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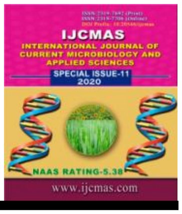

Review Article

Biological Control and its Role in Management of Post Harvest Diseases

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ABSTRACT

Post-harvest diseases cause considerable losses to harvested fruits and vegetables during transportation and storage. Synthetic fungicides are primarily used to control postharvest decay loss. However, the recent trend is shifting towards safer and more eco-friendly alternatives for the control of postharvest decays. Of various biological approaches, the use of antagonist microorganisms is becoming popular throughout the world. Several postharvest diseases can now be controlled by microbial antagonists. Although the mechanism(s) by which microbial antagonists suppress the postharvest diseases is still unknown, competition for nutrients and space is most widely accepted mechanism of their action. In addition, production of antibiotics, direct parasitism, and possibly induced resistance in the harvested commodity are other modes of their actions by which they suppress the activity of postharvest pathogens in fruits and vegetables. Commercial use and application of biological disease control have been slow mainly due to their variable performances under different environmental conditions in the field. To overcome this problem and in order to take the biocontrol technology to the field and improve the commercialization of biocontrol, it is important to develop new formulations of biocontrol microorganisms with higher degree of stability and survival. Future outlooks of biocontrol of plant diseases is bright and promising and with the growing demand for biocontrol products among the growers, it is possible to use the biological control as an effective strategy to manage plant diseases, increase yield, protect the environment and biological resources and approach a sustainable agricultural system.

Keywords

Vegetables, Component, Total production, Post harvest diseases

Introduction

Horticultural crops constitute a significant component of total agricultural production of the country and cover nearly 11.6 million ha area with a total production of over 91 million tonnes per year. The combined annual production of fruits and vegetables is likely to cross 377 million (MT) mark by 2021 from the current level of over 227 MT. Currently over 77MT fruits and about 150 MT vegetables are produced in India and their annual growth rate ranging between 5-6 % respectively (ASSOCHAM 2013).Postharvest decays of fruits and vegetables account for significant levels of postharvest losses. It is estimated that about 20-25% of the harvested fruits and vegetables are decayed by pathogens during postharvest handling even in developed countries (El-Ghaouth *et al.,* 2004; Droby, 2006; Zhu, 2006; Singh and Sharma, 2007). In developed countries, postharvest losses are often more severe due to inadequate storage and transportation facilities. Synthetic fungicides are primarily used to control postharvest diseases of fruits and vegetables (El-Ghaouth *et al.*, 2004; Korsten, 2006; Singh and Sharma, 2007; Zhu, 2006). However, the global trend appears to be shifting towards reduced use of fungicides on produce and hence, there is a strong public and scientific desire to seek safer and eco-friendly alternatives for reducing the decay loss in the harvested commodities (Mari *et al.*,2007). Among different alternatives biological control is best alternative, in which use of microbial antagonists like fungi, bacteria, yeasts are quite promising and gaining popularity (Eckert and Ogawa, 1988; Droby *et al.*, 1991; Wisniewski and Wilson, 1992; Droby, 2006; Korsten, 2006).

Post harvest diseases

The diseases which develop on harvested parts of the plants like seeds, fruits and also on vegetables are the postharvest diseases. The harvested products may get infected on the way to storage or to market or even before their final consumption. The plant parts may get infected in the field, but expression of symptoms may take place later, at any stage before final consumption. The plant products may get infected by microorganisms and cause rotting or decaying by partially or totally. The quantity of plant products becomes reduced due to the above infection. The seeds or grains may get damaged by accumulation of toxic substance, the mycotoxin produced by the infected microorganism.

Classification of Postharvest diseases

Cristensen and Kaufmann (1965) divided the pathogens in to two categories:

Field pathogen

The field pathogens are those, which cause infection during development of plants or their products before harvest.

Storage pathogen

The pathogen which causes infection during storage are the storage pathogen. Symptoms from infection caused by the field pathogen may be very inconspicuous at the time of harvest. In fleshy or juicy fruits and vegetables, infection by field pathogen continues to develop even after harvest.

They may become infected during storage by the same field pathogen(s) or by other pathogen(s).In seeds and grams, the disease caused by field pathogens ceases to develop further soon after harvest. But they may be infected further by other pathogens during storage.

Types of Postharvest diseases

Observations of many investigators indicate that the real cause of the spoilage of vegetables and fleshy fruits in transit and also in storage are due to high moisture, high temperature and injuries caused during marketing. Due to high moisture content and nutrient in harvested vegetables and fruits, they are vulnerable to attack by the pathogenic organisms (Table 1).

Injuries of fruits and vegetables may be caused during harvesting, packing, and transposition they help the pathogen to enter the host and cause damage. But the seeds and grains can be stored for long time due to low moisture content (about12-14%), where most of the pathogens cannot grow favourably. Stackmann and Harrar (1957) divided the pathogenic storage diseases into two categories:

Diseases of stored seeds and grains

Field fungi, like *Alternaria*, *Fusarium*, *Cladosporium*, *Verticillium*, *Helminthosporium*, *Colletotrichum* etc., attack seeds and grains on growing crops, but are unable to grow in storage due to low relative humidity i.e., below 90%. During storage or transit the seeds and grains are damages by the different species of *Aspergillus* and *Penicillium*, which can grow well at a relative humidity range from 70-90%. The commonly available *Aspergillus* species are *A. repens*, *A. ruber*, *A. flavus*, *A. Candidus* etc. *Aspergillus* and a number of other storage fungi invade the embryo of the seeds and grains and they discolour the embryo or seeds as a whole, thereby the germination percentage reduces markedly.

In some cases, spoilage of stored grains and seeds results in drastic increase of temperature up to 70°C or more, which encourage the growth of different thermophilic and thermotolerant fungi such as *Aspegillus fumigatus*, *Absidia*spp., *Mucorpusillus*, etc. In addition to storage fungi, other microorganisms may grow in or on seeds and accelerate the deteriorate process. During breeding period of insects, the moisture content and temperature of seeds increase, thereby rapid growth of the pathogen takes place producing enormous amount of spores. During storage, the fungi produce mycotoxins that cause great damage to both domestic animals and human beings. The important fungi in this respect are *Aspergillus* and *Penicillium*, which produce aflatoxin and other toxins.

Diseases of vegetables and fruits

Different members of Ascomycotina and Deuteromycotina cause the major postharvest diseases of fruits and vegetables which are as below:

Alternaria

Different species of *Alternaria* cause rot of many fresh fruits and vegetables, e.g., black rot of orange, tuber rot of potato, sweet rot of sweet potato, purple blotch of onion, *Alternaria* rot of onion, *Alternaria* rot of cabbage, etc.

Botrytis

It causes grey mold rots of fruits like pear and citrus etc., and vegetables like onion, tomato etc. Every year it causes great economic loss.

Fusarium

It causes different diseases, commonly called pink or yellow molds. Different species of *Fusarium* cause damage to tubers, bulbs, storage roots etc and frequently cucurbits etc. It also causes brown rot of fruits like lemon, orange etc.

Penicillium

Species of *Penicillium* are commonly called blue or green molds, these cause rots of different fruits like onion, sweet potato etc. They also cause spots on different fruits. Under storage, the spotted fruits bear tufts of spores. Though most of the *Penicillium* species prefer relatively high temperature for their growth in storage, they still remain active near freezing temperature at a slow rate. A few species produce ethylene which increase respiration of fruits, thereby it reduces the storage life of the fruits. It also produces patulin, a mycotoxin which directly contaminates the sauces and fruit juices prepared from infected partly rotten fruits.

Sclerotinia

It infects different fruits and vegetables. Most common diseases are cottony rot of lemon, watery soft rot of bean pods, cucurbits etc. Storage diseases like bacterial soft rot of vegetables such as onion, carrot, potato etc, are mainly carried out by different species of *Erwinia*, such as *E. carotovora*,*E. chrysanthemi*etc.

Causes of postharvest diseases

Losses due to postharvest disease may occur at any time during postharvest handling, from harvest to consumption. When estimating postharvest disease losses, it is important to consider reductions in fruit quantity and quality. Losses due to post harvest disease are affected by a great number of factors including:

Commodity type

Cultivar susceptibility to postharvest disease

The postharvest environment (temperature, relative humidity, atmosphere composition, etc)

Produce maturity and ripeness stage

Treatments used for disease control

Produce handling methods

Postharvest hygiene

Virtually all postharvest diseases of fruits and vegetables are caused by fungi and bacteria. In some root crops and brassica’s, viral infections present before harvest can sometimes develop more rapidly after harvest. In general, however, viruses are not an important cause of postharvest disease. Postharvest diseases are often classified according to how infection is initiated. The so-called quiescent or latent infections are those where the pathogen initiates infection of the host at some point in time (usually before harvest), but then enters a period of inactivity or dormancy until the physiological status of the host tissue changes in such a way that infection can proceed. The dramatic physiological changes which occur during fruit ripening are often the trigger for reactivation of quiescent infections. Examples of postharvest diseases arising from quiescent infections include anthracnose of various tropical fruit caused by *Colletotrichium* spp. and grey mold of strawberry caused by *Botrytis cinerea.*

The other major group of postharvest diseases are those which arise from infections initiated during and after harvest. Often these infections occur through surface wounds created by mechanical or insect injury. Wound need not be large for infection to take place and in many cases may be microscopic in size. Common postharvest diseases resulting from wound infections include blue and green mold (caused by *Penicillium* spp.) and transit rot (caused by *Rhizopusstolonifer*). Bacteria such as *Erwiniacarotovora* (soft rot) are also common wound invaders (Table 3).

Many pathogens, such as the banana crown rot fungi, also gain entry through the injury created by severing the crop from the plant. Many of the fungi which cause postharvest disease belong to phylum Ascomycotina and Deuteromycotina as already mentioned above 4 (b). Genera within the phylum Basidiomycota are generally not important causal agents of postharvest disease, although fungi such as *Sclerotium rolfsii* and *Rhizoctoniasolani*, which have basidiomycete sexual stages,can cause significant postharvest losses of vegetable crops such as tomato and potato. While diseases caused by these pathogens are primarily field diseases, the development of symptoms often accelerates after harvest. The major causal agents of bacterial soft rots are various species of *Erwinia*, *Pseudomonas*, *Bacillus*, *Lactobacillus* and *Xanthomonas*. Bacterial soft rots are generally of less importance postharvest diseases of many vegetables, although they are generally of less importance in most fruit. This is because most fruit have a low pH which is inhibitory to the majority of bacterial plant pathogens.

A number of strategies are currently being employed to manage and control postharvest diseases. Losses due to diseases in the field, storage, as well as in transit and market can amount up to 25% of the total production in industrialized and in developing countries damage is often higher, exceeding 50%, because of the lack of adequate storage facilities (Nunes, 2010). There are two principal factors which make plant products more susceptible to spoiling: the high water content in fruit which allows pathogen attack (Harvey, 1978) and the wounds present on the plant organs during storage, often as a resultof harvesting and transportation. Synthetic fungicides are primarily used to control postharvest diseases (Sharma *et al.*, 2009).

However, the use of postharvest fungicides is being increasingly limited because of environmental and toxicological risks. Moreover, the global trend appears to be shifting towards reduced use of fungicides on produce and hence, there is a strong public and scientific desire to seek safer and eco-friendly alternatives for reducing the decay loss in the harvested commodities (Mari *et al.*, 2007). In addition, the repeated and continuous use of fungicides has led to the development of fungal strains (Brent and Hollomon, 2007).

In the last few years, biological control of postharvest disease of fruits has been developed as a promising alternative to chemical control.

Biological control

The term biological control and its abbreviated synonym biocontrol have been used in different fields of biology. In plant pathology, the term applies to the use of microbial antagonists to supress diseases as well as the use of host-specific pathogens to control weed populations (Cook, 1993).

The organism that supresses pathogen is referred to as the Biological Control Agent (BCA). Different modes of action in which the competition for nutrient and space between the pathogen and the antagonist is considered as the major modes of action by which microbial agents control pathogens causing postharvest decay (Droby *et al.*, 1992; Ippolito *et al.*, 2000; Jijakali *et al.*,2001).

In addition, production of antibiotics (antibiosis), direct parasitism and possibly induced resistance are other modes of action of the microbial antagonists by which they suppress the activity of postharvest pathogens on fruits and vegetables (Janisiewicz *et al.*, 2000; Barkai-Golan, 2001; El-Ghaouth *et al.*, 2004)

Competition for space and nutrients

Competition for nutrition and space between the microbial antagonist and the pathogen is considered as the major mode of action by which microbial antagonists suppress pathogens causing decay in harvested fruits and vegetables (Droby *et al.*, 1989; Wilson and Wisniewski, 1989).

To compete successfully with pathogen at the wound site, the microbial antagonist should be better adapted to various environmental and nutritional conditions than the pathogen (Barkai-Golan, 2001; El-Ghaouth *et al.*, 2004).

Competition for space

Rapid colonization of fruit wound by the antagonist is critical for decay control, and manipulations leading to improved colonization enhance biocontrol (Mercier and Wilson, 1994). Thus, microbial antagonists should have the ability to grow more rapidly than the pathogens. Similarly, it should have the ability to survive even under conditions that are unfavourable to the pathogen (Droby *et al.,* 1992). The biocontrol activity of microbial antagonists with most harvested commodities increased with the increasing concentrations of antagonists and decreasing concentrations of pathogen.

For example, *Candida saitona,* was effective at a concentration of 10^7 CFU/ml for controlling *Penicillium expansum* on apples (McLaughlin *et al.,* 1990). In an another study, El-Ghaouth *et al.,*(1998) reported that for *Candidasaitona,* a concentration of 10^8 CFU/ml was better in controlling blue mold (*Penicillium expansum*) on apples. This qualitative relationship however is highly dependent on the ability of the antagonists to multiply and grow at the wound site.

Competition for nutrient

Research work conducted in this mode of action of microbial antagonists supports the hypothesis that competition for nutrients plays a major role in the mode of action of *Pichiaguilliermondii* against *Pencilliumdigitatum* Pers.: Fries) Sacc., in citrus (Droby *et al.,* 1992;). *M.pulcherrima* out competes like *Botrytis cinerea* and *Pencillium expansum* in apple through iron depletion (Saravanakumar *et al.,* 2008). As a result of its ability for supressing postharvest diseases, Kurtzman and Droby (2001) and Grebenisan *et al.,* (2008) have recommended it as potential yeast for controlling fruit rots. Further, non-pathogenic species of *Erwinia,* such as *E. cypripedii* (Hori) Bergey, showed antagonistic activity against various isolates of *Erwiniacaratovora sub sp. caratovora*. The causal agent of soft rot of many vegetables like carrot, tomatoes (*Lycopersicon esculentum* L.) and pepper (*Capsicum annuum* L.), primarily by competing for nutrients (Moline *et al.,* 1999). It has been demonstrated through in vitro studies that microbial antagonists take up nutrients more rapidly than pathogens, get established and inhibit spore germination of the pathogens at the wound site (Wisniewski *et al.,* 1989; Droby *et al.,* 1998).

In general, microbial antagonists are most effective in controlling postharvest decay on fruits and vegetables when applied at concentration of 10^7-10^8 CFU/ml (McLaughlin *et al.,* 1980; El-Ghaouth *et al.,* 2004), and rarely, higher concentrations are required.

Antibiosis

Production of antibiotics is the second important mechanism by which microbial antagonists suppress the pathogens of harvested fruits and vegetables. For instance, bacterial antagonists like *Bacillus subtilis* and *Pseudomonas cepacia* Burkh are known to kill pathogens by producing the antibiotic iturin (Gueldner *et al.,* 1998; Pusey,1998). The antagonism produced by *Bacillus subtilis* was effective in controlling fungal rot in citrus (Singh and Deverall, 1984).

Further *Pseudomonas cepacia* inhibited the growth of postharvest pathogens like *Botrytis cinerea* and *Pencillium expansum* in apple by producing an antibiotic, pyrrolnitrin (Janisiewicz and Roitman, 1998). *Pseudomonas cepacia* was also effective in controlling green mold(*Pencillium digitatum*) in lemon (*Citrus limon* L.) by producing antibiotics.

Table.1 Common post harvest diseases in fruits and vegetables

Commodity	Disease	Pathogen
Apple	Pencillium rot	*Pencilliumexpansum*
Strawberry	Graymold	*Botrytis cinerea*
Banana	Crown rot	*ColletotrichumMusae*
Tomato	Rhizopus rot	*Rhizopus nigricans*
Chilli	Anthracnose	*Colletotrichum capsica*
Grape	Graymold	*Botrytis cinerea*
Pear	Rhizopus rot	*Rhizopus stolonifera*
Tomato	Alternaria rot	*Alternaria alternate*

Table.2 Post harvest losses of fruits and vegetables

Region and country	Commodity	Losses (%)
Kenya	Banana	11.2-50
Tanzania	Sweet potato	32.5-35.8
Cambodia Loas	Yard-longbean	12.2-21.8
Loas Vietnam	Chilli pepper	10.7-16.9
Bangladesh	Litchi	8.0
Pakistan	Tomato Potato Onion	20 12 9.0
Srilanka	Banana	20
Iran	Grapes	13
Ghana	Tomato	20
Egypt	Oranges Tomatoes	14 15
Egypt	Pomegranate Onion	23 19
Jordan	Tomato Eggplant Pepper Squash	18 19.4 23 21.9
Srilanka	Tomato	54
Saudi Arabia	Tomato Cucumber Figs Grapes Dates	17 21.3 19.8 15.9-22.8 15
Cambodia Loas Vietnam	Tomato	24.6 16.9 19.1
Nigera	Tomato Bell pepper Hot pepper	20-28 12-15 8-10

Table.3 Post harvest losses of fruits and vegetables in India

Fruits and Vegetables	Losses (%)
Mango	20-26
Banana	18.3-28.8
Grapes	14.4-21.3
Pomegranate	35.4
Potato	10.5-19.8
Tomato	11-35
Bell Pepper	6.7-17.1
Cabbage	9.4-30.4
Onion	12-30
Cucurbits	52
Cauliflower	12.9-35.1
Citrus	27
Litchi	30
Okra	31
Guava	20

Table.4 Microbial antagonists used for the successful control of postharvest diseases of fruits and vegetables

Antagonists	Disease(pathogen)	Fruits/Vegetables
Candida olephila	Penicillium rot (*Penicilliumexpansum*)	Apple
Bacillus subtillis	Graymold (*Botrytis cinerea*)	Strawberry
Pichiaguillermondii	Crown rot (*Colletotrichummusae*) Rhizopus rot (*Rhizopus nigricans*)	Banana Tomato
Metschnikowia Pulcherrima	Blue mold (*Penicilliumexpansum* Graymold (*Botrytis cinerea*)	Apple
Candida olephila	Graymold (*Botrytis cinerea*)	Tomato
Pichiaguillermondii	Alternaria rot (*Alternariaalternata*)	Tomato
Trichodermaharzianum	Graymold (*Botrytis cinerea*)	Pear
Cryptococcus laurentii	Brown rot (*Monilinafructicola*)	Cherry
Pichiaguillermondii	Anthracnose (*Colletotrichumcapsici* (Syd.) Butler &Bisby	Chillies

Table.5 Commercially available bioproducts for control of post harvest diseases

S.No	Microorganism	Product name	Target pathogens	Fruit	Country
1.	*Aureobasidium pullulans*	Boniproduct	*Penicillium* *Botrytis* *Monilinia*	Pome fruit	Europe
2.	*Bacillus subtilis*	Avogreen	*Cercospora* *Colletotrichum*	Avocado	South Africa
3.	*Candida olephile*	Nexy	*Botrytis* *Penicillium*	Pome fruit	Belgium, EU
4.	*Pseudomonas syringae*	Biosave	*Penicillium* *Botrytis* *Mucor*	Pome Citrusfruit Cherry Potato Sweet potato	United States
5.	*Trichodermaharzianum T-39*	Trichodex	*Botrytis cinerea*	Most of food crops	Bioworks,USA

Although, antibiosis might be an effective tool for controlling postharvest diseases in a few fruits and vegetables, at present emphasis is being given for the development of non-antibiotic producing microbial antagonists for the control of postharvest diseases of fruits and vegetables (El-Ghaouth *et al.,* 2004; Singh and Sharma, 2007).

Researchers are aiming to isolate, evaluate or to develop those antagonistic microorganisms that control postharvest diseases of harvested commodities by the mechanism of competition for space and nutrient, direct parasitism or induced resistance (Droby, 2006).

Parasitism

Parasitism or predation occurs when the antagonist feeds on or within the pathogen, resultingin a direct destruction or lysis of propagules and structure (Bull *et al.,* 1998. Wisniewski *et al.,* (1991) observed that *Pichiaguilliermondii* cells had the ability to attach to the hyphae of *Botrytis cinerea and Pencillium*. After yeast cells were dislodged from the hyphae, the hyphal surface appeared to be concave and there was partial degredation of cell wall of *Botrytis cinerea* at the attachment sites.

Microbial antagonists also produce lytic enzymes such as gluconase, chitinase and proteinases that help in the cell wall degradation of the pathogenic fungi (Chernin and Chet, 2002).

Bonaterra *et al.,* (2003) reported that direct parasitism was a major factor that permitted *Pantoeaagglomerans* (Ewing & Fife) to control *Monilinialaxa* (Aderh. & Ruhl).Honey or *Rhizopus stolonifer* decay on stone fruits.

Thus, strong attachment of microbial antagonist with enhanced activity of cell wall degradation enzymes may be responsible for enhancing the efficacy of microbial agents in controlling the postharvest diseases of fruits and vegetables (Wisniewski *et al.,* 1991). And, attachment of the microbial antagonists to a site enhances their potential activity for the utilization of nutrients at the invasion site; it partly affects the access of the pathogen to nutrients as well (El-Ghaouth *et al.,* 2004).

Induced resistance

Induction of defense responses in the harvested fruits and vegetables by the microbial antagonists has been suggested and is another mode of action of microbial antagonists for controlling postharvest decay in them (El-Ghaouth *et al.*, 1998; Ippolito *et al.*, 2000). For example, *Cyptococcus saitona* induced chitinase activity and formed structural barrier (papillae) on host cell walls in apple against *Penicillium expansum* (El-Ghaouth *et al.*, 1998). Similarly, *Aureobasidium pullulans* caused a transient increase in the activity of 1, 3-gluconase, peroxidase and chitinase enzymes in apple wounds which stimulated wound healing processes and induced defense mechanism against *Penicillium expansum* (Ippolito *et al.*, 2000).

Microbial antagonists induced disease resistance in the harvested commodities by the production of antifungal compounds, as in avocado (*Persea americana* Mill) fruit (Yakoby *et al.*, 2001, and accumulation of phytoalexins, like scoparone and scopoletin in citrus fruits (Rodov *et al.*, 1994). Production of such antifungal compounds by microbial antagonists in the host cells help in inducing defense mechanism and hence provide biocontrol on the harvested commodities.

Characteristics of an ideal antagonist for the post harvest environment

Genetically stable

Effective at low concentrations

Not fastidious in its nutrient requirements.

Ability to survive adverse environmental conditions (including low-temperature and controlled-atmosphere storage).

Effective against a wide range of pathogens on a variety of fruits and vegetables.

Amenable to a formulation with a long shelf life.

Easy to dispense.

Does not produce metabolites that are deleterious to human health.

Resistant to pesticides.

Compatible with commercial processing procedures.

Non-pathogenic to lost commodity.

Basic approaches for using the microbial antagonists

There are two basic approaches for controlling the postharvest diseases of fruits and vegetables which are as:

Natural microbial antagonists

Natural occurring antagonists are those, which are present naturally on the surface of fruits and vegetables, and after isolation, antagonists are used for the control of postharvest diseases (Janisiewicz, 1987; Sobiczewski *et al.*, 1996). Chalutz and Wilson (1990) found that when concentrated washings from the surface of citrus fruit were plated out on agar medium, only bacteria and yeast appeared while after dilution of these washings, several rot fungi appeared on the agar, suggesting that yeast and bacteria may be suppressing fungal growth.

Thus, it indicates that when fruits and vegetables are washed, they are most susceptible to decay than those, which are not washed at all.

Artificially introduced microbial antagonists

Although the first reported use of a microbial was the control of Botrytis rot of strawberry (*Fragaria x ananassa* Duch.) with *Trichoderma* spp. (Tronsmo and Denis, 1977), the first classical work was the control of brown rot of stone fruits by *Bacillus subtillis*. Since then, several antagonists have been identified, and used for controlling postharvest diseases of different fruits and vegetables.

Several microbial antagonists have been identified and artificially introduced on a variety of harvested commodities including citrus, pome, and stone fruits, and vegetables for control of postharvest diseases. *Trichodermaharzianum* Rifai has been effective in controlling anthracnose in banana (Devi and Arumugam, 2005) and rambutan (*Nepheliumlappaceumm* L.) and graymold in grapes, kiwifruits and pears (Batta, 2007).Several microbial antagonists have been patented and evaluated for commercial use, of which ASPIRE, YieldPlus andBIOSAVE-110 are used worldwide for controlling postharvest diseases of fruits and vegetables effectively.

Application methods of microbial antagonists

Generally microbial antagonists are applied by two different ways which are given as:

Preharvest application

In several cases, pathogens infest fruits and vegetables in the field, and these latent infections become major factor for decay during transportation or storage of fruits and vegetables. Therefore, preharvest application(s) of microbial antagonistic culture are often effective to control postharvest decay of fruits and vegetables (Ippolito and Nigro, 2000). The purpose of preharvest application is to pre-colonize the fruit surface with an antagonist immediately before harvest so that wounds inflicted during harvesting can be colonized by the antagonist before colonization by a pathogen (Ippolito and Nigro, 2000).

Although this approach could not become commercially viable, because of poor survival of microbial antagonists in the field conditions, however it has been quite successful in certain cases.

Candida sake CPA-1 reduced blue mold by nearly 50% on wounded apples if the apples were inoculated with antagonist 2 days before harvest and inoculation with *Penicillium expansum* and cold storage for 4 months. Similarly, preharvest application(s) of *Cryptococcus laurentii* and *Candida olephila* reduced storage rots in pear. Field application of *Epicoccumnigrum* was reported to be an effective for controlling postharvest brown rot (*Monilina* spp.) in peaches.

Post harvest application

Postharvest application of microbial antagonists is a better, practical and useful method for controlling postharvest diseases of fruits and vegetables. In this method, microbial cultures are applied either as postharvest sprays or as dips in an antagonist´s solution (Barkai-Golan, 2001). This approach has been more effective than preharvest application of microbial antagonists, and has several successes. For example, postharvest application of *Trichoderma harzianum*, *Trichoderma viride*, *Gliocladium roseum* and *Paecilomyces variotii* Bainier resulted in better control of *Botrytis* rot in strawberries and *Alternaria* rot in lemons than preharvest application(s) (Pratella and Mari, 1993) (Table 4 and 5).

Future prospects

Little attention has been paid to produce the commercial formulation of bio agents

The biocontrol concept should be popularized at University level

Genetic engineering and other molecular tools should offer a new feasibility for improving selection and characterization of biocontrol

Need of mass production, understand their mechanism and also evaluate the environmental factors that favour the rapid growth of bio agents

People turning more health conscious biocontrol seems to be best alternative to disease suppression

Bioagents bring the disease suppression with no environmental hazards

Bioagents needs to be formulated that favour the activity survival of microbe containing in it.

Significant advances and commercially available products shall be made available for postharvest use in future

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