A review on conventional and non-conventional methods to manage post-harvest diseases of perishables

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Abstract: Post harvest diseases are a cause of great economical loss to perishables throughout the world. In this review conventional and non-conventional control methods to manage post harvest diseases of the perishables have been discussed in detail in light of relevant work of past.

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1. Introduction

Fresh fruits and vegetables are considered as an important component of a healthy balanced diet. Fruits are excellent source of dietary fiber, vitamins, carbohydrates and antioxidants and are highly perishable products especially during the post-harvest phase, when considerable losses due to microbiological diseases, disorders, transpiration and senescence can occur. Fruits are perishable by nature and require protection from spoilage during their storage and distribution to give them desired shelf life. Because fruits are now often sold in areas of the world far remote from their production sites, the need for quality as well as extended shelf life for these has also expanded.

Approaching towards the target of second green revolution in India there is an urgent need to unearth new strategies to control post harvest pathogens, so that we can increase the production of fruits and vegetables by improving their shelf life.

In India, post-harvest losses in vegetables at different regions of the country have been estimated, which were for Delhi 7.2-34.7%; Maharashtra 15-20 % and Uttar Pradesh 4-10 % (Madan et al. 1993). In Delhi, the percentage loss reported by the retailers who have permanent shop was around 6.75% and around 8.8% for pushcart vendors.

Post harvest pathogen not only affects the produce quantitatively but also qualitatively. A perusal of literature on the changes occurring during pathogenesis in various fruits clearly revealed that the quantity of various free and bound amino acids and organic acids was altered and a gradual decrease in sugar and ascorbic acid content was observed with the advancement of diseases (Bhargava and Arya 1983; Tripathi and Shukla, 2007).

A number of enabling technologies are available for optimizing product quality through manipulation of nutrition, water and light to minimize post harvest disorders and quality deterioration as well as to optimize carbon assimilation, distribution and accumulation in harvested organs. Common practices used for the control of post-harvest diseases of fruit are controlled atmosphere storage, refrigeration and fungicides (Kader 1992). Among these, chemical treatment ensures product protection, but is permitted for only a few species; in addition, public opinion demands a reduction in the use of chemical products (Caia et al. 1988). This latter issue, along with the appearance of pathogens resistant to fungicides (Spotts and Cervantes 1986) and of iatrogenic diseases (Griffiths 1981), have contributed to arousing increased interest in the development of alternative methods for controlling plant pathogens, capable of integrating, if not totally replacing, synthetic fungicides.

Number of strategies, which are instrumental to control post-harvest decay without the pollution of the environment and risk to public health, are enumerated here.

1.1. Physical agents

Various physical agents have been used to control the advent and spread of the post-harvest diseases of fruits. The action of these agents is either preventive or curative. Low-pressure storage, cold storage and modified atmosphere storage are aimed at preventing the procuration and spread of the disease. Heat and radiation treatments are meant for killing or weakening the quiescent pathogen, thereby improving the shelf life of the fruits.

1.1.2. Low Pressure storage

Storage life is influenced by atmospheric pressure and at low pressure it is extended (Bangerth 1974). Low pressure (180-190 mm) has been reported to reduce fruit ripening (Tolle 1969). At low atmospheric pressure the availability of O_2 for respiration is reduced besides controlled storage, use of fungicides under reduced atmospheric pressure helps from rotting of storage fruits by protecting them (Stenvers and Stork 1977).

1.1.3. Low storage temperature

Low temperature also reduces the ripening and the respiration rates. At 13°C fruits have been kept in the best condition (Tomkins 1963). The temperature requirement for slow ripening depends upon the stage of maturity; green fruit at 15°C, orange green fruit at 10°C and red fruit at 8°C have been kept for a longer period. However, under very low temperature conditions chilling injury is caused and such situation arises below 10°C. Alternations of low and high temperatures, 2°C and 20°C respectively have prolonged storage tissues (Hobson 1981). At ambient temperature fruits can be stored for longer duration.

1.1.4. Modified controlled atmosphere for storage

Modified atmosphere storage or controlled atmosphere (CA) has basically aimed at maintaining (i) A low temperature (ii) An adequate humidity (iii) Low oxygen tension (iv) Low ethylene concentration, around the fruits. The important factors for extending the shelf life are high CO_2 content under low O_2 content conditions (Higasion and Ogata, 1980). The best storage atmosphere is the mixture of 2.5 percent O_2 and 2.5 percent CO_2. Controlled atmospheres (CA) have been demonstrated to reduce fungal growth (Ahmadi et al. 1999). Tian et al. (2001) found that growth of *Monilinia fructicola* (G. Wint.) Honey, both in potato dextrose agar (PDA) and in sweet cherry fruit, declined significantly with increased CO_2 concentrations. CA storage at low temperature has a beneficial effect on various pathological and physiological problems occurring in stored fruit (Rogiers and Knowles 2000; Tian et al. 2002). CA treatment affected both the pathogen and the host. Decay development was retarded because growth, sporulation, and enzyme activity of the pathogen were reduced, and the improved physiological condition of the host enabled it to resist decay more effectively (Sams and Conway, 1987). However, when the fruit was transferred to air at ambient temperature, decay developed rapidly and rendered fruit commercially unacceptable (De Vries-Paterson et al. 1991). This indicated that CA storage could not be used as a sole method for decay control. To optimize the benefit of CA, other methods such as application of biological control agents or a reduced level of fungicides, would be needed to completely prevent decay development and extend the shelf life of fruit (Spadaro et al. 2002). Most perishable fruits are stored at low temperature and are packaged under modified atmosphere (MAP) in order to extend their shelf life. However, these steps do not eliminate undesirable microorganisms from these fruits.

1.2. Heat treatment

Pre-storage heating holds potential as a non-chemical method for control of post-harvest diseases by directly inhibiting pathogen growth, activating the natural resistance of the host, and slowing down the ripening process. Heat treatments are promising and have been used with success in eradication or suppressing the development of fungi on the surface as well as those situated just below the surface as a result of pre harvest infection. Post-harvest curing at 34–36°C for 48–72 h effectively controls citrus decay and reduces chilling injury symptoms (Ben-Yehoshua et al. 1987; Del Rio et al. 1992). In recent years, heat treatments have been used effectively to extend the shelf life of fruits. Research indicates that pre-storage heat treatment at 38°C for 4 days could maintain apple firmness, colour, soluble solids and organic acids while promoting resistance to physiological disorders such as scald and fungal diseases during storage (Conway et al. 1994; Klein and Lurie 1991). The most common heat treatment to fruits is hot water treatment. This treatment is most suitable for such diseases where there is a wide gap in the thermal death point of the pathogen and its host and where the temperature of the water does not spoil the produce by scalding of skin, loss of natural colour, flavour or softening of the flesh part. In some instances, heat treatment has been shown to induce disease resistance in harvested tissue. In lemon fruit, it induced the accumulation of phytoalexin, scoparone, and increased tissue resistance to infection (Ben-Yoshoshua et al. 1988). In spite of interesting possibilities emerging with pre-storage heating, the sensitivity of many harvested crops to heat treatment and the energy required for the treatments may prove to be a liability.

1.2.1. Irradiated Fruits

Emission and propagation of energy through material medium in the form of waves is called irradiation. Both ionizing and non-ionizing irradiation have been used for post-harvest storage studies. Irradiation basically controls the post-harvest diseases by sterilizing the fruits (Abdel-Kader et al. 1968). Salunkhe (1961) has presented a detailed account of the role of irradiation on increasing shelf life of tomato. Tomato fruits irradiated with 10 Krad of 200 KV X-rays at the breaker stage have a longer shelf life (Hannan 1956).

The application of microwave energy provides rapid heating and is widely used in food industry. A number of researches have used microwave energy successfully to reduce bacterial populations on various foods. In stone fruits, heat treatment was found successful (Margossan et al. 1997; Karabulut et al. 2002). Post-harvest heat treatments of fruits and vegetables have been used to control insect disinfestations (Nelson 1996; Ikediala et al. 1999). Microwave treatments were found effective against *Botrytis cinerea* and *Penicillium expansum* (Margossan et al. 1997; Wang et al. 2001; Karabulut and Baykal 2002).

1.2.2. **Ultraviolet light treatment (UV Treatment)**

Recently, low dose of ultraviolet light especially UV-C hormesis have emerged as alternative technology to avoid chemical fungicides. Low dose UV-

C hormesis was shown to induce resistance in post-harvest commodities to harvest decay and to extend shelf life of fruits by delaying the ripening and senescence process. Application of a low dose of UV-C light reduced the development of post-harvest decay in horticulture crops such as onion, sweet potato, apple peach, citrus fruits, bell pepper, tomato, carrot and strawberry. UV-C therapy effectively reduced storage rot 60-90 % compared with 100 % decay for the non-irradiated control. There are evidences that enzymes in host, namely phenylalanine ammonia-lyase, peroxidase and antifungal hydrolases plays important role in induction of defensive responses in fruits. UV-C light has already been tested in many fruit mainly to control post-harvest diseases and delay some ripening associated processes (Douillet-Breuil et al. 1999; D'hallewin et al. 2000). Pre-storage treatment of several post-harvest commodities with low doses of UV-C has been shown to reduce disease development and, in some cases, delay ripening. UV-C treatment controlled natural infection in walla walla onions, sweet potato, tomato, apple, peach and citrus fruit (Droby et al. 1993). Optimum doses of UV-C for the control of post-harvest decay in various commodities occurs in a rather narrow range and appears to vary depending upon the commodity, the type of cultivar and the physiological status of tissue (Stevens et al. 1991; Droby et al. 1993). Optimum UV dose for vegetables, pome, stone and citrus fruit were reported to be in the range 2-10 × 10^4 erg/mm^2 caused skin blemishes and increased the susceptibility of the tissue to decay (Liu *et al*, 1993). Tissue discolouration were also reported in citrus fruit and peaches exposed to injurious UV doses (Ben-Yehoshua et al. 1992)

1.3. Chemical agents

1.3.1. Calcium chloride ($CaCl_2$)

A post-harvest calcium treatment was reported safe and effective methods of improving the quality and shelf life of fresh fruits (Tsantili et al. 2002). Calcium delays ripening and particularly softening by altering intracellular and extra cellular processes (Feruson, 1984). It also reduces disorder and decay losses (Conway 1989; Fallahi et al. 1997).

Immersing the fruit in the solution of $CaCl_2$ at 0.36 M is the most common post harvest treatment, while the majority of calcium studies are referred to apple texture (Conway et al. 1994). However, Calcium affects on other ripening processes of fruits. In pre-climacteric Apples, infiltration of $CaCl_2$ retarded colour changes and decreased ethylene and CO_2 production during at 0°C. Treatments of apples with 0.3 M $CaCl_2$ for 1.5 min increased firmness, but did not affect CO_2 production, Titrable acidity and Soluble solid content (SSC) during storage at 4°C (Duque et al. 1999). Ca immersion increased firmness, but reduced CO_2 production (Luna-Gazman et al. 1999) and prevented colour changes (Lester 1996) in melon discs. In lemons 'Verna', harvested at the colour break state. $CaCl_2$ at 1 mM applied by vacuum infiltration increased firmness and prevented colour changes during 21d of storage at 15°C (Martiniz Romero et al. 1999).

1.3.2. Sodium bicarbonate

Selected organic and inorganic salts are active antimicrobial agents and have been widely used in the food industry. Among these, sodium bicarbonate (SBC) and potassium sorbate are used for controlling pH, taste and texture, and they also exhibit broad-spectrum antifungal activity (Miyasaki et al. 1986; Corral et al. 1988). The potential of bicarbonate salts for the control of post-harvest pathogens has been demonstrated in citrus, carrot, bell pepper and melon (Punja and Gaye 1993; Aharoni et al. 1997; Fallik et al 1997). Sodium bicarbonate at a concentration of 2% (w/v) has potential for controlling *Rhizopus*, *Alternaria* and *Fusarium* decay on 'Galia' and 'Ein Dor' fruits (Aharoni et al. 1997). *In vitro* exposure to Sodium bicarbonate (SBC) reduced mycelial growth of *Rhizopus stolonifer*, *A. alternata* and *Fusarium* spp. The direct and indirect effects of bicarbonate salts on microorganisms have previously been noted (Punja and Grogan 1982; Depasquale and Montville 1990). 1% SBC did not control decay development, while a higher SBC concentration (3%) cause phytotoxicity, which led to a lesser (*C. annum*) in 3% potassium bicarbonate, led to increased weight loss, decrease firmness and further decay (Fallik et al. 1997).

1.3.3. Chitosan

Chitosan and its derivatives, including glycolchitosan, were reported to inhibit fungal growth and to induce host-defence response in plants and harvested commodities. Chitosan, a high molecular weight cationic polysaccharide, is soluble in dilute organic acids, and have been used as a preservative coating material for fruits (El Ghaouth et al. 1991). It has ability to form a semi-permeable film (Bai et al. 1988) and chitosan coating have definite potential to modify the internal atmosphere as well as decrease transpiration losses in fruits. Chitosan coatings have been found to extend the storage life of fresh fruit and that too without causing anaerobiosis (El Ghaouth et al. 1991). Moreover, they have also been reported to reduce decay by inhibiting the growth of several fungi (Allan and Hadwinger 1979; El Ghaouth et al. 1989, 1991).

Recently, chitosan treatment was shown to stimulate defense enzymes and formation of physical barriers in harvested tissue. Chitosan treatment leads to the induction of chitinase, a defense enzyme with great potential to destroy chitinaceous cell wall of fungi (Mauch et al. 1984) and has also been reported to elicit the production of phytoalexin in pea pods (Kendra and Hadwinger 1984). In strawberry fruit, the ability of chitosan to stimulate defence enzymes such as chitinase seems to be expressed more in cut fruits than in intact ones (El Ghaouth et al. 1992). In bell pepper and tomato fruit, the activity of chitinase, chitosanase, and β-1, 3-glucanase appeared to increase upon chitosan treatment (El Ghaouth and Arul 1992). Also, various structural barriers including the formation of hemispherical protuberances along host cell walls and the occlusion of many intercellular spaces with a fibrillar material were observed in chitosan treated bell pepper tissues. While it is not possible to determine exactly the role played by these inducible defensive reactions in control of Botrytis rot, they are more likely to have played a supporting role. This is indirectly supported by the fact that the ingress of the pathogen appears to be restricted directly by chitosan. Expression of such defensive reactions following chitosan treatment could help the tissue restrict fungal colonization, as well as delay the resumption of quiescent infections. In plant tissue, it has been suggested that antifungal hydrolases provided a long term and generalized protection (Kuc 1990). If this is the case, the activity of hydrolytic enzymes in harvested tissue could be manipulated to affect the resumption of quiescent infections, which typically become active when tissue resistance decline. Feeding trials with domestic animals have recently demonstrated that chitosan is non-toxic and biologically safe (Hirano et al. 1990).

1.4. Organic Fungicides

A number of fungitoxic chemicals for controlling postharvest diseases have been developed. These chemicals are mostly used as dilute solutions into which the fruit or vegetables are dipped before storage or as solutions used for washing or hydrocooling of fruits or vegetables immediately after harvest (Sharma and Alam 1998).

Elemental sulphur is used as dust or sprays; borax, biphenyl, sodium-O-phenyl and others are impregnated in the boxes or wrappers coating the fruits. Postharvest fumigation with SO_2 and acetaldehyde is used to eradicate spores and very superficial infections. Benomyl, triforine, dichloran etc. are used as dips, sprays or wax formulations (Eckert and Ogawa 1988).

The application of fungicides to fruits after harvest to reduce decay has been increasingly curtailed due to the development of pathogen resistance to many key fungicides (Bus et al. 1991), the lack of replacement fungicides, negative public opinion regarding the safety of pesticides. Taken together, all these factors have resulted in reframing of government policies which not only allows restricted use of fungicides (Vinas et al. 1991, 1993) but also provides the impetus to develop alternative and effective natural methods of controlling post-harvest diseases.

1.5. Biological agents

1.5.1. Biocontrol

In the recent past, biological control has emerged as an effective strategy to combat major postharvest decays (Wilson and Wisniewski 1989; Korsten et al. 1994) of fruits. However, compared to the long-standing interest in biological control of soil borne pathogens (Cheath et al. 1992), research into biological control of post-harvest decays is still in its infancy. Thus, biological control of post-harvest diseases of fruit and vegetables offers a viable alternative to the use of present day synthetic fungicides (Cook et al. 1999; El-Ghaouth et al. 2003). Today biological control of post-harvest diseases of fruit has become an important field for research (Droby et al. 1998; Sugar and Basile, 2008). Microbial antagonists have been reported to protect a variety of harvested perishable commodities against a number of post-harvest pathogens (Wisniewski et al. 2001). Post-harvest treatment of fruits with microorganisms recovered from fruit surfaces is being developed as an alternative method for control of post harvest diseases of Citrus, Apples, and other fruits and vegetables. A number of yeasts and bacteria have been reported to inhibit post-harvest decay of fruit effectively (Mclaughlin et al. 1992; Fan and Tian 2000). Utilization of antagonistic yeasts as an alternative appears to be a promising technology (Elad et al. 1994; Fan et al. 2002). Several mechanisms have been reported to play a significant role in the biocontrol activity of antagonistic yeasts. Among them, interaction between yeast and post-harvest pathogens is involved. It has been suggested that attachment of the yeast to fungal hyphae and extensive production of an extracellular matrix by yeasts may play a key role by either enhancing nutrient competition or by some other undetermined mechanisms (Wisniewski et al. 1991; Wan and Tian 2002). The modes of action of antagonistic microorganisms may be due to secretion of antibiotics, competition for space and nutrients (Droby et al. 1989; Piano et al. 1997), production of cell wall lytic enzymes (Wisniewski et al. 1991; El-Ghaouth et al. 1998), and induction of host resistance (Arras 1996). Some antagonist-based products are commercially available and others are currently under varying degrees of development (Castoria et al. 2001). However, application of antagonistic microorganisms alone does not always provide commercially acceptable control of fruit decay. Combining antagonists with other post-harvest treatments could increase the performance of biocontrol agents (El-Ghaouth et al. 2000c; Spotts et al. 2002). Few studies have been carried out to evaluate the efficacy of such treatments for conserving viability of cells of post-harvest biological control agent to be of practical use, microbial agents must be formulated as products capable of storage, distribution and application approaches from traditional agrochemicals product design (Rodham et al. 1999). Formulation is necessary in order to present the product in a usable form and in order to optimize the efficacy, stability, safety and ease of application of the product (Rhodes 1993).

In several reports, workers have shown that certain bacteria and yeast strains *(Agrobacterium* *tumefaciens*,. *Aureobasidium pullulans*, *Bacillus polymyxa*, *B. subtilis*, *B. brevis, B. cereus, B. licheniformis, B. thuringiensis, Candida oleophila, C. guilliemondii, C. sake, Cryptococcus laurentii, Debaryomyces hansenii, Enterobacter cloacae, Erwinia herbicola, Klebsiella pneumoniae, Metschnikowia pulcherima, Pichia gulliermondii, Pseudomonas syringae, P. putida, P. cepacia, P. aureofaciens, P. maltophilia, P. fluorescens, Sporobolomyces reseus, Streptomyces noursei*,) for biocontrol of post-harvest fungal decays of citrus and tomato fruits (Droby et al. 1989; Chalutz et al. 1992; Wisniewski et al. 1991; Spadaro et al. 2002) caused by *A. alternata, Botrytis cinerea, Geotrichum candidum, Monilinia fructicola, Penicillium digitatum, P. italichum, P. expansum and Rhizopus stolonifer.*

Microbial antagonists (*Cryptococcus infirmominaturs*, *C. oleophila*, *Pantoea agglomerans*, *B. subtilis*, *P. cepacia* LT-4-12W) were found effective against fungal pathogens (*P. expansum*, *B. ceneria, Rhizopus* sp.) on Pome (Vinas et al. 1998), Stone fruits (Pusey and Wilson 1984), Peach (Wilson et al. 1987), Sweet cherry (Spott et al. 1998), *Prunus persica* var. nectarina and *Prunus persica* (Karabulut et al. 2002).

The addition of Xanthan gum to *A. pullulans*, applied to strawberries against *B. cinerea* increased the bioactivity of bioagent (Ippolito et al. 1998). Certain freeze-drying protective agents and rehydration media enhanced the viability of the antagonist *Pantoea agglomerans*, effective against blue mold and gray mold of pome fruits (Costa et al. 2000).

It has been reported that the capability of *C. sake* to survive a freeze-drying process. They also obtained a dry product with high viability (>80%). Aspire, a product from *C. oleophila*, and Bio-Save, developed from *P. syringae* have been commercialized to control decay caused by *P. italichum* and *P. digitatum..* Avogreen is also commercial product of *B. subtilis* to control diseases caused by *Cercospora* spot and anthracnose of avocado (Janisiewicz and Korsten 2002). Initial results on biocontrol are promising and have been achieved by the application of *B. sphericus, Candida oleophila, C. teneus, Debaryomyces hansenii* against *Alternaria citri*, *Botrytis cinerea, Geotrichum candidum, Penicillium chrysogenum, P. digitatum, P. italicum, Ulocladium* sp. (Sharma 1992; Sharma 1993; Mehrotra et al. 1996; Sharma et al. 1997; Mehrotra et al. 1998; Sharma 2000).

In recent past, several possible biocontrol mechanisms have been recommended as being successful against post-harvest fruit spoilage. These include competition (for nutrients and space), antibiosis, parasitism, induction of resistance in the host tissue and production of volatile metabolites. Information on the mechanisms of action for most of the antagonists is still unfinished because of the lack of information on the complex interactions between the host, the pathogen, the antagonist and the other microorganisms present. Nevertheless, a good understanding of the mechanism of action is essential before developing appropriate formulations and methods of application in order to obtain official approval. In this review information is provided about several mechanisms of biocontrol agents that could provide future biocontrol agents (Jamalizadeh *et al*., 2011).

However, progress has been substantial, the first commercial products have been registered in the United States by the U.S. Environmental Protection Agency and are sold under the names BioSave 100 and 110 and Aspire. Similarly, in South Africa, biocontrol products for the control of fruit diseases are registered [National Department of Agriculture Fertilizer, Farm Feeds, Agricultural and Stock Remedies (Act 36 of 1947)], and sold as Avogreen and Yield Plus.

1.5.2. Effect of other materials on biocontrol

Biological control has advanced greatly during the last few years and microbial antagonists have been reported to control several post-harvest diseases of fruits (Janisiewicz and Roitman 1988; Janisiewicz and Marchi 1992; Wilson and Wisniewski 1994; Vinas et al. 1999). Reliability and cost are two major factors that will determine the feasibility of any biocontrol system (Janisiewicz et al. 1992). Higher concentrations of the antagonist must be applied to achieve a more effective control (Pusey and Wilson 1984; Janisiewicz 1987).

Enhancers are chemicals that serve as a food base for antagonistic microbes or fungicidal to pathogens (Spurr 1994). Therefore, enhancers can be used to manipulate the antagonist populations on fruit and can greatly improve biocontrol levels. Nutritional manipulation has been shown to enhance biocontrol activity of several antagonists. Janisiewicz et al. (1992) reported that L-asparagine and L-proline greatly enhanced biocontrol of *P. syringae* against *P. expansum.* McLaughlin et al. (1990) demonstrated that calcium salts improved the efficacy of yeast biocontrol agents against *P. expansum* and *B. cinerea*. Glycochitosan enhanced the biocontrol activity of *C. saitoana* against control of post-harvest decay on apples and citrus (El-Ghaouth et al. 2000c).

The addition of calcium chloride to the antagonist greatly enhanced biocontrol on apples but less on pears. On apples, the best result was observed with the addition of $CaCl_2$ at 20 mM. Increasing $CaCl_2$ concentration up to 100 mM did not enhance biocontrol more than with 20mM. Wisniweski et al. (1995) reported enhanced biocontrol of *C. oleophila* by adding $CaCl_2$ on apples, but in their study the improvement of biocontrol was achieved with the addition of $CaCl_2$ at more than 90mM. Yeast, *P. guilliermondii* also observed improvement of the biocontrol agent using $CaCl_2$ (Chalutz et al. 1992). Mc Laughlin et al. (1990) found that the ability of $CaCl_2$ to improve biocontrol was dependent on the yeast strain and the concentration of $CaCl_2$ used. Glucose reduced the inhibitory effect of $CaCl_2$ on spore germination of *P. expansum* and *B. cinerea in vitro* (Wisniewski et al. 1995). Concentration of $CaCl_2$ necessary to enhance biocontrol on pears was higher than on apples, pears were at a more mature stage than the apples. The mode of action of $CaCl_2$ at these concentrations could thus base on improved colonization of the antagonist at the wound sit and /or on inhibition of pathogen growth.

Chitosan and its derivatives, including glycolchitosan, were reported to inhibit fungal growth and to induce host-defense responses in plants and harvested commodities (Wilson et al. 1994 and Abadias et al. 2001a). Combining 0.2% glycolchitosan with the antagonist *C. saitoana* was more effective in controlling green mold of oranges and lemons, caused by *P. digitatu*m, and gray and blue molds of apples than either treatment alone (El-Ghaouth et al. 2000a, 2000b). Pretreatment of lemons with sodium bicarbonate further increased control of green mold on the light-green and yellow lemons (El-Ghaouth et al. 2000a). Other fruit coatings may also be useful for further reducing decay when applied with biocontrol agents (Cuppett 1994; Bancroft 1995).

For example, a fruit coating containing sodium salts of carboxy-methyl-cellulose, sucrose esters of fatty acids, and mixed sucroglycerides and soap, commercialized under the name TAL Pro-long, reduced the spread of a range of post-harvest decays of pome fruits (Bancroft 1995). The mechanism of action of TAL-Prolong is not fully understood, but it reduces ripening and extends the natural resistance to invasion by the pathogen in storage. Sucrose esters of fatty acids, and mixed sucro-glycerides and soap, commercialized under the name TAL Pro-long, reduced the spread of a range of post-harvest de-cays of pome fruits (Bancroft 1995). The mechanism of action of TAL-Pro-long is not fully understood, but it reduces ripening and extends the natural resistance to invasion by the pathogen in storage.
Generally regarded as safe (GRAS) substances such as sodium carbonate, sodium bicarbonate, and ethanol reduced conidial germination of *P. digitatu*m, the causal agent of green mold of citrus (Smilanick et al. 1995; Smilanick et al. 1997). Combining treatments of 3%sodium carbonate and the antagonist *P. syringae* ESC-10 was superior to either treatment alone in controlling green mold on citrus (Smilanick et al. 1999). This combination overcomes the significant shortcomings of both individual treatments. The antagonist alone is a poor eradicant and is usually incapable of controlling green mold on fruit inoculated with the pathogen 24 h before treatment with the antagonist. In contrast, the carbonate salts control these infections (Smilanick et al. 1997). Carbonate salts, on the other hand, do not provide persistent protection from re-infection after treatment, whereas the antagonist persists for long periods after application and protects fruit from re-infection. Ethanol at 10%, in combination with ethanol-resistant *S. cerevisiae* strains 1440 and 1749, isolated from wine and ensile acorns, respectively, reduced the incidence of gray mold decay on apples from more than 90% to close to 0%, whereas either treatment alone did not reduce decay (Mari and Carati 1998). The same concentration of ethanol reduced green mold of lemons to less than 5% (Smilanick et al. 1995).

1.5.3. Botanicals

Drawbacks of synthetic chemical methods have increased interest in developing further alternative control methods, particularly those that are environmentally sound and biodegradable. Thus, replacement of synthetic fungicides by natural products (particularly of plant origin), which are non-toxic and specific in their action, is gaining considerable attention. Because of greater consumer awareness and concern regarding synthetic chemical additives, foods preserved with natural additives have become popular. This has led researchers and food processors to look for natural food additives with a broad spectrum of antimicrobial activity (Marino et al. 2001). The plant kingdom represents an enormous reservoir of potential fungicidal compounds that could be useful alternatives to synthetic fungicides. Recently, there have been several attempts to use naturally occurring compounds for the control of post-harvest decay. Plants also produce a variety of essential oils and volatile substances that could have potential as antifungal preservatives for harvested commodities. Both plant essential oils as well as similar compounds in wood smoke have shown promise as natural antimicrobials. Essential (volatile) plant oils occur in edible, medicinal and herbal plants, which minimize questions regarding their safe use in food products. Essential oils and their constituents have been widely used as flavouring agents in foods since the earliest recorded history and it is well established that many have wide spectra of antimicrobial action (Kim et al. 1995; Alzoreky and Nakahara 2002). The advantage of essential oils is their bioactivity in the vapour phase, a characteristic that makes them attractive as possible fumigants for stored product protection.

There have been some studies on the effects of essential oils on post-harvest pathogens (Bishop and Thornton 1997). Some of the essential oils have been reported to inhibit post-harvest fungi in *in vitro* conditions (Sharma 1998; Hidalgo et al. 2002; Sharma and Verma 2004). However, the *in vivo* efficacy and practical activity of only a few of the essential oils have been studied. Some of the essential oils have been reported to protect stored commodities from biodeterioration. There are also some reports on essential oils enhancing the storage life of fruits and vegetables by controlling their fungal rotting (Dubey and Kishore 1988). The potential of essential oils to control postharvest decay has also been examined by spraying and dipping the fruit and vegetables (Tiwari et al. 1988; Dixit et al. 1995). A promising recent development involves incorporating these antimicrobials into packaging materials, rather than the food itself. This concentrates the antimicrobial at the surface of the product, which is where noxious organisms grow and reduces interference from food constituents (Han 2003).

Fumigation of sweet cherries with thymol was effective in controlling post-harvest grey mold rot caused by *B. cinerea* (Chu et al. 1999), and brown rot caused by *M. fructicola* (Chu et al. 2001). Fumigation with thymol at 30 mg l−1 reduced the incidence of grey mold rot from 35% in untreated fruit to 0.5%. Liu et al. (2002) also found that thymol was more effective for controlling brown rot symptoms on apricots, and fumigation of plums with relatively low concentrations such as 2 or 4 mg l^{-1} can greatly reduce post-harvest decay without causing any phytotoxicity. Recently, carvone, a monoterpene, isolated from the essential oil of *Carum carvi* has been shown to inhibit sprouting of potatoes during storage and it also exhibited fungicidal activity in protecting the potato tubers from rotting without altering taste and quality of the treated commodity, and without exhibiting mammalian toxicity (Hartmans et al. 1995; Oosterhaven, 1995). Treatment of oranges with the essential oils of *Mentha arvensis*, *O. canum* and *Zingiber officinale* has been found to control blue mold, thereby enhancing shelf life (Tripathi 2001) Although the fungitoxic properties of the volatile constituents of higher plants have been reported, little attention has been paid to the fungitoxicity of these substances when combined. This information is desirable since the fungitoxic potency of most of the fungicides has been reported to be enhanced when combined (Levy et al. 1986; Pandey and Dubey 1997).

The potential uses of volatile fungicides for the control of post-harvest decay will reside in

1. their safety for human consumption,
2. our understanding of their biological activity and dispersion in harvested tissue, and
3. our ability to develop formulations that allows the delivery of non-phytotoxic concentrations which interfere with fungal developments.

In theory, volatiles that easily diffuse in plant tissue could prove to be useful for the suppression of latent and quiescent infections. In order to accelerate developmental research in natural fungicides, it is imperative that we

1. develop a database of literature pertaining to known plant derived fungicides,
2. devise effective, simple and reproducible bioassay designed to reveal the fungicidal activity of natural compounds and their potential phytotoxicity, and
3. establish structure activity relationship which could help screen promising compounds.

1.6. Integrated Management

Although many alternatives to chemical control have been investigated, none, when used alone, is as effective as fungicides (Conwey et al. 2005). Hot air treatment either reduced or completely eradicated decay of apple fruit caused by *Penicillium expansum* (Leverentz et al. 2000) but the pathogen was not completely eradicated in the case of decay by *Colletotrichum acutatum* (Janisiewicz et al. 2003). Heat treatment, while a good eradicant, has no residual activity (Fallik et al. 1996). The reduction of decay by biological control is generally more variable than for fungicides since biocontrol is affected more by environmental factors. There is also a narrower spectrum of activity than is found with chemical control. Similarly, SBC is not effective in providing protection if fruit are infected after treatment (Smilanick et al. 1999).

Integrating different physical control options such as radiation and ultraviolet illumination was found to be effective against fungi sensitive to low gamma doses such as *Colletotrichum* spp. (Barkai-Golan 2001). Combining physical and chemical alternatives has also been extended to combine radiation and fungicide applications. In this case, both dosages could effectively be reduced to provide cumulative protection (Barkai-Golan 2001; Korsten 2006). Integrating hot water treatments with SBC and fungicides have been known to be effective in reducing decay (Conway et al. 2005; Barkai-Golan 2001).

Combining fungicides in natural or edible waxes has also resulted in increased effectiveness of the products compared to using the products on their own. Korsten et al. (1991) also described successful control of mango postharvest diseases when the antagonist was incorporated into the natural waxes applied on the packing line. Shrink or plastic wrapping the fruit after heat treatments has also proven to be an effective integrated approach (Barkai-Golan 2001). Fungicides used at low concentrations when combined with biocontrol agents have been shown to be effective against several postharvest diseases (Korsten et al. 1993).

Biocontrol products could effectively be integrated with chemicals used at lower concentrations or when used with softer chemicals or disinfectants. Combining chemical elicitors such as chitosan with Bacillus subtilis was found to increase the effectiveness of postharvest biocontrol treatments of *Penicillium* spp. on citrus (Obagwu 2003; Obagwu and Korsten 2003). Other combinations such as calcium salts and sodium bicarbonate with biocontrol agents proved similarly effective (Barkai-Golan 2001; Janisiewicz and Korsten 2002). Adding SBC to the heated or antagonist treated fruit had little effect on decay caused by either pathogen, but on non-heated fruit, it slightly reduced decay caused by *P. expansum* (Conway et al. 2004). An increase in control of decay on oranges caused by *Penicillium digitatum* and *Penicillium italicum* occurred when *Bacillus subtilis* antagonists were combined with SBC (Obagwu and Korsten 2003). Combining SBC with another antagonist also improved decay control of *P. digitatum* on oranges and grapefruit (Porat et al. 2003)

The decay of apples caused by *P. expansum* or *C. acutatum*, was reduced by treatment of heat, heat tolerant antagonistic yeasts or heat in combination with either antagonist. Either heat or the antagonists alone reduced decay caused by *C. acutatum*, but a combination of the two was required to completely eliminate decay caused by this pathogen (Conway et al. 2004). When the antagonist specific for *P. expansum* was combined with one specific for *B. cinerea*, the resulting mixture inhibited the development of both type of lesions (Janisiewiez, 1988). In another study, a combination of yeasts resulted in better control of decay caused by *B. cinerea* than either antagonist alone and also reduced the variability in control levels (Guetsky et al. 2001)

Treatment of heat, radiation and SBC can help to eradicate fungal spores at the time of application, but they do little to protect against future infections. In contrast, the antagonist can help in protection against future infection, but do little to eradicate the inborn infections. Thus, these treatments are complementary to one another when applied in combination, and therefore the combinations are more effective than any individual treatment.

Integrating various methods may provide a more durable, consistent, sustainable and practical solution to producers who utilize the hurdle approach to eliminate pathogens. It is without a doubt that a more sophisticated holistic approach to total product management will ensure quality and safety and provide retailers with the desired extended shelf life.

2. Discussion

Substantial amounts of vegetables and fruits are lost to spoilage after harvest. This thrashing can range from 10-50% depending on the product and country. Currently, synthetic chemicals are the primary resources of checking post harvest diseases of vegetables and fruits. Public worry about food safety, though, increased concern to find out the efficient alternatives to unsafe chemical pesticides to control post harvest diseases of perishables. The eventual aim of recent research programmes in this area has been the advancement and assessment of various alternative control strategies to trim down reliance on synthetic fungicides. At present quite a few promising biological approaches that include the natural plant based antimicrobial substances (volatile aromatic compounds, acetic acid, essential oils, jasmonates, glucosinolates, plant extracts and propolis), the application of microbial antagonists (bacteria, fungi, yeasts), the antimicrobial substances from soil (deoxyfusapyrone and fusapyrone) and the natural animal-based antimicrobial substances like chitosan have been advanced to curb the menaces of post harvest diseases in perishables. Compounds that activate host plant defense responses potentially recommend socio-environmentally potent alternative methods for disease control. Amalgamation of the above complementary techniques could well lead to efficient control of post harvest diseases.

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