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## Wastage of postharvest fruit and its control\*

### By L. E. Rippon

Horticultural Postharvest Laboratory, N.S.W. Department of Agriculture, Gosford.

### Introduction

Postharvest wastage in fresh fruits and vegetables is extensive, and is represented as total loss of a commodity or reduction of its quality and value to varying degrees. Substantial losses during storage, shipment and marketing are the rule rather than the exception, and are much heavier than is generally realized because commodities increase several-fold in unit value in passing from farm to consumer. The added value cost of produce in Australia may represent a fiveto ten-fold increase on farm gate value. Most researchers separate losses into the three categories of mechanical injuries, parasitic diseases and non-parasitic disorders, and although the parasitic diseases are the ones discussed here, any enlightened program of spoilage control must consider all three causes.

As much as we might like to consume our fruits and vegetables as 'tree' or 'garden fresh', it is a fact of life we may not always do so. Often they are produced in areas distant from population centres and frequently they mature at a time of the year when consumer demand is weak or when the market is glutted with the product. These circumstances may necessitate a period of several days, weeks or even months for storage and shipment before the product reaches the consumer. Substantial losses from decay may occur during this period if the product is not treated with an effective inhibitor of microbiological growth and/or stored in an environment that is unfavourable to disease development. Coursey and Booth (1972) and Eckert (1975) have reported that postharvest losses of 25-50% of a crop are not unusual in some tropical countries where refrigeration facilities are not available and appropriate

\*A paper presented at the 2nd Australian Food Microbiology Conference, Sydney, July 1979. chemical treatments are not used. Losses from decay in technologically advanced countries are more difficult to assess because they vary greatly with season, farm and postharvest handling practices. In Australia it is very difficult to find documented evidence of the extent of this spoilage, but from published surveys in the United States (USDA 1965) and decay records of untreated produce in large scale experimental shipments (Eckert 1977), we can get some idea of the magnitude of the problem.

Apart from the obvious loss of one or more units of an edible product, postharvest deterioration may have several less evident consequences such as:

- Partial or total loss of consumer packages with only one or several diseased units.
- Reduced shelf-life (postharvest life) of the product due to accelerated ripening or senescence triggered by the release of the gas ethylene from a few diseased fruits in a package or storage room. Peacock (1973) showed that the green-life of bananas was reduced by the presence of fruit infected with *Colletotrichum musae*, and Wild *et al.* (1976) showed that the presence of lemons



Fig. 1. Collapse of a carton of lemons after storage in the container for six weeks. (Note also the presence of dead buttons).

infected with Penicillium digitatum

significantly increased the ripening rate of other fruit in the same environment.

- Possible contamination of the edible product with a mycotoxin elaborated by the disease-inducing organism. For example, Buchanan et al. (1974) and Sommer et al. (1974) showed that patulin is produced by *Penicillium expansum* in diseased apples and stone fruit, while Boyd and Wilson (1972) showed that furanoterpenoid metabolites were present in sweet potatoes infected with *Ceratocystis fimbriata*.
- Softening of processed fruits by heat-tolerant macerating enzymes, such as those Harper et al. (1972) showed to be secreted by *Rhizopus stolonifer* in incipient infections on apricots and peaches.

Postharvest deterioration causes severe losses of both time and money. After months of good agronomic practices in the production of a crop, deterioration in a few days or weeks can turn an otherwise successful agricultural venture into an economic failure, by wiping out a large investment in harvesting, packaging, storage and transportation, the combined costs of which may be several-fold greater than the total value of the product in the field. Postharvest deterioration is a serious problem not only for the producer or distributor of fresh fruits and vegetables, but also may adversely influence the availability and cost of these commodities to the consumer. Coursey and Booth (1972) in their studies concluded that, as a result of huge postharvest losses of staple root crops and fruits in some developing countries in the tropics, the solution to food shortages is not wholly agronomic in nature. More indirectly, postharvest diseases are often a major consideration in reaching decisions that influence the consumer price: requirements and duration of storage, mode of transportation, possible utilization of costsaving practices such as mechanical harvesting, bulk handling, and consumer packaging. While we enjoy year-round availability of fresh fruits and vegetables, the price we pay often includes the cost of the technology of transporting these commodities over long distances (perhaps from one side of the continent to the other), or holding them in storage for extended periods—for example up to nine months for apples stored under controlled atmosphere.

The most serious postharvest diseases are

those that cause rapid and extensive breakdown of certain fruits and vegetables which are high in moisture and nutriment, often soiling the entire package and causing secondary infections in the advanced stages of the disease. This condition is exemplified by the attack of *Rhizopus* spp. on stone fruits and strawberries, *Penicillium* spp. on citrus and pome fruits (blue and green mould rots), and *Erwinia carotovora* on leafy vegetables and potatoes (bacterial soft rots). It has been estimated that 30% of all fruit decay is caused by species of *Penicillium* and 36% of all vegetable decay by soft rot bacteria (Wiant and Bratley 1948).

The stem-end rots and brown rots are another group of postharvest diseases that are quite serious on specific commodities, but typically the pathogen is restricted to the inner tissues of the host and there is less tendency for the disease to spread after harvest. Examples of economically important diseases in this category are *Monilinia fructicola* on stone fruits (brown rot), *Phytophthora* spp. on citrus fruits (brown rot), and stem-end rots of citrus fruits caused by *Diplodia natalensis*, *Phomopsis citri*, and *Alternaria citri*.

### Nature and cause of postharvest diseases

Ripening fruits and vegetables are susceptible to attack by a variety of pathogenic microorganisms which were not infective during the period of their development on the plant. A plant product that is mature and living should not be viewed, however, simply as a nutritive medium capable of supporting the growth of the wide variety of saprophytic organisms. While extracts of many plant products support the growth of common fungi and bacteria, the living fruit or vegetable is quite resistant to attack by most microorganisms. The extent of microbial deterioration is determined in each individual case by the physiological capabilities of the microorganism and the properties of the specific plant product.

The 'brown rots', exemplified by Monilinia fructicola on stone fruits, Gloeosporium spp. on apples, and Diplodia stem-end rot of citrus fruits, may become a serious problem when a substantial portion of the crop is infected at the time of harvest, but usually the diseased fruits remain firm and the pathogen does not spread readily during storage and shipment. On the other hand the 'soft rots' of fruits and vegetables caused by Rhizopus, Geotrichum, Sclerotinia and Erwinia spp. are a serious group of postharvest diseases which progress rapidly under optimum conditions, and the extracellular enzymes of the pathogens may macerate the fleshy tissues of the host into a watery mass within a few days. In addition, the soft-rot pathogens may spread by contact to adjacent units in the same container, creating pockets of decayed produce.

The development of diseases may be recognized as two stages—infection and symptom expression. These two events may be discrete and separated by several months (latent infections), or they may take place in a continuous sequence if the environment is favourable for development of the pathogen.

# Modes of infection *Latent*

The delay between infection and symptom expression or latent infection is a phenomenon due to a transient resistance of the host to the extensive development of the



**Fig. 2.** Cool storage trial to demonstrate the effect of low temperatures in assisting in the storage of grapefruit that have been treated with fungicide to prevent mould wastage.

pathogenic organism. Certain postharvest diseases may arise from infections on the flower parts of young fruit, in which case the period of latency can be as long as several months. In other diseases, short-term latent infections may be initiated in the field several weeks or less before the crop matures.

Conidia of Botrytis cinerea (grey mould) of strawberries germinate in moisture on flower petals and then move from the necrotic petal into the receptacle (strawberry) and form a latent infection, which becomes the site of disease development after the berries are harvested. Spores of anthracnose (Colletotrichum spp.) of avocado, banana, mango and paw-paw germinate in water on the fruit surface during its development on the plant. The germ tube terminates in a durable quiescent structure known as an appressorium, which gives rise to infection hyphae when the fruit begins to ripen. Diplodia, Phomopsis and Alternaria (stem-end rots of citrus fruits) spp. form a dormant infection in the stem button in the field and become active after harvest and only when the fruit senesces and the button separates from the fruit. Grey mould (Botrytis cinerea), the most serious disease of cool-stored grapes, originates from late-season infections in the vineyard, while latent infections of brown rot (Monilinia fructicola) of stone fruit significantly influence the choice of fungicide to be used to control this disease after harvest (Kable 1971).

### Infection through lenticels

The lenticels of some fruits and vegetables provide entry points for pathogenic organisms that are unable to penetrate the uninjured surface of the host. Lenticel rotting of apples arises from latent infections of *Gloeosporium* spp., and bacterial soft rot of potatoes after harvest arises from contamination with cells of *Erwinia carotovora* at harvest, which remain quiescent until the tubers are subjected to conditions which increase their susceptibility to decay.

### Infection during and after harvest

Unlike those referred to under latent and lenticel infections, most microorganisms responsible for postharvest diseases are unable to penetrate the surface barriers of the host. As a result, injuries arising during and after harvest are the usual point of entry for these wound-invading pathogens, which, as a group, cause the most damaging postharvest diseases. Injury caused by severing fruit from the plant is a common site of invasion by wound pathogens. Good examples of this are crown rot and stem-end rot (*Gloeosporium musae*) of bananas, pedicel rot of pineapple and stem-end rots of mango, paw-paw, avocado and green pepper. The organisms responsible for the most serious postharvest diseases of citrus fruits are green and blue moulds (*Penicillium digitatum* and *P. italicum*), which invade through any injury caused to the rind during harvesting and handling. Under the most favourable conditions, germination and infection beyond control can occur after 24 to 36 hours.

Increases in injury resulting from mechanical harvesting compared with hand harvesting are well documented, and one of the challenges to postharvest pathologists is the control of postharvest diseases incited by wound pathogens in mechanically harvested produce. In addition to providing injuries as sites for infection, this method of harvesting causes bruises and excessive pressures which stimulate latent infections without necessarily rupturing the rind. Examples of this are blue mould (*Penicillium expansum*) of apples and soft rot of potatoes, which are both initiated within the lenticels, when excessive pressure crushes cells around the lenticels.

### Factors influencing disease severity

The capability of a microorganism to initiate a postharvest disease, as well as the final outcome, depends on a number of factors which can be conveniently associated with the microorganism, the host, or the environment.

### Microorganism

For development of a postharvest disease, the microorganism must elaborate enzymes which macerate the host tissue and cause a release of nutrients from the macerated cells which are suitable for growth of the organism. Some fungi, such as Rhizopus stolonifer and Geotrichum candidum, grow very slowly if at all below 10°C, whereas others, e.g. Botrytis spp. and Penicillium spp., are capable of growth at 1°C. Spores of some fungi, for example those that initiate latent infections in the field, must be capable of growth and infection in drops of pure water or dilute nutrients which diffuse from the host surface. Others, like *Penicillium digitatum*, require complex nutrients both for germination and infection.

Plant cells are held together by intracellular materials composed mainly of pectic polysaccharides. Development of a postharvest disease depends upon the capability of the pathogen to secrete enzymes that depolymerize these insoluble pectic polymers, leading to a loss of tissue coherence and separation of the individual cells, a process referred to as tissue maceration. The cells of such tissue increase in permeability and die, allowing diffusion of host metabolites which may be used as substrates for growth by the pathogen.

### Pathogen-host interactions

Each type of fruit and vegetable may be attacked only by a relatively limited and unique group of parasitic fungi, which have nutritional requirements and enzymatic capabilities permitting them to develop extensively in their host's tissue. This specificity is seen with the *Penicillium* genus, in which P. digitatum causes postharvest disease of citrus fruit only, whereas P. expansum is a serious pathogen of apples and pears, but not citrus. P. italicum is more universal in its activity attacking a wide variety of fruit and vegetables. Indeed, even different tissues of the same host may vary in susceptibility to the same isolate of a pathogen. For example, the outer leaves of cabbage are more resistant to attack by Botrytis spp. than the inner leaves, and adaxial surfaces are more resistant than the abaxial surfaces (Yoder and Whalen 1975).

Susceptibility of produce to postharvest decay is influenced significantly by crop maturity at harvest and the ensuing physiological changes that are collectively called ripening. Susceptibility of apples to the blue mould *P. expansum* increases with maturity and ripeness, apparently associated with greater susceptibility to bruising (Wright and Smith 1954), while oranges become more susceptible to invasion by Penicillium moulds with advancing maturity. Resumption of the activity of latent infections during ripening of fruits is a clear case of increasing disease susceptibility with maturation, rather than susceptibility of host to mechanical injury. Alternaria spp. infections of citrus only occur when the fruit passes a threshold of ripeness during storage, while latent infections of banana, mango, paw-paw and avocado fruits by Colletotrichum spp. are seldom a problem until the fruit approaches ripeness. Fusarium dry rot in

potatoes becomes more serious with time in storage (Boyd 1972), and the susceptibility of carrots to *Botrytis* spp. at 5°C similarly increases with storage. There are recorded cases of susceptibility to disease decreasing with maturity. For example, it is known that green tomatoes are more susceptible to bacterial soft rot than ripe tomatoes (Parsons and Spalding 1972), and white potatoes become less susceptible to bacterial soft rot during the first few weeks of storage because the periderm layer matures during this period (Boyd 1972).

### Host

The inherent and variable resistance of fruits and vegetables to postharvest disease might be associated with one or more properties of the host, such as: pH, nutrient availability or water status of the tissue; inhibitors of microbial growth; vulnerability of the cell wall to attack by pectolytic enzymes of the pathogen; and the ability of the host to form morphological or chemical barriers to the development of the pathogen.

*pH, nutrients and water status of the host.* The acidity of the tissue of many fruits may be one of the most important reasons for their general resistance to the bacteria that cause soft rot of many vegetable crops (Lund 1971). Tomatoes, peppers, cucumbers and pears are the few fruits known to be seriously affected by bacterial soft rot. The pH of fruit tissues is less than 5, and this inhibits the growth of most bacteria capable of degrading plant tissues. Vegetable tissues are generally less acid.

Clear cut examples of unique nutrients or growth stimulants in susceptible varieties of fruit and vegetables are rare, but their existence must, however, be considered. *Penicillium digitatum*, a unique pathogen on citrus fruits, requires a complex medium for rapid germination and vigorous hyphal growth. Citrus fruits contain both ascorbic acid and terpenes known to stimulate germination of *P. digitatum* spores (French *et al.* 1978; Pelser and Eckert 1977), as well as a complex of organic nutrients which sustains vigorous growth through the infection process.

Many fruits and vegetables are more susceptible to invasion by pathogens when the tissues are turgid, attributed usually to the pressure of a water film associated with injuries. Slight desiccation, for example, reduces the susceptibility of citrus fruits to *Penicillium digitatum*. In contrast, wilted carrots are more susceptible to *Botrytis* and *Rhizopus* spp. than more turgid roots, the susceptibility increasing when water loss from the tissues exceeds 8%.

Inhibitors of microbial growth. There are two types of inhibitors: preformed compounds and inhibitors synthesized by the host in response to attempted infection or other injuries. Examples of preformed compounds are the tannins and 3,4-dehydroxybenzaldehyde found in bananas (Greene and Morales 1967; Mulvena *et al.* 1969). Their concentration decreases with ripening of the fruit, which is believed to play a role in their increased susceptibility during this period. Antimicrobial compounds synthesized by plant tissue following injury are common, and have been reported in carrots, potatoes and apples in response to fungal infection.

Increase in susceptibility of the host with ripeness. Three factors may be involved in the observed increase in susceptibility of fruits and vegetables to disease during storage. These are (1) a decrease in ability of the host tissue to synthesize microbial inhibitors, such as 6-methyxymellein and benzoic acid, with age of the product in storage, (2) an increase in membrane permeability resulting in the release of nutrients and water into the intercellular spaces, and (3) increase in the susceptibility of the plant cell wall to attack by macerating enzymes of the pathogen.

### Environment

The best postharvest environment for maintenance of fresh fruit and vegetables is that which, firstly, maintains the product in the optimum condition for consumption, and secondly, prevents invasion by microorganisms. Often the same environment satisfies both requirements; sometimes it does not, and some compromise must be made between these conflicting demands.

Temperature. Storage at optimum temperatures for the maintenance of quality in fresh fruits and vegetables is the most important loss-reducing factor in the postharvest environment. The facilities for maintaining proper temperatures during all phases of marketing are sometimes referred to as the cold chain. In some countries the cold

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chain is well developed, although not always used to full advantage, but in others, however, few of the links in the chain exist, and this accounts for the large postharvest losses in such countries (Parpia 1976).

Although many fresh fruits and vegetables, such as apples, oranges, carrots and grapes, keep best at temperatures slightly above freezing, others are sensitive to chilling and suffer physiological disorders if they are not held at moderate temperatures. Many fruits typically of tropical origin, e.g. bananas and mangoes, are damaged if stored below 7.5°C. Vegetables such as green beans, cucumbers, eggplants, certain melons and squash, sweet peppers and tomatoes, are also sensitive to cold and generally must be stored above 7.5°C (Lutz and Hardenburg 1968).

The response of decay-causing organisms to temperature must be considered in efforts to reduce postharvest losses. Lowtemperature storage is the most effective and practical means for delaying development of decay in fruits and vegetables with deep seated infections that cannot be eradicated by other postharvest treatments. The attack on the product by most microorganisms becomes very slow as the temperature is decreased below 5°C. The guiding principle of low temperature storage, therefore, should be to maintain the temperature as low as possible without injuring the commodity.

With rare exceptions, low temperatures do not have a permanent effect upon the pathogen cells; their development is attenuated until later in the postharvest period when the temperature is again suitable for their growth. An organism like *Geotrichum* spp., with a relatively high temperature optimum, can be inhibited almost indefinitely at 0°C, but will rapidly develop disease when the temperature of the product rises. Organisms like *Botrytis*, *Penicillium* and *Sclerotinia* spp. grow slowly at near-freezing temperatures and limit storage life of the product if some other treatment is not used to control them.

Low temperature delays the development of postharvest diseases firstly by inhibiting host ripening, thus prolonging the disease resistance associated with immaturity, and secondly by direct inhibition of the pathogen with a temperature unfavourable to its growth. Each fruit and vegetable variety has an optimum storage temperature that will maintain the desired quality of the product for the maximum time (Burton 1973). *Relative humidity.* The control of relative humidity in the postharvest environment is often as important as the control of temperature. In some situations the effects of the two factors are difficult to separate because the capacity of air to hold moisture varies with temperature.

The relative humidity in the postharvest environment not only affects moisture loss from fruits or vegetables, but also the activity of decay-causing agents. Very high relative humidities generally favour the growth of decay organisms, but do not necessarily result in increased decay losses in fruits and vegetables. In potatoes, for example, high relative humidity promotes curing and healing by suberization, which reduces invasion by microorganisms (Artschwager 1927), while research on the high-humidity storage of Brussels sprouts, cabbage, celery, Chinese cabbage, and leeks also showed that at a low temperature less decay developed at 98–100% relative humidity than at 95% or lower (Lentz and van den Berg 1971). With onions, however, decay increased with higher relative humidity.

At 12°C, germination of the conidia of Botrytis cinerea on the surface of grape berries proceeds rapidly, and the amount of infection increases at relative humidities of 85–95%, with nearly 100% infection above this range, while below 85% the percentage of infected berries is greatly reduced (Nelson 1951).

The effect of humidity on decay is closely coupled with the effects of temperature, and for many commodities relative humidities near saturation result in lower decay only if the temperature is near 0°C (Kurki 1971).

Controlled or modified atmospheres. Whilst good cold-storage practices are vital in reducing postharvest losses, they may be supplemented with controlled (CA) or modified (MA) atmospheres. The inhibitory effects of controlled atmospheres on respiration rates have been well documented (Dewey 1977; Morris *et al.* 1971). The lengthened storage life of apples for example, made possible by cold storage and a controlled atmosphere, allows this fruit to be marketed all the year round.

Certain atmospheres also inhibit the activity of decay organisms. Strawberries shipped by air are much above their optimum holding temperature of 0°C for the major part of the journey. If however, an atmosphere containing 20% carbon dioxide can be maintained within the pallet loads of berries, decay losses caused by *Botrytis cinerea* can be reduced by half when average transit temperatures are in excess of 10°C (Harvey and Harris 1976). Rot of apples caused by *Gloeosporium* spp. increases in atmospheres in which the level of oxygen is reduced to 15 or 7.5%, but 5% carbon dioxide reduces decay levels to about half that of an uncontrolled atmosphere (Lockhart 1969).

Since susceptibility to disease increases with ripeness, it is not surprising that modifying an atmosphere by the addition of ethylene to accelerate ripening increases *Alternaria* rot in tomatoes (Segall *et al.* 1974), and *Diplodia* stem-end rot and anthracnose (*Colletotrichum* spp.) on citrus fruits (Brown and Barmore 1977; McCornack 1972; Smoot *et al.* 1971).

Removal from ambient atmospheres of low concentrations of ethylene formed by ripening fruit or fungi has reduced decay in lemons in modified storage, presumably by maintaining the fruit in an immature, more resistant condition (Wild *et al.* 1976).

### Control of postharvest diseases

'Control' of postharvest diseases has several meanings, which vary with prevailing marketing practices for a given commodity. Fruits such as citrus, apples and grapes have a relatively long life after harvest and the desired control is a reduction in the number of diseased units during the storage life of these fruits. With apples and citrus the main aim of postharvest treatment is to prevent primary infection, whereas with grapes the goal is to prevent the spread of *Botrytis cinerea* from field-infected berries. Strawberries which are infected in the field with *Botrytis* spp., have a relatively short shelf-life and retarding the development of disease in infected berries for several days constitutes satisfactory 'control'. A desired 'control' also for citrus fruits is to prevent sporulation of Penicillium digitatum on the fruit surface, so as to reduce soiling of adjacent fruits.

Several strategies for the control of postharvest diseases are feasible:

- Prevent infection of the product in the field before harvest.
- Reduce the level of pathogen inoculum in the postharvest environment to minimize infection of injured fruit (sanitation).
- Maintain the resistance of the plant product to infection by low temperature storage

and modified atmospheres.

Prevent, eradicate, or attenuate the development of the microorganism in the host by postharvest treatment.

The principles underlying strategies of preventing preharvest infection and reducing the level of pathogen inoculum in the postharvest environment are basic to spoilage control and most of the opportunities in these areas seem to have been exploited. However, major advances in the control of postharvest diseases in the future should be realized through the manipulation of the postharvest environment to maintain the resistance of the product to infection by low temperature storage and modified atmospheres, and by postharvest treatments to prevent, eradicate or attenuate the development of the organism.

### Control of environment

The temperature, relative humidity and gas composition of the atmosphere, are environmental factors influencing



Fig. 3. Nectarines being cooled at 0.5 °C to remove field heat.

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postharvest losses that are well within the capabilities of man to control, and thus provide a set of conditions best suited to each commodity. According to Sommer (1978), temperature management remains the keystone of postharvest pathology and physiology. For perishable commodities the cold chain is often considered the most vital segment in the marketing sequence. Despite many years of experience with refrigerated storage and transport, the full potential for temperature management is seldom reached. All too commonly the removal of field heat from the produce is incomplete, delayed or too slow, so that temperatures are well in excess of the minimum tolerated by the host. A better appreciation is required of the lethal effects of cold on germinating spores or lesions of fungi whose lower limit for growth is above 5°C. In addition, handling injuries tend to lose their susceptibility to infection when fruits and vegetables are cool stored, due possibly to the involvement of stress metabolites and desiccation of the wound. Heat, at the other end of the scale, can also control disease effectively by inactivating deep-seated infections of certain diseases that cannot be eradicated by other postharvest treatments, such as those in deep wounds or latent infections lying quiescent under the surface.

### Chemical treatments

Eckert (1978) believes that major advances in disease control will be in the application of chemical treatments to:

Protect the surface of the product from subsequent infection.

Prevent infection through harvest injuries.

Eradicate or attenuate established infections or deep-seated inoculations in lenticels, stomates and latent infections.

Retard senescence of the plant product, thereby retaining the resistance associated with immaturity.

### (i) Preharvest treatments

Preharvest application of chemicals was shown by several investigators to reduce postharvest decay to a substantial degree. *Rhizopus* rot of peaches after harvest may be significantly reduced by spraying the fruit one week before harvest with dichloran (Luepschen *et al.* 1971; Ogawa *et al.* 1971). Orchard sprays with the 2-substituted benzimidazole fungicide, benomyl, reduced the incidence of brown rot on peaches after



**Fig. 4.** Flood application of fungicide to oranges after the washing operation.

harvest, but had little effect upon *Rhizopus* rot (Kable 1971; Ogawa *et al.* 1968). A similar response has been recorded when oranges sprayed with 300–1000 mg L<sup>-1</sup> benomyl 30 days before harvest showed substantially less *Penicillium* mould two weeks after harvest than non-sprayed fruit (Brown and Albrigo 1972). The persistent activity of the benomyl is due to its systemicity.

As a means of preventing the infection of harvest injuries, the application of fungicides in the field is less desirable than treating the fruit after harvest for two reasons. Firstly only a small portion of the fungicide applied in the field is bound to the harvested product where it can protect subsequent injuries, and secondly the deposit required for postharvest disease control may be removed by washing. Preharvest treatment may be justified where substantial harvest injury is expected or where the crop cannot be treated promptly after harvest. Mechanical harvesting invariably results in a substantial increase in decay starting from superficial injuries, and preharvest sprays may be a desirable

approach to this problem.

Care must be exercised in choosing a preharvest fungicide since there is a high risk that residues will exercise selection pressure on the pathogen, permit the build-up of fungicide-resistant strains and nullify the benefits of a postharvest treatment with the same fungicide. Some investigators have questioned the wisdom of applying preharvest sprays to control postharvest diseases because of this potential problem.

### (ii) Postharvest treatments

Prevention of infection through harvest injuries. Injuries incurred by fruit during harvest and handling are the major sites of infection by wound pathogens. Mechanical harvesting substantially increases such infections. Chemical treatments to control these infections should be applied as soon as possible after harvest. However, in some cases a delay of several hours may significantly increase the effectiveness of the treatment because germinating spores are more sensitive than dormant spores (Eckert 1977). In practice there will always be a few hours between harvest and treatment, so the primary goal is to apply the treatment before the pathogen penetrates too deeply into the host tissue. The maximum period between harvest (or inoculation) and successful treatment varies; 10 hours for peaches inoculated with Rhizopus spp. and held at 25°C, 24 hours for oranges inoculated with green mould (*Penicillium digitatum*) and held at 24°C. In contrast, with potatoes the maximum time may extend to two weeks for control of Phoma spp. by treatment with gaseous sec-butylamine (Graham et al. 1973).

The principal factors influencing the length of this period are temperature, growth rate of the pathogen, resistance of the host tissue, and penetration of the chemical into the host tissue.

Fungicides applied to prevent infection at harvest fall into two categories. The first comprises the water-insoluble compounds such as thiabendazole and benomyl which are applied to produce a uniform coverage of the chemical on the surface of the host. Water-soluble salts such as *sec*-butylamine and sodium orthophenylphenate form the second group. These are applied in a fairly concentrated solution in water. The injuries absorb the ionic solution, but the intact cuticle is impermeable. Thus, lightly rinsing the fruit with water after the treatment, removes the toxic chemical from the surface but leaves a significant residue in the injury sites.

Treatments to eradicate or attenuate established infections. Latent infections can be eradicated in many cases by the application of a systemic fungicide after harvest because the fungus pathogen has been restricted to the epidermal or lenticel region by the resistance of the host, and so the chemical is able to penetrate into these outer layers and inhibit fungal development.

Treatment of fruits and vegetables with heated water at about 50°C has also eradicated established infections which were uncontrollable by chemical agents, but care has to be exercised because the dosages of heat required for disease control are very close to the injury threshold of the host. Some interest has developed in combining heat and fungicides, the temperature being borderline for disease control but well below the injury threshold. The elevated temperature increases the effectiveness of a lower dose of the fungicide dosage.

Maintenance of host resistance by retarding senescence. In addition to lower temperatures and modified atmosphere storage, certain chemical growth regulators can retard ripening and senescence, with significant reductions in postharvest disease. Prominent among these is 2,4-dichlorophenoxyacetic acid (2,4-D), which is used to treat fruit before storage to delay senescence of the button, the usual point of attack by Alternaria citri. Gibberellic acid (GA) may reduce *Penicillium* decay of navel oranges by delaying senescence of the peel, an event which facilitates infection by wound-invading pathogens. Since low temperatures inhibit physiological processes which lead to deterioration in quality as well as senescence of fruits and vegetables, there seems little justification for research on chemical agents to retard senescence of commodities which can safely be stored at temperatures below 5°C.

### Some significant fungicides

A detailed treatment of the useful postharvest fungicides is not possible within the scope of this paper, but reference should be made to some of them because they identify with significant stages in postharvest pathology and disease control.



**Fig. 5.** Application of a water emulsion of wax containing the plant growth regulator 2,4-D to oranges to minimize water loss and preserve a fresh appearance.

Dichloran was developed in the 1960s and provided, for the first time, a highly effective treatment for the control of *Rhizopus* rot of peaches, nectarines and sweet cherries, and *Botrytis* rot of several vegetable crops during long-term cold storage.

sec-Butylamine, developed in the same period, afforded a means of treating citrus fruits immediately after harvest to prevent the infection of injuries by *Penicillium* spp.

Solutions of sodium orthophenylphenate have been used extensively since the late 1950s for control of postharvest diseases of citrus fruits, apples, pears, sweet potatoes and other perishable fruits and vegetables.

The 2-substituted benzimidazole group, which includes thiabendazole, benomyl and thiophanatemethyl, has been extensively evaluated over the past decade, and proved dramatic in its effectiveness. The chemicals have solved the problem of stem-end rot and crown rot of bananas created by the practice of shipping bananas as hands and single fruits rather than as bunches. They have been used widely for the control of citrus green and blue mould (*Penicillium digitatum* and *P. italicum*) and brown rot (*Monilinia*) *fructicola*) in stone fruit. These compounds also provided a breakthrough in postharvest pathology, in that they demonstrated for the first time that infections in the host tissues at harvest could be inactivated by chemical treatments applied to the surface of the product after harvest. Stem-end rot of citrus fruit, lenticel rots of apples, latent infections of anthracnose on bananas and other tropical fruit, and latent infection of *Monilinia* spp. on peaches, have all been controlled by benomyl to an extent not thought possible in earlier times.

However, the euphoria associated with the numerous successful applications of the benzimidazole fungicides to problems of postharvest pathology was dampened with the realization by the mid-1970s that pathogenic fungi could develop resistance to this class of fungicides with comparative ease. The problem calls for the strategic use of these materials, in conjunction with sequential application of fungicides with different mechanisms of action in multistage fruit-handling operations.

### Challenges of postharvest disease control

Advanced technologies in the production, harvesting, packaging, storage, transport, distribution, and retailing of fresh fruits and vegetables can provide consumers with a relatively constant supply of these products throughout the year. The marketing chain from farm to consumer is a complex linkage of segments, each of which is dynamic and changing as it responds to economic, social, political or other pressures. Researchers engaged in the maintenance of quality and prevention of losses must be alert to these changes, because of the ultimate biological effects of physical alterations in one or more links in the chain.

The fresh fruit and vegetable industry has entered an era of dynamic innovation in the areas of mechanical harvesting, consumer packaging, and bulk handling and transportation. Most of the proposals that have been made in these areas will tend to intensify postharvest disease problems either by increasing the opportunity for wound infections or by creating an environment favourable for disease development.

An understanding of the mechanism of the development of fungal resistance, and the production of fungicides with a greater immunity to the development of resistance are needed. Another challenge is that despite substantial progress over the past 15 years in the development of highly effective fungicide treatments, there is as yet no chemical treatment which acts directly against several important postharvest diseases.

Postharvest pathology and wastage control in fresh fruit and vegetables, in Australia at least, is a relatively young science, but, together with research into handling foodstuffs, has made substantial progress. However, with changing technologies, there will be many new challenges to be met.

### **Further reading**

Hall, E. G., and Scott, K. J. (1977). Storage and market diseases of fruit. CSIRO Food Res. Q. (Collected supplements 1–24): 52 pp. 45 refs.

Hall, E. G. (1979). Handling and storing fresh fruit and vegetables in the home. CSIRO Food Res. Q. 39, 56-67.

### References

- Artschwager, E. (1927). Wound periderm formation in the potato as affected by temperature and humidity. J. Agric. Res. (Washington, D.C.) 17, 137-52.
- Boyd, A. E. W. (1972). Potato storage diseases. *Rev. Plant Pathol.* 51, 297–321.
- Boyd, M. R., and Wilson, B. J. (1972). Isolation and characterization of 4-ipomeanol, a lung-toxic furanoterpenoid produced by sweet potato (*Ipomoea* batatas). J. Agric. Food Chem. 20, 428-30.
- Brown, G. E., and Albrigo, L. G. (1972). Grove application of benomyl and its persistence in orange fruit. *Phytopathology* **62**, 1434-8.
- Brown, G. E., and Barmore, C. R. (1977). The effect of



Fig. 6. Trials with benomyl and sodium

orthophenylphenate (SOPP) using oranges inoculated with benomyl-resistant and benomyl-susceptible strains of green mould. ethylene on susceptibility of Robinson tangerines to anthracnose. *Phylopathology* 67, 120-3.

- Buchanan, J. R., Sommer, N. F., Fortlage, R. J., Maxie, E. C., Mitchell, F. G., and Hsieh, D. P. H. (1974). Patulin from *Penicillium expansum* in stone fruits and pears. *J. Am. Soc. Hortic. Sci.* 99, 262–5.
- Burton, W. G. (1973). Environmental requirements in store as determined by potential deterioration. In 'Proc. 7th British Insecticide Fungicide Conf.' Vol. 3, pp. 1037–55.
- Coursey, D. G., and Booth, R. H. (1972). The postharvest phytopathology of perishable tropical produce. *Rev. Plant Pathol.* 51, 751–65.
- Dewey, D. H. (1977). Controlled atmospheres for the storage and transport of perishable agricultural commodities. Mich. State Univ. Hortic. Rep. No. 28.
- Eckert, J. W. (1975). Postharvest diseases of fresh fruits and vegetables — etiology and control. In 'Postharvest Biology and Handling of Fruits and Vegetables'. eds.
  N. F. Haard and D. K. Salunkhe. (Avi Publ. Co.: Westport, CT.)
- Eckert, J. W. (1977). Control of postharvest diseases. *In* 'Antifungal Compounds'. eds. M. R. Siegel and H. D. Sisler. (Marcel Dekker: New York.)
- Eckert, J. W. (1978). Pathological diseases of fresh fruits and vegetables. *In* 'Postharvest Biological Biotechnology' eds. H. O. Hultin and M. Milner. pp. 161–209. (Food and Nutrition Press, Westport, CT.)
- French, R. C., Long, R. K., Latterell, F. M., Graham, C. L., Smoot, J. J., and Shaw, P. E. (1978). Effect of nonanal, citral and citrus oils on germination of conidia of *Penicillium digitatum* and *P. italicum*. *Phytopathology* 68, 877–882.
- Graham, D., Hamilton, G. A., Quinn, C. E., and Ruthven, A. D. (1973). Use of 2-aminobutane as a fumigant for control of gangrene, skin spot and silver scurf diseases of potato tubers. *Potato Res.* 16, 109-25.
- Greene, G. L., and Morales, C. (1967). Tannins as the cause of latency in anthracnose infections of tropical fruits. (*Gloeosporium musarum* in bananas). *Turrialba* 17, 447-9.
- Harper, K. A., Beattie, B. B., Pitt, J. I., and Best, D. J. (1972). Texture changes in canned apricots following infection of the fresh fruit with *Rhizopus stolonifer*. *J. Sci. Food. Agric.* 23, 311-20.
- Harvey, J. M., and Harris, C. M. (1976). Temperature maintenance in air shipments of strawberries to Far Eastern Markets. Bull. Inst. Int. Refrig. Annexe 1976-7, pp. 559-65.
- Kable, P. F. (1971). Significance of short term latent infection in control of brown rot in peach fruits. *Phytopathol. Z.* 70, 173–6.
- Kurki, L. (1971). Moisture in vegetable storage. Acta Hortic. 20, 146-51.
- Lentz, C. P., and van den Berg, L. (1971). Study of factors affecting temperature, relative humidity, and

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moisture loss in fresh fruit and vegetable storages. Cand. Inst. Food Sci. Technol. J. 4, 146-53.

Lockhart, C. L. (1969). Effect of CA storage on storage rot pathogens. Mich. State. Univ. Hortic. Rep. No. 9, pp. 113–21.

Luepschen, N. S., Rohrbach, K. G., Jones, A. C., and Peters, C. L. (1971). Methods of controlling *Rhizopus* decay and maintaining Colorado peach quality. Colorado State Univ. Exp. Stn Bull. No. 547S.

Lund, B. M. (1971). Bacterial spoilage of vegetables and certain fruits. J. Appl. Bacteriol. 34, 9-20.

Lutz, J. M., and Hardenburg, R. E. (1968). The commercial storage of fruits, vegetables, and florist and nursery stocks. U.S. Dep. Agric. Handbook No. 66.

Morris, L. L., Claypool, L. L., and Murr, D. P. (1971). 'Modified Atmospheres -- an Indexed Reference List Through 1969, with Emphasis on Horticultural Commodities'. (Univ. Calif. Press: Berkeley, CA.)

Mulvena, D., Webb, E. C., and Zerner, B. (1969). 3,4-Dihydroxybenzaldehyde, a fungistatic substance from green Cavendish bananas. *Phytochemistry* 8, 393-5.

McCornack, A. A. (1972). Effect of ethylene degreening on decay of Florida citrus fruit. *Proc. Fla State Hortic.* Soc. 84, 270-2.

Nelson, K. E. (1951). Effect of humidity on infection of table grapes by *Botrytis cinerea*. *Phytopathology* 41, 859–64.

Ogawa, J. M., Manji, B. T., and Bose, E. (1968). Efficacy of fungicide 1991 in reducing fruit rot of stone fruits. *Plant Dis. Rep.* 52, 722–6.

Ogawa, J. M., Leonard, S., Manji, B. T., Bose, E., and Moore, C. J. (1971). *Monilinia* and *Rhizopus* decay control during controlled ripening of freestone peaches for canning. *J. Food Sci.* 36, 331-4.

Parpia, H. A. B. (1976). Postharvest losses — impact of their prevention on food supplies, nutrition, and development. In 'Nutrition and Agricultural Development — Significance and Potential for the Tropics' eds. N. S. Scrimshaw and M. Behar. pp. 195-206. (Plenum; New York.)

Parsons, C. S., and Spalding, D. H. (1972). Influence of a controlled atmosphere, temperature and ripeness on bacterial soft rot of tomatoes. J. Am. Soc. Hortic. Sci. 97, 297-9.

Peacock, B. C. (1973). Effect of Colletotrichum musae infection on the preclimacteric life of bananas. Queensl. J. Agric. Anim. Sci. 30, 239–49.

Pelser, P. du T., and Eckert, J. W. (1977). Constituents of orange juice that stimulate the germination of conidia of *Penicillium digitatum*. *Phytopathology* 67, 747-54.

Segall, R. H., Geraldson, C. M., and Everett, P. H. (1974). The effects of cultural and postharvest practices on postharvest decay and ripening of two tomato cultivars. *Proc. Fla State Hortic. Soc.* 86, 246-9.

Smoot, J. J., Melvin, C. F., and Jahn, O. L. (1971). Decay of degreened oranges and tangerines as affected by time of washing and fungicide application. *Plant Dis. Rep.* 55, 149–52.

Sommer, N. F., Buchanan, J. R., and Fortlage, R. J. (1974). Production of patulin by *Penicillium expansum*. *Appl. Microbiol.* 28, 589–93.

Sommer, N. F. (1978). Trends and counter-trends in postharvest disease control. Abstracts of the XXth International Horticultural Congress, Sydney.

U.S. Dep. Agric. (1965). Losses in agriculture. U.S. Dep. Agric. Agric. Handb. No. 291.

Wiant, J. S., and Bratley, C. O. (1948). Spoilage of fresh fruits and vegetables in rail shipments unloaded at New York City, U.S. Dep. Agric. Circ. No. 773.

Wild, B. L., McGlasson, W. B., and Lee, T. H. (1976). Effect of reduced ethylene levels in storage atmospheres on lemon keeping quality. *HortScience* 11, 114-5.

Wright, T. R., and Smith, E. (1954). Relation of bruising and other factors to blue mold decay of Delicious apples. U.S. Dep. Agric. Circ. No. 935.

Yoder, O. C., and Whalen, M. L. (1975). Variation in susceptibility of stored cabbage tissue to infection by *Botrytis cinerea. Can. J. Bot.* 53, 1972-7.





# Changing philosophies in obtaining safe food — international aspects\*

By J. H. B. Christian

CSIRO Division of Food Research, North Ryde, N.S.W.

There are some differences, and of course some similarities, in the problems confronted when attempting to ensure the safety of foods in international trade compared with those encountered in domestic markets.

Problems of major importance when considering imported foods appear to be: Uncertainty about the hygienic conditions prevailing during manufacture in another country,

- Opportunities for mishandling during transportation, e.g. contamination, and undesirable changes in temperature and water activity  $(a_w)$  that may permit microbial growth, and
- The cost of the microbiological monitoring of imported foods.

What are the solutions to such problems? The following have been suggested, and sometimes adopted, by national and international agencies.

(1) An importing country may examine any imported food on any basis it wishes and decide, arbitrarily, on its acceptability. This method, presently employed by some countries, is unacceptable for routine use because different authorities in the one country may give different rulings and because the exporter has no known microbiological criterion at which to aim.

(2) National microbiological criteria can be applied at import. In this case, the organisms of concern in a food, methods of analysis, acceptable limits and sampling plans are all defined. The exporter knows what is expected of his product and mishandling during transportation may be detected. However, the scale of testing by authorities at import that is necessary to give an adequate assurance of safety may be overwhelmingly expensive and an exporter may be obliged to produce to a range of different specifications for different markets.

\*A paper presented at the 2nd Australian Food Microbiology Conference, Sydney, July 1979. (3) International microbiological specifications may be established. These require agreement between countries on all the components of a specification, which is not readily obtained, but should substantially facilitate international trade. The cost of testing remains high.

(4) International codes of practice may be devised, together with microbiological criteria to test whether the hygienic provisions of the codes have been adhered to. These should reduce to a more acceptable level the extent of testing required at import.

(5) A refinement of the latter process is for an importer to lay down a standard, and a code of practice, which is applied by the processor. The importer then accepts with a minimum of spot checking all product certified by the exporter as meeting the standard. Again, an exporter serving several markets may find it necessary to produce to several standards.

At present, virtually all testing for safety is performed at the national level using the approaches of (1) and (2) above. Exceptions are a number of bilateral agreements described in (5). Some combination of (4) and (5) probably offers the best solution.

Two basic steps in the development at the international level of present philosophies in obtaining safe foods were taken by the International Commission on Microbiological Specifications for Foods (ICMSF). Recognizing that uniform methodology was a prerequisite to international specifications, ICMSF published in 1968 a compilation of commonly used methods for the isolation and enumeration of microorganisms in foods. A revised and enlarged edition has since appeared (ICMSF 1978). On the basis of these methods, ICMSF commenced an international comparative testing program, aimed at establishing a set of standard methods. Testing programs were also set up by Sub-Committees of the International Standards Organization and by groups of

EEC countries, and the standardizing activities of the Association of Official Analytical Chemists in the USA became involved also. These endeavours continue and substantial progress has been made, but microbiological methodology is far from static and comparative testing of innovations will always be necessary.

The ICMSF also recognized that standard methods were not enough – a reliable microbiological criterion was not feasible without an appropriate sampling plan. A second book (ICMSF 1974) describes what different sampling plans can achieve, provides data on the probability of acceptance and rejection for numerous sampling plans and suggests sampling plans for some 70 food commodities. The principle that stringency of examination should be related to severity of hazard to be anticipated under normal conditions of use is incorporated in the recommendations. The use of 3-class attributes plans in which the quality of the product in terms of microbial criteria can be divided into the three classes: wholly acceptable, marginally acceptable or defective, was a very important innovation. This recognizes the heterogeneous distribution of microorganisms in foods and the imperfections of techniques for enumerating microorganisms. Variables sampling plans, which show great promise for use within a food plant, are unlikely to be appropriate for foods in international trade. They require a knowledge of the type of frequency distribution of organisms and this will be rare in imported foods.

While great interest was shown in many countries in standardized methods and in ICMSF sampling plans, the body concerned with international food law, the Codex Alimentarius Commission, was not ready, and indeed lacked the mechanism, to receive them. This situation changed in 1972 with Recommendations of the UN Conference on the Human Environment that increased support be given to the Codex Alimentarius Commission to develop international standards for pollutants in foods. Part of the report referred to the setting up of ad hoc Joint FAO/WHO Expert Consultations on Food Microbiology. These were to assist the Commission in establishing internationally accepted methodology for sampling and analysis and to initiate action towards the development of microbiological standards for foods.

The Codex Committee on Food Hygiene was already involved in the preparation of codes of hygienic practice for the production and handling of foods for which no Commodity Committees existed and for approving the hygienic provision in Codes elaborated by Commodity Committees. All of these Codes had in common a lack of microbiological end-product specifications. It was the task of the Consultations to recommend such specifications, and also to draft specifications for foods for which, although no Code of Hygienic Practice existed, there was an evident need in respect of food safety in international trade. Specifications for egg products were dealt with by the 1975 Consultation. Recommendations on frozen precooked shrimps and prawns and on frozen raw froglegs were made by the Second Consultation in 1977, which also considered drafts prepared by other Codex Committees relating to Foods for Infants and Children and Ice Mixes and Edible Ices. The specifications for egg products have already been appended to the Code of Hygienic Practice which is at the final step, Step 9, of the Codex acceptance procedure.

All of the specifications recommended to the Codex so far have included drafts of methods where already agreed methods did not exist. However, it has been accepted that as ISO methods become available they will be inserted into the specifications.

The next development resulted from a request from Sub-Committee 9 of ISO/TC34 that the Second Consultation set out guiding principles for the establishment and application of microbiological specifications for foods. The resulting draft was well received by the Codex Committee on Food Hygiene and government comments were referred back to the microbiologists for further consideration. Unfortunately, the UNEP funds used to support the Consultations had run out. While the Codex Alimentarius Commission agreed in principle to the setting up of a Joint FAO/WHO Expert Committee on Microbiological Specifications for Foods, similar to Expert Committees advising on Additives and Pesticides, this was not feasible at the time. The Commission suggested that, as an interim measure, experts be invited to participate in a working group to convene in Geneva in early 1979 at the expense of governments.

This meeting has now been held. The working group considered further the General Principles for the Establishment of Microbiological Specifications for Foods and this should come before the Codex Committee on Food Hygiene in July, 1979.

The working group had another brief and this I suggest will have an important bearing on the philosophy of obtaining safe foods – to consider the need or otherwise for microbiological criteria for raw foods, taking as examples raw meat and poultry. The conclusion reached was that for both of these products there was no way in which the application of a Code of Practice based on acceptable processing procedures could ensure the absence of pathogens if the animal from which the product was derived was itself contaminated. No government appeared willing to accept the presence of any salmonella in a food, so that criteria permitting c > 0 for salmonella were unacceptable where c is the maximum allowable number of sample units yielding unsatisfactory test results. At the same time c = 0 appeared unobtainable. Hence, microbiological criteria for these foods could not serve any purpose, unless some acceptable antimicrobial treatment could be included in the processing.

What influence this recommendation will have on governments is, of course, not known. However, if it prompts authorities to rethink their attitudes to (1) terminal pasteurization-type processes for raw meats and poultry, and (2) the maximum levels of salmonellae that can be tolerated in raw food, the outcome could be of great interest.

I have dealt mainly with philosophies and developments in the United Nations agencies and subsidiaries. However, the dairy industry, from which much of food microbiology sprang, has also worked in this area for a long period through the International Dairy Federation. Efforts have accelerated over the last three years and recommendations have recently been made of microbiological specifications for dried milk and for ice mixes and edible ices.

The IDF has espoused the concept of 3-class plans and has recommended criteria for dried milk which are, in respect of total plate count and coliform count, very close to those recommended by the NHMRC in Australia. Beyond this point, however, we run into a philosophical confrontation. IDF has decided that Standards, which are mandatory, shall not include limits for pathogenic organisms, although criteria attached to Codes of Practice, which are advisory, may include them. The Codex Food Hygiene Committee's working group, in its general principles, has concluded the virtual opposite, that Standards should consist of limits for pathogens but that any appropriate microorganisms might be utilized in establishing other criteria.

Another problem that remains relates to definitions of the various types of microbiological criteria. Only the 'Standard' is clear cut. Both 'Specifications' and 'Guidelines' may be defined differently by different bodies, e.g. ICMSF, Codex working groups and national regulatory authorities. This should be resolvable, but has in recent times caused increasing confusion.

So much for history. It is probably true to say that the philosophy of providing safe foods has not changed much in the five years since ICMSF published its philosophies. However, the opportunities to implement these philosophies have certainly improved as a consequence of a steady chain of developments. From placing the entire burden of ensuring safe food on microbiological examination at import, the emphasis has been forced back to the conditions of manufacture by the establishment of Codes of Hygienic Practices which in turn become more effective as appropriate end-product specifications are devised and attached to them.

For many products a high level of safety can now be ensured by application of recommended Codes, end-product specifications and import standards. A major challenge now is to increase efficiency and reduce costs by the introduction of simplified and more rapid microbiological methods and the establishment of a system whereby the manufacturers' reports on the microbiological status of their products can and will be accepted with confidence by importers and authorities in importing countries.

### References

ICMSF (1974). Microorganisms in Foods. 2. Sampling for microbiological analysis: principles and specific applications. (University of Toronto Press: Toronto.)

ICMSF (1978). Microorganisms in Foods. 1. Their significance and methods of enumeration. 2nd Ed. (University of Toronto Press: Toronto.)



# Dried grapes — the involvement of lipids in their production

### By D. Barnett

CSIRO Division of Food Research, North Ryde, N.S.W.

### Introduction

For several thousand years grapes have been preserved by drying and the practice probably arose from chance observations. Requirements for stable food supplies in the early Middle East civilizations led, by Roman times, to a considerable trade in dried fruit. Trade in dried grapes was initially impeded by their naturally slow drying characteristics. However, it was discovered more than 2000 years ago that the drying rate could be increased by treating the fruit with a mixture of wood ashes and olive oil. The Romans adopted this method, which was described by Lucea Junius Moderatus Culumella in A.D. 60. A variation of the method is still used in a number of grape growing countries, including Australia. The use of drying oils to reduce drying time, and consequently the risk of rain damage to grapes, is particularly advantageous in Australia where the possibility of rain in the grape growing areas during the autumn drying season is high. Grapes are generally dried to a moisture content of about 16%.

The total world output of dried grapes is about 900 000 t. Australian production ranges from 50 000 to 85 000 t; in 1977 it was 64 467 t (Australian Dried Fruits Control Board 1978). The currants, raisins and sultanas to which these data pertain, are produced in Australia by the following methods. Currants are prepared from the small Zante grape variety and are dried on shaded racks without any pre-treatment. Raisins are produced from the large-berried Gordo Blanco (Syn-Muscat of Alexandria) and Waltham Cross varieties. These grapes may be dipped in hot sodium hydroxide solution before drying on shaded wire racks. They may also be treated with the alkaline oil emulsion used for sultanas (varieties, Sultanina, Thompson Seedless and

Kishmish). This emulsion consists of drying oil (20 g  $L^{-1}$ ) emulsified in aqueous potassium carbonate (25 g  $L^{-1}$ ). In the industry it is commonly called dipping oil because, initially, it was applied to picked bunches by dipping them in a tank containing the mixture. Today, however, it is frequently sprayed on bunches after they have been spread on drying racks (Fig. 1). Oil treatment reduces the time required for drying on shaded racks from about 20 days to 8–10 days (Grncarevic and Radler 1967). A more recent procedure called trellis drying is gaining popularity in Australia; this entails severing the fruiting vine canes, spraying the attached bunches with emulsion and then leaving them to dry on the trellis. The dried berries are then harvested mechanically.

The dried grapes are delivered to packing sheds where they are cleaned, washed and graded. Raisin seeds may be removed by expression, leaving perforations in the skin which cause the fruit to become sticky. Before packaging, sultanas and deseeded raisins are treated with a dressing oil which reduces stickiness and clumping and also gives an attractive gloss to the surface of the fruit (Dried Fruit Processing Committee 1973).

In this review the nature of the grape drying process is considered together with the changes in drying behaviour induced by constituents of the dipping oil. The quality changes brought about by this treatment are outlined. The role of dressing oils, which may have both deleterious and beneficial effects is also discussed.

### Grape cuticular wax

Grapes have a continuous cutin membrane covering the epidermis and this noncellular support carries a fine film of cuticular wax. The wax imparts a characteristic bloom and, because of its hydrophobic nature, provides







an effective barrier against excessive water loss from the berry. Chambers and Possingham (1963) investigated the ultrastructure of the wax by electron microscopy and showed that it consisted of a series of overlapping platelets, each about 0.1  $\mu$ m wide. A more recent scanning electron micrograph (Fig. 2) shows the platelets of sultana surface wax.

Chemical investigation of sultana wax (Radler and Horn 1965) revealed two separable fractions, 'hard' wax (soluble in chloroform) and 'soft' wax (soluble in petroleum ether). These waxes had widely different melting points. It was found that the former consisted mainly of the triterpene oleanolic acid. This material, which was subsequently shown to be ineffective as a water barrier (Radler 1965), constitutes some 70% of the cuticular wax of which there is about 100  $\mu$ g cm<sup>-2</sup> on the grape surface. The 'soft' wax was separated into its constituents by Radler and Horn (1965) and was shown to consist of long chain alcohols, aldehydes, esters, free acids and hydrocarbons. It is this mixture which provides the water barrier. This has been verified by model experiments (Grncarevic and Radler 1967) and selective removal of the 'soft' wax from grapes with petroleum ether vapour (Radler 1965). The cuticular membrane which underlies the wax has no importance as a water barrier. This was demonstrated by the similar drying rates of peeled grapes and those from which the wax had been removed by solvent treatment (Radler 1965). Other results from the same study indicated that drying rate increased with increasing removal of 'soft' wax from grape skins. The 'soft' wax probably prevents water loss from the grape berry by acting as a strongly hydrophobic layer that repels water.

### **Drying oils**

Oils are applied to grapes to shorten the outdoor drying period and thus minimize the risk of fruit being damaged by rain. The



Fig. 1. Spraying grapes with drying emulsion.

increased drying rate also produces a sultana of lighter colour.

The dipping or spraying solutions consist of an emulsion of a mixture of ethyl esters of fatty acids, some free fatty acids and small amounts of sulphonated compounds in alkaline solution. The esters and free acids are derived from tallow. The ethyl esters and associated fatty acids account for about 90% of the oil and have been shown by several investigators (Martin and Stott 1957; Grncarevic 1963; Radler 1964; Ponting and McBean 1970) to be the active material in increasing drying rate. Radler (1964) measured the effect of various solvents, oils, esters, acids and alcohols added to aqueous potassium carbonate (25 g L<sup>-1</sup>), on the drying rate of grapes. Ethyl oleate, stearate and caprylate were found to be most effective. Oleic acid, butyl or amyl oleate and some solvents were also moderately effective. Ponting and McBean (1970) investigated the effect of carbon chain length of the ethyl esters and found that  $C_{14}$ - $C_{18}$  gave the highest relative drying rates.

The exact mechanism whereby the hydrophobic cuticular wax becomes relatively permeable to water has been the subject of much conjecture. A surface effect theory, which is discussed later, was proposed by Chambers and Possingham (1963).

### Role of potassium carbonate

The standard dip or spray solution contains potassium carbonate at a concentration of 25 g L<sup>-1</sup>, which gives a pH of about 11.0. The high pH prevents the



**Fig. 2.** Scanning electron micrograph of untreated grape cuticle wax (× 10 000).

growth of microorganisms in the dip tanks and aids in producing a stable emulsion of the oil in water. Use of potassium carbonate enables the dipping oil to exert a maximal effect on drying rate; however it has been demonstrated that ethyl oleate (the major constituent of dipping oil) alone produces considerable increase in the rate of water loss from grapes (Ponting and McBean 1970).

Use of potassium carbonate solutions, alone, as a drying aid have given various and conflicting results. Grncarevic (1963) found that grapes dipped in potassium carbonate solutions of 20–50 g L<sup>-1</sup> dried about twice as quickly as untreated grapes. Radler (1965), and Ponting and McBean (1970), were unable to repeat this result when they applied potassium carbonate solutions to grapes.

Sodium carbonate is much cheaper than the potassium salt but it is not as effective in increasing drying rate as the latter (Barnett, unpublished data). The reason for this is not clear but it is of interest that Ponting and McBean (1970) found a mixture of oleic acid and sodium carbonate to be almost ineffective in increasing grape drying rate whereas Radler (1964) using oleic acid with potassium carbonate obtained a substantial effect. Commercial drying oils contain substantial amounts of free fatty acids which form soaps with the potassium carbonate. These soaps may facilitate the penetration of esters into the waxy cuticle of the grapes. The differing effects of sodium and potassium carbonates may be due to the physical properties of the soaps formed, potassium soaps being 'softer' and more readily soluble than those of sodium.

### Mechanism of water loss from grapes

Grapes do not have detectable stomata so water loss from the fruit is entirely through the cuticule. Martin and Stott (1957) showed that the wax layer of the Sultana grape berry is a highly effective barrier to the passage of water. The effect of drying oil is to increase the water permeability of this wax layer. It was shown that little or no wax was removed by the dipping treatment (Dudman and Grncarevic 1962) and it was concluded that the wax was modified in some nondestructive way. Electron microscopy of carbon replicas carried out with grape skins before and after drying and dipping (Chambers and Possingham 1963) showed that the wax platelet structure was essentially





Fig. 3. Left, untreated grape with wax bloom evident. Right, grape treated in dipping oil showing wet appearance.

unaltered by the dipping process. The platelets were seen to be pressed more closely to the surface of the dried grapes but they still retained their gross structure. Dudman (1962) and Grncarevic (1963), carried out washing experiments which indicated that treatment with a drying emulsion does not permanently alter the rate of water loss provided that washing of treated grapes occurs within 24 hours of dipping. This led Dudman (1962) to propose that the application of oil had brought about critical surface changes, perhaps rendering the wax more hydrophilic. This was also suggested by the macroscopic observation of the apparently 'wet' appearance of grapes which persists up to 24 hours after dipping (see Fig. 3). Chambers and Possingham (1963) explained the various observations by proposing that dipping emulsions accelerate water loss by eliminating the slow process of vapour diffusion through the very small spaces between wax platelets. The emulsion fills the minute spaces and at the same time converts the normally hydrophobic wax surface to a hydrophilic state (Fig. 4). This theory has been supported by model

experiments in which solvent extracted sultana wax was coated onto a cellulose acetate support and shown to behave similarly to the natural grape cuticle with respect to dipping and washing (Grncarevic *et al.* 1968). Oleanolic acid, which contains hydrophilic hydroxyl and carboxyl groups, did not suppress water evaporation when tested similarly (Grncarevic and Radler 1967). The surface effect theory has been restated (Possingham 1972) in the general context of the dissipation of water from plant cuticular surfaces.

There are a number of criticisms which can be levelled at the above hypothesis:

- Ethyl oleate or similar ester functionalities are neither strongly hydrophilic nor surface active.
- Free fatty acids might be expected to give excellent results by creating hydrophilic surfaces but generally they are not as effective as the esters.
- Surfactants which would be expected to impart increased hydrophilic character to the wax surface failed to increase drying rates above that of untreated controls (Saravacos and Charm 1962; Petrucci *et al.*)





**Fig. 4.** Scanning electron micrograph of emulsion treated grape surface 6 hours after dipping (× 10 000).

1974; Barnett, unpublished data). Reversibility (and 'wet' appearance) of the dipping effect is only observed for about 24 hours after treatment. Drying rate enhancement is still observed when the grape ceases to have a wet appearance and the drying oil has possibly penetrated into the wax.

The precise reason for increased water transmission through cuticular wax of grapes following the application of drying oil remains uncertain despite long usage of the process and extensive research.

### Other effects of dipping oils

### Colour

Grapes which are dried without the application of oil emulsion (naturals) take up to 28 days to reach commercial dryness and also undergo considerable darkening during that time. This darkening is thought to be due to the enzyme polyphenol oxidase present in the grape skin. Faster drying, such as that resulting from the use of drying oil, appears to limit enzyme darkening and produces the familiar light golden Australian sultana (Fig. 5). Studies by Radler (1964) suggested that inhibition of darkening was the result of rapidly increasing sugar concentration. This view is supported by the drying behaviour of a mutant of the Thompson seedless grape, Bruce's Sport, which yields light coloured sultanas in the absence of oil treatment and does not contain polyphenol oxidase but does have adequate substrate phenols.

### Flavour

The use of fatty acid esters to assist in the production of dried grapes introduces the possibility of off-flavours in the fruit. Using pure ethyl and methyl oleate as additives to Thompson Seedless raisins, Guadagni et al. (1975) found that 50% of a taste panel were able to detect the esters at levels of about 200 p.p.m. Amounts of this order may be present on sultanas offered for sale (Barnett 1978) but such fruit is not normally rejected because of off-flavour. Guadagni and Stafford (1979) found that the emulsifiers (sulphonated alcohols etc.) used in commercial oleate preparations can be the most significant source of off-flavour in dried vine fruits. The determination of thresholds for the ester residues is important so that residual levels may be kept as low as practicable even though the taste may not be objectionable to most people.

### **Dressing oils**

Dried grapes are processed to remove grit, stems, cap-stems and damaged fruit. During this processing the fruit is washed and, in the case of most raisins, deseeded. These operations result in skin damage to the fruit which causes stickiness and leads to reduced shelf life. Before final packing, therefore, a dressing oil is sprayed onto the fruit. This is either paraffin oil or a stabilized vegetable oil and the treatment leaves a residue of 0.2–0.3% by weight of oil on the fruit (Dried Fruits Processing Committee 1973).

The reasons for using a dressing oil have been summarized by Goldenberg (1976) as follows:

- unsightly crystallization of sugars on the surface is avoided because oiling reduces drying out of the fruit
- packaging and handling are made more convenient since the dressing oil prevents aggregation of fruit into clumps and gives 'free-running' properties
- ▶ some insect infestation is prevented
   ▶ shelf-life is extended.

The oil should also impart an attractive gloss to the fruit. Paraffin oil has traditionally been used on dried vine fruit but in recent years its use has been discouraged by a number of importing countries. This has led to increasing use of hydrogenated vegetable oils as alternatives. These oils have two major disadvantages





according to Goldenberg (1976). Firstly, their higher melting point leads to difficulties in coating dried grapes, particularly during winter months. This property can also cause fruit to clump together and give a dull appearance in place of the desired gloss. Secondly, there is the strong possibility of the vegetable oils developing off-flavours due to oxidative rancidity. Purified liquid paraffin oil is not subject to these problems and was cleared of any suspicion of toxicological hazard in a recent report (Food Additives and Contaminants Committee 1975). Nevertheless, doubts persist in some countries and research is required to find more acceptable dressing oils than those presently in use.

### Analysis of grape surface lipids

A detailed analysis of grape cuticle wax was made by Radler and Horn (1965). In general it resembles other epicuticular plant waxes (Baker and Martin 1963). Radler and Grncarevic (1964) used thin-layer chromatography (TLC) for the rapid estimation of added paraffin oil on dried grapes. A qualitative study by means of TLC of lipid additives present on a number of foods including grapes was made by Ristow (1968). Drying oil esters on grapes have been determined by gas chromatography (Stafford *et al.* 1974).

Barnett (1978) used TLC, column chromatography and infrared spectroscopy to study the residues arising from the application of lipids to grapes during drying and processing. He showed that ethyl esters are subject to appreciable hydrolysis on the grape surface during drying. Lipid residues can be partially removed by washing; 30-40% of paraffin oil residues were removed from sultanas by a simple water wash (Goldenberg 1976). Up to 40% of drying oil residues (both free fatty acids and esters) can also be removed during washing in the packing house (Barnett 1978).

The ethyl esters of drying oil and the triacylglycerols of the presently used dressing oils are derived from edible fats and oils and consequently have received little attention from regulatory authorities. It is important, nevertheless, that levels of these substances should be kept as low as possible, compatible with good processing practice. This will avoid or reduce the development of off-flavours and minimise public concern about uncontrolled food additives.

### Conclusion

Modification of the waxy cuticle of grapes by lipid treatment in order to accelerate drying has a long history, but a precise explanation of its effect with grapes and some other fruits remains doubtful. Interestingly, a study on the use of drying oil and potassium carbonate was made with lucerne hay (Tullberg and Angus 1972). Potassium carbonate alone increased the drying rate of lucerne thereby allowing more rapid and efficient hay making (Anon 1978). Hence the modification of cuticular waxes to increase the rate of loss of water may well be applicable to a wide range of primary products. For example, it might possibly be utilized to produce a non-toxic herbicidal spray. The splitting of nearly mature fruits following heavy rains might be prevented by increasing water loss from the fruit using an





Fig. 5. Sultanas. Left, dried with emulsion treatment; right, dried with no treatment (naturals).



ester or similar spraying treatment.

World concern about fossil fuel resources is growing and one consequence is the exploration of food processes which are more fuel efficient. In this context solar drying is obviously a highly attractive process and any additional treatment, such as the use of drying oils, which can enhance its efficiency, merits further attention.

The need for a non-toxic, stable and effective dressing oil for dried vine fruits remains unsatisfied.

### References

- Anon (1978). Rural Res. No. 100, 26-28.
- Australian Dried Fruits Control Board (1978). 54th Annual Report.
- Baker, E. A., and Martin, J. T. (1963). Nature (London), 199, 1268-70.
- Barnett, D. (1978). Food Technol. Aust. 30, 498-502.
- Chambers, T. C., and Possingham, J. V. (1963). Aust. J. Biol. Sci. 16, 818-25.
- Dried Fruits Processing Committee (1973), Grape drying in Australia, CSIRO, Melbourne.
- Dudman, W. F. (1962). Aust. J. Sci. 25, 168-9.
- Dudman, W. F., and Grncarevic, M. (1962). J. Sci. Food Agric. 13, 221-4.
- Food Additives and Contaminants Committee Report (1975). The Mineral Hydrocarbon in Food

Regulations 1966. Min. Agric. Fish. Food (HMSO: London).

- Goldenberg, N. (1976). Chem. Ind. 21, 956-7.
- Grncarevic, M. (1963). Am. J. Enol. Vitic. 14, 230-4.
- Grncarevic, M., and Radler, F. (1967). Planta 75, 23-7.
- Grncarevic, M., Radler, F., and Possingham, J. V. (1968). Am. J. Enol. Vitic. 19, 27-9.
- Guadagni, D. E., and Stafford, A. E. (1979). J. Food Sci. 44, 782–4.
- Guadagni, D. E., Stafford, A. E., and Fuller, G. (1975). J. Food Sci. 40, 780-3.
- Martin, R. J., and Stott, G. L. (1957). Aust. J. Agric. Res. 8, 444.
- Petrucci, V., Canata, N., Bolin, H. R., Fuller, G., and Stafford, A. E. (1974). J. Am. Oil Chem. Soc. 51, 77–80.
- Ponting, J. D., and McBean, D. McG. (1970). Food Technol. 24, 85-8.
- Possingham, J. V. (1972). Ann. Bot. (London) 36, 993-6.
- Radler, F. (1964). J. Sci. Food Agric. 15, 864-8.
- Radler, F. (1965). Nature (London) 207, 1002-3.
- Radler, F., and Grncarevic, M. (1964). J. Agric. Food Chem. 12, 266-7.
- Radler, F., and Horn, D. H. S. (1965). Aust. J. Chem. 18, 1059-69.
- Ristow, R. (1968). Dt. Lebensmitt. Rdsch. 64, 322.
- Saravacos, E. D., and Charm, S. E. (1962). Food Technol. 16, 91.
- Stafford, A. E., Fuller, G., Bolin, H. R., and Mackey, B. E. (1974). J. Agric. Food Chem. 22, 478–9.
- Tullberg, J. N., and Angus, D. E. (1972). J. Aust. Inst. Agric. Sci. 38, 314-15.

### News from the Division

### Appointments

Mr I. B. Powell, B.Sc. (Hons) joined DRL as an Experimental Officer, to assist in a program aimed at developing bacteriophageresistant strains of streptococci for the Australian cheese industry.

Mrs D. J. Freeman has been appointed as an Experimental Officer to assist Dr F. B. Whitfield, FRL, in a research program designed to identify the compounds responsible for characteristic flavours in seafoods. Mrs Freeman holds degrees in organic chemistry and in pharmacy. The project is funded by the Fishing Industry Research Fund.

### General



Three staff members of DFR are on extended visits overseas. Dr D. Graham, Leader of FRL's Plant Physiology Group, is currently working in the Botany School of the University of Cambridge, and will later move to the Agricultural Research Council's Weed Research Organization at Oxford. He will return in July 1980. His main interest is in enzymes and metabolic regulation in relation to chilling injury in plants.

Dr D. G. Oakenfull, FRL, is in the Chemistry Department of the University of Kent at Canterbury until February 1980, studying the kinetics of fast reactions in experiments on bile salt micelles.

Dr R. J. Pearce, MRL, is in the U.S.A. for collaborative research on the functionality of proteins, in the Department of Food Science and Nutrition, Ohio State University, U.S.A. and will return in August 1980.

### **Higher degrees**

Mrs K. H. Adams of FRL's Liaison Section has been awarded the degree of Master of Environmental Studies by Macquarie University.

Miss P. L. Conway, Food Safety and Nutritional Quality Group and Mr G. W. Francis, Food Structure Group, received Master of Science degrees from the University of Queensland and Macquarie University, respectively.

### Awards

The Australian Journal of Dairy Technology Award has been conferred on Mr M. J. Coventry of the University of Melbourne, Dr A. J. Hillier and Dr G. R. Jago of DRL for their article entitled 'The metabolism of pyruvate and citrate in the thermoduric cheese starter *Streptococcus faecium (Strep. durans)*'.

The Australian Society of Dairy Technology Silver Medal/Loftus Hills Dairy Science Award was presented to Mr Ron Hill for published work on research into milk proteins. This has been a significant contribution to the understanding of the physical and chemical properties of milk proteins and how they behave in modern manufacturing processes.



### V. Stekly

### Obituary

We announce with regret the death on 10 December 1979 of Vladimir Stekly, always known to his friends as Peter, who was Senior



Technical Officer at the CSIRO Food Research Laboratory, North Ryde, NSW.

Peter was born in Czechoslovakia in 1926, and in 1948 was awarded a Diploma in Horticulture, Food and Wine Technology in Prague. With his wife he came to Australia in 1952 and in 1955 secured a Diploma in Food Technology from Hawkesbury Agricultural College. In 1956 he joined Edgells at Bathurst as Quality Control Officer with specific responsibilities for baby foods, and in 1960 he moved to the CSIRO Division of Food Research where he undertook supervision of the food processing pilot plant, first at the Homebush laboratory and later at the new laboratories at North Ryde.

In 1967 Peter acted as Manager of a Government Food Processing Laboratory set up at Alafua, Western Samoa, with assistance from Australia through the South Pacific Technical Assistance Plan.

Most recently Peter Stekly was attached to the Food Technology Liaison Group of the Food Research Laboratory where his wide experience and great familiarity with food processing operations were of immense value in answering inquiries from the food industry. In the special area of canning technology Peter has regularly lectured to Inspectors of Foodstuffs in the Australian Army and to specialist courses at Hawkesbury Agricultural College. An important recent activity was an in-plant training program for retort operators which involved Peter in visits to 20 canneries in order to ensure that they complied with requirements for the safe processing of canned foods for export.