use.

Ethylene gas can be generated *in situ* or purchased in compressed cylinders. The gas is generated by catalytic conversion of a flammable liquid concentrate (Ethy-Gen) to C_2H_4 gas (11). This is one of the most popular commercial sources of C_2H_4 gas. The other widely used source of C_2H_4 gas is that purchased in compressed cylinders of either pure C_2H_4 or C_2H_4 diluted with an inert gas such as the product called "banana gas" (50, 3. 51).

Postharvest physiologists are aware that ripening fruit produce C_2H_4, but many commercial handlers either are unaware of or ignore this fact. If ripening room operators conscientiously monitored C_2H_4 levels in these rooms, then the use of ripening fruit might be a viable commercial alternative. However, the use of ripening fruit as a source of C_2H_4 generally is limited to home ripening recommendations.

Which source to use? The source of C_2H_4 used by handlers normally is determined by the facilities for treatment, legal considerations. cost, and safety. The use of C_2H_4 gas requires some type of enclosure. The cost of building special ripening facilities with precise temperature and humidity controls is well-justified when repeated ethylene applications are required.

Each handler also must consider the legality of C_2H_4 application (25). Ethylene gas used for plant regulation such as coloration or ripening of fruit and vegetables is regarded legally as a pesticide for regulatory purposes (52). Therefore, it must be registered with the Environmental Protection Agency (EPA) and the appropriate state agencies. Containers of C_2H_4 gas or C_2H_4-releasing liquids must bear EPA approved labeling. including EPA registration and establishment numbers, intended uses. ingredients statement. and appropriate precautionary labeling statements.

Because of the explosive nature of certain mixtures of C_2H_4 and

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example. these recommend- -s specify the type of electrical wiring and piping to be used i -ening rooms. Other state and local fire marshal rules or building codes may apply (46). Generally. these rules all pertain to safety. but this may change. Recently. a bill was introduced in the California state legislature which would have required that tomatoes treated with C_2H_4 be labeled "artificially ripened".

Safety must be a prime consideration in the technology of C_2H_4 application. Ethylene gas is explosive in air at concentrations from 3.1% to 32% (31,000 to 320,000 ppm) (8). To put these values in proper perspective. the minimum explosive concentration (3.1% C_2H_4 in air) is the suggested C_2H_4 concentrations for tomato ripening (24) and citrus degreening (56) by 200 and 6200 times, respectively. One might conclude. therefore. that it would take extreme negligence for the fruit ripening process to be dangerous. However, explosive accidents have ocurred in the past. Safety during C_2H_4 application is dependent upon the operator. C_2H_4 source. and the method of application. The safety advantages afforded by the catalytic generator. and "banana gas" sources of C_2H_4 may be negated by the operator and the method of application. Each ripening room operator should have equipment to measure C_2H_4 concentrations. This is accomplished readily with an inexpensive gas detector kit and C_2H_4 detector tubes obtainable from several companies that sell specialty gases.

Methods of applying C_2H_4 gas. The methods of applying C_2H_4 gas can be placed into 3 categories: shot. trickle, and flow-through. Shot methods employ the rapid injection of C_2H_4 into the ripening room atmosphere. This can be accomplished by weighing he amount of C_2H_4 dispensed from a cylinder, using a premeasured amount (lecture bottle). or timing the delivery of C_2H_4 from a cylinder. The original system described by Denny (15) was a shot method. with the C_2H_4 delivered in a premeasured container. Dispensing C_2H_4 directly from large cylinders can be extremely dangerous and never should be done. Shot methods have the advantage of simplicity, but the disadvantage of requiring frequent room aerations to prevent carbon dioxide (CO_2) buildup which necessitates recharging the room with C_2H_4 following each aeration.

The "trickle" method can be defined as the slow. continuous dispensing of C_2H_4 into the ripening room atmosphere. Usually. compressed C_2H_4 cylinders are equipped with a regulator and flow meters to control the flow of C_2H_4. In practice. the catalytic generator also is a trickle method for introducing C_2H_4. Trickle methods are safer than the shot methods but they still may require periodic room aeration and recharging with C_2H_4.

The flow-through system is a modification of the trickle methods. In principal. the flow-through system supplies a constant. ripening-effective blend of C_2H_4 and fresh outside air which passes over the product and out an exhaust port in the room. The constant air exchange prevents CO_2 accumulation to inhibitory levels and eliminates the need for periodic aeration. The flow-through system has proven to be a safe and efficient method for introducing C_2H_4 for citrus degreening (56) and tomato ripening (42). Sherman and Gull (42) provided detailed instructions for installing a tlow-through system.

Commodity requirements. One of the most important requirements is that the commodity be physiologically mature (capable of continuing normal development when detached from the plant). For some commodities like citrus. legal definitions of maturity exist (57). For others. such as tomatoes, there are no legal definitions of maturity, and it is difficult to determine whether a green fruit is mature or immature based on external appearance. Other important commodity requirements include the desired C_2H_4 concentration. sensitivity to CO_2, and the optimum temperature, humidity. and length of treatment. Local extension recommendations should be consulted for specific commodity information (24, 31, 56).

Protecting harvested products from ethylene

History and economic importance. Some of the earliest investigations of C_2H_4 were related to its detrimental effects on plants and have been thoroughly reviewed (I). Crocker and Knight (12) re- ... of the adv- effects of C_2H_4 on harvested horticultural products continue t- -pear in literature. Risse and Hatton (38) recently reported the detrimental effects of C_2H_4 on harvested watermelons.

It is extremely difficult to assess the economic importance of protecting harvested horticultural products from C_2H_4. Abeles has pointed out the problems of estimating the economic losses caused by C_2H_4 as a component of air pollution (1, 2). Detrimental effects of C_2H_4 during the normal short-term marketing of fruit and vegetables are not well-defined and certainly are secondary to considerations regarding the maintenance of optimum temperature and humidity in the postharvest environment. However. costs to the individual shippers involved can easily run into tens of thousands of dollars when losses do occur from problems like russet spotting of lettuce or yellowing of cucumbers. Economic evaluations of long-term storage responses to C_2H_4 (5, 17, 28) generally have not been reported because investigators have not wanted to speculate about extended season prices. Detrimental effects of C_2H_4 may be most important for ornamental crops where estimates once placed postharvest losses at about 20% due to mishandling. However. mishandling included poor temperature management. and improper humidity. as well as exposure to C_2H_4 (45). In summary. losses caused by C_2H_4 are known to occur, but they are usually quantitatively undefined. A conservative estimate for the United States would be in the tens of millions of dollars annually.

Strategies for protection. The strategies for protecting harvested horticultural products from the detrimental effects of C_2H_4 can be placed into 3 major categories: avoidance, removal. and inhibition. Although these techniques are grouped for convenience it should be remembered that they may overlap and are not mutually exclusive.

Avoidance. Circumvention of undesirable product exposure to C_2H_4 begins with careful harvesting, grading, and packing which includes selecting the desired maturity and avoiding mechanical injury. The 2nd and probably most important step in avoidance is proper temperature management. which should include rapid cooling to the product's lowest safe temperature. This suppresses ethylene production and reduces sensitivity to ethylene (9). Chilling temperatures should be avoided when handling chilling-sensitive commodities. however. because increased C_2H_4 production frequently follows chilling injury (55). Ethylene-sensitive commodities should not be transported. stored. or displayed with C_2H_4-generating commodities. Guidelines suggesting compatible product mixes have been published (33). Given the food distribution system in the United States. it is not possible to avoid undesirable exposure to product-generated C_2H_4 completely. but exposure should be kept to a minimum. Placing ripe tomatoes between iceberg lettuce and cucumbers in retail produce displays may be colorful. but it completely ignores the biology of these products and constitutes unnecessary exposure of the produce to undesirable temperatures and C_2H_4.

Other management steps for avoidance include minimizing the use of internal combustion engines during product handling in enclosed spaces. and following strict sanitation practices to insure that overripe and decaying products are promptly removed.

Removal. Undesirable levels of C_2H_4 in produce storage areas can be removed by simple ventilation with fresh air if the air is not polluted with high C_2H_4 levels. Usually, one air exchange per hour is required to maintain a low C_2H_4 level. Ripening room facilities located in produce distribution centers always should be vented to the outside to ensure that C_2H_4 is not accidently introduced into the storage environment of the distribution center.

C_2H_4 can be scrubbed from the atmosphere by trapping and/or conversion to other products when ventilation cannot be used for removal. A large number of reagents and techniques have been tested over the years (1, 7, 15), but only potassium permanganate is presently in common commercial use. To be effective, $KMnO_4$ must be adsorbed on a suitable carrier with a large surface area. Celite, vermiculite. silica gel. alumina pellets (1), perlite (40). and expanded glass (32) have all been successfully used as carriers. A number of commercial potassium permanganate scrubbers are avaiable in sachets. filters. blankets. and other specialized trapping devices (? . 18. 19). Ethylene is trapped most effectively when air is drawn through the scrubber. Potassium permanganate scrubbers are advantageous because they change from purple to brown as the MnO_4^- is reduced to MnO_2. The major disadvantage of permanganate scrubbers seems to be their expense (7, 40).

Heated catalyst ethylene scrubbers were successfully used during the 1983–1984 storage season in commercial controlled atmosphere apple storages to maintain ethylene concentrations below 2 ppm in New York State. and below I ppm in England (Blanpied, personal communication). Another technology for C_2H_4 removal, which seems promising but has yet to be commercially developed. is the use of UV light (41). Also, there is a potential for biological removal of C_2H_4. Abeles (2) discussed the role of soil bacteria as a sink for atmospheric C_2H_4. Perhaps the C_2H_4-consuming *Mycobacteria* that have been isolated from soils (14) could be genetically engineered to perform satisfactorily in horticultural applications.

Inhibition. Controlled atmospheres (CA) have been used widely for many years for the long-term storage of apples (13) and. to a limited extent. for other commodities. such as pears (44) and cabbage (22). CA systems require special refrigerated, gas-tight structures (16. 29) which allow for precise temperature control and maintenance of the storage atmosphere (primarily reduced O_2 and elevated CO_2). CA storage has multiple effects on the physiology of the commodity (30. 44). but the retardation of ripening is at least partially attributable to the low O_2 atmosphere. slowing C_2H_4 synthesis and action. and the elevated CO_2 inhibiting C_2H_4 action. Very low levels of O_2 (17) and C_2H_4 scrubbing (5, 7) may enhance the storability of products in CA. The use of CA essentially has been limited to long-term stationary storages. but at least one company. now out of business. incorporated CA capability into modern transport containers (21). Modified atmosphere. (a less precise type of CA) have been used for many years on several commodities during transport.

Low pressure storage (LPS) seems to offer potential for prolonging the useful life of horticultural commodities (10. 34). LPS effectively reduces O_2 levels and. in this respect. is similar to CA storage. Storage at subatmospheric pressures also increases the diffusivity of volatile gases, however. including C_2H_4, from the internal atmosphere of the commodity (10). Commercial applications of LPS systems have not been widespread to date. Grumman Allied Industries developed the Dormavac System for hypobaric transporation of perishables (23). but this met with only limited commerical success and was discontinued.

Chemical treatments could be used to protect horticultural products by inhibiting C_2H_4 synthesis and/or action. Yang and Hoffman (58) reviewed many of the known inhibitors of C_2H_4 biosynthesis. Some inhibitors appear to have commercial potential for regulating fruit ripening (6, 54), but their use on food products must be preceded by FDA and EPA approvals. Many ornamental horticultural crops can be protected from the detrimental effects of C_2H_4 by treatment with the anionic complex silver thiosulphate (STS). Veen (53) recently reviewed some of the uses for STS in commercial horticulture.

Methods outlined here for removal of C_2H_4 and inhibition of C_2H_4 synthesis and/or action should not be regarded as panaceas for solving C_2H_4-related problems in commercial horticulture. The best defenses for protecting harvested horticultural products from the detrimental effects of C_2H_4 are the steps outlined under avoidance. When the shelf life of products can be extended further by the methods of C_2H_4 removal or inhibition of its action, then their use may be advantageous. The long term storage of apples in controlled atmospheres with C_2H_4 scrubbing is a good example of postharvest practices which employ avoidance, removal, and inhibition methods for protection from C_2H_4 (7, 17).

Summary

Control of C_2H_4 in the postharvest environment is of great importance in commercial horticulture. Application of C_2H_4 facilitates the orderly marketing of citrus, tomatoes. bananas, mangoes. and honeydew melons. Safe and efficient application methods have been developed and are readily available for commercial use. Detrimental effects of C_2H_4 on harvested horticultural products largely can be avoided by careful management of the postharvest environment, but this management is not always practical in commercial situations. Technologies exist for removing C_2H_4 or inhibiting C_2H_4 action. but there is still considerable room for improvement during the storage and handling of most commodities.

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AUG 09 1999

TECHNICAL ADVISORY PANEL
REVIEW ON ETHYLENE
(FOR PROCESSING CLASSIFICATION)

NATURAL AND SYNTHETIC FORMS

Ethylene is produced naturally by ripening fruits, and also by some microbes.

All other forms of ethylene are synthetic, such as being produced in a variety of ways, as delineated in the NOSB Materials Database.

CRITERIA:

2119(m)(1) - Chemical interactions in organic farming systems:
Not applicable for the purposes of discussing ethylene as a post-harvest material.

2119(m)(2) - Toxicity and persistence in the environment:
Not applicable for the purposes of discussing ethylene as a post-harvest material.

2119(m)(3) and NOSB processing criterion #2 - Consequence of manufacture, misuse, disposal:
Manufacture of synthetic ethylene on a large scale requires significant support and precautionary measures to ensure human and environmental safety. In this regard, impact could be deemed significant, at least in relation to energy input, as well as other material aspects, depending on the case.

Small scale production of synthetic ethylene gas may not necessarily endanger human safety or the environment. As these scenarios vary from one to another, they may have to be evaluated individually.

Use of naturally-generated ethylene from ripening fruits is not a widespread practice, although it does not result in any obvious environmental or safety hazards. As such, it would be an acceptable method under this criterion to use natural ethylene in post-harvest organic systems.

2119(m)(4) and NOSB processing criterion #3 and #4 - Effect on human health and nutrition, preservative effects, replacement of losses during processing:
Significant precautions must be followed to ensure that handling of ethylene gas does not result in human injury, either by explosion or over-exposure. (Far greater risks exist in the actual ethylene factory than in the ripening room.) Naturally-emitted ethylene generally does not exist in high enough concentrations to warrant these considerations. Tanks of compressed gas must always be treated with care, regardless of the source.

The effect of ethylene application to fruits post-harvest does not indicate conclusively negative effects on the nutritive value of the product. Further research is warranted in this area, to further analyze the effects on constituent nutrients in fruits which are ripened naturally versus those treated with supplemental ethylene, be it from a natural or synthetic source. Ethylene application is not a preservative treatment. There is nothing to indicate that synthetic and naturally-occurring ethylene are anything but exactly the same in molecular structure or action (combination products obviously notwithstanding). Furthermore, so far as is known, the ethylene emitted by one type of ripening fruit is the same as that from other ripening fruits. For the time being, it may therefore be safe to conclude that ethylene in and of itself does not pose concerns regarding negative impact on human nutrition.

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2119(m)(5) - Agroecosystem biology:
Not applicable

NOSB criterion #5 - GRAS, residues:
Not listed as GRAS, as this is an EPA-controlled material. No residues are encountered with any of the forms used post-harvest. All excess ethylene applied dissipates into the atmosphere. Pollution of the atmosphere in this regard may need to be monitored in some cases, if deemed excessive.

2119(m)(6) and NOSB processing criteria #1 and #7 - Alternatives to substance, essential need:
Ethylene is not an essential material to add to fruit, as sufficiently mature fruit produces it naturally; the minimum required from outside sources is zero. Lack of use of ethylene from external sources results in less uniform ripening of fruit, and consequent greater losses. Ethylene application has also been shown to improve sprout production. The advantages of using ethylene are undeniable, from logistic, yield, labor, and marketing perspectives.

The alternative to synthetic ethylene is naturally generated ethylene. One conceivable approach is to use stocks of ripening fruit (such as ripening apples, bananas, or other fruit) as "seed sources" of ethylene, which can be used in storage units on a rotational basis with unripe fruit, some of the newly ripened fruit then replacing the original ripe ethylene "generator." This type of system would have limited applications, but there may be other systems which can be developed as well.

There has thus far been no widely-known method to produce and harness large amounts of natural ethylene. There has not been exhaustive research in methods to produce natural ethylene on a large scale. To date, there has been very little motivation in the market to invent this, with efforts rather being placed on expanding the exemptions for use of synthetic ethylene products in organic foods to other products in addition to bananas. Were such a means developed, it could satisfy many of the criteria which are failed by the commonly-used synthetically-generated sources of this material.

Ethylene does have natural sources, albeit generally not in the quantities desired. Although perhaps not as cut-and-dried as some other cases, it would seem that synthetic ethylene fails NOSB processing criterion #1 and #7.

2119(m)(7) and NOSB processing criterion #6 - Compatibility:
The criteria established by OFPA and the NOSB are to serve as guidelines, even in difficult cases like this. Synthetic ethylene fails OFPA criterion 2119(m)(3), and NOSB processing criteria #1, #2, and #7, and for these reasons, also fails OFPA criterion 2119(m)(7) and NOSB criterion #6. Naturally generated ethylene could easily be compatible with organic production systems, and development of an acceptable source or system of natural ethylene should be encouraged.

In and of itself, the role that ethylene plays in food production systems should not disqualify it from organic systems; it is the synthetic nature of the material which is too problematic too warrant approval. It is easy to imagine that strong arguments could made which would support use of synthetic ethylene for a number of agricultural commodities. In all of these cases, the main reasoning is about increasing yields, decreasing labor, and providing the market with commodities which otherwise might be too expensive or rare. If this rationale were extended to other materials and other crops, there might be possibilities to introduce fruits into certain marketplaces which have never been there before, because they either ripened too slowly or too quickly, were too delicate, too small, grew too slowly, or were not worth transporting for any number of other reasons. Exemptions to the OFPA and NOSB criteria should be strongly discouraged in all cases.

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SUMMARY AND RECOMMENDATION:

Although it is not an easy decision to make, synthetic forms of ethylene should be prohibited for post-harvest treatment of all organic products. Naturally-generated forms should be allowed.

COMMERCIAL/FINANCIAL INTEREST:

I unequivocally claim that I have no personal, commercial, or financial interest whatsoever in the this material or the decisions regarding it.

5 August, 1999

Additional Information for Inclusion in TAP Review Folder

Background Information for

Ethlyene, Processing

Index to Ethylene, Processing Background Information for TAP Review from OMRI, September 23,1999

A. Control of ethylene in the postharvest environment. 1985. Sherman. HortSci V 20. Pages 57-60.

B. Fruit ripening with calcium carbide. 1990. Sy and Wainwright. Trop. Sci. Pages 411-420.

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A:

Control of Ethylene in the Postharvest Environment

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The beneficial and harmful effects of ethylene (C_2H_4) on harvested horticultural products arc well documented The purpose or this presentation is to outline the technologies currently available in commercial horticulture for both the application of C_2H_4 to harvested horticultural crops. and the protection of these crops from the undesirable effects of C_2H_4. The ressarct base for this topic cuts across many areas of considerabie postharvest research. including temperature effects. atmospheric modification. and regulation of C_2H_4 synthesis. A complete review of any of these topics is beyond the scope of the presentation. and. in most instances. the reader is referred to recent research reports or comprehensive reviews of the specific subject

Postharvest application of ethylene

History and economic importance. The use of practices to hasten fruit ripening dates to antiquity (35). although it was not known until relatively recently that C_2H_4 was the causal agent. The earliest practices employed the emanations from ripe fruit or the smoke generated from burning combustible products. The use of kerosene stoves for "sweating" or "forced curing" of citrus was a well-established commercial practice by the late 1800s. It was believed that the high temperatures and high humidities in the "sweating" rooms resulted in degreening. Sievers and True (43) demonstrated that degreening was due to some unknown gaseous product from the incomplete combustion of kerosene. Denny (15) provided convincing evidence in 1924, that C_2H_4 was the effective constituent in "stove gas", and described a method for commercially degreening lemons using cylinders of compressed C_2H_4 gas.

Denny's work (15) marked the beginning of the knowledgeable use of C_2H_4 gas in commercial degreening and fruit ripening practices. Harvey (27) published the 1st comprehensive bulletin describing the commercial application of C_2H_4 for ripening bananas, pineapples, dates, Japanese persimmons. tomatoes, hard pears, apples, and muskmelons. Interestingly. one early commercial use for C_2H_4 was for blanching celery (26), a practice which has long since been discontinued.

Rosa (39) predicted that for tomatoes "the acceleration of the coloring process by low concentrations of C_2H_4 is sufficiently great to make it commercially valuable. . .". These words seem especially prophetic when considering the economic importance of C_2H_4 applications in postharvest horticulture today. Although it is not possible to assign an exact monetary value for C_2H_4 use. it is accurate to state that C_2H_4 applications play an important role in the orderly marketing of fresh citrus (excluding limes), tomatoes. bananas, mangoes and honeydew melons Recent reports (20 47 48) indicate that these products have a value of over 1 9 billion dollars in the United States alone

Sources of ethylene There are 3 potential sources of C_2H_4 for commercial use liquids, gases. and ripening fruit Liqutd sources? are C_2H_4-releasing chemicals such as (2-chloroethyl) phosphonic acid, commonly known as ethephon This chemical is registered and widely used for preharvest applications to concentrate maturity or otherwise facilitate harvest of several horticultural crops According to the label. the only presently registered use for postharvest application is for the degreening of lemons in Flonda (49) Therefore. further discussion will be limited to the other sources of C_2H_4 for postharvest use.

Ethylene gas can be generated *in situ* or purchased in compressed cylinders The gas is generated by catalytic conversion of a flammable liquid concentrate (Ethy-Gen) to C_2H_4 gas (11). This is one of the most popular commercial sources of C_2H_4 gas. The other widely used source of C_2H_4 gas is that purchased in compressed cylinders of either pure C_2H_4 or C_2H_4 diluted with an inert gas such as the product called "banana gas" (50, 3, 51).

Postharvest physiologists are aware that ripening fruit produce C_2H_4, but many commercial handlers either are unaware of or ignore this fact. If ripening room operators conscientiously monitored C_2H_4 levels in these rooms, then the use of ripening fruit might be a viable commercial alternative. However, the use of ripening fruit as a source of C_2H_4 generally is limited to home ripening recommendations.

Which source to use? The source of C_2H_4 used by handlers normally is determined by the facilities for treatment, legal considerations, cost, and safety. The use of C_2H_4 gas requires some type of enclosure The cost of building special ripening faciltties with precise temperature and humidity controls is well-justified when repeated ethylene applications are required.

Each handler also must constder the legality of C_2H_4 application (25) Ethylene gas used for plant regulation such as coloration or ripening of fruit and vegetables is regarded legally as a pesticide' for regulatory purposed (52). Therefore, it must be registered with the Environmental Protection Agency (EPA) and the appropriate state agencies Containers of C_2H_4 gas or C_2H_4-releasing liquids must bear EPA approved labeling, including EPA registration and establishment numbers, intended uses, ingredients statement, and appropriate precautionary labeling statements

Florida Agr. Expt. Sta. J. Series No. 5413. Mention of vendors or products does not constitute a guarantee or endorsement of the vendor or product by the Florida Agr. Expt. Sta., and it does not imply its approval to the exclutions about fruit degreening a ipening processes (36, 4). For example, these recommendatio pecify the type of electrical wiring and piping to be used in ripening rooms. Other state and local fire marshal rules or building codes may apply (46). Generally. these rules all pertain to safety, but this may change. Recently. a bill was introduced in the California state legislature which would have required that tomatoes treated with C_2H_4 be labeled "artificially ripened".

Safety must be a prime consideration in the technology of C_2H_4 application. Ethylene gas is explosive in air at concentrations from 3.1% to 32% (31.000 to 320,000 ppm) (8). To put these values in proper perspective. the minimum explosive concentration (3.1% C_2H_4 in air) exceeds the suggested C_2H_4 concentrations for tomato ripening (24) and citrus degreening (56) by 200 and 6200 times, respectively. One might conclude. therefore. that it would take extreme negligence for the fruit ripening process to be dangerous. However, explosive accidents have ocurred in the past. Safety during C_2H_4 application is dependent upon the operator, C_2H_4 source, and the method of application. The safety advantages afforded by the catalytic generator. and "banana gas" sources of C_2H_4 may be negated by the operator and the method of application. Each ripening room operator should have equipment to measure C_2H_4 concentrations. This is accomplished readily with an inexpensive gas detector kit and C_2H_4 detector tubes obtainable from several companies that sell specialty gases.

Methods of applying C_2H_4 gas. The methods of applying C_2H_4 gas can be placed into 3 categories: shot, trickle, and flow-through. Shot methods employ the rapid injection of C_2H_4 into the ripening room atmosphere. This can be accomplished by weighing the amount of C_2H_4 dispensed from a cylinder, using a premeasured amount (lecture bottle), or timing the delivery of C_2H_4 from a cylinder. The original system described by Denny (15) was a shot method. with the C_2H_4 delivered in a premeasured container. Dispensing C_2H_4 directly from large cylinders can be extremely dangerous and never should be done. Shot methods have the advantage of simplicity. but the disadvantage of requiring frequent room aerations to prevent carbon dioxide (CO_2) buildup which necessitates recharging the room with C_2H_4 following each aeration.

The "trickle" method can be defined as the slow. continuous dispensing of C_2H_4 into the ripening room atmosphere. Usually. compressed C_2H_4 cylinders are equipped with a regulator and tlow meters to control the tlow of C_2H_4. In practice. the catalytic generator also is a trickle method for introducing C_2H_4. Trickle methods are safer than the shot methods but rhey still may require periodic room aeration and recharging with C_2H_4.

The flow-through system is a modification of the trickle methods In principal. the flow-through system supplies a constant. ripening-effective blend of C_2H_4 and fresh outside air which passes over the product and out an sxhaust port in the room. The constant air exchange prevents CO_2 accumulation to inhibitory levels and eliminates the need for periodic aeration. The flow-through system has proven to be a safe and efficient method for introducing C_2H_4 for citrus degreening (56) and tomato ripening (42). Sherman and Gull (42) provided detailed instructions for installing a flow-through system.

Commodity requirements. One of the most important requirements is that the commodity be physiologically mature (capable of continuing normal development when detached from the plant). For some commodities like citrus, legal definitions of maturity exist (57). For others, such as tomatoes, there are no legal definitions of maturity, and it is difficult to determine whether a green fruit is mature or immature based on external appearance. Other important commodity requirements include the desired C_2H_4 concentration, sensitivity-lo CO_2, and the optimum temperature. humidity, and length of treatment. Local extension recommendations should be consulted for specific commodity information (24, 31, 56).

Protecting harvested products from ethylene

History and economic importance. Some of the earliest investigations of C_2H_4 were related to its detrimental effects on plants and have been thoroughly reviewed (1). Crocker and Knight (12) premature closing - buds) was caused by 0.5 to 1 ppm C_2H_4. Examples of the advers cts of C_2H_4 on harvested horticultural products continue to appear in literature. Risse and Hatton (38) recently reported the detrimental effects of C_2H_4 on harvested watermelons.

It is extremely difficult to assess the economic importance of protecting harvested horticultural products from C_2H_4 Abeles has pointed out the problems of estimating the economic losses caused by C_2H_4 as a component of air pollution (1, 2) Detrimental effects of C_2H_4 dunng the normal short-term marketing of fruit and vegetables are not well-defined and certainly are secondary to considerations regarding the maintenance of optimum temperature and humidity in the postharvest environment. However, costs to the individual shippers involved can easily run into tens of thousands of dollars when losses do occur from problems like russet spotting of lettuce or yellowing of cucumbers Economic evaluations of long-term storage responses to C_2H_4 (5, 17, 28) generally have not been reported because investigators have not wanted to speculate about extended season prices Detrimental effects of C_2H_4 may be most Important for ornamental crops where estimates once placed postharvest losses at about 20% due to mishandling. However, mishandling included poor temperature management. and improper humidity, as well as exposure to C_2H_4 (45). In summary, losses caused by C_2H_4 are known to occur. but they are usually quantitatively undefined. A conservative estimate for the United States would be in the tens of millions of dollars annually.

Strategies for protection. The strategies for protecting harvested horticultural products from the detrimental effects of C_2H_4 can be placed into 3 major categories: avoidance, removal. and inhibition. Although these techniques are grouped for convenience it should be remembered that they may overlap and are not mutually exclusive.

Avoidance. Circumvention of undesirable product exposure to C_2H_4 begins with careful harvesting, grading, and packing which includes selecting the desired maturity and avoiding mechanical injury. The 2nd and probably most important step in avoidance is proper temperature management, which should include rapid cooling to the product's lowest safe temperature. This suppresses ethylene production and reduces sensitivity to ethylene (9). Chilling temperatures should be avoided when handling chilling-sensitive commodities. however. because increased C_2H_4 production frequently follows chilling injun (55). Ethylene-sensitive commodities should not be transported. stored. or displayed with C_2H_4-generating commodities. Guidelines suggesting compatible product mixes have been published (33). Given the food distribution system in the United States. it is not possible to avoid undesirable exposure to product-generated C_2H_4 completely. bur exposure should be kept to a minimum. Placing ripe tomatoes between iceberg lettuce and cucumbers in retail produce displays may be colorful. but it completely ignores the biology of these products and constitutes unnecessary exposure of the produce to undesirable temperatures and C_2H_4.

Other management steps for avoidance include minimizing the use of internal combustion engines during product handling in enclosed spaces. and following strict sanitation practices to insure that overripe and decaying products are promptly removed.

Removal. Undesirable levels of C_2H_4 in produce storage areas can be removed by simple ventilation with fresh air if the air is not polluted with high C_2H_4 levels. Usually, one air exchange per hour is required to maintain a low C_2H_4 level. Ripening room facilities located in produce distribution centers always should be vented to the outside to ensure that C_2H_4 is not accidently introduced into the storage environment of the distribution center.

C_2H_4 can be scrubbed from the atmosphere by trapping and/or conversion to other products when ventilation cannot be used for removal. A large number of reagents and techniques have been tested over the years (1, 7, 15), but only potassium permanganate is presently in common commercial use. To be effective, $KMnO_4$ must be adsorbed on a suitable carrier with a large surface area. Celite, vermiculite, silica gel, alumina pellets (1), perlite (40), and expanded glass (32) have all been successfully used as carriers. A number of commercial potassium permanganate scrubbers are available in sachets. filters. blankets. and other specialized trapping devices (37. 18. 19) Ethylene is trapped most effectively when air is drawn through the scrubber. Potassium permanganate scrubbers are advantageous because they change from purple to brown as the MnO_4^- is reduced to MnO_2. The major disadvantage of permanganate scrubbers seems to be their expense (7. 40).

Heated catalyst ethylene scrubbers were successfully used during the 1983–1984 storage season in commercial controlled atmosphere apple storages to maintain ethylene concentrations below 2 ppm in New York State, and below 1 ppm in England (Blanpied, personal communication). Another technology for C_2H_4 removal. which seems promising but has yet to be commercially developed. is the use of UV light (41). Also, there is a potential for biological removal of C_2H_4. Abeles (2) discussed the role of soil bacteria as a sink for atmospheric C_2H_4. Perhaps the C_2H_4-consuming *Mycobacteria* that have been isolated from soils (14) could be genetically engineered to perform satisfactorily in horticultural applications.

Inhibition. Controlled atmospheres (CA) have been used widely for many years for the long-term storage of apples (13) and. to a limited extent. for other commodities. such as pears (44) and cabbage (22). CA systems require special refrigerated. gas-tight structures (16. 29) which allow for precise temperature controi and maintenance of the storage atmosphere (primarily reduced O_2 and elevated CO_2). CA storage has multiple effects on the physiology of the commodity (30. 44). but the retardation of ripening is at leas: partially attributable to the low O_2 atmosphers. slowing C_2H_4 synthesis and action. and the elevated CO_2 inhibiting C_2H_4 action. Very low levels of O_2 (17) and C_2H_4 scrubbing (5. 7) ma! enhance the storability of products in C.4 The use of C.4essentially has beer limited to long-term stationan storages. but a: least one company now out of business. incorporated CA capability into modem transport containers (21). Modified atmospheres (a less precise type of CA) have been used for man! years or, several commodities during transport.

Low pressure storage (LPS) seems to offer potential for prolonging the useful life of horticultural commodittes (10. 34). LPS effectively reduces O_2 levels and. in this respsct. ts similar to CA storage. Storage at subatmospheric pressures also increases the diffusivitp of volatile gases. however. including C_2H_4. from the internal atmosphere of the commodity (10). Commercial applications of LPS systems have not been widespread to date. Grumman Allied Industries developed the Dormavac System for hypobaric transporation of perishables (23), but this met with only ltmited commerical success and was discontinued.

Chemical treatments could be used to protect horticultural products by inhibiting C_2H_4 synthesis and/or action. Yang and Hoffman (58) reviewed many of the known inhibitors of C_2H_4 biosynthesis. Some inhibitors appear to have commercial potential for regulating fruit ripening (6. 54), but their use on food products must be preceded by FDA and EPA approvals. Many ornamental horticultural crops can be protected from the detrimental effects of C_2H_4 by treatment with the anionic complex silver thiosulphate (STS). Veen (53) recently reviewed some of the uses for STS in commercial horticulture.

Methods outlined here for removal of C_2H_4 and inhibition of C_2H_4 synthesis and/or action should not be regarded as panaceas for solving C_2H_4-related problems in commercial horticulture. The best defenses for protecting harvested horticultural products from the detrimental effects of C_2H_4 are the steps outlined under avoidance. When the shelf life of products can be extended further by the methods of C_2H_4 removal or inhibition of its action, then their use may be advantageous. The long term storage of apples in controlled atmospheres with C_2H_4 scrubbing is a good example of postharvest practices which employ avoidance, removal, and inhibition methods for protection from C_2H_4 (7, 17).

Summary

Control of C_2H_4 in the postharvest environment is of great importance in commercial horticulture. Application of C_2H_4 facilitates the orderly marketing of citrus, tomatoes. bananas, mangoes, and honeydew melons. Safe and efficient application methods have been developed and are readily available for commercial use. Detrimental effects of C_2H_4 on harvested horticultural products largely can be avoided by careful management of the postharvest environment. but this management is not always practical in commercial situations. Technologies exist for removing C_2H_4 or inhibiting C_2H_4 action, but there is still considerable room for improvement during the storage and handling of most commodities.

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REVIEW

Fruit ripening with calcium carbide

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Abstract *The use of calcium carbide as a method of ripening fruits is reviewed. The technique is a traditional method of ripening and degreening fruit which is still widely practised in developing countries, where ethylene is either unavailable or too expensive. Calcium carbide ripens fruit by the release of acetylene, which has been shown to act in a similar way to ethylene on unripe fruit, though higher concentrations of acetylene may have to be used. The techniques of using calcium carbide in various countries including India, the Philippines, Senegal and Brazil are described. Results of work using calcium carbide to generate acetylene or bottled acetylene gas to ripen mango, banana, citrus, melons, plums and peaches are detailed. Practical considerations are discussed, including the risks to health through the toxicity of the calcium carbide and its by products; also the explosive nature of acetylene as compared to ethylene, as well as methods of producing acetylene from calcium carbide.*

Keywords: fruit ripening, calcium carbide, acetylene, ethylene.

Introduction

Climacteric fruits are often harvested in a mature but unripe condition and then subsequently allowed to ripen. The climacteric fruits will ripen naturally, but this may happen slowly, at a time which is not predictable, and/or unevenly. To overcome these problems, fruit such as banana, mango and avocado can be ripened artificially by exposing the fruit to either ethylene or certain similar gases such as acetylene or propylene for a short period which initiates the ripening process.

The fruit then ripen rapidly, synchronously and at a predictable time. Almost all internationally traded bananas are now initiated to ripen by exposure to ethylene (Proctor and Caygill 1985); yet because of the expense and/or unavailability of ethylene in developing countries, the low-cost technique of producing acetylene from calcium carbide to ripen fruit is still widely practised.,

Though the technique of generating acetylene from calcium carbide has been used for many years, it is poorly documented. The purpose of this review therefore is to collate known information about the use of calcium carbide for ripening fruit and to identify any existing problems in the technique.

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Calcium carbide is widely used in the chemical industry for the manufacture of neoprene, chloroethylenes and vinylacetate (Geesner 1977). When placed in water or exposed to moist air the formation of acetylene and calcium hydroxide with the evolution of heat occurs (Geesner 1977). Acetylene liberated from calcium carbide (CaC_2) acts on some fruits causing them to ripen in a similar manner to ethylene, and numerous studies have been undertaken to compare the ripening properties of ethylene and acetylene, as detailed below.

Comparison of ethylene and acetylene as initiators of fruit ripening

With bananas, ethylene initiates ripening at 0.01, 0·1 and 1 ml l^{-1}, but at 0.01 ml l^{-1}, acetylene failed to initiate ripening; at 0·1 ml l^{-1} the peel remained green and the soluble solids content was lower although a climacteric rise in respiration was induced; and at 1 ml l^{-1}, ripening was only slightly slower than when ethylene was used (Thompson and Seymore 1982).

These findings support those of Pantastico and Mendoza (1970) who found that bananas treated with acetylene take longer to ripen than those exposed to an equivalent concentration of ethylene. Banana fruit were ripened satisfactorily with acetylene but the fruit had a lower total of soluble solids than those ripened with ethylene (Kaltenbach, 1939).

With mango, results were similar to those obtained with banana where ethylene at concentrations of 0·01 ml l^{-1} and above and acetylene at 1·0 ml l^{-1}, initiated ripening. At 0.01 ml l^{-1} acetylene caused limited softening but had no effect on the other ripening changes; whilst at 0·1 ml l^{-1}, acetylene showed only delayed ripening compared with ethylene (Medlicott *et* al. 1987).

Ethylene is not always available in many developing countries or when it is, the cost is too high. For example in the People's Democratic Republic of Yemen, ethylene is generated from a proprietary imported product used to ripen bananas. The cost of ethylene produced in this manner was 50 times greater at £0.5 litre^{-1} than acetylene generated from locally available calcium carbide (Smith and Thompson 1987).

Use of calcium carbide in fruit ripening

The use of calcium carbide to generate acetylene to colour and ripen fruits has been known for many years and was the subject of a review 50 years ago (Kaltenbach 1939). Numerous reports have been published on the use of calcium carbide in commercial practice and/or in experimental investigations in an extensive number of countries and on a wide range of fruit species (Table 1).

Stevenson (1954) working in Australia states that 1 lb of solid calcium carbide with water yields 5 cubic feet of acetylene, or one-fifth of a pound of carbide will yield sufficient acetylene to give a concentration of 1 part of acetylene to 1000 parts of air in a chamber of 1000 cubic feet.

Table I. The fruits and countries where acetylene, generated from calcium carbide, has been used to ripen fruits

Fruit species	Country	Source
Banana	Australia	Stevenson 1954
	Egypt	Salem *et al.* 1976
	India	Singh *et al.* 1975
	India	Khan *et al.* 1977
	Philippines	Pantastico and Mendoza 1970
	Philippines	Bondad 1971
	Philippines	Espanto 1984
	South Africa	Malan 1953
	Sudan	Seymour 1984
	Taiwan	Chiang 1955
	USA	Hartshorn 1931
	Yemen	Smith and Thompson 1987
Mango	Brazil	Sampaio 1981
	Costa Rica	Valerde *et al.* 1986
	India	Subramanham *et al.* 1972
	India	Nagaraj *et al.* 1984
	India	Mann 1974
	Malaysia	Berwick 1940
	Philippines	Tirtosoekotjo 1984
	Senegal	Sy *et al.* 1989
	South Africa	Marloth 1947
Citrus	Australia	Stevenson 1954
	Australia	Prest 1932
	Philippines	Bondad 1971
	Philippines	Acena and Macatangay 1957
	South Africa	Marloth 1935
Tomatoes	Australia	Stevenson 1954
	Morocco	Kaltenbach 1939
	Philippines	Bondad and Pantasico 1971
	USA	Rosa 1925
Pawpaws	Australia	Stevenson 1954
Plums	South Africa	Putterill 1938
Melons	South Africa	Rattray 1940
Peaches	South Africa	Davies and Boyes 1940

The recommended way to introduce acetylene into a chamber is to put a large vessel of water in the chamber and place carbide into the water and close the chamber door. The chamber should be ventilated and then immediately re-charged with acetylene three times a day.

In India, one of the world's largest mango producing countries, the tradition of ripening early season fruits with calcium carbide is well established. In a trial undertaken by Mann (1974) to study different doses of calcium carbide, mature hard green fruits of

Dashehari mango were used. The fruits were packed together with calcium carbide and covered tightly with newspaper so as to prevent the leakage of acetylene. Calcium carbide was moistened by a drop of water before packing so as to release the gas. The fruits ripened within 8 days and those fruits ripened with 2 g of calcium carbide per 4–5 kg of fruits were assessed by organoleptic evaluation, to develop the most desirable taste aand flavour (Mann 1974). With the cultivar Keitt in Costa Rica (Valerde *et al.* 1986), a rate of 3–5 g of calcium carbide per kg of fruit was recommended to obtain ripening in 8 days.

Nagaraj *et al.* (1984), in an observational trial in India, used ventilated wooded boxes where they placed 2 g of calcium carbide per kg of fruit, the standard commercial rate of application. The fruits were covered with straw and craft paper. After 96 hours, the calcium carbide packets were removed from the boxes and the uncovered fruits were kept in a separate ventilated room.

This treatment gives uniform and attractive yellow skin colour, and causes rapid softening of fruits. The authors report that the technique is commercially exploited for ripening and marketing mangoes, and the calcium carbide treatment significantly reduced the number of days required to attain edible ripe fruit compared with untreated fruit.

In the Philippines, ethylene is not readily available and is difficult to apply without placing the banana fruits in an enclosed room or container (Bondad 1971). The most popular method of ripening bananas consists of treating a stack of fruit with calcium carbide in a 200 litre drum, cut to a convenient height and lined with three to four plastic sacks laid flat against the bottom and sides (Espanto 1984). The sacks are also used to cover the stack. Calcium carbide wrapped in newspaper is placed in the drum under seven to eight spirally-arranged hands.

Another method consists of simply enclosing the same number of hands with carbide in a sack. In either case, treatment is usually carried out over a 24-hour period. Farmers use about 250 g of carbide per sack or drum, though no indication of the quantity of fruit this ripens is given (Espanto 1984).

A more controlled ripening of bananas, used in Egypt, involved the use of air tight rooms with an initial temperature of 25°C, a relative humidity of 90–95% and an acetylene concentration of 1 ml l^{-1} (Salem *et al.* 1976).

In Senegal, calcium carbide is often used by wholesalers and farmers to ripen fruit, mainly mango and banana. Fruit are harvested mature green and ripened with calcium carbide. Two similar methods are used. The first involves placing calcium carbide wrapped in cloth or newspaper in the bottom of a basket made of palm leaves, locally known as a 'damba' (Figure 1), and then covering the basket with a piece of sack or strong craft paper in order to increase the internal basket temperature and relative humidity during the ripening initiation process.

The baskets are placed in a closed room for 2 or 3 days. Fruits are then selected from these baskets when they have developed a yellow skin colour and offered for sale. The second method consists of forming a large stack of fruits (1 to 2 tons) in a corner of a room (Figure 2) and putting calcium carbide in several places in the stack and covering the fruits with craft paper and securing with rope. As in the first method, fruit are left for 2 or 3 days then selected for sale.

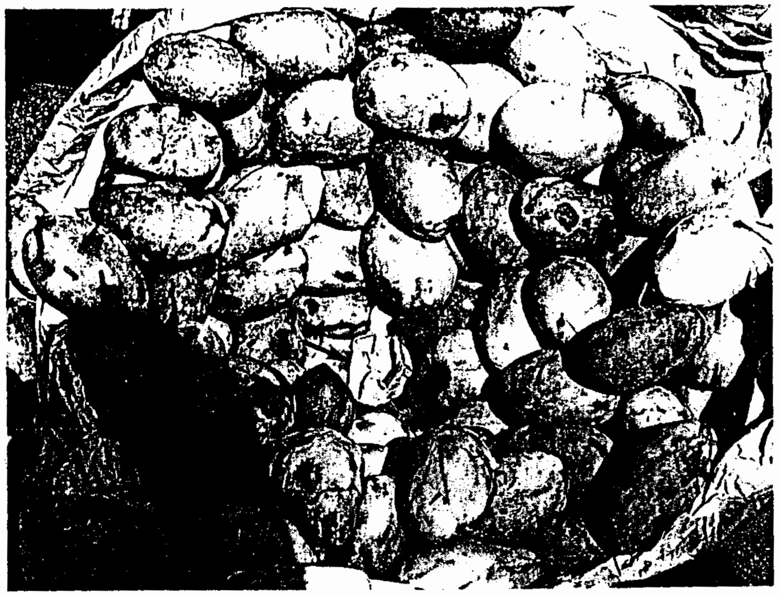

Figure I. The ripening of mangoes in a paper lined basket by the inclusion of calcium carbide which is wrapped in paper (arrowed)

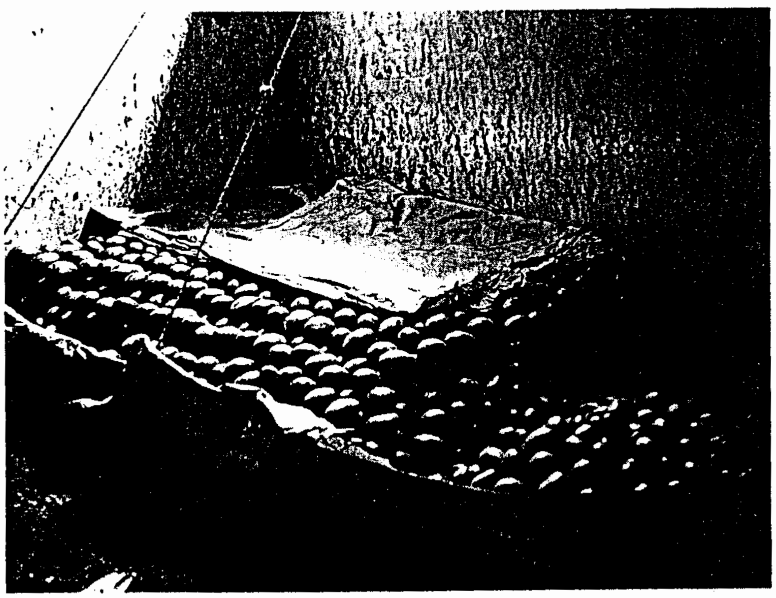

Figure 2. Ripening mangoes by stacking in a corner of a room and exposing them to acetylene produced from calcium carbide

The fruits ripened in these ways are not always of good eating quality; although they have good skin colour, the ripening process is not complete and the fruit have a high acidity and lower sugar content.

In Brazil, some exporters are using calcium carbide treatments prior to export or sale in local markets, with the aim of improving peel colour developnient (Medlicott 1986). This treatment generally involves stacking boxes of fruit under tarpaulin covers with calcium carbide placed in containers of water.

No standard amount ofcalcium carbide was used, although treatment was always for 12 hours after which the tarpaulin was removed. Treatment was only used at the beginning of the season, and the technique increased the rate of ripening and resulted in fruit of excellent peel colour, but of poorly developed flavour.

In Malaysia, mangoes are picked slightly unripe and artificially coloured by means of calcium carbide. This is generally done in soap boxes or baskets lined with banana leaves on which the calcium carbide is sprinkled; the receptacle is filled to the top and the fruit covered with more leaves on which more calcium carbide is scattered. After two or three days, the colour of the fruit becomes uniform, though the taste is insipid, and the consignment is ready for sale (Berwick 1940).

A component of ripening that is readily detectable is the loss ofchlorophyll in skin. In citrus, the degreening of the skin is of primary importance and acetylene in a few instances has been shown to be effective. The temperature and relative humidity have to be regulated and 30–35°C and 85–90% are recommended, respectively (Acena and Macataugay 1957). It was found that the period of exposure to acetylene for the most efficient degreening was between 36 and 120 hours, depending on cultivar.

Marloth (1935) found perfectly satisfactory degreening of oranges with one part of acetylene in 1000 parts air for an exposure period of 60 hours. However, for grapefruit and lemons, the fruit was liable to burn, so a lower concentration was suggested. The efficiency of calcium carbide was dependent on the amount of impurities present and this was variable, depending on the source of carbide.

The conclusion made by Marloth (1935) was not to recommend the use of carbide to colour citrus; in South Africa, the future use of calcium carbide to colour citrus would be restricted to the few instances where ethylene was not available. There are few other reports of acetylene being used to degreen citrus and its use does not seem to have been widespread.

Problems associated with using calcium carbide to ripen fruit

Calcium carbide is produced by combiningcalcium oxide and carbon (both readily available materials) in a furnace; it releases acetylene when combined with water (Reid 1985). The danger of explosion with acetylene is reported by many investigators. Acetylene has a garlic odour and is a flammableand explosive gas (Geesner 1977). It is flammable in air at lower concentrations than ethylene and its minimum auto-ignition temperature in air is 305°C.

According to Thompson and Seymour (1982), the suitability of acetylene as a ripening agent depends on its efficiency at concentrations well below this minimum explosive level. Both ethylene and acetylene gases can be explosive in air at high concentration, acetylene being slightly more dangerous to use than ethylene. Therefore, the lower the exposure concentration required, the safer it will be.

Impurities are present in commercial calcium carbide samples. Some of these could be dangerous to human health. For instance phosphorous hydride (PH,) and arsenic hydride (AsH_3) are contained in calcium carbide (Delpierre 1974). These components might be found in the calcium hydroxide formed during the liberation of acetylene.

In most major traditional methods, calcium carbide is in direct contact with the fruit during the ripening process, and the fruit is often not washed before eating. Therefore, the danger of contaminated fruit increases, although tropical fruit like bananas have a thick peel which is discarded before eating the pulp. However arsenic hydride (AsH_3) is soluble in organic solvents and in oil and fat (Delpierre 1974). Dissolving of AsH_3 in the wax of the fruit peel may therefore occur.

Data from the literature indicates that calcium carbide contains numerous impurities and shows that the same amount of acetylene is not always liberated from the same weight of calcium carbide. For instance, in Australia, 1 g of calcium carbide released 312 ml of acetylene (Stevenson 1954); whilst Seymour (1984) found in The Sudan that 1 g of calcium carbide gave 240 ml of acetylene; and in Senegal, Medlicott (1986a) calculated that the volumes liberated per gram of calcium carbide, whilst showing some variation, averaged approximately 150 ml.

As well as differences in the composition of the carbide, another possible explanation may be the difference in size of calcium carbide pieces used, as the smaller lumps have a higher surface area and will release acetylene more rapidly. For experimental control of this variable, Bondad and Pantastico (1971) passed the calcium carbide through a sized mesh.

Obtaining a known and predictable quantity of acetylene from calcium carbide does, however, appear unlikely. Also the imprecise nature of defining the quantity or the weight of calcium carbide put with the fruits and the duration that the fruits are exposed to acetylene, seem to be aspects where there are major differences in techniques and are possible causes for unreliable results.

An improvement in the traditional methods of using calcium carbide which would reduce the health risk would be non-contact use of calcium carbide with the fruits. For this, the simplest method would be to put calcium carbide in a container with enough water in an isolated part of the ripening room.

Another adaption on this principle reported by Prest (1932) is to put the carbide in a suitable vessel and arrange water to drip onto the carbide from a second vessel. Where available, specific acetylene generators such as those used for mine and bicycle lamps (Malan 1953) could be used. These work on the same principle of water dripping onto carbide, but are in an enclosed container and the acetylene escapes through a small hole which, in the case of the lamps, is ignited.

Another technique could be to put the calcium carbide in a closed container where moist air is blown over the calcium carbide and then acetylene is released in a constant flow into the ripening room. Comparative experiments carried out at the Institute de Technologie Alimentaire, Senegal, on banana ripening by direct contact (calcium carbide wrapped in newspaper and placed at the bottom of the fruit baskets) or by placing the carbide in a separate container with water, showed an earlier colour break in the direct contact; but those fruits nearest the calcium carbide were more coloured and ripened earlier than those on the top of the basket.

However, after 7 days, fruits from direct and indirect exposure to calcium carbide had the same full yellow peel colour (Sy *et al.* 1985). In another investigation on ripening 'carabao' mango with calcium carbide, Tirtosoekotjo (1984) concluded that the quality of carbide treated fruits was considerably affected by such factors as fruit maturity, the carbide to fruit ratio and the manner in which the fruit were treated.

These studies were orientated towards a determination of the best set of conditions for using calcium carbide to ripen mango fruit.

Earlier, Harvey (1928) stated that acetylene generated from calcium carbide was convenient but that the gas had a disagreeable odour and produced a noticeable flavour in treated fruits. Acetylene is also toxic to the fruit and may be disagreeable to those who handle it. Even so, the problem of taint from using acetylene is not widely reported by other users.

Future areas of investigation

The use of calcium carbide to produce acetylene to ripen fruit does have disadvantages compared with ethylene, as described above. However, in developing countries, the unavailability of high cost of ethylene often precludes its use and, therefore, the use of calcium carbide to generate acetylene to ripen fruit is seen as a practice that is valuable and set to continue in the future. However, there are two topics that require further investigation for the improved use of calcium carbide. Firstly, there is a need to have a better understanding of the ripening response of tropical fruit to acetylene.

This requires investigating a range of factors, including the concentration and exposure time to acetylene, the optimum stage of maturity of the treated fruit, the variation of response between different cultivars of the same species, the organoleptic quality of treated fruit and the treatment of fruit at the high ambient temperatures often encountered in situations where calcium carbide is regularly used.

Secondly, the need to have a safe, controlled delivery system for acetylene from calcium carbide. This would involve the development of a low cost acetylene generator which should be applicable to the wide range of users, would prevent direct contact between the fruit and calcium carbide and yet produce a consistent and known quantity of acetylene to initiate ripening.

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C:

TI: History and development of fruit differentiation, growth and ripening control in pineapple.
AU: Williams,-DDF
AD: Maui Pineapple Co., Haliimaile, HI 96768, USA. PY: 1987
SO: Proceedings of the Plant Growth Regulator Society of America. 1987, 413-422; 14 ref. Lincoln, Nebraska, USA; Plant Growth Regulator Society of America.
AB: A review of the history of Cayenne pineapple growing in Hawaii. In addition to discussing the effects of climate and elevation, it mentions the use of plant growth regulators for enlarging or ripening the fruit, and in propagation and the control of flowering.

TI: Physico-chemical changes during flower bud differentiation in pineapple (Ananas comosus, L. Merr.).
AU: Ahmed,-F; Bora,-PC PY: 1987
AD: Assam Agric. Univ., Jorhat-13, India.
SO: Indian-Journal-of-Plant-Physiology. 1987, 30: 2, 189-193; 13 ref.
AB: Pineapple plants of the cv. Kew, raised from slips planted in July and November of the previous year, were treated in August and January, respectively, with several chemicals. Applications of Ethrel [ethephon] at 100 p.p.m., of Ethrel at 25 p.p.m. mixed with 0.04% $CaCO_3$, and of urea at 2% were very effective in enhancing the synthesis of different metabolites (sugars, ascorbic acid, proteins and nucleic acids). The duration of flower bud differentiation was shorter in summer than in winter.

TI: Regulation of flowering in pineapple the year round.
AU: Bose,-TK; Aich,-K; Mitra,-SK; Sen,-SK PY: 1983
AD: B.C.K.V., Kalyani 741 235, India.
SO: Indian-Agriculturist. 1983, 27: 4, 331-336; 11 ref.
AB: The effects of monthly planting between 15 July and 15 November and treatment with calcium carbide (20 g/litre), Ethrel (ethephon] (0.25 ml/litre) or NAA (20 mg/litre) 335 days after planting, on flowering, yield and fruit quality in the cv. Kew were studied. Treatment with ethephon or calcium carbide between April and July induced flowering 22 to 30 days after treatment, but the period from treatment to appearance of inflorescence gradually increased from August to November. Sequential planting and application of chemicals regulated the time of harvest and a year-round cropping was achieved. Among the chemicals, calcium carbide generally induced flowering better than ethephoff. Fruits from the NAA treatment were heavier whereas calcium carbide and ethephon induced better quality.

TI: Effect of different chemicals on flowering and fruit development in pineapple.
AU: Santha,-KK; Aravindakshan,-M; Namboodiri,-KM PY: 1983
AD: Kerala Agricultural University, Trichur, India.
SO: South-Indian-Horticulture. 1983, 31: 4/5, 181-186; 12 ref.
AB: In trials over 2 seasons, 14 different treatments were applied to plants (19-month-old initially) of the cv. Kew. The best flower induction in both seasons was obtained with Ethrel [ethephon] at 250 p.p.m., Ethrel at 50 p.p.m. + urea at 2% + calcium carbide at 0.04% or Planofix [NAA] at 200 p.p.m. The fruits were largest after treatment with Planofix at 20 p.p.m. None of the treatments improved fruit quality.

TI: Studies on response of growth regulators to induce flowering in pineapple [Ananas comosus (L). Merr.].
AU: Prasad,-A; Roy,-SK PY: 1987
AD: Dep. Hort., Chandra Sheklar Azad Univ. Agric. & Technol., Kanpur 208 002, India.
SO: Symposium on Himalayan horticulture in the context of defence supplies. 1987, 53. Tezpur, India.
AB: Plants of the early flowering cv. Kew were treated with CCC [chlormequat]. Ethrel [ethephon], NAA, GA3 or PCPA [4-CPA]. The best induction of early flowering was obtained with Ethrel. The best ages for treatment were 12 months for the plant crop and 3 months for the ratoons.

TI: Changes in chemical composition of the Kew cultivar of pineapple fruit during development.
AU: Kermasha,-S; Barthakur,-NN; Inteaz-Alli; Mohan,-NK; Alli,-I PY: 1987
AD: Dep. Food and Agricultural, McGill Univ., Ste. Anne de Bellevue, Que, Canada.
SO: Journal-of-the-Science-of-Food-and-Agriculture 1987, 39: 4, 317-324; 17 ref.
AB: The chemical composition of the Kew cultivar of Indian pineapple (Ananas cosmosus) was studied after 65 days (premature), 100 days (early mature), and 150 days (late-mature) from flowering. The fruit was chemically analysed for sugars, amino acids and minerals. Total soluble carbohydrates increased from 6.02 to 9.24%, and protein content decreased from 0.69 to 0.21%, during the fruit development process. The vitamin C content decreased from 20.4 to 11.1 mg/100 g edible fruit during development. Fructose, glucose and sucrose

together were about 5.0% (pre-mature), 2.4% (early mature) and 1.2% (late-mature) of fresh weight. The contents of iron, boron and zinc increased during development. The major minerals in the mature fruit were potassium 31.33, phosphorus 3.13, calcium 3.92, sodium 2.63, iron 3.22, Zn 0.55 and copper 0.06 mg/100 g fresh sample. The content of most amino acids decreased during development; only aspartic acid showed a considerable increase in the late-mature fruit.

TI: Development of a heat unit model of pineapple ('Smooth Cayenne') fruit growth from field data.
AU: Fleisch,-H; Bartholomew,-DP PY: 1987
AD: Department of Agronomy & Soil Science, Hawaii University, Honolulu, HI 96822, USA.
SO: Fruits. 1987, 42: 12, 709-715, 763-766; 14 ref.
AB: Climatic and flower-development data from 13 plantations of pineapple cv. Smooth Cayenne were collected over a period of more than 2 years. For each of the 5 phenophases from floral induction to fruit maturity a minimum temperature based on field data was determined. A heat-unit model based on air temperature adequately descnied development in all cases, indicating that flower and fruit development depended mainly on temperature.

TI: The influence of planting material, spacing and Ethrel on pineapple development, production and quality.
AU: Mwaule,-YW PY: 1985
AD: Serere Research Station, P.O. Soroti, Uganda.
SO: Acta-Horticulturae. 1985, No. 153, 63-68; 22 ref.
AB: The effects of 3 types of planting material (crowns, slips or suckers), 3 single row spacings (wide (90 X 90), medium (90 X 60) or narrow (60 X 60 cm)) and 2 hormone levels (with or without Ethrel [ethephon] at 50 ml per plant) were investigated in field trials using the cv. Smooth Cayenne. Forcing with ethephon significantly reduced mean fruit weight, slip and sucker production, planting-to-harvest period and harvest duration but increased fruit acid and soluble solids contents. Spacing had little influence on the parameters measured. There were significant interactions between ethephon and planting material on the planting-to-harvest period and harvest duration (which were shortened); and also between ethephon and spacing on soluble solids (which were highest at medium spacing).

TI: Influence of time of planting, slip size and plant age for the floral induction of Smooth Cayenne pineapple in the Reconcavo Baiano. I. Vegetative growth, offset production and natural flowering.
OT: Influencia da epoca de plantio, tamanho da muda e idade da planta para a inducao floral do abacaxi 'Smooth Cayenne' no Reconcavo Baiano. I. Crescimento vegetativo, producao de mudas e florescimento natural.
AU: Reinhardt,-DHRC; Costa,-JTA; Cunha,-GAP-da PY: 1986
AD: CNPMF, Bahia, Brazil. LA: Portuguese
SO: Fruits. 1986, 41: 1, 31-41; 30 ref., 2 fig.
AB: A trial was carried out from 1980 to 1983 with two slip sizes (35-44 cm and 25-34 cm), 4 planting dates (29 January, 18 April, 22 July and 5 November) and floral induction (with calcium carbide) at 7, 9, 11 or 13 months. Flowering of control plants occurred when plants reached a certain size, at different times of year, and was apparently enhanced by short days, low night temperatures, poor insolation and water stress. Vegetative growth depended on moisture availability. Offset production was proportional to plant size at the time of floral induction. Slip production was also enhanced by low temperature and poor insolation.

TI: Influence of ethephon and urea on the flowering of pineapple plants (Ananas comosus (L.) Merrill 'Cayenne').
OT: Influencia do ethephon e ureia no florescimento de plantas de abacaxi (Ananas comosus (L.) Merrill 'Cayenne').
AU: Fahl,-JI; Carelli,-MLC; Franco,-JF LA: Portuguese
AD: Secao de Fisiologia, Instituto Agronomico, 13.100 Campinas, SP, Brazil.
SO: Abstracts of the XIV Brazilian congress on herbicides and herbaceous weeds (SBHED) and the VI congress of the Latin American Weed Association (ALAM), Campinas, 1982. unda, 161. Campinas, Sao Paulo, Brazil.
AB: Pineapple plants were sprayed 13 months after planting with 475 and 950 p.p.m. ethephon ± 2% urea. The addition of urea significantly increased early flowering. With the high rate of urea + ethephon, 100% flowering was achieved 74 days after treatment while with the low rate of ethephon alone it occurred 116 days after application.

TI: Don't force pineapple plants too early.

AU: Dalldorf,-DB PY: 1985
AD: Agricultural Research Station, East London, South Africa.
SO: Information-Bulletin,-Citrus-and-Subtropical-Fruit-Research-Institute. 1985, No. 151, 7.
AB: Data are presented from trials with Smooth Cayenne pineapples on the effects on yields of applying NAA or ethephon to plants or suckers of differentsizes. With the plant crop, optimal yields of 80 t/ha were obtained when a flower inductant was applied when the D-leaf length was 63.9 cm, and D-leaf and plant weights were 71.6 and 1700 g, respectively. With suckers, forcing when the D-leaf length was 63.9 cm, D-leaf and plant weights were 40.3 and 1020 g, respectively, and average fruit size was 1298 g, resulted in yields of 108 t/ha from an average of 2 suckers/plant.

TI: Effect of artificial induction of flowering in pineapple.
AU: Balakrishnan,-S; Nayar,-NK; Mathew,-V PY: 1980
AD: College of Horticulture, Trichur, Kerala, India.
SO: Agricultural-Research-Journal-of-Kerala. 1980, 18: 2, 158-161; 5 ref.
AB: Plants raised from suckers (PRS) were treated with 100 p.p.m. Ethrel [ethephon] + 2% urea + 0.04% $CaCO_3$ at 14, 15, 16, 17 or 18 months after planting, and plants raised from crowns (PRC) were similarly treated at 16, 17, 18, 19 or 20 months after planting. In PRS, treatment at 18 months gave the highest percentage (96.07) of flowering (78.57% in the control) and in PRC, treatment at 17 months gave the best results, namely, 91.78% flowering, with 28.57% in the control. Data are tabulated on leaf and fruit characteristics.

TI: Pineapple flower inductants. Too much?
AU: Dalldorf,-DB PY: 1983
AD: Agricultural Research Station, East London, South Africa.
SO: Information-Bulletin,-Citrus-and-Subtropical-Fruit-Research-Institute. 1983, No. 135, 4.
AB: In trials with a plant crop of pineapples (cv. Smooth Cayenne), the effects were compared of NAA at 5, 10 or 20 p.p.m. and ECP (ethephon) at 250, 500 or 1000 ml/ha, each applied once or followed by a second application of NAA at 10 p.p.m. 10 days later in November, January and March. Preliminary results indicated no significant differences between treatments in their effects on average fruit weight, yields and the percentage fruit harvested. There appeared to be no advantage in using more than 500 ml ECP/ha.

TI: Influence of pH and urea on the action of 2-chloroethylphosphonic acid in inducing flowering in pineapple.
OT: Influencia do pH e da ureia na acao do acido 2-cloroetilfosfonico na inducao floral do abacaxi.
AU: Velez,-AML-de; Cunha,-GAP-da PY: 1983
AD: Universidade Federal da Bahia, Cruz das Almas, BA, Brazil. LA: Portuguese
SO: Pesquisa-Agropecuaria-Brasileira. 1983, 18: 11, 1199-1205; 18 ref.
AB: In trials with the cv. Smooth Cayenne, the addition of urea (2%) and/or $Ca(OH)_2$ (to raise the solution pH to 10) to ethephon at 50-100 p.p.m. did not appreciably improve flower induction compared with ethephon alone. It is suggested that, in view of the cost, the 50 p.p.m. ethephon rate (which gave 80.6% flower induction) is adequate.

✓TI: The control of flowering and planning production in pineapple plantations. Its results.
OT: La maitrise de la floraison et la planification de la production dans les exploitations d'ananas. Ses consequences.
AU: Lacoeuilhe,-J-J PY: 1983
AD: IRFA, BP 153, Martinique, French West Indies. LA: French
SO: Fruits. 1983, 38: 6, 475-480.
AB: The possibility of controlling the date of flowering makes it possible to base the date of harvest on demand and prices. A table showing data to be considered, the sequence of cultural operations and the choices available is presented. Effects on efficiency, increasing profits and diversification are discussed.

Crops

HORTSCIENCE 24(6):983–984. 1989.

Sensory Differences between Bananas Ripened Without and With Ethylene

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Additional index words. flavor, texture, color, *Musa* ×*paradisiaca*

Abstract. **A descriptive analysis panel used the terms fruitiness, greenness, and softness to score the flesh of bananas (*Musa* ×*paradisiaca* L.) ripened either with ethylene or allowed to ripen naturally to the same skin color. Fruit that ripened naturally were considered more fruity, less green, and softer than ethylene-treated fruit.**

Controlled ripening of bananas is a marketing practice in many countries and usually involves the application of ethylene (≈25 $ml \cdot m^{-3}$) to green but mature fruit held in controlled temperature rooms (15 to 20C). The skin color is routinely used by both market operators and consumers to determine when the fruit are ripe (Wills et al., 1989). The assumption that the sensory attributes of the flesh of ethylene-ripened bananas are the same as those of fruit allowed to ripen naturally seems to prevail. This equivalence, however, is disputed by many consumers who claim that fruit allowed to ripen naturally are of superior eating quality. While information is available on the identification of flavor volatiles (McCarthey et al., 1963; Nursten and Williams, 1967) and there has been some attempt to identify optimum storage conditions (Vakis, 1981; Abdullah, et al. 1985), a review by Marriott (1980) highlights that little is known about the effects of ripening conditions on the eating quality of bananas.

Although work has been carried out to identify optimum storage conditions, the criteria of optimum eating quality have not been determined with standard sensory evaluation techniques. For example, in a study by Peacock (1980), a panel of 10 assessors was used to evaluate ethylene-treated bananas ripened at various temperatures. The selection criteria for panelists were not specified and the influence of skin color was not assessed, in that liking for skin appearance and the eating quality of flesh appear to have been determined on the same bananas in the same sitting. Thompson and Seymour (1982) have reported that bananas are equally palatable when treated with either acetylene or ethylene, but, similarly, they did not present the sensory methodology or quantitative sensory profiles.

We, therefore, used a trained analytical sensory panel to examine whether fruit ripened with ethylene had a similar eating quality as fruit allowed to ripen unassisted to the same skin color.

Eight panelists who regularly consume bananas were trained in the technique of descriptive analysis described by Piggott (1984). Training was carried out with commercially available (i.e., ethylene-treated) 'Cavendish' bananas at ripeness stages 5, 6, and 7 as determined by comparison of skin color with a commercial banana ripening guide (Commw, Ind. Sci. Res. Org., 1971). Bananas at stage 5 are predominantly yellow, but the tips remain green; at stage 6, bananas are full yellow and, at stage 7, the first signs of brown spotting are apparent. Bananas were peeled immediately before use, so that panelists at no time saw the banana skins. During four 45-min sessions held within 2 weeks, panelists, as a group, determined three consensus terms (i.e., fruitiness, greenness and softness) that could be used to differentiate the sensory attributes of bananas at the various stages of ripeness and they individually scored these attributes on a 150-mm line scale. Panelists were then tested using banana pieces (three) representing the three stages of ripeness they had been trained to evaluate. Pieces were of uniform length (30 mm) and shape (cylindrical) and were randomly selected from a representative sample taken from ≈20 bananas. Testing occurred on three occasions and on each occasion two of the possible three stages of ripeness were randomly selected for presentation (i.e., two of 5 vs. 6, 6 vs. 7, 5 vs. 7). Two panelists were consistently unable to give scores that identified the stage of ripeness and they were therefore eliminated from the panel and their results were not considered for inclusion in Table 1.

Descriptor words and scores for commercially ripened bananas at various stages of skin color ripeness are shown in Table 1. The flavor terms chosen by panelists are contained in the exhaustive list of terms suggested by Harper et al. (1968) and the scores are consistent with composition and texture changes known to occur during ripening: softening, synthesis of flavor compounds (McCarthy et al., 1963; Nursten and Williams, 1967), and presence of a green taste, usually attributed to hex-3-enol, which disappears as ripening progresses (Charles and Tung, 1973; Marriott, 1980). The scores also indicate that panelists used different regions of the scale; however, the order of scores for each panelist is consistent with the order of ripeness.

Panelists were then presented with fruit that had been ripened with or without ethylene, with both samples at skin color stage 6. Bananas ripened unassisted were obtained from a commercial shipment of green fruit before, and ethylene-treated fruit were obtained after, commercial gassing (Flemington Markets, Sydney) and allowed to ripen at 20C. About 300 bananas were held in storage to allow selection of stage 6 fruit. Fresh uniform samples were prepared and presented on five different days as previously described. The sensory scores were tested by analysis of variance and also converted to ranks for comparison with Kramer's rank sum tables. This procedure follows the recommendation by O'Mahony (1986) that sensory data should be analyzed by both parametric and nonparametric methods. The data (Table 2) show that, while natural- and ethylene-ripened bananas were of the same skin color, the sensory attributes for the flesh differed. Fruit allowed to ripen unassisted were consistently ranked as more ripe than ethylene-ripened fruit. Mean scores over five replicates for commercial bananas are essentially the same as those obtained during the training period, indicating that the panel as

Table 1. Scores obtained at the end of training for sensory attributes of bananas at various identified stages of ripeness. Each score is the distance (mm) along a 150-mm line scale anchored by little fruitiness–very fruity, no green–strong green, very firm–soft. Individual scores are means from two replicates.

Attribute	Individual panelist scores						Mean
				Stage 5			
Fruitiness	35	7	22	21	30	10	21
Greenness	120	138	98	135	135	138	126
Softness	30	34	32	33	40	57	38
				Stage 6			
Fruitiness	75	35	55	66	50	34	53
Greenness	80	105	85	101	102	85	93
Softness	65	44	51	89	55	73	63
				Stage 7			
Fruitiness	120	98	88	106	65	77	92
Greenness	30	64	32	47	22	20	36
Softness	115	88	116	117	95	89	103

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Tablc 2. Sensory evaluation by six panclists of five rcplicatcs of untreated and cthylcnc-trcatcd bananas at skin color 6. Values in brackets indicate the median rank order of data. Each score is the distance (mm) along a 150-mm line scale anchored by little fruitiness-very fruity, no grccn–strong grccn, very firm–soft.

Atlributc	Panclist	Untreated	Ethylcnc-treated
Fruitiness	1	81.8 ± 13.4 (1)	45.8 ± 24.1 (2)
	2	105.6 ± 12.6 (1)	35.2 ± 25.3 (2)
	3	108.3 ± 12.6 (1)	75.3 ± 22.7 (2)
	4	118.4 ± 20.8 (1)	62.0 ± 37.7 (2)
	5	110.8 ± 28.1 (1)	68.2 ± 38.7 (2)
	6	86.6 ± 28.9 (1)	49.4 ± 30.7 (2)
	Mean[z]	102 a	55 b
Grccnncss	1	67.2 ± 17.8 (2)	104.2 ± 24.1 (1)
	2	45.6 ± 21.8 (2)	93.2 ± 51.6 (1)
	3	37.3 ± 16.6 (2)	70.8 ± 40.1 (1)
	4	39.8 ± 45.7 (2)	100.0 ± 34.7 (1)
	5	35.0 ± 24.1 (2)	73.2 ± 37.5 (1)
	6	43.0 ± 20.1 (2)	103.6 ± 31.2 (1)
	Mean	48 a	94 b
Softness	1	80.4 ± 18.3 (1)	44.6 ± 23.4 (2)
	2	95.4 ± 14.5 (1)	52.0 ± 28.7 (2)
	3	110.5 ± 10.9 (1)	80.5 ± 37.5 (2)
	4	115.4 ± 28.1 (I)	63.2 ± 46.8 (2)
	5	102.8 ± 26.0 (1)	78.0 ± 43.3 (2)
	6	80.4 ± 13.2 (1)	60.8 ± 15.2 (2)
	Mean	97 a	63 b

[z]Different letters for means indicate significant difference between means at $P < 0.01$.

a whole performed consistently throughout the test.

It seems that exogenous ethylene causes the skin and flesh to ripen out of phase, with ripening in the flesh occurring more slowly. Two previously reported biochemical studies also allude to this possibility. When the rate of ripening (i.e., skin color change) is accelerated by increasing storage temperature, Rippon and Trochoulias (1976) found that flesh softening is inhibited. Also, Vendrell and McGlasson (1971) suggested that in the absence of exogenous ethylene, peel ripening depends on ethylene produced by the flesh. If consumers who complain that "fruit doesn't taste as good as it used to" are basing the time of optimum eating quality on skin color of fruit ripened without ethylene, then they are correct in their complaints that ethylene-ripened bananas have a different taste. It might be expected that fruit ripened with ethylene would, at a more advanced skin color, attain an eating quality similar to that of fruit allowed to ripen unassisted. It would seem that, for marketing purposes, the banana industry should gather more information on consumer preferences so that the desired level of ripeness can be provided at the point of sale. It would also be useful to determine whether a significant segment of the population preferred bananas ripened without ethylene and whether the range in ripeness offered to consumers might be extended.

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E:

B. *Ultraviolet Light*

Ethylene will absorb UV light in the range from 100 to 190 nm with a maximum at 162 nm (CAL66494). The primary products of photolysis are hydrogen (24%), acetylene (50%), *n*-butane (23%), and ethane (3%) (NOY64226). The reaction between ethylene and UV light represents the primary method by which ethylene is destroyed in the upper atmosphere. Irradiation of ambient polluted air samples with sunlight for 6 hours reduced reactive hydrocarbons to levels found in uncontaminated air. Ultraviolet light was the active component. Irradiation of 0.045 μl/liter ethylene with UV light for 24 hours reduced the levels to 0.012 μl/liter and no ethylene was destroyed in the dark (STE67147). Ultraviolet light degrades ethylene by several mechanisms. In air, UV light will generate ozone from oxygen, which in turn will oxidize ethylene. Ultraviolet light, at wavelengths below 190 nm, will decompose ethylene directly, and atomic oxygen, indirectly. This was shown by the fact that ozone did not accumulate in low-oxygen atmospheres and ethylene removal was similar in air and low-oxygen atmospheres (SHO86176)

III. Ethylene Levels in Soil and Its Degradation by Microorganisms

Soil ethylene levels are often higher than those observed in the air. In the air, levels are reduced by diffusion, UV light, and oxidation. In soil, darkness, limited diffusion, and loss of organisms which decompose ethylene can result in its accumulation. In addition, soils contain biogenic and chemical sources of ethylene and other hydrocarbons such as methane, ethane, and propane (VAN83519)

Ethylene levels in soil atmospheres can be in the 10-μl/liter and above range (PER81320, MEE83631, DOW72325, SMI74217, CAM79199). Most of these values are associated with soils which were waterlogged (DOW72325, LEY78347, MEE83631), compacted (CAM79199), treated with organic materials (BUR75055), or contained diseased plants (DEM71040). Normally, ethylene oxidation by the ethylene consumer *Mycobacterium paraffinicum* is efficient enough to remove most of the ethylene in soils.

Plant damage has been associated with high ethylene levels in the soil. Workers have reported damage to vineyards (PER81320), potato (CAM79199), tulip (DEM71040), and grain crops (LEY78347, PRI76177). However, Kays and Nicklow (KAY74166) reported no effect on tomato plants grown in soil containing up to 16 μl/liter ethylene. Several workers have noted high ethylene levels in potted soils (ISH84157, HAR78721, SHE76009, ORC85407). In these reports, soils were air dried before use. Drying soil has been shown to destroy ethylene consumers (COR75085, GOO78193). While it is likely that drying destroyed *M. paraffinicum*, this has never been specifically tested. The loss of dehydration-sensitive organisms such as ethylene consumers should be taken into consideration when interpreting results with air-dried soils. Ethylene consumption is faster in the presence of oxygen. High ethylene levels observed in flooded soils may be due to the reduced activity of *M. paraffinicum* by low-oxygen tensions.

A. *Factors Controlling Ethylene Production by Soil*

The sources of ethylene encountered in soils are only partially understood. Bacteria and fungi have been isolated from soil, and they produce ethylene when methionine was added to the culture medium (ARS88728, BAB84559, LYN72045, LYN75576, PRI76343). However, there is no evidence showing that methionine levels in soil are high enough to account for the synthesis of ethylene by these organisms. Because of this, there is some question concerning the role of methionine-dependent organisms in ethylene production in anaerobic or waterlogged soils.

Methionine-independent ethylene producers such as *Penicillium* have been isolated from soil (CON75115). The role of *Penicillium* in ethylene production during anaerobic conditions in the soil needs to be examined further because ethylene production was inhibited under anaerobic conditions. Other organisms may play a role in soil ethylene production.

Bacterial methionine-independent ethylene production occurs primarily in aerial plant pathogenic bacterium like *Pseudomonas* (FRE64313). A survey by Fukuda (FUK84363) indicated that *Corynebacterium aquaticum* also produced ethylene. In spite of these reports, large numbers of soil bacteria capable of ethylene production have not been described. Methionine-independent ethylene production by fungi is more frequently observed. In two surveys of hundreds of fungal cultures, about 25% were capable of ethylene production (ILA68357, FUK84363)

Many workers have assumed that soil ethylene was biogenic. It is also possible that soil ethylene is produced chemically during the decomposition of microorganisms and complex organic molecules. Ilage (ILA72009) made the observation that several soil fungi produced ethylene during cell autolysis. Ethylene production has also been observed when oven-dried (GOO78193, HUN80022, HUN82267) or autoclaved (ISH84157, NAK80359) soils are moistened and incubated. In addition, antifungal and antibacterial antibiotics had no (LYN83415) or limited (FRA85422) ability to inhibit soil ethylene production.

The production of ethylene by soil following the addition of ACC, methionine, and lipids (BAB84559, FRA85416, ISH87069) has been reported. The amount of free ACC or methionine in soil is not known, so their contribution to soil ethylene remains to be established.

Lipids may be a source of soil ethylene. Extraction of soil with solvents isolated various lipids but no free amino acids (ISH87069). Under aerobic and anaerobic conditions, enzymes extracted from soil produced ethylene from these lipids (ISH87069). These workers concluded that soil lipids are degraded by a combination of microbial, enzymatic, and nonenzymatic reactions involving peroxides and free radical reactions.

B. *Role of Soil Ethylene in Plant Growth*

The presence of ethylene in the soil atmosphere can influence numerous facets of root growth and development. These processes are discussed in greater detail earlier in this volume. To evaluate the consequences of physiologically active levels of ethylene in soil we must enumerate some of its effects on roots. Ethylene can influence root elongation, branching, formation of adventitious roots, aerenchyma development, root hair formation, orientation to gravity, root nodulation, mycorrhizal development, and disease development. As yet no work has been done on the effect of ethylene on water or metabolite translocation.

IV. Ethylene Levels in Wat

Paddies and bogs are aquatic areas for agricultural production. In addition, lakes and estuaries are important ecological areas. There is little data available con- cerning the levels of ethylene dissolved in these aquatic systems. Some phys- iological effects of ethylene on aquatic angiosperms, such as the promotion of elongation, are an important part of their adaptation to this unique environment (OSB84167). Submergence also initiates other changes associated with flood stress such as aerenchyma, hypertrophy, increased formation of adventitious roots, reduced lignification, and changes in leaf anatomy (RID87053). The 10,000-fold reduction in ethylene diffusion in water compared to air must be considered when evaluating the role of ethylene in aquatic environments (MUS72093). The concentration of ethylene (assuming equilibration with air) in ditch water in Holland was reported to vary from 0.015 to 0.05 μl/liter while that of tap water was 0.007 μl/liter (ELZ80225). *Lemna gibba* growing in ditch water with ethylene levels greater than 0.02 μl/liter was reported to develop gibbous (swollen on the underside) fronds. Fronds grown at low ethylene levels were flat on the lower side.

TABLE 8.3.
Recommended Ethylene Levels

	Environmental air standards (μl/liter)[a]	
Location	1-Hour maximum	8-Hour maximum
Rural	0.25	0.05
Residential	0.5	0.1
Commercial	0.75	0.15
Industrial	1.0	0.2

Source: ANO68627.
[a] 1 μl/liter = 1150 μg/m³.

V. Air Quality Standards

There are no national standards for ethylene levels. In California, the recc mended levels are 0.5 μl/liter for 1 hour, or 0.1 μl/liter for 8 hc (ANO62001). In 1968, the American Industrial Hygiene Association recc mended the ethylene levels listed in Table 8.3. (ANO68627). These recomm dations are reasonable and should prevent most crop loss, except that in sensi floral greenhouse crops.

A. *Control Strategies to Reduce Ethylene Emissions*

Attempts to control hydrocarbon emissions from auto exhaust also rec ethylene levels in the atmosphere. There are no attempts to specifically coi ethylene from point sources such as automobiles, fires, or petrochemical pl manufacturing products from ethylene.

CHAPTER 9

The Role of Ethylene in Agriculture

I. Introduction

A. Early History

Ethylene was unknowingly employed in ancient agriculture. One example was the practice in the Middle East of gashing figs on the tree to promote fruit growth and ripening (see Chapter 4, this volume) (GAL68178). In China weighted lids were placed on growing bean sprouts to promote hypocotyl thickening and crispness (S. F. Yang, personal communication). In both cases stress ethylene was responsible for the desired plant behavior. Long before ethylene was recognized as the active agent, emanations from ripe fruit or smoke were used to hasten fruit ripening (KNI13337, COU10007, CHA36292, MIL47335) or promote flowering (ROD32005, TRA40521, GON24015). The first intentional use of ethylene in agriculture was to degreen citrus fruits (DEN23000). This was followed by its use to blanch celery (HAR25001) and ripen fruit (HAR28001).

B. Approaches for Developing Uses

The options available for the manipulation of ethylene production and activity are the subject of this book and are summarized in Figure 9.1. Ethylene can be added to or removed from the gas phase. Its synthesis can be promoted or inhibited, and its action inhibited. These concepts have been discussed earlier (MOR82029). The discovery that methionine was the precursor of ethylene led to the identification of AOA and AVG as inhibitors of its synthesis. Cobalt ions and

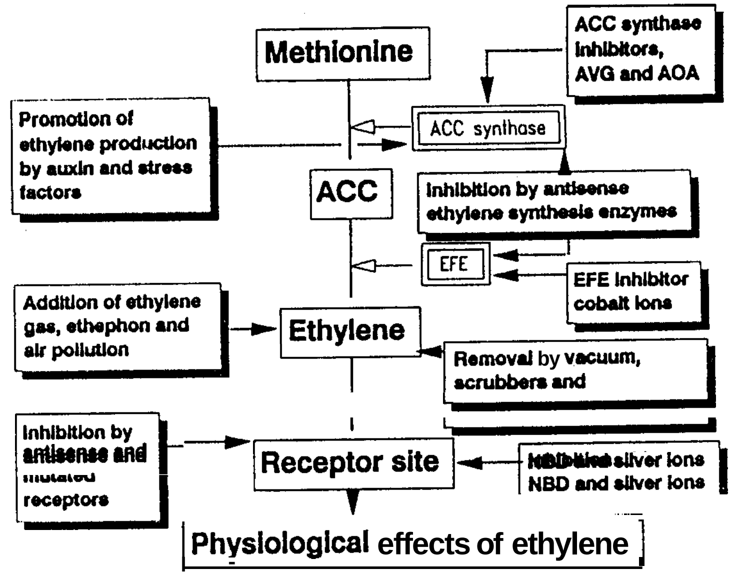

Figure 9.1. A model for the options available to manipulate behavior of plants or plant products (fruits, flowers, etc.) by manipulating ethylene physiology. In addition to adding ethylene or an ethylene generator such as ethephon, agriculturists can promote or inhibit the synthesis of ethylene, remove the gas which is formed, and promote or inhibit the action of the gas which is present.

other compounds are used to inhibit EFE. Silver and 2,5-norbornadiene are two compounds that block ethylene action. These topics are discussed earlier in this book. The ability to manipulate ethylene levels, its production, and its action makes it unique among plant hormones.

Genetic engineering introduces the possibility of altering or adding genes related to ethylene physiology (YAN85041). This strategy applies to genes involved in the synthesis or action of the gas. Ethylene promoters can be attached to other genes whose regulation would be of interest. Ethylene receptors associated with specific target cells such as the abscission zone (OSB89103) may also be amenable for regulation. The use of antisense and truncated polygalacturonase mRNAs have indicated the potentials of altering ethylene-mediated processes (SMI88724).

There are two ways of identifying uses for ethylene in agriculture. The first is to use the known effects of ethylene as a "shopping list." The second is to look at a specific crop and identify ethylene-induced effects which might be useful. Both approaches have been used in the development of ethephon as an agricultural tool (DEW71364). Recent advances in understanding the biochemistry of ethylene should be exploited. Some examples might include methods to control the synthesis of MACC or its breakdown. The development of techniques to isolate EFE and the gene responsible for its synthesis offers the opportunity to inhibit ethylene production in tissues such as flowers, whose lives we wish to prolong. The introduction of ethylene-insensitive mutants or the use of silver may uncover new roles of ethylene in plants.

II. Ethylene as an Indicator of Plant Maturity

A. Indication of Maturity

The rise in respiration and ethylene evolution indicates the initiation of ripening in mature climacteric fruits. Recognition of the date of niaturity is important in assuring high-quality fruit. Immature fruit are prone to physiological disorders such as scald and overmature fruit have reduced shelf life. Harvest before the climacteric is considered best for apples destined for long-term storage. Normally, fruit harvest is initiated on the basis of horticultural characteristics, experience, and economic factors. The development of inexpensive portable gas chromatographs (see Chapter 2, this volume) raises the possibility of using ethylene as a maturity indicator. Monitoring of internal levels of fruit ethylene could be used to decide about the disposition of each day's harvest. A table has been published relating ethylene levels to recommended disposition; long-term controlled atmosphere (CA) storage, short-term conventional storage, or fresh market (DIL82019).

Efforts have been made to use ethylene levels in apples as a method of predicting maturity and optimum harvest dates. While the initiation of the climacteric could be detected, most workers have concluded that it was not a practical way to predict optimum harvest dates (BLA86465, BLA87061, CHU84129, CHU88226). The major obstacles were variability in the data and the effort required to get accurate measurements. Other problems were that internal ethylene levels at harvest did not always predict the quality of fruit after storage (KNE89403). Fresh market quality and the rate of ethylene production were not always correlated; apple fruit-.with intermediate rates were considered as having the highest quality by a taste panel (SAL83303).

B. Prediction of Maturity Date

Determination of the induced ethylene climacteric can predict the onset of the natural climacteric (LIU78388). This effect has been used to develop a test to predict fruit maturity. The procedure is based on the ability of exogenous ethylene to induce the accumulation of endogenously produced ethylene (LIU78388, DIL85353). The procedure predicts the autogenous ethylene climac
teric a week in advance and allows preclimacteric apples to be harvested and he
in CA storage successfully (DIL89001, DIL89409).

Daminozide and Maturation

Daminozide (SADH, Alar) delayed autocatalytic ethylene production ar
ripening by about 10 days when applied to apple trees (LOO71350, DIL8535
DIL89001). Alar came to be widely used on apples intended for long-ter
storage. The scheduling of harvesting and handling of apples was adjusted to t
use of this chemical. Since Alar has been withdrawn from use in the Unit
States, the accurate prediction of the initiation of the ethylene climacteric a
assessment of the maturity state of apples at actual harvest is even more critic
Alternative methods based on bloom date and temperature during the growi
season have been described (ABE84429).

III. Ethylene Fumigation

A. Degreening

The first practical use of ethylene was to degreen citrus (DEN230(
DEN24757). The practice was quickly adopted for other fruit and its impact c
be evaluated from comments in a speech in 1940 by Dr. H. G. Knight, forn
Chief of the U.S. Bureau of Agricultural Chemistry and Engineering, defendi
the costs and benefits of research. Knight said "the treatment bleaches out t
predominant green color and leaves the orange a beautiful natural yellow. T
chemical investigations leading to the development of this treatment, which
now in rather general use, cost the taxpayers of the country about $4,000 anc
estimated to be worth about $4,000,000 a year to producers of citrus fruits
Florida alone and about the same amount to producers in Californ
(AN04108s, CRO48139). The U.S. Department of Agriculture published cir
lars with detailed instructions for degreening operations (WIN55001).

Ethylene continues to be used before the marketing of fresh citrus, exc
limes (SHE85057). It is a routine practice which is recognized by agencies wh
regulate pesticide usage and insurance codes (SHE85057). Descriptions a
engineering specifications for commercial facilities to degreen citrus fruit
available (ANO64001). Localized recommendations are available from Univ
sity and County Extension personnel (WAR73001). Ethylene sources for
greening include catalytic gas generators, compressed ethylene, and compres
ethylene diluted with an inert gas (e.g., "banana gas") (SHE85057). Ethylene
applied by "shot," "trickle," and "flow-through" methods. Control of the c
centration is required for optimum activity, to limit the cost of materials, anc

prevent explosions. The explosive range of ethylene in air is 3.1 to 32%. The 3.1% cconcentration exceeds the level recommended for citrus fruit degreening by 6200 times. Instructions for installation of flow-through systems have been published (SHE81001).

B. Ripening Fruit

Ethylene was used unknowingly to ripen bananas in East Africa. Green bananas used to make beer were placed in the sun for 3 hours and then buried in a banana leaf-lined pit for 3 days. The pit was often warmed by a fire before the bananas were buried (MAS38362). Ripening bananas in fire-warmed pits was also practiced by Samoans (MAS56051). Von Loesecke (VON49001) quoted a marketing report stating that artificial ripening of bananas was widely practiced in India. Fruits were exposed to smoke and warmth in a confined space. It is likely that many of these practices took advantage of ethylene in smoke and restricted ventilation to hasten ripening (SIM59001).

When ethylene produced by kerosene stoves was shown to be the active agent for degreening citrus, these stoves were also used to ripen mature-green bananas (MIL47335). Soon, ethylene itself was used to ripen bananas near the point of retail sale. This eliminated the heat generated by the stove and the decay it encouraged (MIL47335).

The techniques used to ripen bananas in the United States, Great Britain, and Australia have been described (SIM59001). They have not changed appreciably since the 1930s. Early workers recommended ethylene for promoting ripening of tomatoes, pineapples, cantaloupes, dates, jujubes, persimmons, pears, mangoes, pomegranates, peppers, avocadoes, honeydew melons, apples, plums, papayas, cherimoyas, plantains, chicory, and endive (CHA27135, CHA36292, HAR28001, HIL46001, MIL47335, ROS25315). Ethylene was also used to loosen hulls of mature walnuts harvested in the husk (CHA36292). It also promoted dehiscence of pecan nut shucks (FIN36074). However, pecans are not usually harvested in the shuck.

More recent guides on the use of ethylene to ripen tomatoes (GUL81001, SHE81001) and honeydew melons (KAS70001) are available. Jahn has pointed out that 100 μl/liter ethylene is a saturating dose, and higher levels are unnecessary (JAH75018). A formula based on ACC content of the fruit, ethylene concentration, and exposure time needed to ripen bananas has been published (INA88561). Currently, in the United States, ethylene-enhanced ripening is used in the marketing of tomatoes, bananas, mangoes, and honeydew melons (SHE85057).

Acetylene is used as a substitute in countries where ethylene is not readily available. It is used at concentrations of 0.1% in air (THO82407). It is explosive

in air at 2.5% and higher. This means that the margin of safety for acetylene is less than that for ethylene.

C. Bean Sprouts

Ethylene has been used in the commercial production of bean sprouts. About half of the sprouts currently produced in California are gassed with ethylene (S. F. Yang, personal communication). The system was recently studied in Japan where a combination of 50 μl/liter ethylene and 600 μl/liter CO, applied 1 hour per day improved sprout quality (TAJ85317).

D. Flowering of Geophytes and Pineapple

Ethylene has been used to promote flowering of geophytes. Ethylene accelerated the flowering of iris bulbs (STU66019). Iris, freesia, and narcissus geophytes are treated with ethylene to break dormancy and promote flowering (DEM86085, SCI-182173, UYE83091). Since flowers compete with the formation of new bulbs, flowering is undesirable for bulb production. Bulb development was promoted and flowers were blasted when tulip bulbs were treated with 0.5 μl/liter ethylene for 3 days before planting (KAM76101).

Ethylene was shown to induce flowering in pineapple (ROD32005). Acetylene was also effective, and because of its greater solubility in water, was applied in the field as an aqueous solution (COL35078, LEW37532). Acetylene was generated by mixing calcium carbide with water and was used in pineapple growing regions (COO42093). The use of acetylene was eventually replaced by auxins (CLA42536) and ethephon.

E. Seed Germination

Striga hermonthica and ***Striga asiatica*** (witchweed) are phanerogamic parasites of sorghum and millet. They cause serious crops losses in Africa and in an infested area in North and South Carolina (EPL81951). Seeds, which are normally dormant, germinate near host roots which produce strigol, a germination stimulant (EGL82013). The ***Striga*** seedlings then invade the host species. Ethylene was found to be a germination stimulant for ***Striga*** (EGL7058 EPL75433). A control program in North Carolina based on the injection of ethylene into the soil was instituted (EPL81951) and several thousand hectares were treated (EGL82013). The objective was to cause suicidal germination of seed in the absence of obligate host roots. Three annual applications of ethylene in sequence along with control of escaped plants achieved eradication in treated areas. Partial eradication has been achieved on sites in Africa with the same treatments (BEB86694).

F. Curing Tobacco Leaves and Defoliation

Ethylene has been used to promote curing of tobacco leaves after harvest (CRO48139) and to defoliate nursery stock after digging (MIL4001I). Ethylene is still used for these purposes either in the form of the gas or as ethephon.

G. Latex Flow

In 1912 Kamernum secured a British patent on scraping the bark of rubber trees to increase the yield of latex (DIC76088). It was suggested that plant hormones induced by scraping were causal agents (BAP39017). Various hormones were tested and the auxin analogue, 2,4-D was found to be effective (CHA51167). Its use became standard practice. Subsequently, it was suggested that auxin-induced ethylene production played a role in 2,4-D action (MAX67548, MOR62420) and that ethylene was the causal agent for sustained latex flow (DIC76088). This was verified by applying ethylene and compounds which stimulate its production to the tapping panel (ABR68291, DIC76088). Ethephon was soon used to improve latex yields. The Rubber Research Institute of Malaysia developed "Ethad," a molecular sieve material saturated with ethylene and then enshrouded with a viscus mixture of oil and grease (DIC76088). Its effect was similar to ethephon in some tests.

IV. Chemicals That Generate Ethylene

A. Ethephon

1. Discovery

Some early examples of compounds used to release ethylene or acetylene are Omaflora (a formulation of β-hydroxyethyl hydrazine) and calcium carbide. Omaflora (CAT65027) and calcium carbide (COL35078) were used to promote flowering in pineapple.

Ethephon was first synthesized by the Russian workers Kabachnik and Rossiiskaya (KAB46295). The observation by Maynard and Swan (MAY63596) that it produced ethylene was not exploited. The discovery that ethephon was an ethylene generator was not the result of a specific search for an ethylene-generating compound. Ethephon was included in a group of compounds synthesized by the GAF Corporation (Wayne, New Jersey) and tested for growth regulator activity by Amchem Products, Inc. (COO68974, MOR82029). The appearance of treated plants plus the chemistry of ethephon suggested that it was converted into ethylene. Ethylene was released for testing by Amchem Products, Inc, in 1967 (ANO67001) and papers describing some of its effects quickly appeared (EDG68064, MOR69337, WAR69156, YAN69203). Commercial development proceeded rapidly (DEW71364).

2. Ethephon Chemistry and Ethylene Release

The class formula for the 2-chloroalkylphosphonic acids (MAY63596) is:

$$R—CH(Cl)—CH_2—PO_3H_2$$

The hydrolysis at pH 5 and above produces chloride, phosphate, and ethylene (MAY63596). It was suggested that a chemical mechanism involving nucleophilic attack on the phosphate dianion by water was responsible for ethylene production (MAY63596, YAN69203). This resulted in the elimination of chlorine and the formation of phosphate.

$$Cl—CH_2—CH_2—PO_3H_2 + OH^- \rightarrow CH_2{=}CH_2 + Cl^- + H_3PO_4$$

Quantitative evolution of ethylene was obtained by treating ethephon with excess base at 75°C for 10 minutes (COO68974). The yield of ethylene and phosphate were equal and when $[^3H]_2O$ was present, $[^3H]$ did not appear in the olefin, indicating that dehydrohalogenation was not a part of the reaction (YAN69203).

3. Ethephon Residue Tolerance

Published toxicological data for ethephon indicate that the levels in milligrams per kilogram per day which had no observable effect are: 90-day feeding (dog), 5; 3-generation reproduction study (rat), 75; teratology (rabbit), 50; neurotoxicity (chicken), 1000; and 2-year oncogenicity (rat), 1.5 (ENV85105). Based on these studies, the acceptable daily intake of ethylene is 7.5 mg/kg/day. This value adjusted to a 60-kg human with a 100-fold safety factor becomes 4. mglday. The residue tolerance factor for ethephon on wheat and barley is 0.1 ppm

As discussed earlier, ethylene does not have a physiological effect on micro organisms, insects, or animals at the μl/liter levels used to alter plant growth Ethephon should break down in aqueous animal fluids in a manner similar to th occurring in plants. Its conversion to ethylene minimizes it as a hazard.

4. Ethephon Analysis

Ethephon can be analyzed by gas chromatography (BAC70730) an $[^{31}P]$NMR (LAN86577). The GC method is the more sensitive of the tw Ethephon labeled with $[^{14}C]$ has been used to study its absorption, translocatio and metabolism.

5. Absorption, Translocation, and Metabolism

The breakdown of ethephon to ethylene occurs primarily on the leaf surfa (BEA88784). $[^{14}C]$Ethephon was'translocated throughout walnut seedlings 1 no metabolites were detected (MAR72051). On grapes, breakdown and absorption was slow (62% of the ethephon was recovered after 7 days) and, within the berries, only [^{14}C]ethephon was detected. Ethephon breakdown on leaves was rapid (WEA72013). Most of the [^{14}C]ethephon applied to peach fruit was broken down and only a small portion was absorbed (LAV74097). Ethephon absorbed by basal leaves of apricot shoots moved into the fruit on the shoot. The reverse did not occur (GIU81033). Exposure of cherry fruit to ethephon for 1 hour resulted in a 30% reduction in fruit removal force (BUK71777). This illustrates its rapid uptake and later conversion to ethylene. Ethephon applied to *Poa pratensis* and *Avena sativa* shoots was still releasing ethylene after 25 days (VAN78639). In a study with squash, cucumber, and tomato a 50% conversion of [^{14}C]ethephon to ethylene occurred in 7 days (YAM71606). Most of the remaining label was still in the form of [^{14}C]ethephon. However, a derivative of [^{14}C]ethephon was observed in squash seedlings. An unidentified metabolite of [^{14}C]ethephon was also detected in cherry leaves (GIL75290).

In contrast to the small amount of metabolism in most species, ethephon has been found to be converted into nonvolatile products such as 2-hydroxyethylphosphonic acid in rubber trees. Between 39% (ARC73535, AUD73634) and 19% (AUD76183) of [^{14}C]ethephon applied to rubber trees was converted to nonvolatile products. Rubber tree cells in suspension culture also metabolized [^{14}C]ethephon to nonvolatile products (AUD78329).

6. Uses of Ethephon in Agriculture

Ethephon is regarded as "liquid ethylene," and is probably the most widely used plant growth regulator in agriculture. It is registered in the United States for many preharvest and harvest-aid processes (Table 9.1.). In 1985, L.D. Page of the Union Carbide Corporation (Fremont, California) estimated that over 50% of the ethephon used in the United States was applied to field crops such as wheat, barley, and cotton (MOR86375). Ethephon is used in large amounts in tropical regions in the production of coffee, pineapple, rubber and sugarcane (Table 9.2.). It is also used on horticultural and ornamental crops in both the United States and Europe. Reports describing new uses for ethephon continually appear and point to new opportunities for its use.

a. Seed Germination Ethephon has been used to break the dormancy of peanut (KET77042), geranium (ROG87165), and witchweed seeds (EGL70586). Ethephon applied to the soil reduced the number of witchweed shoots. As a result, its host sorghum exhibited increased height and flowering (BAB83125).

b. Sprouting of Bulbs Ethephon has been used to overcome dormancy of gladiolus (HAL70427) and *Liatris* (KER84163) corms. In addition to promoting sprouting of gladiolus, ethephon also increased the effectiveness of fungicides, apparently by increasing their penetration into the corms (SIM72369).

Ethylene in Plant Biology 2nd ed. 1992
FB Abeles, PW Morgan & ME Saltveit, Jr.
Academic Press 414 pp.

TABLE 9.1.
Registered Uses for Ethephon in the United States[a]

Purpose	Crop	Product[b]
Preharvest		
Promote lateral branching	Azalea, geranium	Florel®
Increase bud hardiness and delay bloom	Sweet cherry	Ethrel®
Initiate uniform flowering	Pineapple, bromeliad	Ethrel®, Flor
Promote flowering on young trees	Apple	Ethrel®
Modify flowering and sex expression, early fruit set	Cucumber, squash, pumpkin	Florel®
Thin flowers and promote return bloom	Apple	Ethrel®
Prevent preharvest fruit drop and promote fruit color	Apple	Ethrel®, Fru N[c]
Prevent lodging, shorten stems	Wheat, barley	Cerone®, Fl
Harvest aid		
Hasten yellowing and reduce curing time	Tobacco	Ethrel®
Promote and/or hasten uniform ripening, color development, and maturity	Cherry, table grape, raisin grape, pepper, blackberry, tomato, apple, boysenberry, pineapple	Ethrel®, Flo
Promote fruit abscission and nut hull dehiscence	Cherry, apple, blackberry, cantaloupe, walnut	Ethrel®, Flo
Promote mature fruit dehiscence and enhance defoliation	Cotton	Prep®
Promote defoliation, remove shoots	Rose, tallhedge buckthorn, apple nursery stock, dwarf and leafy mistletoe	Florel®

[a] Information taken from specimen labels in product label guide, Rhone-Poulenc Agricultura Company, 1990, for Cerone®, Ethrel®, Prep®, and Florel®, which are all registered trade n
[b] Ethrel for Flue Cured Tobacco®, and Ethrel, Plant Growth Regulator® are sold in 2 pound ethephon per gallon. Cerone® and Ethephon for pineapple and sugar cane contain 4 pounds ethephon per gallon. Prep® contains 6 pounds of ethephon per gallon. Florel® is sold in 0. pounds of ethephon per gallon.
[c] Active ingredient, 1-naphthaleneacetic acid, sodium salt.

c. Growth Retardation The capacity for ethephon to inhibit stem el tion has been used to increase hardiness in seedlings used for transpla Ethephon-treated tomato seedlings had reduced height and leaf area but s leaf number and dry weight as controls (WOO87133).

Ethephon treatment also increased frost tolerance and the survival r tomato seedlings after transplanting (LIP82400). Transplanting of tobacco

Basic physiological studies of ethylene action were overshadowed when it was shown that ethylene had the commercially important property to ripen fruit (DEN23000). For the next twd decades, researchers concentrated on the applied aspects of ethylene action instead of on its synthesis and mode of action.

Denny showed that ethylene promoted ripening of lemons at concentrations as low as 0.2 µl/liter, while the maximum effect was observed at 5 µl/liter. Oxygen was required for ethylene action, and an increase in respiration was noted 1 days after ethylene treatment. He also noted that ethylene was safe for humans and did not represent a fire hazard at these parts-per-million levels (DEN24757). The cost of treating a railroad car with ethylene was estimated to be $1.00 U.S. (CHA24339). Ethylene was later shown to stimulate the ripening of cucumbers, tomatoes, apples, and mandarins (IVA30262). Ethylene treatment was also shown to increase respiration and levels of invertase in these fruits. Attempts to understand ethylene action through its induction of specific enzymes and other metabolic changes were reported by Regeimbal and Harvey on pineapple (REG27117) and with Vacha on banana (REG27357).

The citrus and banana shippers were the first to use the degreening properties of ethylene (MIL47335). Ethylene was also used to remove astringency from persimmons, to ripen pears, and to loosen walnut hulls. Concern was voiced about the residual effects of ethylene treatment. It was suggested that its use constituted a fraud by allowing inferior fruit to be sold at a higher price. Researchers showed that ethylene-treated fruit were equivalent in quality and healthfulness to naturally ripened fruit and that employees working with ethylene in packing houses were not exposed to hazardous conditions (CHA34152).

At about the same time that ethylene was discovered to affect plants, it was also being tested for its anesthetic properties in animals (LUC23851). The first successful use of ethylene to cause anesthesia was in 1885 when Luessem used a 75% ethylene : 25% air mixture to anesthetize mice.

The story of the early discovery of ethylene as the active principle in illuminating gas has received much attention. Crocker and Knight (CRO08259) and Harvey and Rose (HAR15027) reviewed the early history starting in 1863. Chace outlined the development of the commercial use of ethylene to color fruits (CHA34152). In 1933, Crocker reviewed the general effects of ethylene and summarized the work from the Boyce Thompson Institute (CRO32295). This was followed by numerous reviews (e.g., LAA34691, BUR62265, KAY87077).

IV. Ethylene Production by Plants

The use of ethylene to manipulate the growth and development of agricultural crops was investigated in the 1920s and 1930s. Meanwhile, the possibility that it was an endogenous growth regulator was questioned for many years. Experiments with bioassays provided preliminary evidence that ethylene was produced by plants. Cousin's report in 1910 was the first to document that the ripening of bananas was promoted by gases produced by other plants, in this case, oranges (COU10007). He stated that, "emanations from oranges stored in a chamber were found to have the effect of bringing about premature ripening of bananas if these gases were passed through a chamber laden with this fruit." Since it is now known that oranges are not as active in producing ethylene as other fruit, it is possible that the effect observed was from fruit infected with *Penicillium*.

In 1932, Elmer showed that volatile substances from ripe apples and pears inhibited the normal growth of potato sprouts (ELM32193). In 1933, Kidd and West noted that ethylene stimulated the respiratory climacteric in apples. They also observed that emanations from apples mimicked the ethylene effect on seedling growth, and suggested that apples make ethylene (KID33055). In 1933, Botjes showed that volatile compounds from ripe Winesap apples caused leaf epinasty of tomato seedlings, and the triple response of etiolated peas. He concluded that apples produced ethylene since the effects were similar to those observed when ethylene was applied to the plants (BOT33207). In 1934, Kidd and West tested several fruits for their ability to evolve gases which would cause a climacteric rise in respiration in apples. Effective fruits were bananas, pears, peaches, and tomatoes. Ineffective were grapes, oranges, and senescent, abscising leaves. Ventilation and subatmospheric levels of oxygen reduced the rate of ripening. Applied ethylene had no effect without oxygen, but induced the climacteric when the fruits were returned to air (KID34051). In 1934, Kessler placed ripe apples and seedlings in bell jars. He observed that root and stem growth of these seedlings was abnormal. Theoretical considerations led him to conclude that the effect was due to minute quantities of ethylene produced by the fruit (KES34853).

These and other studies performed with bioassays indicated that ethylene was produced by plants. Conclusive chemical proof that ethylene was a natural product of plants was provided by the English scientist Gane in 1934 (GAN34008). The comparatively crude technique available required collecting the gases evolved from 60 pounds of ripening apples for 4 weeks.

A few years later, in 1940, Biale reported that gases produced by lemons infected with *Penicillium italicum* stimulated respiration, degreening and pedicel abscission in sound lemons (BIA40458). These gases were also produced by *Penicillium* grown in culture and were thought to include ethylene. Also in 1940, Miller, Winston, and Fisher used tomato epinasty and other bioassays to show that *P. digitatum* produced ethylene. This observation accounted for the increase in ethylene production observed from diseased versus sound oranges (MIL40269).

As evidence accumulated concerning its biological activity, some scientists began referring to ethylene as a plant hormone. For ethylene to be considered a plant hormone, it was felt that certain criteria must be met. In addition to promoting ripening, ethylene should be produced by ripening fruit (GAN34008, ISA38307, KES34853). Furthermore, if it was controlling ripening, its production should increase before ripening started.

In 1935, Gane provided chemical evidence that fruits other than apples produced biologically active levels of ethylene. In addition, he suggested that they responded to it and that it played a role in ripening (GAN35351). Bioassays showed that ripening tomato fruit produced ethylene. It was also shown that the rate of production increased with ripening of tomatoes (HEI53397). In 1957, Workman and Pratt, using relatively insensitive manometric techniques, provided evidence that an increase in ethylene production was associated with ripening and the respiratory climacteric in tomato (WOR57330).

For some workers, support for a hormonal role for ethylene hinged on showing that its concentration increased before ripening. Biale, Young, and Olmstead considered ethylene a byproduct of ripening since an increase in production was detected after the start of ripening (BIA54168). With the advent of gas chromatography some of these issues could be resolved. It was now possible to show that an increase in ethylene production preceded, or coincided with, the respiratory climacteric and other parameters associated with fruit ripening.

This view of hormonal control was simplistic. Hormonal control of physiological events can also result from a change in the sensitivity of the tissue to the hormone which is already present. The issue of sequential changes in ethylene production and subsequent response is not as acute as it once was. We now know that some ripening parameters can be temporally separated from the increase in respiration and ethylene production. The ripening of many kinds of fruit is regulated by two factors. These are the internal concentrations of ethylene and the sensitivity of the tissues to the gas as they age and mature. Moreover, this picture is complicated because applied ethylene can age the tissue and make it more sensitive to a second exposure of ethylene. The question of a regulatory function for ethylene in stress and senescence is still the subject of investigation.

The use of the term hormone for ethylene is now an accepted practice. However, careful consideration of the definition of the word leads one to think that its use is not completely accurate. The term hormone was coined by animal scientists. They used it to describe endogenous chemicals, active in low amounts, transported from a site of production to a site of action, to bring about a desired physiological response. For all practical purposes, ethylene is produced by all cells all the time, and transport is not a concern. However, there is little point in originating a new name for a gaseous plant regulator since only one is known. The word hormone is adequate as long as the distinction concerning translocation is remembered. However, the precursor of ethylene, 1-aminocyclopropane-1-carboxylic acid (ACC), is soluble, and can be translocated in plants. Interorgan translocation of ACC was first described for the root-to-shoot transport in tomato (BRA80322). Translocation of ACC within flowers has be suggested to play a role in floral senescence (WOL90837). Ethylene levels xylem sap of Norway spruce in May were found to correspond to those o solution in equilibrium with 70 μl/liter ethylene (EKL90190). Ethylene levels dormant wood a month earlier were near zero.

The physiological significance, or evolutionary advantage, of a gasec hormone is hard to assess. One advantage is that degradation is not essenti offering an advantage in economy in the regulation of hormone levels. Unl other hormones which are rendered biologically inactive by enzymatic transf mations, ethylene simply diffuses away from the plant. In the soil it is consum by microorganisms and in the air by UV light or ozone. It is difficult to envis the evolutionary sequence of events, or selection pressures, that made an unsa rated two-carbon gas a hormone. Reconstruction of the forces and events wh led to the acquisition of biological activity by ethylene, or other hormones, not been actively pursued by plant taxonomists or physiologists.

V. Discovery of the Plant Hormones

The discovery of another hormone, auxin, took place during the same period that of ethylene. Early work by Charles Darwin in the 1880s demonstrated t the tip of a grass coleoptile was the site of light perception in phototropism. T Hungarian scientist Paal found that the tip provided the material required for growth phenomena observed in the subtending portion of the coleoptile. Betwe 1900 and 1910 he reported that removal of the coleoptile tip stopped elongat of the remaining portion of the coleoptile. Replacing it caused the coleoptile resume growth. Placing the tip on one side of the decapitated coleoptile cau growth on that side. At about the same time, the Danish scientist Boysen-Jen reported that phototropism did not occur if a strip of mica was inserted betw the illuminated tip and the rest of the coleoptile. Replacement of the impervi mica with a block of gelatin caused the plant to regain phototropic sensitiv This demonstrated that the tip provided a growth promoter. Chemical identifi tion of the growth substance stemmed from the work of Fritz Went. In 1926 Dutch graduate student reported that tips of oat coleoptiles secreted a substa into agar blocks that replaced the physiological effectiveness of intact coleop apices. The substance was called auxin. The chemical structure of auxin was determined directly from these plant extracts but rather from urine. Kogle and associates isolated, purified and identified indoleacetic acid (IAA) in 1934. T showed that this component of urine mimicked the physiological effectivenes coleoptile tips. In 1935 K. V. Thimann identified IAA as a component of pl tissues.

Research with other plant hormones was active in the years after the disc

CHAPTER 3

The Biosynthesis of Ethylene

I. Nonbiological Synthesis

Ethylene can be produced by numerous nonbiological chemical reactions. Many organic substances will produce ethylene when subjected to heat, oxidation, light, or ionizing radiation. The presence of ethylene in smoke and illuminating gas led to the discovery of its biological activity (see Chapter 1, this volume).

Culture filtrates from microorganisms can evolve ethylene. Ethylene formation by cell-free extracts of *Escherichia coli* have been reported (INC86151). Similarly, culture filtrates of the ectomycorrhizal fungi *Hebeloma crustuliniforme* and *Laccaria laccata* evolved ethylene that was nonenzymatic in nature (GRA80340). Illuminated and sealed flasks, devoid of tissue, but containing agar and culture medium accumulated 0.5 μl/liter ethylene after 17 days (LEO87223).

Nonbiological model systems that produced ethylene were used during the search for the biosynthetic pathway. Chemical systems comprised of ascorbate and copper were used to study the synthesis of ethylene and other hydrocarbons from peroxidized linolenic acid (LIE64343). These systems led to the serendipitous discovery that methionine enhanced ethylene production.

It was shown that carbons 3 and 4 of methionine gave rise to ethylene in both the chemical system and apple tissue (LIE65449, LIE66376). Ethylene was also produced from carbons 3 and 4 when a mixture of methionine, flavin, and pea extract was illuminated (YAN66739). Other analogues of methionine could be used in this system. The ease with which ethylene was produced from these compounds points out the major problem with nonbiological systems; they do not

exhibit the specificity characteristic of living tissue. Cell-free systems have been used to study ethylene production from methionine, methional, and α-keto-γ-methylmercaptobutyric acid (KMBA, the deaminated product of methionine) (BEA70641, FUK89173, YAN67274, YAN69360).

Ethylene production from KMBA has been used to measure peroxidase or ligninolytic activity (YAN69360). The ligninase of *Phanerochaete chrysosporium* is a peroxidase. It was reported to produce ethylene from KMBA in the presence of hydroxy radicals (HO•) (KEL82423). The following pathway has been proposed for the production of hydroxy radicals from oxygen (FUK89107).

$$2Fe(II) + 2O_2 \rightarrow 2O_2^- + 2Fe(III) \text{ (superoxide production)}$$
$$2O_2^- + 2H^+ \rightarrow H_2O_2 + O_2 \text{ (dismutase reaction)}$$
$$H_2O_2 + Fe(II) \rightarrow HO^\bullet + Fe(III) + OH^- \text{ (Fenton reaction)}$$
$$KMBA + HO^\bullet \rightarrow C_2H_4 + CO_2 + \text{methanethiol(?)}$$

The enzyme responsible for generating ferrous ions is an NADH:Fe(III) EDTA oxidoreductase (FUK89107). The mechanism is:

$$NADH + 2Fe(III) \xrightarrow{\text{NADH:Fe(III)EDTA oxidoreductase}} NAD^+ + 2Fe(II)$$

The requirement for Fe(III)EDTA could be replaced with $K_3Fe(CN)_3$ and 2,6-dichlorophenol indolephenol. Ethylene production depends on a source of KMBA. The system will not produce ethylene from ACC or α-ketoglutarate. Some bacteria contain a transaminase which converts L-methionine to KMBA. The enzyme uses various 2-oxo acids as cosubstrates but 2-oxoglutarate had the highest activity (SHI89489). Bacteria which do not produce ethylene after the addition of methionine may lack either the transaminase or the oxidoreductase (OGA90287).

II. Biological Synthesis

A. *Bacteria*

Ethylene does not appear to play a role in bacterial growth and development. A few soil-borne species such as *Mycobacterium paraffinicum* can grow on ethylene and convert it into ethylene oxide. The biochemistry of ethylene oxidation and its role in controlling ethylene levels in the soil atmosphere is discussed in Chapter 8, this volume.

Ethylene-producing bacteria may have enhanced disease-inducing capacity compared to other bacteria. While most living plant tissue is resistant to microbial attack most of the time, all dead or dying plant materials are susceptible to decay. The role of ethylene in microbial attack may be to promote senescence and thereby facilitate growth of the organism. Certain strains of *Pseudomonas* confirmed by showing that inhibitors of ethylene synthesis and action delay ripening. Some examples of these studies are cited in Table 6.2.

In the nonclimacteric strawberry, AVG, silver, and 2,5-norbornadiene had no effect on ripening as measured by anthocyanin accumulation (GIV88402).

II. Ripening Enzymology

A number of enzymes have been shown to increase during ripening. For example, some of the enzymes associated with ripening in apple include: exo-polygalacturonase (BAR78213), malic enzyme, carboxylase, diaphorase, cytochrome C reductase (JON65158), α-amylase (KIT80277), chlorophyllase (LOO67245, RHO67001), β-cyanoalanine synthase (MIZ87031), and RNase (RHO67001). However, in another report, malic enzyme activity was found not to change (KNE89403). Most of the information available on ripening enzymes is that concerned with softening, respiration, and ethylene production. As described in greater detail later the two major softening enzymes are cellulase and polygalacturonase. However, hemicellulases may also play a role. The increase in respiration has been shown to be due to an increase in enzymes from the alternative pathway of respiration. The effect of ethylene on this pathway has been discussed earlier. Details on specific fruits are included in the following discussions.

The increase in ethylene production during ripening is due to an increase in ACC synthase and EFE. An increase in these enzymes during the ripening of apple (MAN86495, MOR86173, PLI89241), apricot (SAW89633), avocado (SIT86130), pear (KNE87724), and tomato (BRE87476) has been observed. Enzymes associated with the synthesis of methionine do not appear to be involved. Adding methionine usually has no effect on ethylene production (TER85467).

Treating fruit, especially immature fruit, with ethylene can promote the synthesis of EFE (AGI87144, INA86348, SIT86130, TER85467). An increase in EFE has also been observed in nonripening *nor* tomatoes (LIU85891). During normal ripening ACC synthase activity typically increases at the same time (SAW89633) or before (BRE87467) EFE activity. Added ethylene reverses this sequence so that an increase in EFE occurs at the same time or prior to the formation of ACC synthase. This reversal has been observed in avocado (STA91921), banana (INA86348), tomato, and cantaloupe (LIU85407). These observations indicate that ethylene has a more immediate effect on regulating EFE than precursors in the ethylene biosynthesis pathway. The increase in EFE after ethylene treatment is not limited to fruit tissue. The same effect has been reported in vegetative tissue and is a part of the autostimulation of ethylene production observed in pea (SCH89331), winter rye (IEV90221), and carnation (OVE90368). Inhibition of EFE levels has also been reported for some species. This response may play a role in autoinhibition of ethylene production (BRE84869, SAW89633). A loss of EFE activity during the later stages of ripening in fruits such as avocado results in an accumulation of ACC in postclimacteric tissue (SJA85105).

ACC produced during ripening can be conjugated with malonic acid to form MACC. This reaction is catalyzed by D-amino acid malonyltransferase. Malonyltransferase has the ability to conjugate D-amino acids, α-amino-isobutyric acid and ACC with malonyl-CoA to form *N*-malonyl-amino acids. The K_m for malonyltransferase is 170 m*M* compared to 66 μ*M* for EFE, suggesting that MACC is a minor sink for tissue ACC (MAN86495). Accumulation of MACC is usually associated with postclimacteric fruit (MOR86173). Treatment of unripe tomato fruits with ethylene has been shown to increase levels of malonyltransferase (LIU85891).

Studies with two-dimensional gel electrophoresis (BAK85167, LAY90850) and differential cDNA cloning (MAU87177) have indicated that many proteins are involved in ripening. Decreases as well as increases in the synthesis of proteins, for which most of the functions are not known, have been demonstrated. Details concerning the ripening of specific fruits are presented below.

III. Ripening in Fruits of Commercial Interest

A. *Bromeliaceae*

Pineapple *Ananas comosus* Merr.

Postharvest respiratory drifts for six stages of development ranging from dry flower to senescence indicated that pineapple is a nonclimacteric fruit. Pineapple were shown to have internal ethylene levels of 80 to 1140 μl/liter. Application of ethylene to fruit did not increase ripening of mature fruit as measured by a change in respiration or chemical composition (DUL67059). However, fruit color is manipulated with ethephon in commerce. The nonclimacteric nature of this fruit may be due to its anatomy. The fruit is a syncarpium formed by the coalescence of a thickened rachis, spiny-toothed bracts, and abortive ovaries.

B. *Musaceae*

Plantain *Musa paradisiaca* L.
Banana *Musa paradisiaca* var. *sapientum* L.

Bananas demonstrate the typical influence of tree factors and an aging requirement before ripening. Fruit left on the tree will remain green for 7 weeks while those removed from the stem ripen in 2 weeks (BUR65200). Following harvest, the time required to respond to ethylene decreases as the fruit ages. Before the onset of natural ripening, fruit are sensitive to the level of ethylene (0.2 μl/liter) found in preclimacteric fruit (BUR65200). The term green life was defined as the time it took harvested green bananas to ripen (PEA72137). It was shown that ethylene hastened aging by shortening green life in these fruit (PEA72137).

Ripening of banana slices has been inhibited by silver ions (SAL78472). Slices of green banana infiltrated with IAA and 2,4-D did not ripen when treated with ethylene (VEN69601, WAD71165). This included the inhibition of yellowing of the peel as well as softening of the pulp. However, 2,4-D did not prevent ethylene from promoting respiration (VEN69601). This observation suggests ethylene-induced respiration in preclimacteric tissues is mediated by a mechanism distinct from that associated with ripening. The ripening of banana can be separated into changes occurring in the peel and those occurring in the pulp. Treatment of fruit slices with kinetin and gibberellin resulted in delayed peel yellowing but normal pulp softening. Apparently, yellowing and softening are under the control of different juvenility factors.

Most of the increased respiration and ethylene production associated with banana ripening occurs in the pulp (KE088048). Chlorophyll degradation in the peel was inhibited if the peel was removed from the pulp. Treating the isolated peel with ethylene stimulated degradation. It was suggested that in banana, pulp ethylene is a major factor controlling peel yellowing and pulp softening (KE088048). These observations support the idea that climacteric ethylene, associated with ripening, facilitates rapid and uniform changes in fruit. Thus, bananas ripen from the inside out and flesh ripening precedes peel yellowing. Taste panels indicated that ethylene-treated fruit were harder and less fruity than naturally ripened fruit. Externally applied ethylene reverses the ripening process so peels yellow before the pulp softens. Criteria for selecting fruit in the store for consumption should consider this shift in relative rates of ripening for ethylene-promoted ripening versus natural ripening (SCR89983). Superficial brown spots develop on peel tissue late in ripening. The development of these spots was shown to be inhibited by ethylene (LIU76684). This effect of postclimacteric ethylene was blocked by treating fruit with silver ions.

The rise in ethylene production in bananas is associated with an increase in enzymes involved in ethylene biogenesis. Using ethylene to promote ripening, it was found that the last enzyme in the pathway, EFE, was induced in 4 hours. The first enzyme in the pathway, ACC synthase, was induced 8 hours later (INA86348).

During ripening, starch, which represents 23% of the fresh weight of green fruit, was converted into sugar (17% of fresh weight), CO_2, organic acids, and protein. These changes are accompanied by a reduction in pectin and tannic acid

(CHI85561). During the climacteric, starch-to-sugar carbon conversion increased 100-fold, while carbon conversion to CO_2 increased fivefold (BEA89436). It was calculated that 0.6 moles of CO_2 were produced per mole of sucrose formed during ripening (HUB90201). The ATP requirement for sucrose production was between 28 and 83% of the total produced. This suggests that the increase in respiration is needed to produce ATP used in starch-to-sugar conversion. Three enzymes involved in starch-to-sugar conversion which increased during ripening were starch phosphorylase, acid invertase, and sucrose phosphate synthase. The phosphorylase converted starch and inorganic phosphorous into glucose-phosphate and fructose-6-phosphate. Outside the starch grain uridine triphosphate (UTP) reacts with glucose-1-phosphate to form uridine diphosphate glucose (UDPG). In the presence of sucrose phosphate synthase the UDPG, fructose-6-phosphate, and glucose-6-phosphate will generate sucrose-6-phosphate. The latter is dephosphorylated to sucrose and then hydrolyzed to glucose and fructose (HUB90201). Fructose-2,6-bisphosphate plays a pivotal role in carbohydrate metabolism and is utilized in both gluconeogenesis and glycolysis. Studies of the enzymes involved in the synthesis of fructose-2,6-bisphosphate indicate a rise in ATP-dependent phosphofructokinase, a decrease in pyrophosphate-dependent phosphofructokinase, and no change in fructose-1,6-bisphosphatase (BEU87277, BEU89436, MER87579).

Other enzymes which increase during banana ripening are peroxidases (CAN90223), acidic phosphatases (KAN89251), and chlorophyllases (LOO67245). Pectinmethylesterase was found to decrease during ripening (KAN89251).

C. *Moraceae*

Fig *Ficus carica* L.

The first example of the exploitation of ethylene to induce ripening was described in the Bible. The prophet Amos was described as a gasher and gatherer of figs. Sycamore fig (*Ficus sycomorus*) was cultured in the eastern Mediterranean region for its wood as well as its fruit. It was found that gashing mimicked the action of wasps when they exited pollinated fruits. Gashing figs was shown to promote stress ethylene production which subsequently promoted ripening (GAL68178, ZER72378).

The "fruit" of the fig is a synconium. Fertilization of these flowers results in the formation of achenes inside a fleshy body of receptacle tissue. Growth in the fig takes place in 3 distinct stages. Two stages, I and III, of rapid growth are separated by stage II, one of slow growth. Ethylene inhibits fruit growth in I, the cell division period, and stimulates growth in stages II and III. The onset of stage III and a respiratory climacteric rise was preceded by or concomitant with a rise in ethylene production. This, together with the fact that exogenous ethylene adjacent mesocarp. Cellulase appeared in both parenchyma and oil cells concomitantly with wall breakdown. Immunogold detection of cellulase by electron microscopy revealed labeling associated with endoplasmic reticulum, plasmodesmata, and cell wall during the latter part of the climacteric (DAL89033).

E. Rosaceae

1. Pear *Pyrus communis* L.

Exposure to low temperature stimulates ethylene synthesis and ripening of pears. The low-temperature effect has been observed in stored fruit or those attached to the tree. This chilling requirement is usually observed in late maturing pear cultivars (KNE83207). Cultivars which show this requirement include D'Anjou and Passe-Cmssane. The ripening lag of Bosc and Conference can also be reduced by cold storage (BLA85520). Other cultivars such as Barlett do not exhibit a cold storage requirement to induce ripening. Low-temperature storage (0°C) requirements for D'Anjou were 45 days (BLA85520) and 84 days for Passe-Crassane (MOR85353). Once the cold requirement has been met, ripening is similar to that of other climacteric fruit. Ethylene treatment of Passe-Crassane at 15°C could replace the effect of a cold storage treatment (HAR87505). The low-temperature requirement for pear ripening is similar to other processes such as seed stratification and bud chilling. The mechanism by which low temperature acts to initiate these processes is unknown.

The role of aging in ethylene action in pears has been demonstrated (HAN68807, WAN72009). IAA and 2,4-D retarded ripening (FRE73006), while the antiauxin α(*p*-chlorophenoxy)isobutyric acid accelerated the onset of chlorophyll degradation, softening, and CO, production (FRE73380). The action of this antiauxin was independent of ethylene since it depressed ethylene evolution. The role of RNA and protein synthesis was demonstrated by showing that inhibitors of these processes inhibited ripening. Both actinomycin D and cycloheximide were shown to inhibit ripening (DIL79055, KNE87724).

Ethylene-enhanced ripening in pears includes an increase in sucrose, reducing sugars, and soluble pectin, and a loss of chlorophyll, starch, and firmness (HAN39145). Softening was associated with the dissolution of the middle lamella and gradual disintegration of fibrillar material throughout the cell wall. In fully ripe fruit most of the fibrillar arrangement in the cell wall was lost (BEN79197). The fractionation of pectic polysaccharides from the juice of ripening Bartlett pears gave two types of polyuronides. The major type was a homogalacturan whose molecular weight decreased upon ripening. The other type comprised heteropolymers composed of various amounts of arabinose, rhamnose, and galactose. Apparently these polymers are degraded by different mechanisms as pear fruit ripen (DIC89394).

Both cellulase (BEN79500) and polygalacturonase (ATT85665, BEN79500) have been shown to increase during pear ripening. The polygalacturonase in pears possesses both endo-polygalacturonase (BAR82248) and exo-polygalacturonase (DIC89394) activity. The endo-polygalacturonase had a pH optimum of 4.5 while the exo-pol'ygalacturonasehad a pH optimum of 5.5 and was activated by Ca(II) ions (PRE76349). As in other fruits, pectinmethylesterase activity was present in mature firm fruit. Workers have reported that this enzyme either increased (BAR82248) or declined during ripening (BEN79500).

2. Apple *Malus sylvestris* Mill.

Numerous cultivars of apples are used in commercial fruit production. Similarly, numerous patterns of ripening in terms of the climacteric have been reported. In general, earlier maturing cultivars produce more ethylene than later maturing ones (HAN45631, NEL38037). In a study of eleven apple cultivars, internal ethylene levels were around 0.1 μl/liter until they reached the optimal harvest date. At that point the rate of ethylene production increased dramatically (about 1000-fold) for some cultivars such as McIntosh, Spartan, Northern Spy, Golden Delicious, and Mutsu, moderately (about 100-fold) for Empire and Delicious, or slightly (about 10-fold) for Jonagold and Idared (CHU84129) (see Figure 6.2.).

The slow rate of softening and water loss in many apple cultivars are major factors in their ability to retain their quality for up to 6 months in refrigeration and 12 months in controlled atmosphere (CA) storage. The discovery that low O, (about 3%) and high CO, (about 5%) increased the storage life of apples was made by Kidd and West (KID34051). These concentrations are similar to those used today in commercial CA storage. It has been suggested that low O_2 and high CO, blocks ethylene action (BUR67144). However, it may be more appropriate to think of the CA gas phase as narcotic or mildly toxic to fruit tissue. Controlled atmospheres retard ethylene production and cell wall degradation, without causing irreversible or obvious physiological damage (KNE88499, PLI89173, STO88089). The concentrations of O, and CO_2 used in CA storage are selected by trial and error. Injury to fruit has been reported if O_2 levels are too low or CO, levels are too high (LAN84113, STO88089). Low O_2 levels will inhibit respiration and promote fermentation. The increased levels of ethanol in fruit following low O, storage may also play a role in delaying ripening (STO89149). Apples removed from CA storage exhibited a loss of metabolic activity. These fruit have a reduced ability to produce ethylene (LI085489) and have lost some response to ethylene as measured by a promotion of respiration (KNE88504). In spite of a lack of understanding of how CA storage works, it is used to extend the storage life of approximately half of the present domestic apple crop.

Cell wall composition has been shown to change during ripening. A loss of structure and sugars in cell walls undergoing autolytic degradation has been stage III promoted fruit growth (DAN78589) and ripening (ROM71134). However, the final growth of the fruit is not under ethylene control since treatment of stage III peaches with silver or AVG had no effect on fruit size (MIL88119). The role of ethylene during stage III was presumed to be associated with color development, softening, and abscission.

Softening in peaches involves a combination of polygalacturonase, cellulase (PRE77172), and other as yet unknown oligosaccharidases. Clingstone peaches have an exo-polygalacturonase, while freestone cultivars have both exo- and endo-polygalacturonase (PRE78415).

F. *Oxalidaceae*

Carambola *Averrhoa carambola* L.

Star fruit or carambola are considered nonclimacteric fruit. Ethylene and production were lower in ripening fruits than in green ones. Dissolved onds steadily increased with ripening (OSL83229). Ethylene had no effect on range color development or other aspects of ripening (LAM87181).

G. *Rutaceae*

Orange *Citrus sinensis* Osbeck
Lemon *Citrus limon* Burm. f.

Citrus are different from other climacteric fruits in that commercial harvest ccurs after the fruit are mature. Mature citrus produce only trace amounts of hylene and exhibit level rates of respiration (AHA68099, MCM72235, OU62416). This observation lead early workers to suggest that citrus were onclimacteric. As more information became available it was shown that ethene increased respiration, caused a loss of chlorophyll, and promoted abscison of the pedicel button in both immature and mature citrus (EAK70334, AK88559, TAK84314).

A reexamination of the relationship between ethylene production, respiraon, yellowing, and abscission has caused a number of workers to suggest that rious citrus species are climacteric. The patterns of ethylene production and spiration observed after premature fruit are harvested are similar to those hibited by other climacteric fruit (AHA68099, MIZ88273). Increased ethylene oduction has been associated with fruit abscission (BEN70144) and yellowing US80917) on the tree. Seasonal changes of endogenous ethylene levels in ee species of *Citrus* fruits attached to trees were investigated (SAW81935). gh levels of internal ethylene (1000 µl/liter) were observed in young fruit in y, and they were associated with premature abscission of small fruit. In ptember, ethylene levels decreased to the 0.1-µl/liter level. Ethylene levels

rose again in October and November (time of normal color break) to levels of 2 to 10,000 µl/liter. Removal of the fruit from the tree resulted in rapid reduction of internal ethylene to 0.1 µl/liter.

Ethylene applied as an ethephon spray was shown to advance the ripening of oranges as measured by the decrease in chlorophyll and the increase in carotenoid content (ABB84127). The chlorophyll-degrading system in the peel of tangerine and calamondin was shown to require an exogenous supply of ethylene to keep it functioning at a high level (PUR81854). Chlorophyll degradation ceased when the fruit were removed from ethylene and placed in hypobaric conditions (0.2 atmosphere). At 1 atmosphere chlorophyll degradation continued for 24 hours. Ethylene also induced chlorophyllase activity in the peel of these fruits. However, the authors concluded that chlorophyllase may not be the sole or primary enzyme used in chlorophyll degradation. Chlorophyll degradation stopped after ethylene was removed but chlorophyllase activity remained elevated. Chlorophyll degradation is reversible in citrus. Senescence in citrus peel is reversible. The regreening of yellow peels has been cited as an example of this phenomenon (BIA64880). Gibberellin promotes regreening and has been used to facilitate "on-tree" storage of fruit.

The morphology of citrus peel can be influenced by ethylene. Ethephon promoted the formation of "peel puffing" in Satsuma mandarin (*Citrus unshiu*). Peel puffing was characterized by an increase in the ratio of peel weight to fruit weight and a decrease in peel specific gravity. The degree of peel puffing was reduced by treating the tissue with AVG, or by storage of fruit in the presence of $KMnO_4$ (MAO83238). Peel puffing may be similar to the morphology of increased albedo formation seen when citrus are grown in hot, dry climates.

H. *Anacardiaceae*

Mango *Manfigera indica* L.

Ripening of mango is associated with orange coloration, decreased flesh firmness, and the conversion of starch into sugar (KAR39597). The changes in ethylene production and respiration were followed for fruits allowed to ripen on the tree. Ethylene production was high following pollination and decreased as the fruit grew. An increase in both ethylene and respiration was associated with ripening and abscission of the fruit (AKA73381). Fruits harvested at the mature and half-mature stages developed good quality characteristics, but immature fruits showed only limited changes during ripening. Treatment with ethylene initiated ripening at all three maturity stages, although ripening remained incomplete in immature fruits (MED88153). The minimum time required for ethylene to have an effect was 4 hours. Between 4 and 8 hours the effect of the gas was maximal (MED90426). An increase in both catalase and peroxidase activity have been associated with mango ripening (MAT69308).

leaves or subsequent growth of the tree. If olive leaf abscission exceeds 25% then flower bud development for next year's crop is reduced (SUN82957). The ripening of olive was considered to be climacteric on the tree and nonclimacteric after harvest (MAX60275). Ethephon promoted ripening as shown by an increase in anthocyanins, respiration, and abscission (RUG82835). An increase in cellulase was noted during fruit softening and when the skin color went from green to black (HER90022).

T. Solanaceae

I. Eggplant ***Solanum melongena*** var. ***esculentum*** **Ness.**

Eggplant fruits exhibit a climacteric pattern of ripening (SIG84097, SIG84117). Cultivars with small, white, oval fruits where shown to have the longest shelf life. Ethylene reduced storage life of the fruit to one-half that of the controls.

2. Tomato ***Lycopersicon esculentum*** **Mill.**

The tomato has played a major role in ripening research. These berries grow year-round and several ripening mutants are available for genetic analysis. In addition, extensive experience with its biochemistry, and advances in molecular biology and tissue culture have facilitated its use as an experimental model. Slices of tomato pericarp exhibit typical ripening changes such as color development, and increased respiration and ethylene production (GEE88107, SAL78472) (see Figure 6.3.).

As a horticultural crop the tomato has been selected for its usefulness as human food. Most studies concerning ripening tacitly assume that changes in color, texture, and flavor are major concerns in the physiology of the fruit. However, a variety of patterns occur during ripening of other tomato species. Fruits that have a climacteric, change color, and then abscise included *Lycopersicon esculentum*. Other species abscise before ripening. ***Lycopersicon chilense*** and ***L. peruvianum*** bear fruits that do not soften until they fall from the vine. Ethylene production starts after fruit drop. Chlorophyll content falls during development and the mature fruit are whitish without carotenoids. Some fruits such as *Solanum pennelii* soften on the vine while green and then abscise. In *L. hirsutum*, distinguishable external ripening changes are not observed. The fruit remain hard and green, and remain attached to the plant after they have shown an increase in ethylene production. These fruits exhibited the largest rates of ethylene production at the climacteric, in the order of 2000 µl/kg/day or about tenfold greater than L. *esculentum* (GRU81428).

Ethylene has been shown to be required for the development of competence

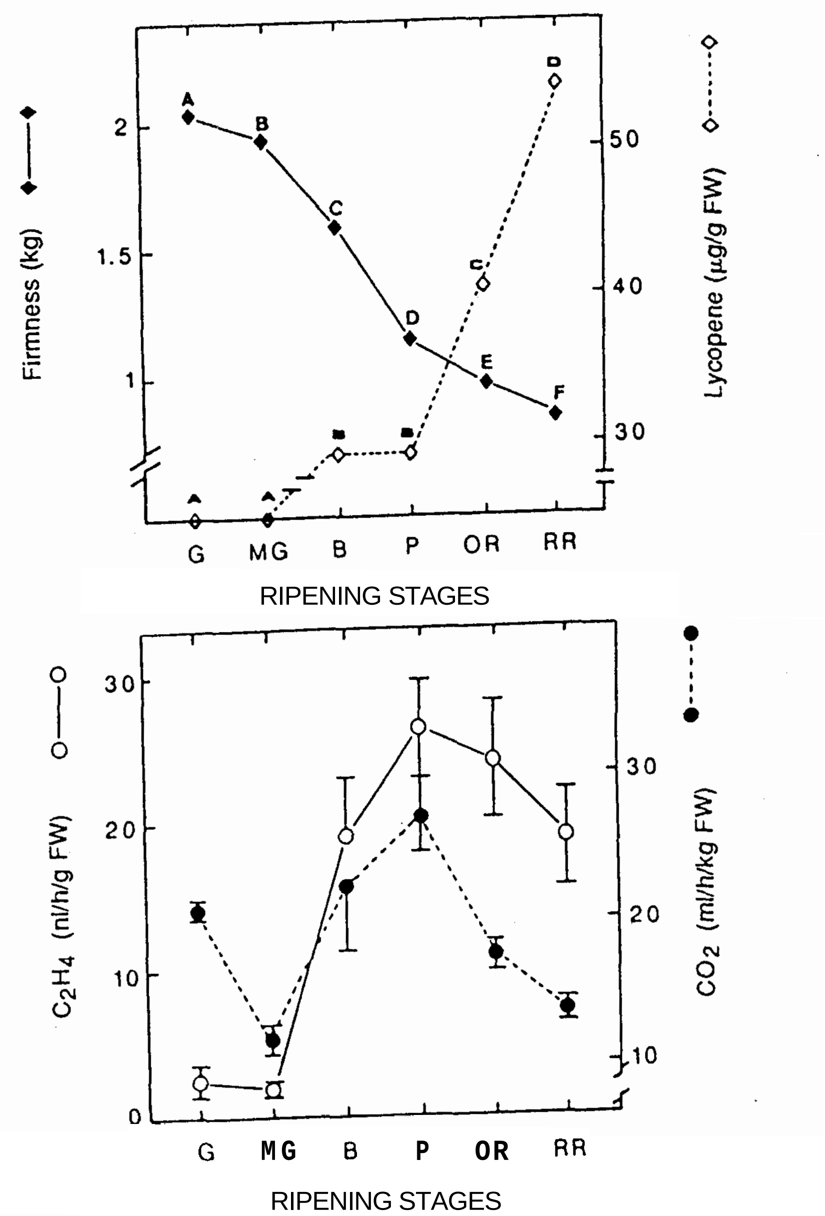

Figure 6.3. Changes in firmness (♦), color development (◊), respiration (●), and ethylene p duction (O) during tomato ripening. Ripening stages G, green; MG, mature green; B, breaker; P, p OR, orange; and RR, red ripe. Courtesy of ROT89340.

to 1 μl/liter and caused "Jonathan breakdown," a postharvest disorder of apples characterized by flesh browning beneath the peel (GUR87777).

The decline of citrus has been rioted near sites located adjacent to landfills (BAU91165). The symptoms observed include leaf abscission, inhibition of growth, and death of the trees. An analysis of the soil atmosphere in an area 20 m from the landfill with damaged trees indicated the presence of 10 μl/liter ethylene, 50% CO_2, and 1% O_2. The soil atmosphere in an area 95 m from the landfill with normal trees had no ethylene, 0.1% CO_2, and 20% O_2.

2. Losses in Greenhouses

The discovery that ethylene regulated plant growth was the result of research investigating the reasons why illuminating gas damaged greenhouse plants. Much of the early literature in this field consists of reports that ethylene in greenhouses damaged carnations (CRO13277), orchids (DAR63761), and other ornamental plants (KRO37000). Typical sources of ethylene were the incomplete combustion of heating fuels, operation of gasoline engines near the greenhouse, and combustion products of insecticide foggers.

3. Shipping Containers

Loss or damage to plants in containers has been associated with the accumulation of ethylene. This was evident as early as 1942 when shippers of holly wreaths suspected that ethylene in storage rooms was responsible for premature defoliation (MIL42000). Damage to carnations (SMI64092), Kentucky bluegrass sod (KIN82638), and ornamental plants (MAR79320) has also been described.

4. Storage facilities

Storage areas may be the primary site of economic loss due to ethylene damage. At this point of crop production all of the economic expenditures have been made, the potential for damage is greatest, and stored produce itself is the source of the gas which will cause the damage. The fact that the storage facility is an enclosure only worsens the problem because biogenic ethylene will now have a chance to accumulate in the gas phase. Levels in apple storage atmospheres range from 100 μl/liter to 700 μl/liter (POR53434, JOH62137). Lower, but still potentially damaging values of 0.05 μl/liter have been reported in floral stores (HAN69004).

Premature ripening, senescence, and abscission are the primary problems observed in storage. Some examples of ethylene-induced damage include bud and stem kill in nursery stock (CUR52104), and induction of bitter-tasting phenolic substances (isocoumarins, also known as 6- or 8-methoxymelleins) in parsnip (SHA88912), sweet potatoes (BUE75018, KIT87037), and carrots (COX73881). Other forms of ethylene-induced damage in storage include loss of

flesh firmness in watermelons (RIS82946) and induction of russet spotting in lettuce (LIP87854).

Without specific studies on produce shelf life it is difficult to tell if ethylene removal would be of benefit. As discussed elsewhere, ethylene removal is not always beneficial.

The problem is due to the difficulty in trying to reduce internal ethylene levels from a tissue which is constantly producing the gas. In addition to the diffusion gradient, internal ethylene is controlled by the rate of synthesis and its loss by diffusion through the epidermis. This means that the only way to reduce internal levels is to reduce the rate of synthesis or increase the rate of diffusion. The rate of synthesis can be reduced by lowering the temperature, though care has to be taken to avoid damage to chilling sensitive crops. The rate of diffusion can be increased by hypobaric storage (BUR66314). Under reduced pressure the free mean path of diffusion, and mobility of ethylene, is increased.

The benefit of ethylene removal is dictated by the sensitivity of the tissue to the gas. There is no point in removing ethylene if the physiological process has advanced to the point where it is not under the control of externally applied or internally generated ethylene. The benefit of ethylene removal can be measured empirically, or by measuring the rate of the process when its synthesis or action is blocked by specific inhibitors.

II. Ethylene Degradation in the Atmosphere by Activated Oxygen and Ultraviolet Light

A. *Activated Oxygen*

Ozone is produced by reaction of oxygen with ultraviolet light. The products of the reaction between ozone and ethylene are water, carbon dioxide, carbon monoxide, and formaldehyde (SCO57171). One of the side effects of this reaction is that ethylene air pollution can reduce ozone pollution. A reduction in ozone levels from 0.15 μl/liter to 0.1 μl/liter was noted near automobile traffic (MEN76080).

Atomic oxygen is formed by the reaction between nitrogen dioxide, sunlight, and ozone. The products of the reaction between ethylene and atomic oxygen are carbon monoxide, ethane, propylene, acetaldehyde, propanal, butanal, hydrogen, ethylene oxide, and dioxyketene (LEI61000).

Hydroxy radicals are formed by the reaction of ozone with sunlight in the troposphere. Ethylene and other compounds are destroyed by reactions with these hydroxy radicals (RAS86169).

This background information will be sent separately.