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Diseases and pests

Pests

Many pests attack fruit and vegetables in the field that can be carried through to the postharvest period. They are mainly insects, but other organisms may be involved including nematodes. In white yams, Thompson *et al.* (1973) found infection with the parasitic nematode *Pratylenchus* sp., probably *P. coffeae* (Loof.), in the field, which could cause tissue just below the skin to become necrotic. They showed that this necrosis could increase during ambient storage, because the nematodes increase in numbers. Storage at 13 °C resulted in a rapid decrease in nematode numbers in the tubers and thus prevented spoilage. When bananas were packed into export cartons in the field, there was occasional infestation of the boxed fruit by insects, mainly cockroaches and ants. Where this was a problem, it was controlled by fumigation with the broad-spectrum insecticide permethrin, usually during ripening in the importing country (Thompson and Burden 1995). Black widow spiders (*Latrodectus hesperus* Chamberlin & Ivie) have been found in grapes exported from California. Yong-Biao *et al.* (2008) exposed Thompson Seedless and Flame Seedless to different O₂ levels at different temperatures. They found complete control of the spiders after ultra low O₂ at 0.5% O₂ or lower at 1 °C and also at 2% O₂ at 15 °C both for 24 h. In 2% O₂, as temperature was increased from 1 to 15 °C, spider mortality increased from 0

to 100%. Two types of thrips attack banana fruit: the flower thrips (*Frankliniella* spp., *Thrips florum*) and red rust thrips (*Chaetanaphothrips* spp.). Flower thrips lay their eggs just below the surface of the skin of fruit that are less than 2 weeks old. The result is a small raised pimple, which can be felt on the surface particularly between the fingers. Rust thrips actually feed on the surface of the newly developing fruit usually between the fingers, especially where they are touching. Their feeding action results in light yellow patches that later turn a reddish brown colour, but at this stage, there are few, if any, rust thrips remaining. Yong-Biao (2011) investigated postharvest control of western flower thrips (*Frankliniella occidentalis*) in lettuce. They vacuum cooled seven cultivars of Iceberg lettuce and stored them at 2 °C for up to 5 days, then exposed them to ultra low O₂ (0.003% O₂) at 10 °C for 2 days. Lettuce stored for 2 days followed by 2 days ULO treatment was effective in controlling western flower thrips. Three of seven cultivars sustained injury to heart leaves by the ULO treatment but this was minimal. Lettuce that had been stored for 2, 3 or 5 days before the ULO treatment tolerated the ULO treatment with no significant quality reduction compared with untreated controls. Magnetic resonance imaging has been used to obtain images of insect damage in pears (Chen *et al.* 1989).

The most important insects that attack fruit are the fruit flies as their presence not only affects the palatability of the fruit but can affect their international

marketability. These are dealt with under different fruit and vegetable sections of this book. Insect infestation has resulted in extensive legislation from many countries to control importing of fruit containing fruit flies. Fruit flies of major concern particularly in fruit exported to countries such as Japan, USA and Australia are (Vijaysegaran 1993):

- *Anastrepha* – South America, Central America and the West Indies.
- *Ceratitidis* – Africa, but has spread throughout the world except Asia.
- *Rhagoletis* – South America, Central America, North America and Europe.
- *Dacus* – mostly Africa.
- *Bactocera* – Asia, Australia and South Pacific.

Diseases

The genesis of postharvest diseases is usually in the field and can be influenced by the environment in which the crop is grown. For example, high calcium levels have been associated with reduction in postharvest disease levels, and a reduction in the nitrogen: calcium ratio has been strongly correlated with increased resistance of apple fruit to postharvest disease (Sugar *et al.* 1992). Other preharvest variables, including fruit position on the plant, growing season and water deficit, have marked effects on the postharvest characteristics and susceptibilities of fruit and vegetables (Sugar *et al.* 1992).

Postharvest diseases of fruit and vegetables are caused by infections with fungi and bacteria. Virus infections can cause diseases, which manifest themselves postharvest, but these are rare. Fungi and bacteria can exist as either parasites or saprophytes. The former feed from living matter and the latter from dead organic matter. Parasites can be obligate or facultative. Obligate parasites can only survive on living hosts, generally only attack a limited range of hosts and are very difficult or impossible to grow on artificial culture media. Facultative parasites can adapt to live on dead organic matter, often have a wide host range and can easily be cultured on artificial media. Most fungi require an acid (pH 2.5–6) environment in which to grow and develop, whereas bacteria thrive best in neutral conditions and only a few species can grow at levels below pH 4.5. Bacteria therefore do not

usually infect fruit, which are acidic. The classification of fungi are in constant review, but generally they may be unicellular or multicellular in the case of yeasts, but most fungi are made up of hyphae that are multicellular filamentous thread-like structures, which can form a dense fluffy mass, called mycelium. Many of the fungi that cause postharvest disease belong to the phylum Ascomycota and the associated fungi Anamorphici. In Ascomycota, the asexual stage of fungus, called the anamorph, is usually more frequently found in postharvest diseases than the sexual stage called the teleomorph. More comprehensive references are Snowdon (1990), Coates *et al.* (1995), Agrios (1997) and Waller (2001).

There are two basic types of diseases in plants. Non-parasitic diseases that are also called physiological disorders where there is no other organism involved and are caused by internal physiology, climatic conditions or soil conditions. The second are diseases associated with infection by organisms. These can be divided into saprophytic diseases. Parasitic diseases are also called pathogenic disease and affect the host plant by absorbing food from the host cells thus weakening the plant, secreting toxins, enzymes and growth substances, which can damage or kill the plant cells or by blocking the translocation of food and water. Some pathogenic microorganisms are fungi, bacteria, nematodes, mycoplasmas, protozoa, viruses, viroids and prions. Infection results in symptoms, which can be used for diagnosis. Sometimes, different organisms can cause the same symptoms, and sometimes, the same organism may give different symptoms depending on the crop, environment, time in the growth cycle and site of infection. Symptoms can be localised affecting only the part of the plant that is infected, e.g. leaf, fruit. They can affect the whole plant where the organism infects one part of the plant and can then spread to all parts of the plant or where one part of the plant is infected, but the symptoms are shown in another part, e.g. the roots are infected and result in wilting of the leaves and stem, which are not actually infected.

In order to be certain exactly the cause of an infection, a diagnostic series of steps must be carried out. The most commonly used is called Koch's postulate where:

- Pathogen is isolated from diseased plant.
- Pathogen is cultured.

- Pathogen is inoculated into healthy plant of the same species and cultivar.
- The same disease symptoms are observed.
- Pathogen is isolated and is the same as in two above.

For infection to take place, there must be the presence of pathogen, presence of suitable host plant at an appropriate stage of crop growth and favourable weather conditions. The most common way of infection is through damaged tissue but many organisms can penetrate through living tissue.

Barriers to infection

Plants have natural defences to infection. Physical barriers include the cuticle and the periderm, both of which may also contain chemical defence mechanisms. The cuticle is a good barrier against bacteria and especially viruses. Many fungal pathogens exude enzymes, which attack the cuticle and epidermis. Stomata, lenticels and hydrothodes are natural breaks on plant surfaces where infection can occur. Chemical barriers may exist on or in the plant or may be produced as a result of infection. They are often phenols called phytoalexins. In some cases, toxins are always present, e.g. in the scales of coloured onion bulbs and terpenes, resins and latex in some plant tissue. Plant cells may also be able to detoxify chemicals given out by the fungi or bacteria.

Phytoalexins

These are toxic substances stimulated to be produced in substantial quantities in plants as a result of infection with pathogenic microorganisms, mechanical injury or chemical injury to the plant. Phytoalexins are produced by healthy cells adjacent to damaged or necrotic cells in response to chemicals produced by damaged cells. Most are toxic to and inhibit the growth of fungi and many are toxic to bacteria, nematodes and other organisms. Pisatin is a phytoalexin in pea and is toxic to humans. Pisatin has been found that the pods of peas which had been stored in an atmosphere containing ethylene. Peas stored with levels of 0.2–20 ppm ethylene produced pisatin levels of about three times those of pods stored where ethylene was eliminated. Production of phytoalexins is stimulated in the host by elicitors given out by fungi.

These elicitors are generally high molecular weight substances in the fungal cell walls.

Physiological barriers

Some plants cells die as a response to infection, which isolates the infection, especially in response to infection by obligate parasites. This is because it forms an area of dead material around the fungus. It is called hypersensitivity.

Morphological barriers

The morphology of the plant can affect infection, e.g. in the fungus *Claviceps purpurea* that causes Ergot in cereals. In Rye, the spores settle on the open flower and germinate, such as pollen, on the stigma and infect the flower that way. Barley is self-pollinated and modern cultivars keep their flowers closed almost all the time so they are not infected.

Epidemiology

Epidemiology is the study of factors affecting the outbreak and spread of infectious diseases. An epidemic is where a pathogen spreads to and infects many individuals over a large area in a relatively short time. Epidemics develop as a result of susceptible host plants, virulent pathogen, a favourable environment over a prolonged period and human activity, e.g. monocropping. Epidemics can be controlled to some degree by legislation, breeding disease resistant plants, environmental control, chemical fungicides, biological control and cultural control.

Legislation

There is a strong aversion in many consumers to treatment of fruit and vegetables with chemical pesticides. This has resulted in increasing legislation on their use, especially after harvest. Chemical pesticide residues in fruit and vegetables are restricted and controlled by legislation. There is a United Nations body set up to provide a scientific basis for legislation called the World Health Organization (WHO) Expert Group on Pesticide Residues with the Food and Agriculture Organization (FAO) Panel of Experts on Pesticide Residues (JMPPR). In Europe, the DG VI (Agriculture)

of the European Commission has the responsibility of harmonising chemical pesticide residues in food. With the introduction of Single European Market on 1 January 1993, a first list of 55 chemicals was produced that could be applied to foods. The list is constantly being updated and it contains the maximum residue levels (MRLs) for different chemicals on different crops. It provides directives for MRL, and these are communicated in directives issued in the Official Journal of the European Communities. Where no MRL exists for a certain crop and chemical, it is subject to a default level (LOD). European Union legislation is effectively removing most postharvest fungicides and insecticides from use on fruit and vegetables. This means that for a wide range of tropical crops, there are only a few permitted compounds and for some crops, none. For producers in developing countries, it is important to try every possible means of non-chemical protection to avoid problems on entry to the export markets.

COLEACP in Paris were given the task of developing an action programme to the European Commission. This has three main purposes:

- To set up a communication programme to raise awareness of the European Union harmonisation programme for pesticide regulations and to provide accurate, regularly updated and easily accessible information in a user-friendly format on MRL for tropical and minor crops produced in Asia, Caribbean and Pacific (ACP) countries and a database.
- Where MRL have been set by default at LOD, to search for available data on essential crop/chemical combinations in order to submit applications for the setting of practical MRL for GAP use in these crops. Where data are missing or inadequate, trials will be set up to establish the missing data.
- To set up mechanisms (and work with other organisations where possible) to build the capability of ACP exporters and exporter associations to develop sustainable systems of crop production, minimising pesticide use through integrated crop management and integrated pest management to comply with EU (and the USA) regulations, also to protect the health of farm workers and preserve the environment.

Control of pests and diseases can be greatly helped by appropriate legislation. Legislation can be used

to control and prevent plant material coming into a country that might be infected with pests or diseases. In other cases, material is heat treated or fumigated, but this are not permitted in many countries because of residues that may be harmful to consumers. An example of legislation being used in disease control is brown rot disease of potato, caused by the bacterium *Pseudomonas solanacearum*. It was first recorded in southern Europe in 1940 and was observed in Sweden in 1972 and in England in 1992. The largest outbreak occurred in the Netherlands in 1995, mainly through heavily infected seed tubers. To avoid introduction of this dangerous quarantine disease into other countries, the European Commission implemented the phytosanitary regulations on *P. solanacearum* in November 1995. Control measures governed by national and European Union plant health legislation rely on accurate detection and reporting of brown rot outbreaks and distribution of the pathogen, prevention of importation of potatoes from known infected areas and restrictions on use of infested land and irrigation water for potato production.

Other strategies for legislative control of plant diseases include:

- Responsibility for prevention and control is for the country where the disease occurs.
- Countries where the disease occurs should report outbreaks as soon as possible.
- Early and sure detection and diagnosis of the pathogen is essential.
- Adequate prevention and control measures should be taken after an outbreak.
- Production of healthy planting material (e.g. seed tubers) should be guaranteed and surveys and (laboratory) checks for (latent) infections performed.
- Education of scientific personnel, farmers, traders, extension service and the public is essential.

Mode of infection

Devising the correct method of control of a particular disease on a particular fruit or vegetable must take into account the identification of the organism involved and knowledge of the ecology and pathogenicity of the organism. Infection can occur before, during or after harvest, and development of

disease may be intimately associated with the physiological status of the fruit or vegetable (Coates *et al.* 1995).

Preharvest fungal infections

Conidia of *Botrytis cinerea* on the surface of necrotic strawberry flower parts germinate in the presence of moisture. The fungus colonises the necrotic tissue and then remains quiescent in the base of the floral receptacle. Several months later, when fruits are harvested, infections develop as a stem end rot in ripe fruit. Mango stem end rot caused by *Dothiorella dominicana* is one example of a postharvest disease arising from endophytic colonisation of fruit pedicel tissue. In this case, the fungus colonises the pedicel and stem end tissue of unripe fruit, where it remains quiescent until fruit ripening commences. Stem end rots of citrus caused by *Lasiodiplodia theobromae* and *Phomopsis citri* result from quiescent infections in the stem button of fruit. These infections can be initiated at any stage of fruit development and remain quiescent until the button begins to separate from the fruit during abscission. Examples of postharvest diseases that can arise from late season infections include brown rot of peach (caused by *Monilinia*), yeasty rot of tomato (caused by *Geotrichum*) and sclerotium rot of various vegetables (caused by *Sclerotium rolfsii*). Some pathogens infect through natural openings such as stomata, hydrothodes and lenticels (Coates *et al.* 1995).

Resistance to infection

Phytoalexins are only produced in response to pathogen invasion, although in some cases, they can be elicited by certain chemical and physical treatments. For example, non-ionising ultraviolet-C radiation is known to induce production of phytoalexins in various crops. UV treatment of carrot slices induces production of 6-methoxymellen, which is inhibitory to *Botrytis cinerea* and *Sclerotinia sclerotiorum* (Coates *et al.* 1995).

Natural antifungal compounds present in fruit tissue may be involved in regulating the quiescence of *Colletotrichum* infections. In avocados, antifungal dienes are present in the peel of unripe fruit at concentrations inhibitory to *Colletotrichum gloeosporioides*, the avocado anthracnose pathogen.

During ripening, levels of these dienes decline to sub-fungitoxic levels coincidentally with the development of anthracnose symptoms on fruit. Preharvest fungal infections of mangoes by *Glomerella cingulata*, the conidial stage *Colletotrichum gloeosporioides*, cause anthracnose disease. The spores of this organism may infect fruit, germinate and form appressoria that remain quiescent on the skin of the fruit until the fruit begins to ripen. The fungus then invades the cells of the fruit by hyphae growing from the appressorium resulting in the anthracnose disease. Fruit, especially those that are to be exported, are harvested at a stage of maturity where the fungus has not penetrated the fruit but is firmly fixed to its surface at the resistant appressorial stage. Therefore, fruits, which look perfectly healthy at harvest, may develop the disease symptoms postharvest. A similar effect was reported by Coates *et al.* (1993a), where spores of *Glomerella cingulata* have been shown to germinate on developing avocado fruit, form an appressoria and a short infection peg that penetrates about 1.5 µm into the skin. The fungus then remains quiescent until harvest, when antifungal dienes in the skin of avocado fruit break down due to degradation by lipoxygenase activity. The fungus then resumes growth and invades the avocado fruit to cause postharvest rots (Prusky *et al.* 1983, 1995). They also reported that it has been shown that breakdown of the dienes was delayed by controlled atmosphere storage, hypobaric storage and treatment with antioxidants.

Environment

A wide range of preharvest factors influence the development of postharvest disease. These include the weather (rainfall, temperature, etc.), production locality, choice of cultivar, cultural practices (pesticide application, fertilisation, irrigation, planting density, pruning, mulching, fruit bagging, etc.) and planting material. These factors may have a direct influence on the development of disease by reducing inoculum sources or by discouraging infection. Alternatively, they may affect the physiology of the produce in a way that impacts disease development after harvest. For example, the application of certain nutrients may improve the 'strength' of the fruit skin so that it is less susceptible to injury after harvest and therefore less prone to invasion by wound pathogens.

(Coates *et al.* 1995). In the field, planting crops in an environment that is less suitable to the microorganism can be used. Examples include growing mangoes where there is a strong prevailing wind can eliminate disease and good ventilation control in greenhouses can reduce disease.

Hygiene

In the case of postharvest diseases that arise from preharvest infections, practices that make the crop environment less favourable to pathogens will help reduce the amount of infection that occurs during the growing season. For example, in tree crops, pruning and skirting can increase ventilation within the tree canopy, making conditions less favourable for fungi and bacteria. Removal of dead branches and leaves entangled in the tree canopy is also an important way to minimise inoculum build-up. In many diseases, overhead irrigation can encourage pathogen spread and infection; trickle or micro-sprinkler irrigation systems may be more appropriate. As many pathogens are soil-borne, minimising contact of leaves and fruit with the soil is desirable. Inoculum for infections occurring after harvest commonly originates from the packing shed and storage environments. Water used for washing or cooling produce can become contaminated with pathogen propagules if not changed on a regular basis and if a disinfectant such as chlorine is not incorporated. Water temperature can also be an important factor in the transfer of inoculum in some situations. For example, tomatoes harvested during hot weather may have a higher temperature than the water used to wash them. In this scenario, inoculum present in the washing water can be taken in by the fruit tissue, causing higher levels of diseases such as bacterial soft rot. Rejected produce, which has not been discarded from the packing shed or storage environment, provides an ideal substrate for postharvest pathogens. 544 Packing and grading equipment, particularly brushes and rollers, which is not cleaned and disinfected on a regular basis, can also be a major source of inoculum. Containers used for storing and transporting fruit can harbour pathogen propagules, particularly if recycled a number of times without proper cleaning (Coates *et al.* 1995).

Harvest and handling fungal infections

In some diseases, infection occurs during and after harvest through the wound created by severing the fruit from the plant as in banana crown rot. Many common postharvest pathogens are unable to directly penetrate the host cuticle and infection can occur through mechanical injury during harvesting and handling as well as insect injuries.

Infection of lychee fruit by yeasts commonly occurs through insect injuries that are difficult to see at harvest. Some chemical treatments used after harvest, such as fumigants used in insect disinfestation and disinfectants such as chlorine, may also injure produce if applied incorrectly. Various types of physiological injury such as chilling and heat injury can predispose produce to infection by postharvest pathogens. For example, the incidence of alternaria rot in pawpaw, apple and various vegetable crops is increased by exposure to excessive cold (Coates *et al.* 1995).

Non-chemical methods of disease control

Breeding

Within a natural population of plants of the same species, there will be differences in the susceptibility to disease, which can be exploited in breeding and selection. Microorganisms can also mutate or, from natural variation, can infect resistant plants. Genetic modification (GM) has great potential for disease control and can be used by inserting a gene into a crop that gives it resistance to a particular disease. However, there is resistance to the method from the public of many countries. In September 2003, over 50 countries signed the Cartagena Protocol on biodiversity so that any country can refuse to import any genetic modified organism (USA refused to sign the Protocol)

Ionising radiation

Ionising radiation is another physical treatment that can be used after harvest to reduce disease in some commodities. Like heat, commodities must be able to tolerate the doses of ionising radiation required to achieve disease control. Some commodities are surprisingly tolerant. For example, strawberries can

Table 28 Effects of controlled atmosphere storage conditions on the decay levels of tomatoes harvested at the green mature stage (source: adapted from Parsons *et al.* 1974)

Storage atmosphere	After removal from 6 weeks at 13 °C (%)	Plus 1 week at 15–21 °C (%)	Plus 2 weeks at 15–21 °C (%)
Air (control)	65.6	93.3	98.6
0% CO ₂ + 3% O ₂	2.2	4.4	16.7
3% CO ₂ + 3% O ₂	3.3	5.6	12.2
5% CO ₂ + 3% O ₂	5.0	9.4	13.9

tolerate the doses of radiation required to effectively control grey mould. In other commodities, however, abnormal ripening, tissue softening and off-flavours can result from applying ionising radiation at doses lethal to the target pathogens. Poor consumer acceptability of food irradiation coupled with high treatment costs pose additional limitations to the widespread use of this technology at the present time (Coates *et al.* 1995).

Considerable interest exists in controlling postharvest diseases and pests of crops without using chemical fungicides. There are several alternative strategies, some of these are dealt with in the following.

Hot water treatment

Hot water dipping of mangoes to control anthracnose can result in increased levels of stem end rot (caused by *Dothiorella* spp.) (Coates *et al.* 1995).

Storage

The natural resistance of fruit and vegetables to disease declines with storage duration (Coates *et al.* 1995).

Ethylene

Exposure of parsnips to low levels of ethylene in cold storage caused bitterness that was probably due to accumulation of the phytoalexin xanthotoxin (8-methoxypsoralen) (Johnson *et al.* 1973).

Carbon dioxide

In a review of the fungistatic effects of high CO₂, Kader (1997) indicated that levels of 15–20% could retard decay incidence on cherry, blackberry, blueberry, raspberry, strawberry, fig and grape. In

celery stored at 8 °C, disease suppression was greatest in atmospheres of 7.5–30% CO₂ with 1.5% O₂, but there was only a slight reduction in 4–16% CO₂ with 1.5% O₂ or in 1.5–6% O₂ alone (Reyes 1988). Pretreatment of Italia grapes with 20% CO₂ for 48 h before storage reduced decay during subsequent storage at 0 °C and 90–95% r.h. for 30 days in both types of grape. It had a marked effect during the first 10 days of cold storage on organic grapes (Massignan *et al.* 1999). Summer Red nectarines tolerated air enriched with 15% CO₂ atmosphere for 16 days at 5 °C. Development of brown rot decay in fruits inoculated with *Monilinia fructicola* 24 h before storage was arrested. After 3 days' ripening in air at 20 °C, the progression of brown rot disease was rapid in all inoculated nectarines, demonstrating the fungistatic effect of CO₂ (Ahmadi *et al.* 1999). Eris and Akbudak (2001) found that peaches stored at 0 °C and 90% r.h. had lower levels of decay in 5% CO₂ and 2% O₂ than in air. Control of infections by the anthracnose and crown rot pathogen of bananas by exposure to 15% CO₂ or 1% O₂ resulted in a reduction in growth of the pathogen but not complete inhibition (Al Zaemey *et al.* 1994). Also that level of CO₂ is toxic to the fruits, so this does not appear to be an alternative to fungicides.

Oxygen

Reports on the effects of reduced O₂ levels in storage on the development of diseases of fruit and vegetables have shown mixed results, but some positive effects have been reported. Parsons *et al.* (1974) showed considerable reduction in disease levels on tomatoes in controlled atmosphere storage with most of the effect coming from the low O₂ levels with little additional effect from increased CO₂ levels (Table 28). Exposure of organically grown Ziv bananas to 2% O₂ atmosphere at 20 °C for either 48 and 72 h immediately after harvest was effective in reducing crown

rot decay after simulated transport, ethylene ripening and shelf life, but less so than 0.2% thiabendazole fungicide treatment (Pesis *et al.* 2001).

Curing

Many root crops have a cork layer over the surface that is called a periderm. This serves as a protection against microorganism infections and excessive water loss. This layer can be broken or damaged during harvesting and handling operations, so curing is essentially a wound healing operation to replace the damaged periderm. Sealing citrus fruits in plastic film bags and then exposing them to 34–36 °C for 3 days resulted in the inhibition of *Penicillium digitatum* infection and reduced decay and blemishes through lignification and an increase in antifungal chemicals in the peel of the fruit without deleterious effects on the fruit (Ben Yehoshua *et al.* 1989a, 1989b). In potatoes, there are some indications that the fatty acids may also act as phytoalexins and the peroxides and epoxides may have toxic effects on infectious agents (Galliard 1975).

Biological control

Microbial antagonists are applied either before or after harvest, but postharvest applications are more effective than preharvest applications. Mixed cultures of the microbial antagonists appear to provide better control of postharvest diseases over individual cultures or strains. Similarly, the efficacy of the microbial antagonist(s) can be enhanced if they are used with low doses of fungicides, salt additives and physical treatments such as hot water dips, irradiation with ultraviolet light, etc. (Sharma *et al.* 2009).

In most reported cases, pathogen inhibition is greater when the antagonist is applied before infection taking place. For this reason, control of quiescent field infections (e.g. *Colletotrichum gloeosporioides*) using postharvest applications of antagonists is often more difficult to achieve than control of infections occurring after harvest (e.g. *Penicillium* spp.). Unless an antagonist has eradicator activity or has some effect on host defence responses, field applications are often necessary to achieve control of quiescent infections. In South Africa, both field and postharvest applications of *Bacillus subtilis* and *B. licheniformis* suppressed anthracnose (caused by

Colletotrichum gloeosporioides) and stem end rot (caused by *Dothiorella* spp. and other fungi) development in avocado. A number of modes of action are involved in these pathogen-antagonist interactions, including site exclusion, nutrient and space competition and antibiotic production. In Australia however, only field applications of a non-antibiotic producing strain of *Bacillus* sp. have shown potential for the control of anthracnose in avocado. There are a number of reports in the literature concerning the biological control of wound pathogens in various fruit and vegetables. To be effective against wound pathogens, an antagonist must be able to successfully colonise wound sites to the exclusion of the pathogen. Microbial antagonists are being used including *Debaryomyces hansenii* Lodder & Krejer-van Rij, *Cryptococcus laurentii* Kufferath & Skinner, *Bacillus subtilis* (Ehrenberg) Cohn, and *Trichoderma harzianum* Rifai. Biocontrol products including Aspire, BioSave and Shemer have also been developed and registered (Sharma *et al.* 2009). Antagonists that act against postharvest pathogens by competitive inhibition at wound sites include the yeasts *Pichia guilliermondii*, *Cryptococcus laurentii* and *Candida* spp. (Coates *et al.* 1995). The bacteria *Streptomyces* and *Pseudomonas* can attack *Pythium* and other fungi.

Microbial antagonists

Microbial antagonists are used to control several postharvest diseases. Although the mechanism(s) by which microbial antagonists suppress the postharvest diseases is still unknown, competition for nutrients and space is most widely accepted mechanism of their action. In addition, production of antibiotics, direct parasitism and possibly induced resistance in the harvested commodity are other modes of their actions by which they suppress the activity of postharvest pathogens in fruit and vegetables (Sharma *et al.* 2009). Microbial antagonists are applied either before or after harvest, but postharvest applications are more effective than preharvest applications. Mixed cultures of the microbial antagonists appear to provide better control of postharvest diseases over individual cultures or strains. Similarly, the efficacy of the microbial antagonist(s) can be enhanced if they are used with low doses of fungicides, salt additives and

physical treatments such as hot water dips, irradiation with ultraviolet light, etc. At the international level, different microbial antagonists such as *Debaryomyces hansenii* Lodder & Krejer-van Rij, *Cryptococcus laurentii* Kufferath & Skinner, *Bacillus subtilis* (Ehrenberg) Cohn and *Trichoderma harzianum* Rifai are being used. Biocontrol products such as Aspire, BioSave, Shemer, etc., have also been developed and registered.

Yeasts

It has been known for many years that some microorganisms found on the surfaces of growing plants can attack potential fungal pathogens. Some yeasts have been shown to be effective. Aspire is a commercial formulation of the yeast *Candida oleophila* registered for postharvest application to citrus for the control of green mould (*Penicillium digitatum*). Its mode of action appears to be that it competes with the pathogen for nutrients released by injuries. The yeast *C. guilliermondii* was shown to be antagonistic towards *Penicillium* spp. (McGuire 1993) and could be successfully incorporated into the commercial citrus waxes FMC 705, FMC 214 and Nature Seal, where it was shown to survive for 2 months at 12 °C. In trials by Brown *et al.* (2000) with Hamlin and Valencia oranges, colonisation by *C. oleophila* of puncture-related injuries that either encompassed oil glands or individually ruptured glands was achieved within 1–2 days at 21 °C. Ruptured oil glands were colonised more effectively if treated 7 h after injury rather than immediately as peel oil was toxic to *C. oleophila* but not to *P. digitatum* spores. Colonisation of puncture injuries by the yeast was comparable after 2 days at 21 and 30 °C, but no colonisation occurred at 13 °C. Growth rates of the yeast were similar in waxed and non-waxed fruits. *C. oleophila* colonised punctures more uniformly than individually damaged oil glands, and provided more effective control of *P. digitatum* originating at punctures than at oil gland injuries. Incubating treated fruits at 30 °C for 2 days before storage at 21 °C enhanced the control of *P. digitatum*.

Arras and Arru (1999) also evaluated the fungitoxic activity of antagonist yeasts against postharvest citrus fruit diseases. Trials were first performed in the laboratory using the yeasts *Pichia guilliermondii* (strain 5A), *Candida oleophila* (strain 13L) and

Rhodotorula glutinis (strain 21A), and Aspire. *P. guilliermondii* 5A strongly inhibited *P. italicum* on artificially wounded Oroblanco (a hybrid between pummelo and grapefruit) fruits with Aspire coming second. More than 200 yeasts were selectively isolated from microbial populations on the surface of different fruits by Lima *et al.* (1998) and tested against various postharvest fruit pathogenic fungi. At 20 °C, the antagonists significantly reduced rot incidence and showed a wide range of activity on different host–pathogens combinations. Two of the yeasts grew in culture at temperatures ranging from 0 to 35 °C. In assays performed *in vitro* at 24 °C, the antagonists showed low sensitivity towards several fungicides common on fruit and vegetables. Antagonistic epiphytic yeasts were isolated from the surface of mandarin orange in Turkey and grapefruit in Israel by Kinay *et al.* (1998). Among the mandarin orange isolates, three were effective in inhibiting green mould. Among the grapefruit isolates, several yeasts showed high efficacy against green and blue mould and sour rot caused by *Geotrichum candidum*. At the wound site, the population of yeasts increased gradually and maintained a high concentration during 1 week of storage at 20 °C. Teixido *et al.* (1999) found that preharvest application of the antagonistic yeast *Candida sake* on Golden Delicious apples was less effective against *Penicillium* rot *Penicillium expansum* than postharvest treatment. No advantages in biocontrol were observed when cold-stored apples were treated with the yeast antagonist both preharvest and postharvest.

The inclusion of a *Bacillus* species or a rose coloured yeast isolated from lemon leaves in either carnauba or polyethylene wax on citrus fruits was not effective in postharvest disease control (Visintin *et al.* 1996).

Manso and Nunes (2012) isolated yeasts from the surface of Bravo de Esmolfe apples cultivated in North Portugal and found that the most effective in controlling diseases was *Metschnikowia andauensis*, strain NCYC 3728 (PBC-2). In semi-commercial trials in cold storage, the reduction of blue mould *Penicillium expansum* was 90%. Rapid colonisation of fresh apple fruit wounds was observed during the first 24 h of cold storage, followed by a significant population increase during the first 15 days of storage and then the population remained stable until the end of storage. The broad spectrum of action of *M. andauensis* PBC-2 was evaluated

with effective control being achieved against *Rhizopus stolonifer*, *Penicillium expansum* and *Botrytis cinerea*, on Rocha pears and on different apple cultivars and against *P. digitatum* and *P. italicum* on mandarins and oranges.

Other fungi

The yeast-like fungus, *Aureobasidium pullulans* (isolate LS-30), displayed antagonistic activity against *Botrytis cinerea*, *Penicillium expansum*, *Rhizopus stolonifer* and *Aspergillus niger* on Italia grapes, and *B. cinerea* and *P. expansum* on Royal Gala apples (Castoria *et al.* 2001). Yam tubers that were sprayed with a conidiospore suspension of *Trichoderma viride* in potato dextrose broth showed a large reduction in the range and number of microflora, including pathogens, on the tuber surface during 5 months storage in a traditional yam barn (Okigbo and Ikediugwu 2001).

Bacteria

Several types of bacteria that attack fungi have been isolated from plants. *Bacillus* spp. on their own or combined with a fungicide could be used to control postharvest diseases of mangoes, avocados and litchi. *B. subtilis* and *B. licheniformis* isolated from mango leaves and applied to fruit as a warm water dip controlled anthracnose levels. The effects were enhanced by the inclusion of either benomyl or prochloraz to the dip. *Bacillus* spp. isolated from leaves and fruit of avocados were more effective in controlling anthracnose and stem end rot of avocados when applied as a postharvest dip than prochloraz applied in the same way. The combination of *B. subtilis* and prochloraz was more effective than when they were applied separately. *B. stearothermophilus*, *B. megaterium* and *B. licheniformis* isolated from the phylloplane of litchi were more effective in reducing fruit browning and postharvest decay of litchi than benomyl when the treatments were applied as warm water dips. Good results have been found in the control of anthracnose in mangoes and avocados with *Bacillus subtilis*. While not giving complete control, it may have an application in reducing the levels of chemical fungicide application (Korsten *et al.* 1993a, 1993c). *In vitro* studies of *Bacillus stearothermophilus*, *B. megaterium* and *B. licheniformis* isolated from litchi trees

showed that they effectively inhibited growth in 11 postharvest pathogens of litchi fruit. Postharvest dips with these antagonists were shown to be more effective in controlling decay, postharvest infection and fruit browning than a warm water plus benomyl treatment (Korsten *et al.* 1993b).

Pseudomonas syringae strains ESC-10 and ESC-11 produce syringomycin and control green and blue moulds of citrus caused by *Penicillium digitatum* and *P. italicum*, respectively (Bull *et al.* 1998). Isolates of *Pseudomonas syringae* NSA-6 and MA-4 reduced brown rot *Monilinia fructicola* to 28 and 73%, respectively, from 98% in the inoculated control after 5 days incubation at 22 °C. Both isolates reduced rhizopus rot *Rhizopus stolonifer* to 5 and 8% from 53% in the inoculated control after 5 days incubation. Isolates MA-4 and NSA-6 suppressed brown rot from 63 to 30% and from 95 to 71–81%, respectively, after 3 and 4 days incubation at 22 °C. The use of 0.5% calcium chloride in the soak suspension significantly improved the activity of *P. syringae* but the use of 'peach wax' (Decco 282) increased brown rot incidence and negated the beneficial effect of calcium chloride (Zhou *et al.* 1999). Silimela and Korsten (2000) tested plastic caps with an added inner wool lining impregnated with copper oxychloride or the bacterial antagonist *Bacillus licheniformis* covering individual fruit. They found that they had little effect in controlling disease under field conditions when the disease pressure was low. *Pseudomonas* sp. isolates, associated with the skin of banana, inhibited germination and appressoria formation *in vitro* of conidia of *Colletotrichum musae* (de Costa and Subasinghe 1998). It also inhibited the growth of *Fusarium* spp., *Botryodiplodia* spp. and *Ceratocystis paradoxa* by 30–42%. Crown rot was significantly reduced using the antagonistic bacteria as a postharvest dip.

Bacillus thuringiensis has been used for controlling insects for many decades. Its postharvest use in controlling tuber moth (*Phthorimaea operculella*) in stored potatoes was described by Das *et al.* (1998).

Ultraviolet light

When air is passed through UV light, some of the oxygen molecules are broken down for an instant to atomic oxygen and ozone. This can have the effect of destroying pathogens when the fruit or vegetable is passed through the light. Stevens *et al.* (1997)

found that it appeared to be only partially effective in practice, and it was concluded that UV-C and yeast biological control agents could be used together as an alternative to chemical control of some storage diseases. Romanazzi *et al.* (2006) reduced grey mould incidence and severity. However, other workers have had more positive results in UV-C alone, reducing postharvest decay on strawberries (Nigro *et al.* 2000, Pombo *et al.* 2011) and mangoes (González-Aguilar *et al.* 2001) and bacterial counts on fresh-cut baby spinach leaves (Escalona *et al.* 2010), watermelon cubes (Artés-Hernández *et al.* 2010) and white mushrooms (Guan *et al.* 2012). UV-C irradiation has also been reported to have other effects. Jiang *et al.* (2010) reported that UV-C irradiation resulted in maintenance of firmness and enhanced antioxidant capacity in shiitake mushrooms. Kim *et al.* (2010) found that UV-C irradiation maintained the quality of strawberries. Kasim and Kasim (2012) found that it prevented yellowing in fresh-cut cress but increased electrolyte leakage due to tissue damage. Obande *et al.* (2012) describes a mobile UV-C unit that was conveyed between the rows of tomato plants in a commercial glasshouse and generally found that fruit treated while still on the plant softened and changed colour more slowly after harvest than those not treated.

Irradiation

Irradiation at 0.05–0.06 kGy may be used to inhibit sprouting and extend shelf life of fresh ginger (Wu and Yang 1994, Mukherjee *et al.* 1995). However, irradiation at these low levels decreased volatile content of fresh ginger, which was perceived by sensory analysis after 5 months in storage (Wu and Yang 1994). Irradiation of dried rhizomes at doses of 5–10 kGy prevented mould and bacteria growth (Dubey and Tiwari n.d.). Xuotong *et al.* (2003) suggested irradiation of modified atmosphere packs of fresh-cut iceberg lettuce with γ irradiation at 0.5–1 kGy to slow oxidation processes and to improve sensory properties. Sprout suppression of onion bulbs can also be achieved by irradiating the bulbs with doses of 20–30 Gy. This level completely prevented sprouting if applied as soon as possible after harvest (Chachin and Ogata 1971). Lu *et al.* (1987) reported that low doses of UV-irradiation induced resistance in onions to postharvest rots. The use of γ irradiation to control sprouting and increase the length of storage time

of potatoes has been proposed as an alternative to cold storage or the use of chemical sprout suppressants. The effects of different levels of γ irradiation on seven potato cultivars were investigated in relation to chlorophyll and glycoalkaloid synthesis on subsequent exposure to light after a period of storage. There were significant genotype differences between cultivars in their response to γ irradiation, with some cultivars exhibiting dramatically reduced levels of glycoalkaloid synthesis compared with others. Also, cultivars responded differently to variable irradiation levels (Dale *et al.* 1997). Irradiation of at least 35 Gy can be used to suppress sprouting in potatoes (Burton and Hannan 1957, Ranganna *et al.* 1997). Sparrow and Christensen (1954) showed that irradiated tubers did not sprout during 15 months storage at 4.4 °C. A dosage rate of 100 Gy was found to be adequate, but lower rates of about 35 Gy can be used to delay sprouting without the side effects which were observed on tubers exposed to higher doses (Burton and Hannan 1957). Dallyn and Sawyer (1959, quoted by Salunkhe *et al.* 1991) demonstrated interactions between temperature and dosage of irradiation. They also showed that a dose of 100 Gy was effective in preventing sprouting of potatoes in storage for 6 months at 10 and 21.1 °C, but 50 Gy was equally effective when the tubers were stored at 4.4 °C. Leszczynski *et al.* (1992) also showed that irradiation inhibited sprouting throughout 6 months storage at 4, 7 or 13 °C. Irradiation can affect tuber quality. Russet Burbank tubers were irradiated with 0, 50, 100 or 200 Gy of γ rays and then stored for 3 months at 10 or 15.5 °C. Irradiation decreased reducing and non-reducing sugar contents after storage. Irradiation at 200 Gy decreased reducing sugars from 1.7 to 0.9%. Irradiation at 100 Gy dosage caused the least changes (5% decrease) in non-reducing sugar content (Badshah *et al.* 1990).

The cultivars Desiree and Metal tubers were irradiated with 0, 50, 100 and 500 Gy γ rays and stored at 5 or 20–25 °C for 6 months. The control samples of Desiree showed higher respiration rates than the irradiated tubers (Alwakdi *et al.* 1991). The cultivars Janka, San and Bobr tubers were exposed to γ radiation (150 Gy) and together with untreated tubers were stored for 6 months at 4, 7 or 13 °C and 85–90% r.h. Irradiated tubers were lower in starch content, but higher in sugar content, especially sucrose. Irradiation increased susceptibility to

enzymic browning. Chip quality did not depend on irradiation but on storage temperature. The highest dry matter losses occurred in non-irradiated tubers stored at 13 °C for 6 months and the highest starch losses in irradiated tubers stored at 4 °C (Leszczynski *et al.* (1992)). Although the law may permit this, there is consumer resistance and economic factors against its use and it is rarely applied commercially.

Irradiation at doses of between 7.5 and 15 krad inhibited sprouting of *D. rotundata* during 6 months storage without inducing adverse changes in acceptability or physiological properties (Vasudevan and Jos 1992). They also reported differences in varietal responses to γ irradiation. Osunde (2008) reported that an average dose of 120 Gy and a dose rate of 114 Gy h⁻¹ were applied to the cultivar Asana and stored for 6 months in a barn or on the ground; results showed that irradiation reduced sprouting in both storage types. However, there was less rotting in the non-irradiated yams stored on the ground than the irradiated ones. Also, food products made from irradiated yams were judged to be better in quality than those made from the non-irradiated ones (Vasudevan and Jos 1992).

Irradiation of king oyster mushrooms with 1 kGy was most effective in extending storage in polystyrene trays covered with PVC film for 4 weeks at 5 ± 1 °C. Hunter L values (lightness) increased after irradiation and remained high throughout the storage period in the irradiated samples. One kilogray irradiated samples maintained an overall better texture than those not irradiated or those irradiated with 2 or 3 kGy (Akram *et al.* 2012). Fernandes *et al.* (2012) studied the effects of γ radiation at 0, 0.5 and 1 kGy on Saffron Milkcap *Lactarius deliciosus* during storage for up to 8 days at 5 °C. They found that irradiation and cold storage did not significantly affect the physical properties but there was a slight decrease in redness in those that had been irradiated.

Organic volatiles

Some organic volatile compounds produced by fruit are thought to have an effect on fungi. The inhibitory effect of (E)-2-hexenal, a volatile component of many fruits, especially after wounding, was studied for its potential for controlling mould development on fruits during storage by Archbold *et al.* (2000). One hundred millilitre of the compound was placed in 1-L

low density film wrapped clamshell containers with 150 g of either the blackberry cultivar Chester Thornless or the grape cultivar Flame Seedless and stored at 2 °C for 7 days. Following removal of the over-wrapped film and chemical from the containers and transfer to 20 °C, mould development was reduced at 20 °C following 14 days of exposure to (E)-2-hexenal in 2 °C storage.

Essential oils

Essential oils may provide alternatives and supplements to conventional antimicrobial additives in foods. Elgayyar *et al.* (2001) evaluated essential oils extracted from anise, angelica, basil, carrot, celery, cardamom, coriander, dill weed, fennel, oregano, parsley and rosemary against *Listeria monocytogenes*, *Staphylococcus aureus*, *Escherichia coli* O:157:H7, *Yersinia enterocolitica*, *Pseudomonas aeruginosa*, *Lactobacillus plantarum*, *Aspergillus niger*, *Geotrichum* and *Rhodotorula*. Oregano essential oil showed the greatest inhibition. Coriander and basil were also highly inhibitory to *E. coli* O:157:H7 and to the other bacteria and fungi tested. Anise oil had little inhibitory effect on bacteria but it was highly inhibitory to the fungi. There was no inhibition of either fungi or bacteria with carrot oil.

Arras and Usai (2001) in *in vitro* studies found that *Thymus capitatus* essential oil at 250 μ l L⁻¹ had fungitoxic activity against *Penicillium digitatum*, *P. italicum*, *Botrytis cinerea* and *Alternaria citri*. The fungitoxic activity of *T. capitatus* essential oils at 75, 150 and 250 ppm on healthy orange fruits, inoculated with *P. digitatum* by spraying and placed in 10-L desiccators, was weak at atmospheric pressure (3–10% inhibition at all three concentrations). In vacuum conditions (0.5 bar), conidial mortality on the exocarp was high at 90–97% at all three concentrations. These data proved not to be statistically different from treatments with thiabendazole at 2000 ppm.

Nitrous oxide

Qadir and Hashinaga (2001) showed that fumigation with a mixture of 80% N₂O with 20% O₂ delayed the appearance of disease and reduced the lesion growth rate on fruit. This response to N₂O was dose- and time-dependent. This suppression of decay by N₂O treatment was thought to be a direct inhibitory effect

on fungal growth rate and/or increased resistance of host tissue. The fruit and diseases tested included Fuji apples, inoculated with *Alternaria alternata* and *Penicillium expansum*, Toyonoka strawberries with *Botrytis cinerea*, *Fusarium oxysporum* f. sp. *fragariae* and *Rhizopus stolonifer*, Satsuma mandarin with *Geotrichum candidum*, Momotaro tomato with *F. oxysporum* f. sp. *lycopersici*, Fuyu persimmon with *Colletotrichum acutatum* and seedling guava with *R. stolonifer*.

Sodium carbonate

Immersion of lemons in a 3% solution of sodium carbonate plus hot water treatment at 52 °C was as effective as imazalil treatment against *Penicillium digitatum* (Lanza *et al.* 1998).

Jasmonates

Methyl jasmonate is a plant growth regulator and there is evidence that jasmonates can also affect microorganisms. Droby *et al.* (1999) showed that postharvest application of jasmonic acid and methyl jasmonate reduced decay caused by the green mould *Penicillium digitatum* after either natural or artificial inoculation of Marsh Seedless grapefruit. The most effective concentration of jasmonates for reducing decay in cold-stored fruits or after artificial inoculation of wounded fruit at 24 °C was 10.5 mol L⁻¹. They suggest that the effect was indirect by enhancing the natural resistance of the fruits to *P. digitatum* at high and low temperatures. Exposure of cultures of *Aspergillus flavus* to methyl jasmonate vapour by Goodrich-Tanrikulu *et al.* (1995) inhibited aflatoxin production. The amount of aflatoxin produced depended on the timing of the exposure. Yao and Tian (2005) found that preharvest sprays of sweet cherry fruit with 0.2-mM methyl jasmonate reduced lesion diameter of *Monilinia fructicola*, but had little inhibitory effect on mycelial growth and spore germination.

There has been considerable research on the physiological properties of jasmonates, for example in mangoes where Gonzalez-Aguilar *et al.* (2000a, 2000b) showed that treatment with methyl jasmonate may prevent chilling injury symptoms without altering the ripening process. Droby *et al.* (1999) found that jasmonic acid and methyl jasmonate effectively

reduced chilling injury in Marsh Seedless grapefruit after cold storage. Meir *et al.* (1996) suggested that methyl jasmonate might mediate the plant's natural response to chilling stress, and by its application might provide a simple means to reduce chilling injury in chilling-susceptible commodities such as avocado, grapefruit and capsicums.

Cai *et al.* (2011) found that methyl jasmonate enhanced activities of ascorbate peroxidase, glutathione peroxidase and glutathione-S-transferase and they suggested that methyl jasmonate can regulate the ascorbate and glutathione metabolism and has important roles in alleviating oxidative damage and enhancing chilling tolerance in loquat fruit.

The combination of exposure to hot air at 38 °C for 12 h and then 1 µmol L⁻¹ methyl jasmonate at 20 °C for 24 h vapour treatment reduced chilling injury and maintained fruit quality during storage at 0 °C for up to 5 weeks plus 3 days at 20 °C for shelf life. The effects of the treatment compared to those not treated included: higher percent of extractable juice, TSS was 25.3% higher and total acid was 62.5% higher, vitamin C and total phenolic contents and higher activities of PAL, SOD and PG, and lower activities of PPO and POD (Jin *et al.* 2009).

Acibenzolar

Acibenzolar (S-methyl benzo [1,2,3]thiadiazole-7-carbothioate) is a chemical activator of induced systemic acquired resistance. When it was applied to strawberry plants at 0.25–2.0 mg a.i. ml⁻¹ to harvested strawberry fruit by Terry and Joyce (2000), it delayed the development of grey mould by about 2 days during storage at 5 °C.

Acidification

Thompson *et al.* (1992) found that in *in vitro* studies, both the germination and growth of *Colletotrichum musae* was greatly reduced at pH 3 in 15 °C. Al Zaemey *et al.* (1993) studied fruit coating and organic acids in *in vitro* control of *C. musae*, but although reductions in the growth of the fungus were observed, there was no complete control.

Coatings

Banks (1984) described a fruit coating called Tal Prolong, which consisted of sucrose esters of fatty

acids and carboxy methylcellulose and showed some reduction in levels of disease on apples coated with Tal Prolong compared to none treated fruit. Banana crowns coated with Semperfresh, which is similar to Tal Prolong and is also made up of sucrose esters of fatty acids and carboxy methylcellulose, showed delayed development of crown rot caused by infection with *Colletotrichum musae*. This effect could be enhanced by the inclusion of organic acids to the coating material (Al Zaemey *et al.* 1993).

Fungicidal chemicals

A large number of chemicals are applied to crops to control fungi that cause diseases. Laws to protect the consumer strictly govern their use, and these laws may vary between countries although most countries conform to the Food and Agriculture Organization of the United Nations and the World Health Organisation regulations. Some chemicals are permitted to be applied to crops only before harvest and the interval of time that must take place between application and harvesting is usually specified. Others are also allowed to be applied postharvest, usually with strict regulations on the maximum permitted residue, the MRL that can remain in the crop. Residue levels of chemical fungicides in a crop are related to the concentration of the chemical used, but also to the time of the crop in storage and the formulation of the fungicide (Table 29).

Fungi are constantly changing, and where a particular chemical fungicide is constantly used, strains of the fungus may develop which are tolerant to that chemical. This commonly occurs with the benzimidazole group of fungicides. Benomyl, which

is a member of the benzimidazole group, was used to control postharvest diseases of yams (Thompson *et al.* 1977). However, during commercial application, a rot caused by infection with *Penicillium sclerotigenum* was frequently observed on the tubers. In *in vitro* tests, this organism was found to be tolerant to benomyl (Figure 54). This tolerance was confirmed in *in vitro* tests, but the organism was highly susceptible to the fungicide imazalil, which gave good control of the disease (Table 30).

The application of some chemicals can actually lead to an increase in the level of disease. An example

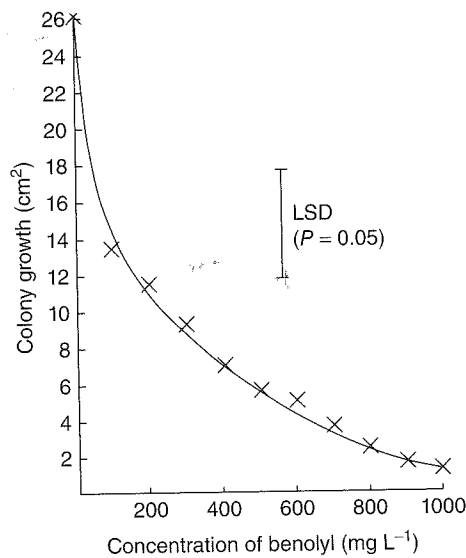


Figure 54 Colony size of *Penicillium sclerotigenum* on potato dextrose agar containing different concentrations of benomyl fungicide after 14 days at 25 °C. (Source: Plumbley *et al.* 1985. Reproduced with permission of John Wiley & Sons Ltd.)

Table 29 Residue levels in microlitres per litre after storage at 21 °C on the skin and in the pulp of citrus fruits of benomyl applied at 0.5 g L⁻¹ in water or in oil emulsion (source: adapted from Papadopoulou Mourkidou 1991)

Storage period	Water	Oil
<i>Residue in the skin</i>		
1 day	0.17	0.83
8 days	0.14	0.52
21 days	0.01	0.13
<i>Residue in the pulp</i>		
1 day	Not detectable	0.01
8 days	0.01	0.03
21 days	0.05	0.14

Table 30 Effects of benomyl and imazalil on the growth (in cm²) of a benomyl-tolerant strain of *Penicillium sclerotigenum* on yam tubers (*Dioscorea cayenensis*) during storage at 20 °C (source: adapted from Plumbley *et al.* 1984)

Treatment	Storage time in days			
	7	14	21	28
Control	1.1	2.2	3.2	3.7
Benomyl (500 ppm)	0.8	1.9	2.1	3.5
Benomyl (1000 ppm)	0.6	1.3	2.4	3.0
Imazalil (500 ppm)	0.0	0.0	0.0	0.0
LSD (p=0.05)	0.56			

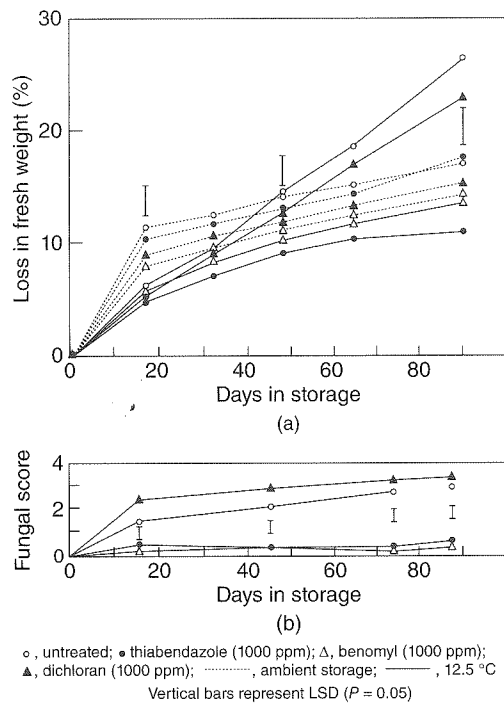


Figure 55 Effects of fungicide treatment on (a) weight loss and (b) the level of surface fungi on stored yams. (Source: Thompson *et al.* 1977.)

of this is the postharvest application of dichloran to control postharvest diseases of yams (Thompson *et al.* 1977). The reason for the increase (Figure 55) was probably due to synergy between the various organisms that were infecting the tuber.

Control of bacteria by chemicals is not normally necessary and where it is practised, it usually achieved by dipping or washing the crop in a solution containing active chlorine at 75–125 $\mu\text{L L}^{-1}$. Household bleach is often used as the source of chlorine. Abu Baker and Abdul-Karim (1994) showed that dipping sapota fruit in the fungicides benomyl, captafol or captan not only reduced the fungal population on the fruit but also the bacterial population. Most of these chemicals are toxic to human beings; therefore, great care must be observed during handling the chemicals, especially before they are diluted, and appropriate protective clothing must be worn.

Chemical application methods include:

- Dipping or spraying the crop in a solution or suspension of the chemical, this may be in hot

water to enhance control. The crop may be passed below a shower of the diluted chemical. This is called cascade application.

- Applying the chemical as a spray but producing very fine particles and then charging them to give a fine even coat of chemical over the crop.
- Applying the chemical as a dust, usually the active chemical diluted with an inert powder such as talc.
- Volatile chemicals can be applied as a vapour that is circulated in a confined space containing the crop. Some chemicals need to be heated to aid circulation, and this process is called thermal fogging. In other cases, chemicals that have a high thermal stability can be burnt and the smoke introduced into the store.
- The chemical may be absorbed onto a pad made of suitable material such as absorbent paper. These pads are placed over cut surfaces such as the cut crown of a hand of bananas. In this case, the pad absorbs latex from the cut surface which also helps keep the pad in position.

Dipping

The crop or part of the crop is immersed in water containing an appropriate concentration of a chemical that is toxic to the fungi, which are known to cause disease on that crop. However, the chemical must be either not toxic to the crop or have acceptable levels of toxicity. The residue of the chemical remaining in the crop after treatment must be at a level that will not endanger public health. In order to improve the effectiveness of these dips, additives may be included in the formulation. These include wetting agents, which reduce the surface tension and allow a better coating of the chemical on the crop, and acids, such as citric acid, which lowers the pH of the fungicide and can make it more effective. In some cases, the area to be protected can be targeted. In pineapples, infection of the fruit is commonly through the cut fruit stalk. Dipping this area directly after it has been cut is normally sufficient to control postharvest disease. This allows for reduced levels of fungicide application, which may thus leave a lower residue and costs less in chemical used. The effectiveness of the fungicidal treatment may be enhanced by heating the water into which the fruit are dipped. In the control of mango anthracnose, postharvest dips in fungicides in cold water rarely reduced levels

of infection; but when fungicide was applied in hot water, complete control could be achieved. The optimum recommended conditions varied between different research workers, but $500 \mu\text{L}^{-1}$ benomyl in water at 55°C for 5 min appeared generally effective without damaging the fruit.

Sprays

Spraying fruit and vegetables with a diluted fungicidal spray postharvest may be more effective than dipping. This is because where the crop is dipped, the fungicide concentration may be reduced if the crop has been washed and is still wet. Also, fungicide may be preferentially removed from the mixture when the crop is removed. Additionally, many fungicidal chemicals are formulated in a way that they are not in solution, but in a fine suspension. This can result in a concentration gradient from the top of the tank to the bottom unless the suspension is frequently agitated. This is less likely to occur with sprays, but it is important with sprays to ensure that the crop is evenly coated with the chemical.

Knapsack sprays (Figure 56) are sometimes used postharvest, but more frequently, the spray is built into a grading line in a packhouse. In this case, the crop is passed over rollers that constantly revolve the fruit to ensure all sides are evenly exposed to the spray. The cascade applicator is a modification of this method where the fungicide is applied as a curtain of liquid under which the fruit pass. This method is good for bananas where the cut crowns are placed



Figure 56 Postharvest application of a fungicide to bananas using a knapsack sprayer.

upwards, because this is the area where the fungi are likely to attack the fruit (Thompson and Burden 1995). It is less useful in fruits such as citrus where it is important to have an even coating of chemical all over the fruit surface.

Electrostatic sprays

Breaking up the pesticide solution into fine droplets and then giving them an electric charge was first developed for field sprays. The main advantages of this system are that they give increased uniformity of application, enabling the application rates to be reduced without loss of biological activity (Law 1982). The principle on which they work is that the particles all have the same electrical charge and thus repel each other. They are attracted to earth, which in this case is the crop, and can form a thin even coat. This method was demonstrated as having a postharvest application by Cayley *et al.* (1987), who developed it for postharvest applications of various fungicides on potatoes. The tubers were passed below an electrostatic sprayer on a roller table. Subsequent work by Anthony (1991) demonstrated its effectiveness in controlling crown rot on bananas with low levels of fungicide. Application methods for bananas were similar to those described by Cayley *et al.* (1987).

Dusting

Various dusts can be applied to crops postharvest to control postharvest diseases. Wood ash is commonly used or sometimes lime (calcium hydroxide) for disease control in stored yams (Coursey 1967, Ogali *et al.* 1991, Thompson 1972a, 1972b). Fungicidal chemicals diluted in an inert carrier, such as talc, are available for postharvest use and dust formulations have been used on potatoes as they are being loaded into store. The advantage of dusts over some sprays is that the crop is still dry after application. Some vegetables may be affected by bacteria when wet, so the result of the application of water-diluted fungicides could be an increase in bacterial diseases. The disadvantage of dusts is that it is difficult to achieve complete coverage over the surface of the crop.

Fumigation

Various fumigants have been applied to crops, postharvest, for various purposes. Their use is strictly

Table 31 Effects of chemical application method on the level of crown rot disease on bananas (where 0 = no infection and 7 is crown completely covered), percentage of hands with pedicel rot and hands that were subjectively considered still marketable (source: adapted from Genay 1991. Reproduced with permission of Cranfield University)

Days in storage	Level of infection		Hands with pedicel rots (%)	Commercially marketable hands (%)	
	5	10	10	5	10
Microstat	2.4b	4.9a	66b	88bc	29a
Cascade	2.8b	4.8a	57ab	71ab	17a
Crown pads	0.8a	3.9a	36a	100c	63b
Untreated	3.0b	5.3a	70b	67a	20a

Figures followed by the same letter were not significantly different ($p=0.05$).

controlled by legislation that is constantly changing. Sulphur dioxide is used for controlling postharvest diseases of grapes (Pentzer and Asbury 1934; Couey and Uota 1961; Ryall and Harvey 1959), litchis (Jurd 1964; Saucó and Menini 1989; Underhill *et al.* 1992; Milne 1993; La Ongsri *et al.* 1993; Tongdee 1993) and snap beans (Henderson and Buescher 1977). Acetaldehyde fumigation can be also be used on grapes (Avissar *et al.* 1989).

Paper pads or paper wraps, impregnated with diphenyl fungicide, were commonly applied to citrus fruit. The chemical vaporised slowly, protecting the fruit from fungal infection.

Sprout suppressants can be applied to root crops in store by fumigation. Tecnazene was reported to be applied as a fumigant in bulk stores, but because of the technical difficulties of application; contractors normally apply it (Burden and Wills 1989). 2-Aminobutane could be used as a fumigant in stored potatoes (Graham and Hamilton 1970). Fumigating oranges with 2-AB was also recommended for postharvest disease control (Eckert 1969).

Chemical pads

Paper pads impregnated with a fungicidal chemical were developed for the banana industry in the Windward Islands. These are called crown pads and they are used to prevent fungal infections on the cut crowns of fruit. Normally, postharvest disease control is achieved by dipping or spaying the fruit with

fungicide when it is taken to the packhouse. However, when bananas are dehandled in the field and packed directly into export cartons, it poses problems of how to protect the cut surface of the crown of the hand from infection. Applying these pads to the cut surface solved this. The pads are made from several layers of soft paper previously soaked in a fungicide (often thiabendazole) and then dried. The pad also absorbs latex given out from the cut surface and prevents it staining the banana fingers. Potassium aluminium sulphate may be added to the pads, which helps coagulate the latex. It was claimed that this system contributed to an increase of 35% in exportable quantities (Hope Mason 1984). The crown pads proved very effective in controlling crown rot (Genay 1991), and the fungicide residue levels in the banana are likely to be small (Johanson *et al.* 1989). However, the pads were difficult to apply using the technique described by the manufacturer, and in Genay's experiments, they were secured by rubber bands to ensure complete and even contact between the cut crowns and the pads. In observations by the author of commercial packs, this even contact between crowns and pads is not always achieved, so these excellent results would not always be achieved in commercial practice (Table 31). This may be reflected in the fact that crown pads are not now used as a method of crown rot control in the major banana producing countries. A British company, Harcloth of Bury Lancashire, has patented the crown pad system on bananas.

