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Abstract

Pre- and postharvest disease control for ornamental plants is mainly provided via fungicide or bactericide application. However, disease control with conventional chemical compounds carries the risk of resistance development by new pathogen races. Additionally, there is increasing public concern over fungicide usage in terms of human and environmental risk. For this reason, and over the past 20 years, researchers developed novel postharvest disease management strategies for cut flowers and other ornamentals. For example, the generally recognized as safe host defense inducers may provide an alternative solution to socially and environmentally less desirable control using conventional fungicides. There are also various biological agents and microorganisms that affect disease development via antagonism and, in many cases, help in integrated disease management (IDM) strategies. However, most of those biotic and/or abiotic agents have not yet been put into practice by growers, who merely rely on chemical control. The current chapter offers an overview on postharvest disease management of various pathogens infecting ornamental plants and cut flowers.

Keywords

Ornamentals • Cut flowers • Postharvest pathogens • Vase solutions • Chemical control • IPM • Elicitors

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1 Introduction

The majority of postharvest diseases lead to permanent loss of a plant's ornamental value. Although this is not relevant to the major issue of food safety, it is relevant to significant economic losses for growers and wholesalers (Darras et al. 2004). Postharvest diseases of ornamental plants and cut flowers destroy approximately 10–30% of the total crop yield produced worldwide (Dole and Wilkins 2004). Postharvest diseases usually cause these losses by reducing the quality or shortening the shelf life of ornamentals or the vase life of cut flowers. The fungi and bacteria causing postharvest diseases usually attack healthy, living tissue, which they disintegrate and rot. Often, however, other fungi and bacteria follow them and live saprophytically on the tissues already killed and macerated by the former pathogens (Agrios 1997).

Before applying treatments to control postharvest decay, it is crucial to recognize the concept of postharvest disease development. Postharvest phase is related to products that are ready for consumption (fruits and vegetables) or ready to be used (ornamentals) by consumers. Therefore, postharvest disease implies to the damaging of the final product at that phase. Apparently, the infection process starts in the greenhouse during cultivation, but disease symptoms develop postharvest. These symptoms are revealed as decay of the root system, as damage on leaves or on flowers, or as premature wilting and dieback of cut flowers held in water.

The current chapter reviews key conventional and novel disease control strategies successfully used during the last 20 years for the management of the most common postharvest diseases of ornamental plants and cut flowers.

2 Most Common Postharvest Pathogens Infecting Ornamental Plants and Cut Flowers

Postharvest diseases are caused by a relatively small number of *Ascomycetes* and Fungi Imperfecti and by few species of *Basidiomycetes* and bacteria (Agrios 1997). The bacteria are of the genera *Dickeya*, *Pectobacterium*, *Xanthomonas*, and

Pseudomonas. Microorganisms that develop and infect after harvest express weak pathogenic ability and are saprophytic in nature, although they infect via natural openings and wounds found on the mature plant's epidermis when conditions are favorable (Agrios 1997).

Postharvest diseases develop on plant tissue during storage and/or at transportation or after the consumer's purchase. During this period, the floricultural products may or may not show disease symptoms of infections that started in the greenhouse during production (i.e., latent infection). For example, *B. cinerea* conidia germinate on flower petals in water droplets and penetrate the epidermis with or without infection structures (Jarvis 1977; Salinas et al. 1989). Then, infection becomes invisible/latent until harvest, and symptoms develop slowly during storage at low temperatures and at high relative humidity (Elad 1988; Darras et al. 2006a). Infection is observed as either gray mold or as hypersensitive flecking on the host (Salinas et al. 1989; Salinas and Verhoeff 1995; Darras et al. 2006a). This latent period may last for up to 6 weeks depending on postharvest conditions, tissue maturity, and host resistance (Jarvis 1977).

2.1 Gray Mold (*Botrytis cinerea*)

2.1.1 Morphology, Geographic Occurrence, and Impact

B. cinerea Pers. belongs to the class *Deuteromycetes* and the phylum *Ascomycota*. The disease caused by *B. cinerea* is called gray mold or *Botrytis* blight. *B. cinerea* mycelium has the typical characteristics of the phylum *Ascomycota* (Jarvis 1977). Extension from the germinated spore occurs at the hyphal apex and growth of the hyphae increases over time. The conidiophores are tall, dark colored, and irregularly or dichotomously branched. The conidia are hyaline or pigmented, ellipsoid to obovoid – globoid – and usually continuous, with 1–3 septa (Ellis and Walter 1974). Conidia germinate in nutrient solutions and less readily in water to usually form 1 to 5 germ tubes (Jarvis 1977). The fungus is pathogenic to most of the cultivated ornamental potted plants and cut flowers. In the Netherlands, substantial quantities of *B. cinerea*-infected rose, *B. cinerea*-infected gerbera, and *B. cinerea*-infected freesia flowers are rejected by sellers in the UK at certain times of the year (Darras et al. 2004). Such rejections result in economic losses to both growers and wholesalers and make professional collaboration problematic.

2.1.2 Infection Process, Symptoms, and Host Range

Infection by *B. cinerea* occurs when a single conidium germinates, forms a germ tube, and penetrates an epidermal cell (Darras, et al. 2006a). Fungal hyphae then colonize adjacent cells resulting in visible lesions (flecks or specks) (Darras et al. 2006a). Gray mold on gerbera and freesia flowers is observed as small necrotic, dark-brown lesions, “spots.” Similar symptoms developed in the laboratory under controlled conditions following artificial inoculation of gerbera or freesia inflorescences at temperatures ranging from 4 to 25 °C (Darras et al. 2006a; Salinas and Verhoeff 1995). *B. cinerea* infects rose (*Rosa hybrida*) flowers and produces necrotic

spots or blister-like patches on petal surfaces (Pie and de Leeuw 1991; Williamson et al. 1995). Infection has been described by Elad (1988) as restricted, brown, volcano-shaped lesions. *B. cinerea* damages phylloclades of *Ruscus* (*Ruscus aculeatus*) by causing small, dark, water-soaked necrotic lesions encircled by a faint halo. These lesions later become brown without growing in size (Elad et al. 1993). *B. cinerea* is also pathogenic to Geraldton waxflower (*Chamelaucium uncinatum*), an Australian native plant which holds a high ornamental and commercial value (Joyce 1993; Tomas et al. 1995). Geraldton waxflower sprigs showed increased abscission of flowers from their pedicels after inoculation with *B. cinerea*. Symptoms of gray mold on lisianthus are reported as crown rot, damping off, stem cankers, and stem, leaf, and flower blights. Gray mold also can cause postharvest deterioration of flowers (Wegulo and Vilchez 2007). Botrytis blight can be a very destructive disease for growers of cut liliiums (*L. tigrinum*, *L. speciosum* var. *rubrum*, *L. regale*); by gathering information on genotypic resistance and making it available to growers, resistant and superior cultivars can be grown for commercial purposes or in private or public gardens, resulting in reduced losses from the disease and less reliance on fungicide applications (Daughtrey and Bridgen 2013). In a series of field trials from 2008 to 2011, the resistance of *Lilium* spp. against *B. elliptica* infection was evaluated (Daughtrey and Bridgen 2013). The most susceptible cultivars (70–94% spotted or scorched leaves) were Chianti, Connecticut King, Côte d’Azur, and Gironde and for *L. tigrinum* the cvs. Pink, Menorca, Red Alert, Royal Fantasy, Sweet Kiss, Vermeer, and White Heaven. In 2009, *Lilium* spp. plants with less than 21% symptomatic leaves were those of cvs. African Queen, Casa Blanca, Cobra, Conca d’Or, Le Rêve, Mozart Zanlazart, Sorbonne, and Time Out. In 2011, the most severely affected (70–100% symptomatic leaves) *Lilium* spp. plants were those of cvs. Chianti, Connecticut King, Cote d’Azur, Gironde, and Red Alert (Daughtrey and Bridgen 2013).

B. cinerea disease symptoms on geranium (*Pelargonium zonale*) plants have been described by Strider (1985) as flower blight, leaf blight, and stem rot. Botrytis blight, of scented geranium (*Pelargonium* sp.), caused by *B. cinerea*, is one of the most destructive diseases affecting geranium production causing stem, leaf, and flower blight. The disease symptoms are characterized by gray, fuzzy sporulating lesions commonly observed under humid conditions. Martinez et al. (2008) reported the infection of *Pelargonium x hortorum*, *Euphorbia pulcherrima*, *Lantana camara*, *Lonicera japonica*, *Hydrangea macrophylla*, and *Cyclamen persicum* by *B. cinerea*.

2.1.3 Biology and Epidemiology

Infection of gerbera (*Gerbera jamesonii*), freesia (*Freesia hybrida*), and rose (*Rosa hybrida*) plants occurs inside the glasshouse during crop cultivation, but symptoms develop during storage or transportation after passing through a latent period (Darras et al. 2004; Elad 1988; Harkema et al. 2013; Salinas and Verhoeff 1995). Favorable and/or fluctuations in temperature and relative humidity (RH) after harvest result in rapid disease development (Darras et al. 2006a; Harkema et al. 2013; Salinas et al. 1989; Williamson et al. 1995). For example, infection of *Fuchsia hybrida* inflorescences after artificial inoculation appeared in less than 24 h at 12 °C and

80–90% RH. Even at 5 °C, disease symptoms were evident in a saturated atmosphere (ca. 100% RH) within the first 24 h of incubation. Moreover, clear differences in *Botrytis* damage were observed between wet and dry long-term transport of cut roses of vars. Red Naomi, Aqua, and Avalanche at low temperatures and at high RHs (Harkema et al. 2013). *Botrytis* developed during dry or wet transport, but infection was significantly lower in dry transport conditions. These findings showed that dry transport of roses has a significant positive effect on product quality with special focus on *Botrytis* development (Harkema et al. 2013).

2.2 *Alternaria* Species

2.2.1 Morphology, Geographic Occurrence, and Impact

Alternaria spp. belong to the Fungi Imperfecti, from class *Hyphomycetes*, from order *Moniliales*, and from family *Dematiaceae* (Rotem 1994). Some species of *Alternaria* are the asexual anamorphs of the ascomycete *Pleospora*, while others are speculated to be anamorphs of *Leptosphaeria*.

Alternaria species are mainly saprophytes. However, some species have acquired pathogenic capacities collectively causing disease over a broad host range (Rotem 1994; Thomma 2003). *Alternaria* diseases are quite common on ornamental crops, including landscape plants and cut flowers. The most common species of *Alternaria* found is *A. alternata* (infecting vinca, dahlia, gerbera, hibiscus, and geranium) (Simmons 1992; Thomma 2003). In most other cases, the species of *Alternaria* have not been identified. Spores of *Alternaria* spp. are dark brown to black and appear in black masses on leaf and petal spots. These are generally spread by watersplash or air movement. In some plants, such as zinnia (*Zinnia* sp.), the infection originates from contaminated seeds (Rotem 1994).

2.2.2 Infection Process, Symptoms, and Host Range

In general, *Alternaria* species are foliar pathogens that cause a relatively slow destruction of host tissues through the reduction of photosynthetic potential. Infection leads to the formation of necrotic lesions, which sometimes have a target-like appearance due to growth interruptions caused by unfavorable conditions and diurnal cycles (Rotem 1994; Thomma 2003). The fungus resides in the center of the lesion, which is surrounded by an un-invaded chlorotic halo, a symptom that is commonly observed for the infection process of necrotrophic pathogens. This zone is created by the diffusion of fungal metabolites like toxins (Agarwal et al. 1997; Rotem 1994; Tewari 1983). Members of the genus *Alternaria* frequently cause quiescent infections in which the fungus enters the tissue and remains dormant until conditions favor infection development (Rotem 1994).

A. alternata caused leaf spots on different geranium species (e.g., *Pelargonium domesticum*, *P. peltatum*, and *P. scented*). The symptoms were brown, irregular spots with increasing diameters (Furukawa and Kishi 2001). These spots turned yellow until they covered the whole leaf area. Similar spots occurred on the flower petals. On petunia, the disease was characterized by small spots which were initially water

soaked (Agarwal and Gupta 1984). These spots turned reddish brown or black, were roughly circular, and reached 3–4 mm in diameter. Their centers were frequently tan to white.

Alternaria leaf spots on impatiens were small (less than 1 mm in diameter), initially water soaked that turned reddish brown with tan centers (Wolkan and Grego 2004). They reached 3–4 mm in diameter and were round. Spots frequently merged to affect most parts of the leaf.

A. dianthi and *A. dianthicola* each cause diseases on carnation and pinks that are recognized by the gray-brown leaf and petal spots with purple margins. Black spore masses were sometimes observed in those spots (Arbeláez 1987).

Infection of *Zinnia acerosa* plants in Dallas, TX, field plots appeared as small, brown flower spots, which enlarged until they covered whole petals, causing conspicuous flower blighting (Colbaugh et al. 2001). Microscopic examination of lesions from infected flower blossoms indicated the presence of short-beaked, cylindrical spores near the smaller lesions on flower petals. Isolation from symptomatic flower petals consistently yielded cultures of an *Alternaria* sp. strain with long chains of conidia.

2.2.3 Biology and Epidemiology

In general, *Alternaria* species are favored by wet and warm conditions. Conidia germinate in just 35–45 min when temperatures range between 28 and 30 °C (Rotem 1994). Tissue penetration and infection occur in 12 h at 10 °C, 8 h at 15 °C, and 3 h at 22 °C (Rotem 1994). Epidemiological studies on safflower (*Carthamus tinctorius* L.) plants infected by *A. carthami* showed that the percentage of leaf and flower area with infection symptoms was greatest at 25 °C, while the frequency of stomatal penetration was greatest at 30 °C (MacRae et al. 1984). Additionally, a long period of wetness (i.e., >6 h) was needed for symptoms to develop. Safflower plants that were kept wet for less than 6 h did not develop symptoms.

2.3 Bacterial Species

Bacteria are invariably present whenever fleshy plant tissues are rotting in the field or in storage (Agrios 1997). Bacterial rot of plants is characterized by soft and watery tissue; cellular debris also frequently oozes from infected cracks. In many soft rots, the bacteria involved are not plant pathogens but rather live saprophytically as secondary parasites (i.e., in tissues already killed by pathogens and environmental causes or in weakened senescent tissues that are unable to resist microbial attack) (Agrios 1997). This is basically the case for harvested ornamental material. Usually, natural defense responses of harvested products are low and sharply decrease. During this period, bacteria may grow and macerate mature and susceptible tissues. However, some bacteria may attack and infect living tissues and cause soft rots in the field or in storage. The most common bacterial genera causing postharvest rots of ornamentals are *Dickeya dadantii* (syn. *E. chrysanthemi*), *Pectobacterium*

carotovorum spp. *carotovorum* (syn. *E. carotovora* pv. *carotovora*), *Xanthomonas campestris*, and *Pseudomonas fluorescens*.

2.3.1 *Dickeya* and *Pectobacterium* Species

Dickeya and *Pectobacterium* and closely related bacteria are straight rods, 0.5–1.0 by 1.0–3.0 μm , and are motile by means of several to many peritrichous flagella (Agrios 1997). These bacteria are the only plant pathogenic bacteria that are facultative anaerobes. Some do not produce pectic enzymes and cause necrotic or wilt diseases (the “amylovora” group), whereas the “carotovora” group has strong pectinolytic activity and causes soft rots (Agrios 1997). Soft rot caused by *Pectobacterium carotovorum* spp. *carotovorum* has a significant negative impact on the flower bulb industry (van Doorn et al. 2011). Bulbous ornamentals such as *Hyacinthus*, *Dahlia*, *Iris*, *Muscari*, *Freesia*, and *Zantedeschia* are susceptible to soft rot infections that damage plant tissue and decrease flower production. In Malaysia, kalanchoe (*Kalanchoe gastonis-bonnierei*) can be infected by *P. carotovorum* spp. *carotovorum* during cultivation in the greenhouse or during postharvest storage or transportation (Golkhandan et al. 2013). Recent experiments by Gracia-Garza et al. (2004) showed the effects of phosphorous addition to soilless mixes or to nutrient solutions on soft rot incidence caused by *P. carotovorum* spp. *carotovorum* on calla lilies (*Zantedeschia* spp.). Soft rot incidence increased by up to 51% when a soilless mix was amended with superphosphate in comparison to the unamended soilless mix. In contrast, addition of phosphorous in the nutrient solution met the phosphorous needs of the plant without enhancing the soft rot. The plant height, fresh mass, and number of flowers per plant were greater in calla lilies irrigated with nutrient solution containing phosphorous than no phosphorous treatments (Gracia-Garza et al. 2004).

2.3.2 *Xanthomonas* Species

The cells are straight rods, 0.4–1.0 by 1.2–3.0 μm , and are motile by means of a polar flagellum (Agrios 1997). Growth on agar media is usually yellow, and most are slow growing. All species are plant pathogens and are found only in association with plants or plant materials (Agrios 1997). Different *Xanthomonas* species have produced large-scale crop damages and losses in the past. For example, the bacterium *X. campestris* pv. *dieffenbachiae* wiped out the whole anthurium (*Anthurium andraeanum*) crop production in the French Antilles in the 1980s (Anais et al. 2000). In 1972, Hayward described the disease caused by *X. dieffenbachiae*, on cv. Kansako Red of *A. andraeanum*, as angular, pale brown, necrotic spots, 1–3 mm with marked chlorotic halos. Scales of bacterial exudate occurred on the under surface of older leaves, while on younger leaves the scales were less obvious, and the lesions were dark brown to black and more extensive (Hayward 1972). The pathogen *X. campestris* was found to be pathogenic on *Syngonium* plants (*Syngonium podophyllum*), and the symptoms of the disease were distinct from those infected by *X. campestris* pv. *syngonii* (Dickey and Zumoff 1987).

2.3.3 *Pseudomonas* Species

The pseudomonads are straight to curved rods, 0.5–1.0 by 1.5–4 μm . They are motile by means of one or many polar flagella (Agrios 1997). Most pathogenic *Pseudomonas* species infect plants. *P. fluorescens* produces yellow-green, diffusible, fluorescent pigments on a low-iron medium. Temperature and relative humidity both play important roles in *Pseudomonas* disease development. For example, the number and size of lesions formed on chrysanthemum and pelargonium leaves and stems after *P. cichorii* infection increased with increases in temperature from 16 to 28 °C but were greatly inhibited at temperatures >28 °C (Jones et al. 1984). Lesions on leaves of both plant species tested constantly expanded under high-moisture conditions. Different isolates of *P. cichorii* were found to be pathogenic to geranium (*P. hortorum*) (Engelhard et al. 1983), different magnolia species (*M. grandiflora*, *M. macrophylla*, *M. soulangeana*, and *M. tripetala*) (Mullen and Cobb 1984), chrysanthemum (*Dendranthema grandiflora*) (Jones et al. 1990), and schefflera (*Schefflera arboricola*) (Chase and Brunk 1984). In greenhouse-grown impatiens, *P. syringae* infections were recorded as large, water-soaked, necrotic lesions on leaves (Cooksey and Koike 1990). However, no reactions occurred after inoculation of *Begonia* \times *semperflorens-cultorum*, *Dieffenbachia maculata*, or *Calendula officinalis* with the pathogen.

2.3.4 Stem Blockage of Cut Flower Stems by Bacteria

Bacterial occlusions can cause interruption of water uptake and wilting problems in cut flowers. A high number of bacteria were detected using light microscopy at the cut surface of dahlia flowers (*Dahlia variabilis*) placed in water for a number of days (van Doorn 1997). Ultrastructural investigations of cut roses also showed that a population of bacteria at the cut surface was responsible for vascular occlusions (van Doorn et al. 1991). For most cut flower species, bacterial populations in holding solutions are relatively small at the grower's storage but increase during handling at the auctions and retailers (Hoogerwerf and van Doorn 1992).

Microorganisms found inside the xylem of flower stems or growing in water or in holding solutions resulted in a decrease in hydraulic conductivity of the stems, especially in the basal stem segment (van Doorn and De Witte 1991). The development of occlusions in the stems of rose cut flowers was correlated with the increasing numbers of microorganisms in stems (Florack et al. 1996). Postharvest increase in stem flow resistance was dependent on the presence of microorganisms in cv. Forever Yours roses (Marousky 1969). High bacterial counts in stems were also correlated with vascular occlusion in the petioles of *Adiantum raddianum* fern fronds (van Doorn et al. 1991). Not only bacteria but also yeasts and filamentous fungi can lead to vascular blockage (Put and Clerckx 1988). For example, in carnation (*D. caryophyllus*), increasing numbers of bacteria and yeasts in the vase solutions resulted in a significant decrease in flower vase life (van Doorn et al. 1995; Zagory and Reid 1986). Among those microorganisms that negatively affected vase life of carnations, *Pseudomonas* bacteria were the most devastating. Moreover, solution turbidity attributed to bacterial growth in the vase was negatively correlated with both the vase life ($P < 0.01$; $r^2 = -0.48$) and the vase solution usage ($P = 0.05$; $r^2 = -0.65$) of cut cv. Baccara roses (Pompodakis et al. 2004).

Reduction of the vase life of cv. Sonia rose was found after inoculations of 10^6 cfu/mL with *Bacillus subtilis*, *Enterobacter agglomerans*, *P. fluorescens*, or *P. putida* (Put and Jansen 1989). The vase life of “white sim” carnations was also reduced when the vase water was inoculated with 5×10^8 cfu/mL suspensions that contained a mixed bacterial population of *Pseudomonas* spp., *Acinetobacter calcoaceticus*, and *Alcaligenes* sp. (van Doorn et al. 1995). Numbers of bacteria in the vase water of gerbera flowers that showed a curvature of more than 90° were found to be 10^6 and 10^8 cfu/ml in the cvs. Liesbeth and Mickey, respectively (van Doorn et al. 1994). Bacterial counts reached a maximum of 10^7 cfu/ml after a few days of vase life in roses, carnations, tulips, and chrysanthemums (van Doorn 1997). When cut cv. Sonia roses were put in vases for 1–4 days, the number of bacteria in the basal 5 cm of the stems was linearly correlated with the number of bacteria in the vase solution, indicating that bacteria in the flower stems contaminated the vase solution (van Doorn and De Witte 1991). Bacteria found in the vase water belonged to the *Achromobacter*, *Alcaligenes*, *Bacillus*, *Escherichia*, *Flavobacterium*, *Micrococcus*, and *Pseudomonas* genera. In the vase water in which rose stems were held, the predominant bacterium found was *Pseudomonas*, while *Enterobacter* was a minor accompanying genus (Florack et al. 1996).

The effect of bacteria on vase life is mainly physical (van Doorn 1997). Hydraulic conductivity of rose stems placed in water containing either 5×10^9 or 2×10^7 cfu/ml of either living or dead bacteria declined rapidly (De Witte and van Doorn 1992). Occlusion by bacteria does not depend on their physiological activity or any defense response of the plant (van Doorn 1997). Bacterial extracellular polysaccharides and globular proteins can also result in vascular blockage (De Witte and van Doorn 1992). When bacterial growth was eliminated, vascular occlusion was still apparent due to the produced extracellular polysaccharides (van Doorn 1997). Vascular blockage was also found in the basal end of the stems of cut roses when the extracellular polysaccharides from *P. cepacia* were added to sterile water (De Witte and van Doorn 1992). A thick layer of material consisting of bacteria with associated extracellular polysaccharides was often found to cover the xylem at the cut surface after a few days in the vase (van Doorn 1997).

Microorganisms through their pectic enzymes may also block the xylem cells, inducing numerous loose vessel fragments (Put and Rombouts 1989). When pectolytic enzymes, such as pectate lyase produced by *P. fluorescens* and polygalacturonase produced by *Kluyveromyces fragilis*, were added to vase water of “Sonia” roses, water relations were disturbed. This may be due to enzymatic degradation of the structures of xylem vessels (Put and Rombouts 1989).

3 Management

In most cases, management of postharvest diseases should be applied before harvest in the greenhouse or in the field (Darras et al. 2006a, 2007). This is crucial as pathogens complete their life cycles in the soil or the growing media. Inoculum loads are always present inside the greenhouses and may infect host tissue when

conditions are favorable. Preharvest infections may become latent and develop postharvest in a compatible host-pathogen interaction (Agrios 1997; Darras et al. 2006b). For that reason, sources of infection should be eliminated, and preventive management has to be applied by means of fungicide treatments. However, caution should be given as common postharvest pathogens such as *B. cinerea*, *Penicillium* spp., and *Alternaria* spp. may develop fungicide resistance. Careful processing of the final product is also critical as it minimizes mechanical damage and significantly reduces subsequent wastage due to postharvest fungal or bacterial infection.

3.1 Chemical Agents (Fungicides/Bactericides)

Various fungicides are used for the pre- and postharvest control of pathogens (Table 1). The best-known chemical classes for *B. cinerea* control include the chlorobenzenes, benzimidazoles, and dicarboximides (Panagiotarou and Chrisaugi 1998). Grinstein et al. (1997) tested pyrimethanil and prochloraz + Zn + folpet against *B. cinerea* infecting rose flower stems. Prochloraz + Zn + folpet controlled infection by increasing plant cover density. In contrast, pyrimethanil + Zn + folpet was better in preventing *B. cinerea* disease when rose petals were exposed to vapors, and no direct contact with the fungicide was used. It was concluded that *Botrytis* blight was delayed when cut rose flower bunches were wrapped in packing paper strips or cellophane bags and previously sprayed with pyrimethanil (Grinstein et al. 1997).

In field evaluations, the fungicides hexaconazole, carbendazim, propineb, chlorothalonil, mancozeb, and the biotic agents *P. fluorescens*, neem oil and garlic clove extracts were tested against *A. alternata* infecting chrysanthemum plants (Kumar et al. 2011). Hexaconazole applied at 0.1% was the most effective against the disease. Chrysanthemum plants treated with hexaconazole showed the lowest disease index (i.e., 4.49) compared to the control plants followed by those treated with chlorothalonil at 0.2% and mancozeb at 0.2%. Furthermore, plants treated with hexaconazole recorded the highest yield of 76.25 quintals/ha (Kumar et al. 2011).

According to Nagrale et al. (2012), preventive sprays of gerbera plants grown in the greenhouse with a range of recommended fungicides for *A. alternata* were more effective compared to curative applications. Preventive sprays of 0.6% Bordeaux mixture, 0.1% tricyclazole, and 0.1% iprodione + carbendazim reduced *A. alternata* disease by up to 95.85%, 96.59%, and 95.88%, respectively, compared to the untreated control plants (Nagrale et al. 2012).

Preplant copper-based dips of calla lily (*Zantedeschia elliottiana*) rhizomes of cv. Yellow were used to prevent plant losses that resulted from latent field infections by *P. carotovorum* (Blom and Brown 1999). More than 90% of the *P. carotovorum* inoculated rhizomes collapsed within 5 weeks after potting. Within the non-inoculated rhizomes, those treated with copper-based compounds showed significantly lower bacterial soft rot percentages than with a quaternary ammonium compound or potassium peroxymonosulfate + sodium chloride +

Table 1 Chemical compounds used to control postharvest pathogens of florists' crops

Pathogen	Host	Application	Chemical or active ingredient	Reference
<i>A. alternata</i>	Chrysanthemum	Preharvest	Hexaconazole, carbendazim 12% + mancozeb 63%, propineb, chlorothalonil, mancozeb, carbendazim 25% + iprodione 25%	Kumar et al. 2011
	Gerbera	Preharvest	Various chemicals	Nagrle et al. 2012
<i>B. cinerea</i>	Various crops	Preharvest/ postharvest	Sulfur, phthalimides, thiram, chlorothalonil, diethofencarb, prochloraz, and tebuconazole	Panagiotarou and Chrisaugi 1998
	Various crops	Preharvest/ postharvest	Folpet, captan, chlorothalonil, dichlofluanid	Ogawa et al. 1977
	Geraldton waxflower	Postharvest	Pyrimethanil, iprodione	Taylor et al. 1999
<i>Leucospermum</i>		Postharvest	Benomyl, carbendazim + flusilazole, chlorothalonil, cyprodinil + fludioxonil, fenhexamid, iprodione, and pyrimethanil	Bezuidenhout et al. 2010
	Rose	Postharvest	Prochloraz, iprodione, dichlofluanid	Elad 1988
	Rose	Postharvest	Picro-cupric-ammonium formate	Hammer and Marois 1989
		Postharvest	Tebuconazole, polyoxin D, and polyoxin B	Elad et al. 1993
<i>P. carotovorum</i>	Calla lily	Preharvest	Quaternary ammonium compound, copper, potassium peroxymonosulfate + sodium chloride + other ingredients	Blom and Brown 1999

other ingredients during the first 6 weeks of forcing. During the remainder of the forcing period, there were no differences in weekly losses of rhizomes (Blom and Brown 1999).

3.2 Environmental Factors, Ethylene Inhibition, and Irradiation

3.2.1 Storage and Transport

Differences in *Botrytis* damage to cut roses of vars. Red Naomi, Aqua, and Avalanche were observed between wet and dry transport at low temperatures and high RHs (Harkema et al. 2013). *Botrytis* developed during dry or wet transport, but infection was significantly lower in dry transport conditions. These findings suggested that dry transport of roses had a significant positive effect on product quality by eliminating *Botrytis* development (Harkema et al. 2013).

Packaging of cut flowers during storage or transport may affect postharvest disease development. For example, package design (size and location of ventilation holes) had a significant effect on the proportion of flowers of rose cv. Sweet Promise with spotting caused by *B. cinerea* (van der Sman et al. 1996). Boxes with large ventilation holes that facilitated effective air ventilation around the packed flowers helped in RH reduction and avoidance of condensed water dispersion on the buds and, therefore, eliminated the chance of conidial germination and consequent infection. This resulted in a maximum of 42% infected flowers, whereas using commercial boxes resulted in 62% infected flowers (van der Sman et al. 1996).

3.2.2 1-Methylcyclopropane (1-MCP)

1-MCP is an ethylene inhibitor used to prevent the detrimental effects of ethylene to climacteric fruits, vegetables, and cut flowers (Serek et al. 1995). In recent research it was shown that 1-MCP may be used to control postharvest *Botrytis* blight of ornamental plants and cut flowers (Table 2). For example, 3.62 μL 1-MCP/L significantly reduced *B. cinerea* infection of *D. caryophyllus* cv. Idra di Muraglia and infection of *Cyclamen persicum* cv. Hallows Bianco Puro Compatto petals (Seglie et al. 2009, 2012). Furthermore, 1-MCP treatments in cyclamen at different rates ranging from 0.38 to 3.62 μL /L slowed *B. cinerea* infection by up to 28 days. Using the lowest concentration of 0.38 μL /L, 1-MCP slowed *B. cinerea* development on *R. hybrida* cv. Ritz by up to 3 days compared to the control flowers. It was concluded that 1-MCP can limit *B. cinerea* development, but its effect was depended on plant species and 1-MCP concentration (Seglie et al. 2009). This funding was

Table 2 Postharvest 1-MCP application on different ornamentals for management of *Botrytis cinerea*

Host	Treatment	1-MCP concentration	Reference
Carnation	Postharvest	0.38–3.62 μL /L	Seglie et al. 2009, 2012
Cyclamen	Postharvest	0.38–3.62 μL /L	Seglie et al. 2009, 2012
Rose	Postharvest	0.38–5.34 μL /L	Seglie et al. 2009; Favero et al. 2015

contradicted by a very recent study on *R. hybrida* cv. Avant Garde flowers which reported that 1-MCP did not protect cut roses artificially inoculated with *B. cinerea* (Favero et al. 2015).

3.2.3 Irradiation Treatments (Gamma, Ultraviolet-C (UV-C))

Gamma irradiation has been used as an alternative nonchemical treatment for pest and postharvest disease control in over 40 countries (Hallman 2011) (Table 3). Ionizing irradiation has been used for many years to sterilize plant and flower surfaces by killing the pests without damaging the plant tissues. Among its advantages is that treatment with gamma irradiation may improve the vase life of cut flowers (Cia et al. 2007). The effects of irradiation are dependent on fungal development, treatment dose, moisture condition, composition of treated products, and storage conditions. Although doses as low as 0.2 kGy were sufficient to protect against the most harmful insects, it was not possible to fully control postharvest fungal diseases of cut flowers (Blank and Corrigan 1995). Gamma irradiation at 4.0 kGy showed complete inhibition of spore germination and mycelial growth of *B. cinerea* in vitro (Chu et al. 2015). On the other hand, in vivo, antifungal activity of gamma irradiation against *B. cinerea* on cut rose flowers of vars. Shooting Star and Babe was dose dependent (Chu et al. 2015).

UV-C irradiation is another type of physical energy used to control postharvest spoilage. Exposure to low doses of UV-C can reduce storage rots in fruits, vegetables, and flowers (Darras et al. 2010a, 2012a; Wilson et al. 1997) (Table 3). This disease reduction was attributed to direct germicidal effects of UV-C on the pathogen and/or to defense response induction in the exposed host tissue (Darras et al. 2012a, 2015). Previous research has shown that exposure to low doses of UV-C (e.g., 0.5–2.5 kJ/m²) may reduce *B. cinerea* postharvest infection on gerbera and freesia flowers by up to 70 and 75%, respectively (Darras et al. 2010a, 2012a, b). This reduction was attributed to direct fungicidal activity, although reductions of 55 and 24% were recorded as a result of an induced defense response. Additionally, UV-C irradiation at 2.5 kJ/m² reduced lesion size caused by *B. cinerea* on leaves of geranium (*P. hortorum*) plants by up to 50%, compared to the nonirradiated control plants (Darras et al. 2015). These disease reductions were the possible result of induction of defense which peaked 24 h post irradiation. Such defense responses were often associated with increases in polyphenol oxidase (PPO) and/or phenylalanine ammonia lyase (PAL) activities (Darras et al. 2012a).

Table 3 Irradiation treatments applied postharvest to various ornamentals for management of *Botrytis cinerea*

Host	Treatment	Doses	Reference
Freesia	UV-C – postharvest	0.5–10 kJ/m ²	Darras et al. 2010a
Geranium	UV-C – postharvest	0.5–10 kJ/m ²	Darras et al. 2015
Gerbera	UV-C – postharvest	0.5–10 kJ/m ²	Darras et al. 2012a, b
Rose	Gamma – postharvest	0.2–4 kGy	Chu et al. 2015

3.2.4 Organic and Inorganic Compounds

There is a significant amount of published research on alternative organic and/or inorganic compounds used as postharvest treatments to control diseases of ornamentals and cut flowers (Table 4). For example, sodium hypochlorite (NaOCl), a chlorine-containing compound, has been used to control bacterial and fungal contamination on fruits and vegetables, on processing equipment, and in flower vase solutions (Suslow 1997; van Doorn et al. 1990). Its strong oxidizing activity has a broad-spectrum antimicrobial impact. When NaOCl is dissolved in water, it ionizes to Na^+ and the hypochlorite ion in equilibrium with hypochlorous acid (HOCl), the active moiety (Dychdala 1983). HOCl damages microbe cell membranes, proteins, and nucleic acids by oxidative degradation upon contact (McDonnell and Russell 1999). A postharvest dip of 200 $\mu\text{L/L}$ NaOCl for 10 s at 20 °C provided significant control against *B. cinerea* infecting rose flowers of cvs. Akito and Gold Strike (Macnish et al. 2010). In this experiment, Clorox[®] Ultra household bleach solution was more effective than the laboratory grade NaOCl. Additionally, when solution pH was reduced from 9.7 (unadjusted) to 7.0, efficacy of NaOCl was considerably improved. Treating cv. Gold Strike rose flowers in this pH-adjusted NaOCl solution resulted in drop in the level of infection on petals to lower levels to those recorded after postharvest dips with the conventional fungicides fludioxonil, copper, cyprodinil, and cyprodinil + fludioxonil. Applying NaOCl prior to a 3- or 10-day commercial shipment also provided the most consistent *B. cinerea* disease control for a wide range of rose cultivars compared to conventional fungicides (Macnish et al. 2010). Woltering et al. (2015) have shown that irrespective of the type of packaging and shipment conditions, roses that received a pre-shipment treatment with 100–150 mg/L NaOCl showed significantly lower *Botrytis* disease compared to the non-NaOCl-treated roses. The NaOCl treatment was generally more effective than comparable treatments with commercial fungicides (Woltering et al. 2015).

Calcium sulfate (CaSO_4) preharvest treatments were used to control postharvest *B. cinerea* infection on cut rose flowers (Capdeville et al. 2005). Application of 10–20 mM CaSO_4 24 h prior to harvest resulted in disease severity reduction by up to 86%. In the non-inoculated assay, the maximum reduction was 86%, while in the inoculated assay, it was 76% (Capdeville et al. 2005).

GA_3 pulsing and spraying treatments at 20 mg/L for 24 h suppressed *B. cinerea* in detached rose petals of cvs. Mercedes and Sonata (Shaul et al. 1995). Since the

Table 4 Recent research on the effects of organic and inorganic compounds used to control *Botrytis cinerea* on ornamentals

Host	Treatment	Concentrations	Reference
Rose	NaOCl – postharvest	100–200 mg/L	Macnish et al. 2010; Woltering et al. 2015
Rose	CaSO_4 – postharvest	10–20 mM	Capdeville et al. 2005
Rose	GA_3 – postharvest	20 mg/L	Shaul et al. 1995
Geraldton waxflower	S-carvone – postharvest	0.64 and 5.08 mM	Hu et al. 2009

studied concentrations of GA₃ were nontoxic to the fungus in vitro, the authors suggested that GA₃ probably activated natural defense responses. Although GA₃ was suggested by Shaul et al. (1995) as a potential alternative to conventional chemical fungicides, no further research on this topic is available.

S-carvone used in concentrations of 0.64 and 1.27 mM significantly reduced *B. cinerea* conidial germination in vitro on glass slides (Hu et al. 2009). Additionally, S-carvone used at concentrations of 2.54 and 5.08 mM inhibited *B. cinerea* spore germination and suppressed mycelial growth on agar. However, S-carvone treatments at 2.54 and 5.08 mM applied to cut waxflowers of cv. My Sweet Sixteen (*Chamaelucium floriferum* × *C. uncinatum* × *C. axillare*) sprigs generally gave no control of *B. cinerea* infection and on flower abscission (Hu et al. 2009).

3.3 Biological Agents/Microorganisms

Over the past 100 years, research has repeatedly demonstrated that phylogenetically diverse microorganisms can act as natural antagonists of various plant pathogens (McSpadden Gardener and Fravel 2002) (Table 5). The interactions between microorganisms and plant hosts can be complex. Interactions that lead to biocontrol can include antibiosis, competition, induction of host resistance, and predation (Cook and Baker 1983). When testing bacterial and fungal isolates from the environment for biocontrol activities, between 1 and 10% showed at least some capacity to inhibit the growth of pathogens in vitro. However, fewer isolates can suppress plant diseases under diverse growing conditions, and still fewer have broad-spectrum activity against multiple pathogenic taxa. Nonetheless, intensive screens have yielded numerous candidate organisms for commercial development. Some of the microbial taxa that have been successfully commercialized and are currently marketed as EPA-registered biopesticides in the United States include bacteria belonging to the genera *Agrobacterium*, *Bacillus*, *Pseudomonas*, and *Streptomyces* and fungi belonging to the genera *Ampelomyces*, *Candida*, *Coniothyrium*, and *Trichoderma* (McSpadden Gardener and Fravel 2002). Screening is generally a critical step in the development of biocontrol agents. The success of all subsequent stages depends on the ability of a screening procedure to identify an appropriate candidate. Primary screens for new biocontrol microbes are still undertaken, and it seems likely that continued prospecting will be required to diversify the potential applications of biocontrol as well as replace more widely used biocontrol products before resistance develops (Larkin and Fravel 1998).

Table 5 Biological agents used for the management of *Botrytis cinerea* infecting ornamentals

Pathogen	Host	Biological agents	Reference
<i>B. cinerea</i>	Cyclamen	<i>U. atrum</i> , <i>G. roseum</i>	Kohl et al. 1998
<i>B. cinerea</i>	Geranium	<i>R. glutinis</i> PM4, <i>B. subtilis</i> , <i>T. harzianum</i> , <i>G. catenulatum</i>	Buck 2004; Elmhirst et al. 2011
<i>B. cinerea</i>	Geraldton waxflower	<i>Pseudomonas</i> sp. 677	Beasley et al. 2001

In regard to ornamentals, *Pseudomonas* sp. 677 significantly reduced conidial germination and retarded germ tube elongation of *B. cinerea* (Beasley et al. 2001). None of the yeasts or fungal isolates tested were found effective in reducing conidial germination or retarding germ tube elongation in vitro, but several significantly inhibited *B. cinerea* growth in vivo. *Fusarium* sp., *Epicoccum* sp., and *Trichoderma* spp. were the most antagonistic in vivo. Of the isolates tested on waxflower, *Pseudomonas* sp. 677 was the most antagonistic against *B. cinerea* by delaying waxflower abscission by up to 3 days (Beasley et al. 2001).

The yeast *Rhodotorula glutinis* PM4 significantly reduced the development of lesions caused by *B. cinerea* on geranium, although efficacy varied between trials (Buck 2004). Treatment with *R. glutinis* in combination with azoxystrobin or trifloxystrobin at 7.5 µg a.i./mL significantly reduced lesion development, compared to treatment with the yeast or the fungicide alone (Buck 2004). Vinclozolin at 250 or 500 µg a.i./mL, in combination with *R. glutinis* PM4, significantly reduced the development of lesions caused by a *B. cinerea* isolate resistant to vinclozolin. Mancozeb did not increase biocontrol efficacy, while thiophanate methyl negatively affected it. Additionally, all combinations of *R. glutinis* PM4 with azoxystrobin, trifloxystrobin, or vinclozolin provided highly effective and consistent *B. cinerea* disease control not acquired with fungicide or yeast treatments alone (Buck 2004). The biological agents *B. subtilis*, *T. harzianum*, and *Gliocladium catenulatum* significantly reduced *B. cinerea* blight of geranium plants (Elmhirst et al. 2011). Reductions in lesion numbers of geraniums treated with *B. subtilis*, *T. harzianum*, or *G. catenulatum* were higher compared to the fungicide Captan 80 (Elmhirst et al. 2011).

Ulocladium atrum and *G. roseum* were used in preharvest trials in commercial greenhouses to control *B. cinerea* blight in cyclamen (Kohl et al. 1998). Both *U. atrum* and *G. roseum* were effective in reducing *B. cinerea* infection on the petioles. The study showed that both agents colonized all treated parts of the cyclamen providing adequate management of the disease via antagonism (Kohl et al. 1998).

3.4 Elicitors of Defense Responses

3.4.1 Jasmonic Acid (JA) and Methyl Jasmonate (MeJA)

The efficacy of MeJA has been tested postharvest on potted ornamental plants and on cut flowers (Table 6). Most tests were carried out in recent years and are still in progress. For example, pre- and postharvest treatments with MeJA on cut Geraldton waxflowers conferred variable protection against postharvest infections by *B. cinerea* (Dinh et al. 2007). JA and MeJA provided systemic protection to various rose cultivars (e.g., Mercedes, Europa, Lambada, Frisco, Sacha, and Eskimo) against *B. cinerea* (Meir et al. 1998). MeJA applied as a postharvest pulse significantly reduced *B. cinerea* lesion size on detached rose petals. In the same study, MeJA at 100–400 µM showed in vitro antifungal activity on *B. cinerea* spore germination and germ tube elongation. Similarly, a postharvest MeJA pulse, spray, or vapor treatment at 200 µM, 600 µM, or 1 µL/L, respectively, significantly reduced *B. cinerea* petal

Table 6 Recent research on elicitors of defense responses tested against postharvest pathogens of ornamentals

Pathogen	Host	Elicitor	Reference
<i>Alternaria</i> sp., <i>B. cinerea</i>	Geraldton waxflower	SA, MeJA, ASM	Beasley 2001; Zainuri et al. 2001; Eyre et al. 2006; Dinh et al. 2007
<i>B. cinerea</i>	Freesia	ASM, MeJA	Darras et al. 2005, 2006a, b, 2007
	Peonies	MeJA	Gast 2001
	Rose	ASM, MeJA	Meir et al. 1998

specking on cut *F. hybrida* inflorescences of cv. Cote d'Azur (Darras et al. 2007). Darras et al. (2005) demonstrated that MeJA vapor was more effective in reducing *B. cinerea* disease on freesia flowers at 20 °C than at 5 or 12 °C. At 20 °C, disease severity, lesion numbers, and lesion diameters were reduced by 58, 50, and 48%, respectively (Darras et al. 2005). Moreover, 1–100 µL/L MeJA postharvest vapor treatment reduced *B. cinerea* development on cut Geraldton waxflower cvs. Purple Pride and Mullering Brook sprigs (Eyre et al. 2006). Application of vapor MeJA to fresh cut peonies resulted in the lowest disease severity and in an improvement of vase life compared to the untreated controls (Gast 2001).

MeJA applied preharvest had variable responses against postharvest *B. cinerea* infection. MeJA was very effective in suppressing the development of postharvest *B. cinerea* disease for greenhouse-grown freesias when applied as a spray at 0.2 mM (Darras et al. 2006b). Dinh et al. (2007) reported that multiple sprays of ≤1000 µM MeJA to field-grown plants significantly reduced *B. cinerea* on Geraldton waxflower cv. My Sweet Sixteen cut sprigs that were non-inoculated or artificially inoculated with the pathogen. MeJA, applied as leaf spray at 10 mM, completely inhibited *P. carotovorum* development in calla lily (*Z. aethiopica*) leaves and afforded a long-lasting effect (Luzzatto et al. 2007). MeJA was more effective than acibenzolar-S-methyl (ASM) in long-term control suggesting that the defense response of calla lily against *P. carotovorum* involved the SA-signaling pathway in the short term, but the jasmonate-/ethylene-signaling pathway was required for durable protection (Luzzatto et al. 2007).

3.4.2 Acibenzolar-S-Methyl (ASM) and Salicylic Acid (SA)

ASM and SA have been applied preharvest on flowering crops to control postharvest decay caused by different pathogens. Preharvest SA sprays at 2000 µg/mL to Geraldton waxflower cv. CWA Pink plants reduced flower disease incidence caused by *Alternaria* sp. and *Epicoccum* sp. (Beasley 2001) (Table 6). However, SA treatment did not suppress postharvest *B. cinerea* infection on flowers of this cultivar. While SA may induce resistance, it can also be directly toxic to pathogens. Direct SA toxicity has been shown against *Cladosporium cladosporioides* and *B. cinerea* (Beasley 2001; Zainuri et al. 2001). Moreover, phytotoxicity was observed to treated Geraldton waxflower leaves (Beasley 2001). ASM has been

used both pre- and postharvest on freesia vars. Cote d'Azur and Dukaat to control postharvest *B. cinerea* infection (Darras et al. 2006b, 2007). Postharvest disease reductions on freesias treated with ASM ranged from 30 to 45% compared to the untreated controls (Darras et al. 2006b). Similar reductions (i.e., 26–44%) were recorded the second year of the glasshouse trials. However, no significant reductions were recorded when ASM was applied to var. Cote d'Azur freesia inflorescences postharvest, although significant reductions in fungal colony diameters and germ tube elongation were recorded in vitro against *B. cinerea* (Darras et al. 2007). It was suggested that a considerable time lag was needed for induction of defense responses by the host.

3.5 Antimicrobial Compounds Used in Vase Solutions

The use of antimicrobial and other compounds in the holding water at storage has been found to extend the vase life of various cut flowers (Damunupola and Joyce 2008; Hoogerwerf and van Doorn 1992; Knee 2000). A broad range of biocides has been used to prevent the proliferation of microorganisms in the vase solutions; however, their antimicrobial activity may be confounded by their other physico-chemical effects (Damunupola and Joyce 2008; Knee 2000). In this respect, the response of cut stems to biocides may vary between cut flower or foliage type, the specific microorganism involved, and other vase solution ingredients.

3.5.1 Hydroxyquinoline (HQ)

The effectiveness of HQ as a biocide in cut flower handling solutions has been known for decades (Damunupola and Joyce 2008; van Doorn 1997). Hydroxyquinoline sulfate (HQS) and citrate (HQC) are the most commonly used HQ compounds in flower handling (Halevy and Mayak 1981; Loubaud and van Doorn 2004; van Doorn 1997). Additionally, 8-HQS promotes stomatal closure (Burge et al. 1996), while both HQS and HQC promote flower longevity by acidifying the holding solution (Halevy and Mayak 1981). Generally, the quinoline esters are acidic (Weinberg 1957) and therefore may inhibit stem plugging by reducing solution pH. Since physiological plugging is mediated enzymatically (van Doorn 1997), the presence of 8-HQC may influence the activity of enzymes by altering the pH (Marousky 1969, 1971). Marousky (1972) considered that while 8-HQ compounds could help prevent microbial occlusion, their ability to reduce vascular blockage may be due to their ability to inactivate enzyme systems through pH adjustment.

3.5.2 Silver Nitrate (AgNO_3), Silver Nanoparticles (SNP), and Silver Thiosulfate (STS)

Silver is typically applied as the nitrate salt (i.e., AgNO_3) (Ketsa et al. 1995; van Doorn et al. 1991). It can act as antimicrobial agent (van Doorn et al. 1990), as an inhibitor of aquaporins in plants (Niemietz and Tyerman 2002), and/or as an ethylene-binding inhibitor during ethylene synthesis and action (Beyer 1976;

Serek et al. 2006; Veen 1979). Pretreatment with AgNO_3 as a pulse at 170 mg/L for 30 min was highly effective against bacterial growth in the basal stem segments of cv. Sonia roses (van Doorn et al. 1990). However, the effectiveness of AgNO_3 as a biocidal agent was highly variable as numbers of bacteria in the 5 cm basal were found to be $<10^2$ cfu/g FW in one experiment, but 1.9×10^6 cfu/g FW when repeated (van Doorn et al. 1990). AgNO_3 significantly increased vase life of *Viburnum tinus* inflorescences and reduced flower abscission, although it negatively affected flower opening and inflorescence fresh weight (Darras et al. 2010b). The effect of AgNO_3 in combination with other vase solution ingredients on the vase life and bud opening of cut *Dendrobium* flowers of cv. Pompadour was investigated by Ketsa et al. (1995). AgNO_3 plus glucose was not as effective in increasing bud opening and vase life as HQS plus glucose. However, HQS plus glucose was less effective in controlling microbial growth (i.e., 10^6 – 10^8 cfu/L and 10^4 – 10^5 cfu/L, respectively) (Ketsa et al. 1995). Research into the mechanism of the inhibitory action of Ag^+ on microorganisms revealed that the expression of cellular proteins and enzymes essential for ATP production was inactivated upon treatment, and DNA lost its replicative ability (Yamanaka et al. 2005).

Silver thiosulfate (STS) is used widely in the cut flower industry as dip, spray, or pulse treatment (Altman and Solomos 1995; Joyce 1993; Joyce and Beal 1999; Slater et al. 2001; van Doorn and Cruz 2000). The ability of STS to inhibit ethylene action was utilized to prolong vase life of various cut flowers (Cameron and Reid 1983; Joyce 1992; Mor et al. 1984; Premawardena et al. 2000; Yapa et al. 2000). Application of Ag^+ as STS to the flowers substantially reduced binding activity by substitution of Cu_2^+ (Beyer 1976). Cu_2^+ is involved in enzymatic reactions related to the biosynthesis and action of ethylene (Himelblau and Amasino 2000). In flowering stems that are sensitive to ethylene such as the *Heuchera sanguinea*, STS reduced flower abscission and prolonged vase life (Han 1998). However, STS may not be as effective in reducing the population of bacteria in vase solutions. For example, van Doorn et al. (1991) found that STS used at 656, 1312, or 2624 mg/L for 4 h was not effective in reducing the numbers of bacteria in petioles of *Adiantum raddianum* fronds, while AgNO_3 at 12.5 and 25.0 mg/L minimized them to zero. Furthermore, Hoogerwerf and van Doorn (1992) reported that STS did not control bacterial populations in solutions holding various cut flowers during transportation from growers to the retailers.

Pure colloidal silver nanoparticles (SNP) are potent and broad-spectrum antimicrobial agents. In the cut rose cv. Movie Star, pulsing 10 mg/L SNP plus 5% sucrose for 24 h and subsequently holding samples in 0.5 mg/L SNP plus 2% sucrose significantly delayed vascular blockage caused by bacterial contamination, and stomatal conductance was inhibited. This resulted in improvement of water balance in cut roses and significant increases in vase life by up to 11.8 days compared to the controls (Lü et al. 2010). Effects of SNP as a dip or spray pretreatment on the subsequent vase life and physiology of cut *Lilium* Oriental hybrid cv. Siberia and *Lilium* Asiatic hybrid cv. Dream Land were investigated by Kim et al. (2004). The vase life of *Lilium* “Siberia” florets was extended with a treatment comprised of 0.1% SNP plus natural chitosan. While ethylene production continuously decreased

and the climacteric-type respiration increased on the fifth day, these processes were not differentially affected by the various treatments. Furthermore, the antibacterial action of SNP and the mechanisms involved have not yet been fully elucidated, and interactions between SNP particles and bacterial species need to be studied (Pal et al. 2007). When cut gerbera flowers of cvs. Double Dutch and Red Explosion were held in a solution of 6 mg/L SNP, their vase life increased by up to 7.8 days compared to the controls (Oraee et al. 2011). At the same time, bacterial populations in the holding solutions decreased significantly.

3.5.3 Chlorine Compounds (NaOCl, DICA, ClO₂)

Many active chlorine (Cl) compounds with antimicrobial properties are commercially available. For example, the sodium salt of dichloroisocyanuric acid (Na-DICA) is a common slow-release (stabilized) chlorine compound used for sterilization of water in swimming pools (Jones et al. 1993; Joyce et al. 2000; Xie et al. 2007). Both DICA and household bleach or sodium hypochlorite (NaOCl) are extensively used in experimentation and flower handling (Akoumianaki–Ioannidou et al. 2010; Darras et al. 2010b; Faragher et al. 2002; He et al. 2006; Knee 2000; van Doorn et al. 1990). Chlorine activity involves the oxidation of cellular components in microorganisms, including essential enzymes in cell membranes and protoplasm (Bloomfield and Arthur 1989; Dychdala 1983). Five to 10 mg/L free available chlorine (FAC) units helped control bacteria in fresh, clean postharvest solutions (Xie et al. 2007). When chlorine compounds were added to vase water, a portion of the chlorine was consumed by water impurities (chlorine demand), which included inorganic reducing agents, like Fe₂⁺, Mn₂⁺, NO₂⁻, and H₂S, as well as organic compounds, like amino acids. The chlorine atom ceases to maintain oxidizing capacity upon reduction to chloride (Cl⁻), and so its disinfectant property is lost or reduced (Dychdala 1983). Consequently, chlorine levels in postharvest vase solutions may decrease rapidly with time. High initial concentrations can be used to meet chlorine demand but may be phytotoxic (van Doorn et al. 1990). Joyce and Beal (1999) suggested that if symptoms of phytotoxicity are evident, then 0.5–1.0% (w/v) sucrose plus 25 mg Cl/L may be appropriate to reduce phytotoxic damage as well as to extend vase life. Chlorine dioxide (ClO₂) is another compound recently used in experimentation. The addition of 2 or 10 μL/L ClO₂ to clean deionized water extended the vase life of *Alstroemeria peruviana* cv. Senna, *Antirrhinum majus* cv. Potomac Pink, *D. caryophyllus* cv. Pasha, *G. jamesonii* cv. Monarch, *Gypsophila paniculata* cvs. Crystal and Perfecta, *L. asiaticum* cv. Vermeer, *Matthiola incana* cv. Ruby Red, and *R. hybrida* cv. Charlotte flowers by 0.9–13.4 days (7–77%) compared to the controls (i.e., 0 μL/L ClO₂) (Macnish et al. 2008). The beneficial effects of ClO₂ treatment were associated with the reduction of aerobic bacteria in vase water and on cut surfaces of flower stems.

DICA has been recommended as a vase solution biocide along with other ingredients, such as citric acid, sucrose, etc. (Akoumianaki–Ioannidou et al. 2010; Darras et al. 2010b; Jones 1991; Knee 2000), although concerns have been expressed over the use of combinations of chlorine compounds with citric acid (Faragher et al. 2002). There may be a decrease in the disinfecting efficiency of

chlorine with the decrease in pH (Dychdala 1983); moreover, chlorine compounds can corrode metal containers. The low chlorine concentrations used by researchers in experiments (e.g., 10 mg/L) (Ligawa et al. 1997) can be inappropriate for use by industry in handling solutions and consumers in vase solutions, where higher concentrations are required (e.g., 50 mg/L) (Jones 1991). In contrast, using DICA at 200 mg/L plus 2% sucrose was found to be effective in prolonging vase life of *Nerium oleander* inflorescences (Akoumianaki–Ioannidou et al. 2010). DICA alone at 100 mg/L did not increase the vase life of cut *Viburnum tinus* inflorescences (Darras et al. 2010b) but significantly increased vase life, flower opening rate, and flower fresh weight of cut *N. oleander* inflorescences when used at 200 mg/L plus 2% sucrose (Akoumianaki–Ioannidou et al. 2010). Knee (2000) found that DICA used alone at 200 mg/L was effective in extending vase life of cut alstroemeria, dianthus, and rose flowers by 2.6, 2.9, and 1.5 days, respectively, as compared to the controls.

3.5.4 Acidifiers

Acidic solutions provide an unfavorable environment for bacteria to develop and also help in rehydration of cut flowers. Water flow increased through rose stem segments with decreasing pH from 6 to 3 (Durkin 1979). In cut cv. Baccara roses, decreasing solution pH from 8 to 6 enhanced flower water relations, fresh weight maintenance, and vase life (Pompodakis et al. 2004). The positive effect of low pH was attributed to the reduction of microbial populations (Halevy and Mayak 1979; Pompodakis et al. 2004). Furthermore, low pH retarded stem blockage of roses in bacteria-free water (Marousky 1971). Bacterial growth was suppressed at low pH, although a population of yeasts and filamentous fungi developed over time (van Doorn 1997). Citric acid at 50 µg/L plus 2% sucrose had significant positive effects on vase life and flower opening scores of *N. oleander* inflorescences independent of storage temperatures tested (i.e., 2, 5 or 20 °C) (Akoumianaki–Ioannidou et al. 2010). Additionally, 50 µg/L citric acid plus 2% sucrose protected the flowers from low-temperature injuries (LTI) when observed at temperatures of 2 °C. It was suggested that the exogenous supply of sucrose might have prevented LTI in oleander-flowering stems, while compounds such as citric acid, DICA, or methanol may have facilitated the absorption and delivery of sucrose into cells and enhanced the ability of inflorescences to be stored at low temperatures (Akoumianaki–Ioannidou et al. 2010).

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