

**MANAGEMENT OF MAJOR PRE AND POSTHARVEST FUNGAL DISEASE (ANTHRACNOSE) OF MANGO FRUITS – A REVIEW**

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**Abstract** Mango, *Mangifera indica* L. having family “Anacardiaceae”, is an important fruit in Physical and Biological Sciences. It is a prominent tropical and subtropical fruit produced all over the world. It is a popular fruit in the national as well as an international market due to its exquisite flavor and high nutritional caloric value. However, mangoes are susceptible to a variety of illnesses at all phases of growth, from the plants in the nursery to the fruits in storage or transit. The mango tree, and especially the fruit, is home to a wide range of infections, including fungus, which may be important causes of fruit rot. *Colletotrichum* species cause anthracnose, a serious disease affecting various crop species, causing fruit blemishes and financial losses. Historically, physical treatments and synthetic fungicides were used, but research into sustainable protection strategies is needed to reduce hazardous chemicals. Pre-harvest and postharvest infections can impair fruit quality and result in significant losses. The visible exterior symptoms frequently appear after ripening, which is commonly associated with the edibility of the fruit and causes significant losses during storage. The economic consequences of such postharvest losses exceed the field losses. So far, chemical-based treatments have dominated mango disease control. However, because of growing worry over residual toxicity caused by the widespread use of synthetic fungicides and the emergence of resistance in pathogen populations, attention has shifted to nonchemical techniques. Biological agents and vegetal derivatives have shown effectiveness, but crucial problems must be resolved before they can be integrated into practical crop protection plans. Biotechnology-based strategies, including RNA interference and genome editing technologies, have shown potential for secure anthracnose control. However, despite their high specificity and potential side effects, several issues need to be addressed before they can be integrated into IPM procedures to prevent *Colletotrichum* spp. disease.

**Keywords:** Mango, anthracnose, *Colletotrichum*, pre and post-harvest, quality, management

### Introduction

One of the most widely consumed fruits is mango (*Mangifera indica* L.) in the world's tropics and subtropics (Durán Zuazo *et al.*, 2019). The fruit's high-calorie content and exceptional flavor make it a highly sought-after choice globally (Spina *et al.*, 2024). India dominates mango production, accounting for 56% of the global total, alongside China, Mexico, Thailand, Indonesia, Pakistan, Nigeria, Philippines, Brazil, Egypt, and Haiti (Mitra, 2014). Mangos are a significant part of the diets of all over the nations, providing flavor and improving living standards in areas with severe nutritional

deficiencies and inadequate living standards (Sanchez Garcia, 2024). However, a variety of diseases or insect pest infestations in mango production at every stage of growth, from the seedling, growth, development, storage as well as transportation. Field diseases cause crop losses, while postharvest infections directly contribute to losses in both the local and export markets (Atanda *et al.*, 2011). According to Mukherjee and Litz (2009), mangoes flavor, taste, and appeal have also improved the quality of life in areas of the world where nutritional shortages and low-income living

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settings. Crop losses are caused by field illnesses, whereas losses in the domestic and export markets are directly related to pre and postharvest infections (Singh *et al.*, 2021; Abbas *et al.*, 2024ab).

Fruits especially mangoes are highly susceptible to pathogenic fungi due to their high moisture content and nutritional value, making them highly perishable and requiring immediate sale. Postharvest losses can range from 17% to 36% (Hailu and Derbew, 2015). Fruit rot, caused by infections like fungus, is a global issue, particularly in mango trees and fruit. To reduce losses, the primary method is to prevent these illnesses, using chemicals for prevention and management, but residues can pose health and environmental risks (Abd Murad and Zainudin, 2017). Therefore, controlling and severity of disease below the threshold while adhering to environmentally safe, financially feasible, and conveniently operable processes is the challenge facing mango production, and disease control in particular. The integrated disease management (IDM) strategy aims to control postharvest disease of mango fruit by utilizing various conventional and modern techniques and procedures, rather than relying solely on fungicides (Shukla *et al.*, 2020; Abbas and Rehamn 2021). This review paper focuses on the etiology of disease, the conducive weather conditions, and control methods.

### **Major postharvest disease of mango (Mango Anthracnose)**

#### **Occurrence and Economic Importance**

The most dangerous disease that is prevalent in all mango planting locations of anthracnose, which is brought on by *Colletotrichum gloeosporioides* (Penz). It significantly hinders the growth of the mango export industry (Dofuor *et al.*, 2023). In every country where mangoes are cultivated, this disease is the main factor restricting fruit output. It is particularly prevalent in areas with high humidity throughout the cropping season when the disease prevalence can approach 100% (González-Fernández and Hormaza, 2020). Any stage of fruit growth can be affected by the disease (Jeevanantham *et al.*, 2024). The infection is usually caused by deep necrotic (premature death of cells) lesions on various organs, such as flowers, leaves, shoots, and mummifies immature fruit. Infections frequently result in severe postharvest fruit losses as well as decreased tree vigor and output. The postharvest stage of the disease is the most destructive and economically significant, causing instant marketable fruit loss. The infection typically starts on immature twigs and leaves, progressing to flowers, causing blossom blight, inflorescence destruction, and fruit set prevention (Singh and Sharma, 2018).

#### **Biology and interaction with their Hosts**

There are around 200 fungal species in the genus *Colletotrichum*, which are loosely grouped into

fifteen complexes species (Talhinhas and Baroncelli, 2021; Junaid an Gokce, 2024; Rehamn *et al.*, 2024). Many of them are pathogens of significant crops, causing anthracnose, a serious illness with significant financial consequences. That's why *Colletotrichum* species have been ranked among the top ten fungal diseases in the world (Dean *et al.*, 2012; Bordoh *et al.*, 2020b). Waller (1992) evaluated the prevalence of *C. gloeosporioides* on tropical fruit crops as well as a variety of perennial and other cash crops. Isolates differ in their pathogenicity and host range in addition to their physical characteristics in culture. Colonies on PDA have dense to sparse lawns of aerial mycelium and range in color from pale to dark gray. Conidia are cylindrical, obtuse-ended, hyaline, and single-celled. When they reach maturity, they have an orange, slimy mass and are formed on irregular acervuli similar to conidiophores colour light brown. Conidial acervuline survives for extended lengths of time, even under unfavorable weather circumstances, and acervuline grows in the form of lesions on fruit leaves, and primary branches. The fungus is heterothallic, and while it is easy to induce the teleomorph culture, appressoria, typically lobed, is rarely found in the wild (Chethana *et al.*, 2021; Sami *et al.*, 2023; Arshad *et al.*, 2024). Since plant infections require warm temperatures and high relative humidity to initiate, they are more common in worldwide tropical as well as sub-tropical locations and a smaller amount common in temperate latitudes (Freeman *et al.*, 1998; Tripathy *et al.*, 2021). Wetness of the leaf surface, light intensity, and rainfall duration and intensity have also been found to be favorably connected with the infectious process (Banya *et al.*, 2020). These factors make crops in areas with high precipitation rates that never truly dry out between rainstorms especially vulnerable to *Colletotrichum* infestations. All *Colletotrichum* species exhibit similar interaction with early-stage host tissues when it comes to fruit infection (Wharton and Diéguez-Uribeondo, 2004) Conidiospores, which spread from infected vegetation or insect pests, keep to the external surface via hemicellulose mucilage, germinate, and frequently infect through specialized structures like appressoria (Jeger and Plumbley, 1990; Prusky *et al.*, 2000; Nazzaro *et al.*, 2017; de Oliveira Filho *et al.*, 2021). Infections can occur through various means like stomatal opening, lenticels, lesions, or through scar tissue abscission (Maraite, 1987; Zulfiqar *et al.*, 1996; Nash and Lucas, 1999; Siddiqui, 2018). The process can be intracellular hemi biotrophy or intramural necrotrophy, with specific host-pathogen interaction and infection strategies found in *C. acutatum*, which show four distinct colonization pathways (Peres *et al.*, 2005). *C. capsici* and *C. circinans* are characterized by an early, asymptomatic, 24-hour stage of subcuticular,

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intramural necrotrophy, during which the pathogen develops between the periclinal and anticlinal walls of the epidermal cells (intramural development). These species then swiftly spread throughout the tissues of the host, causing damage (Roberts and Snow, 1984; Pring *et al.*, 1995).

#### Disease Symptom

The most significant or vulnerable mango disease, anthracnose, fungus *Colletotrichum gloeosporioides* is the cause of this (Kamle and Kumar, 2016). Fruit rot, angular leaf spots, and some blossom blight are the symptoms of this disease. Tiny black to dark-brown irregular patches on the panicles are the first sign of an infection. Their development, clumping, and destruction of the blooms can significantly lower yield output. The initial symptoms of anthracnose infections are tiny, angular, brown to black dots on leaves (Bassanezi *et al.*, 2002; Mohammed, 2013; Muntala *et al.*, 2020). Depending on the climatic conditions at the time, blossom blight can range from a small infection to a severe infection of panicles.

Young, infected fruits get black specks, and then they get smaller and fall off. During marketing, shipping, and storage, mature-stage infected fruits cause large losses and transfer the fungus into storage (Lopes and Berger, 2001). Postharvest anthracnose on fruit surfaces is a round, brown-to-black lesion with an ambiguous border. Large fruit illnesses rarely result in lesions, but the fungus settles and remains inactive until the fruit matures. Dark, depressed circular lesions appear and spread out, sometimes encasing the fruit. Tearstains may cover large portions of the fruit (Pehamberger *et al.*, 1987). In many cases, the fungus may even penetrate the flesh, even if it only produces lesions on the peel. As the infection progresses, the fungus produces acervuli, and many conidia, which might be orange or salmon pink in color, form on the lesions (Zhang *et al.*, 2015; Spadaro *et al.*, 2019).



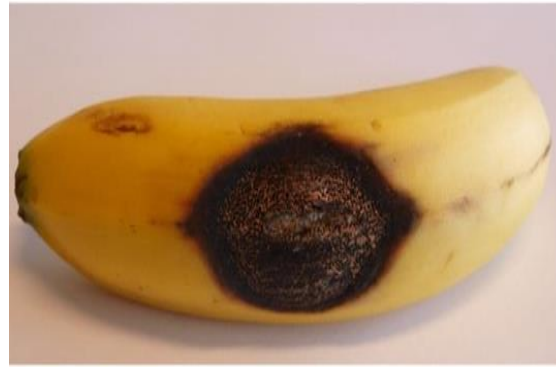
(a)



(b)



(c)



(d)

**Figure 1.** Indications of anthracnose on several fruits (a) Strawberry fruits with sunken brown lesions; (b) pear fruits with dark lesions encircling orange conidia masses; (c) banana fruits with dark lesions; and (d) mango fruits with dark, advanced lesions that resemble circular dots.

Temperature, humidity, precipitation, foggy circumstances, or heavy dews during the blooming period all affect the damage that the anthracnose infection causes. About 25°C is the ideal temperature for anthracnose infection (Moral *et al.*, 2012; Uysal and Kurt, 2017; Morkeliūnė *et al.*, 2021). Serious blossom blight is caused by persistently rainy

conditions during blossoming. It takes 12 hours of relative humidity over 95% for *C. gloeosporioides* to infect and grow on mango fruit. Conidia, however, may endure for one to two weeks at humidity levels as low as 62% until germinating when exposed to 100% relative humidity. Generally speaking, temperatures between 20 and 30°C are ideal for

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infection. Within this range, isolates of *C. gloeosporioides* from various locales varied significantly in the ideal temperature needed for germination and appressorium development (Arauz, 2000). In ripe fruits and injured tissues, infection spreads more quickly (Singh and Sharma, 2018).

Depending on the plant tissue, anthracnose symptoms can vary greatly and result in significant losses in fruit production (de Oliveira Filho *et al.*, 2021). For example, strawberry plants, which are vulnerable to several *Colletotrichum* species, might show severe signs of anthracnose in every area of their body (Smith, 2008; Higuera *et al.*, 2019). In nurseries and the field, anthracnose can cause up to 80% and over 50% of plant and fruit losses, respectively, for this crop (Higuera *et al.*, 2019; Sreenivasaprasad and Talhahas, 2005; Marian *et al.*, 2020). Generally speaking, symptoms on stems or leaves vary from darker patches on branches to tiny greyish-brown sunken areas. In contrast, anthracnose produces black lesions around pink or orange conidia masses on fruits, which can progress into tissue necrosis under the right circumstances (Freeman *et al.*, 1996; Saxena *et al.*, 2016). Infected blooms, on the other hand, seem dry and rotten with impeded fruit development. In any event, fruit destruction during postharvest storage is the disease's greatest impact.

In this case, symptoms may lead to persistent deterioration and a decline in aesthetic and quality standards, which might result in significant financial losses (Arauz, 2000; Lakshmi *et al.*, 2011). For example, anthracnose has a significant impact on banana fruits, causing losses of up to 30–40% of the marketable crops (Maqbool *et al.*, 2010). The disease's harm even spreads to other vulnerable hosts. In this regard, *Colletotrichum* spp. significantly severely reduce the papaya's shelf life in India, the world's largest producer papaya fruit (Tripathy *et al.*, 2021), resulting in losses of up to 93% (Paull *et al.*, 1997; Darshan *et al.*, 2019). Similar to this, anthracnose is a serious post-harvest disease that affects mango fruits, particularly in areas where the climate favors infections, and it can affect up to 100% of output (Arauz, 2000). The primary cause of post-harvest anthracnose is field infections that go dormant until circumstances that are conducive to pathogen germination and growth occur (Lakshmi *et al.*, 2011; Freeman and Shabi, 1996; Nelson, 2008). Because climacteric fruits like bananas, guavas, avocados, pears, mangoes, and papayas undergo physiological and biochemical changes during the ripening stage as a result of cell wall re-modelling and degradation, the disease is especially severe for these fruits (Botelho *et al.*, 2016; Uluisik and Seymour, 2020). However, even for non-climateric crops like dragon fruit, strawberries, and citrus, anthracnose poses a

significant threat (Masyahit, 2009; Deng *et al.*, 2015; Martínez-Blay *et al.*, 2020).

#### **Disease Cycle of anthracnose**

According to Arauz, 2000 infected and defoliated branch terminals, mature leaves, blighted peduncles, dead stems, and sick twigs linked to the trees are the pathogen's primary sources of survival (Figure 2). The pathogen grows on young tissues and immature fruits. Spores germinate and pierce the epidermis and cuticle to spread throughout the tissues. Infections pierce the cuticle of mature fruits but do not develop until the climacteric fruits start to ripen (Ando, 2009; Singh *et al.*, 2022). When circumstances are right, it generates spores, which act as infection centers for the next bloom. As the fungus continues to grow throughout the season, several disease cycles may arise (Peritore, 2020). Tropical circumstances, however, allow for the year-round production of new spore supplies. According to studies on viability, 70% of the fungus's spores that were formed in acervuli on the twigs were still alive. The fungus survived for 14 months on damaged leaves (Prakash and Misra, 1993; Prakash and Misra, 1999).

#### **Management of Mango Anthracnose**

Field management, postharvest treatments, or ideally a mix of both can be used to control postharvest anthracnose. Management plans need to be economical, efficient, and safe for the environment, agricultural workers, and customers (Coglianese and Lazer, 2003; Robertson and Swinton, 2005). Any management plan used to control mango anthracnose should concentrate on both preharvest as well as post-harvest management. The most significant alternative and environmentally friendly strategies that have been recently investigated to combat anthracnose will be reviewed in this review after a brief overview of the traditional methods used to manage the disease, with an emphasis on their potential use at the pre as well as the post-harvest stage. We will pay special attention to new biotechnology-based approaches that have minimal risk of adverse environmental and human health impacts and high specificity (Figure 3).

#### **Pre-harvest Measures**

##### **Cultural control**

The prevalence of *C. gloeosporioides* inoculum sources and the disease's propensity for spreading quickly in the right circumstances make many conventional phytosanitary measures less effective. Examples of effective field management of *Colletotrichum* infections that are impacted only by actions meant to reduce inoculum sources are few, even though general orchard cleanliness has a position in integrated disease control. Enhanced understanding of the pathogen strains' specificity might facilitate the development of efficient phytosanitary measures (Catara *et al.*, 2021). Since the development of the disease depends on wetness

and high relative humidity, orchards should ideally be established in areas with a clearly defined dry season to enable fruit growth in environments that are not favorable to disease development (Berrie, 2019). Anthracnose can be controlled by adjusting the blooming timing for fruit set and growth and development take place during dry circumstances, which concentrates on off-season output during lucrative market windows. A potential disease prevention tactic in tropical regions is to control blooming such that fruit develops during the season with the least amount of rainfall (Nath et al., 2019;

Bhattacharjee et al., 2022). Even in the absence of any management measures, Throughout the dry season, fully ripe mangoes can have a very low prevalence and severity of mango anthracnose. Proper watering and fertilizer administration are crucial to maintaining tree vigor since plant vigor is crucial in preventing twig infection (Tewari, 2008; Mathews et al., 2014).

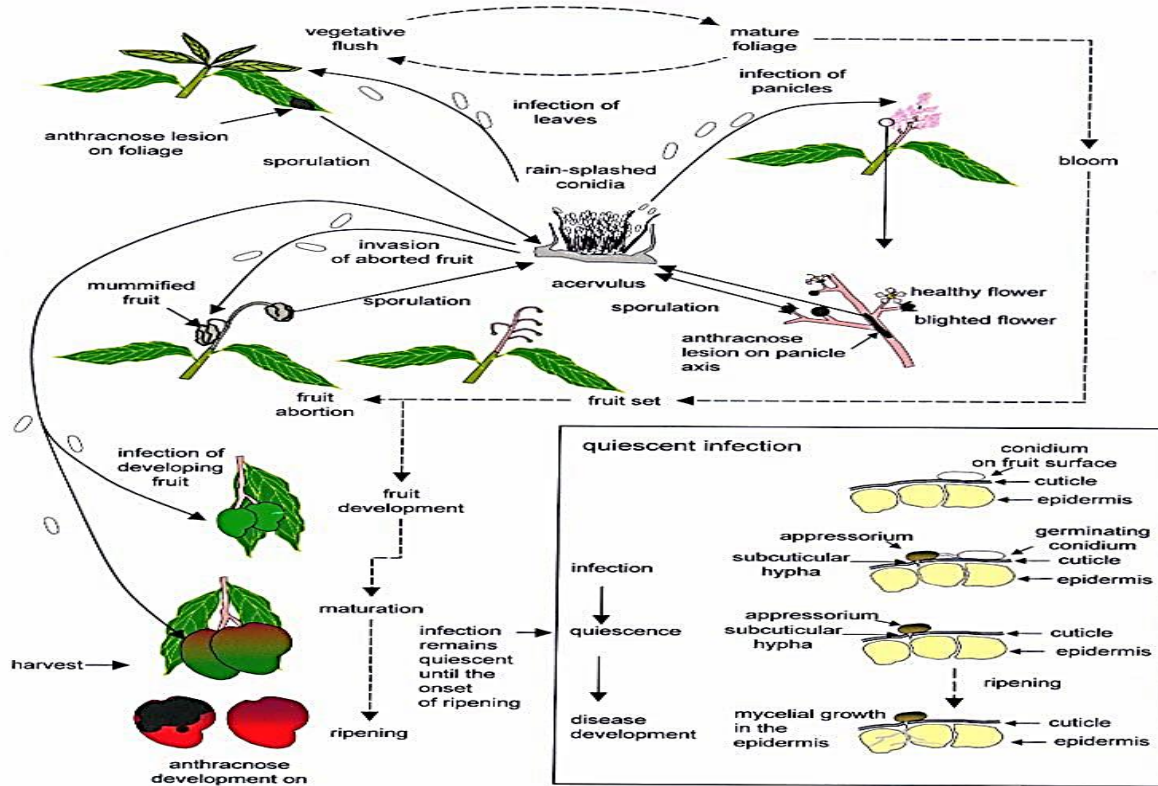


Figure 2. Anthracnose Disease Cycle (Arauze, 2000)

**Resistant varieties**

Mango anthracnose has not been consistently controlled by resistance. This is partially due to cultivars' varying responses to the disease depending on the locale. Anthracnose can affect any commercial mango cultivar, however, some are more or less vulnerable than others (Thiribhuvanamala et al., 2017). Compared to cultivars like Irwin, Kent, or Edward, Tommy Atkins and Keitt are less vulnerable. None of the cultivars now in production are highly resistant to being cultivated in a humid climate without the use of certain fungal treatments (Dodd et al., 1997). Even in the less picky local markets, the amount of anthracnose that appears in less susceptible cultivars like Tommy Atkins under severe disease pressure is still undesirable in a commercial setting due to market beauty requirements (Arauz, 2000).

**Biological control**

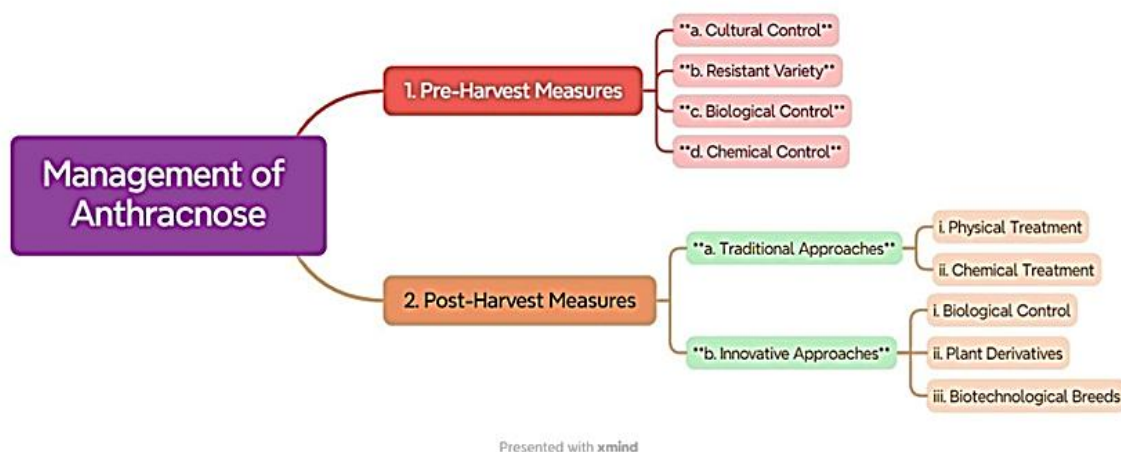
This method suppresses plant diseases by employing microorganisms. By using current resources, biological control aims to boost agricultural yields while preventing the emergence of chemically resistant infections. The biological regulation of anthracnose has not been extensively studied. With the longest experience in this field, Lise Korsten's team has concentrated on employing the food-safe, gram-positive *Bacillus licheniformis* bacteria, which is resistant to desiccation. Either by itself or in conjunction with fungicides, 10 - 25 °C often results in slight decreases in disease (Govender and Korsten, 2006). Significant decreases in Gram-negative bacteria and other modifications have also been seen but with less publicity (Vivekananthana et al., 2004).

**Chemical control**

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The effectiveness of the products, the restricted number of products on the market, and the pesticide laws in the countries of production and destination all limit the usage of fungicides. Copper fungicides are often the most widely accepted (Lamichhane *et al.*, 2018; Tamm *et al.*, 2022). According to Ploetz (2009), during periods of strong disease pressure and phytotoxicity, copper fungicides are frequently less effective on mangos. In the field, anthracnose was well controlled by the fungicides febran, mancozeb,

and dithiocarbam (Akem, 2006). Mango fungicides with after-infection action include prochloraz, timidazole, and benzimidazoles. Benomyl has been used in calander-based spray regimens to delay the emergence, frequently in conjunction with protectant fungicides resistance in the pathogen population. After infection periods, it has also been used to get rid of spray (Arauz, 2000). Prochloraz has been utilized as an eradicator spray or as a protective agent.



**Figure 3.** Strategies for managing pre and post-harvest anthracnose. Both conventional tactics and new techniques were investigated to enhance sustainability in the agri-food industry

**Postharvest measures**

Reducing the fruit's quiescent infection level has been the main objective of postharvest control of mango anthracnose (Arauz, 2000). Although plant-to-plant transmission of anthracnose occurs before or after harvest, postharvest prevention of latent infection is often necessary and used, especially if the fruit is to be kept or transferred to a different area (Dodd *et al.*, 1997). This study updates recent alternative and environmentally friendly strategies for combating anthracnose, focusing on biotechnology-based solutions with high specificity and minimal risk of adverse environmental and human health consequences, especially at the post-harvest stage.

**Traditional Approaches**

**Physical method of control**

They use eco-friendly techniques such as hot water dips of harvested things, forced-air dry heat, and vapor heat that have been developed to prevent *Colletotrichum* growth. However, because they may influence the quality of the fruit, Hot water dips, for instance, have been linked to deterioration in nutritional and organoleptic features, resulting in a decrease in shelf life (Lum and Norazira, 2011; Maringgal *et al.*, 2019; Zhang *et al.*, 2019).

Many of the world's leading mango-producing nations effectively employ hot water treatment.

There is a straightforward and efficient method for minimizing anthracnose deterioration in fully ripe, green mangoes. It works well to eradicate dormant fungal diseases that have taken root on the cuticle, under it, and inside the pedicel. The degree of infection and storage temperature affect how well a treatment works. Unless used in conjunction with fungicides, postharvest fruit dips in hot water are considered modestly effective against mango anthracnose, particularly when the disease pressure is high (Kefialew and Ayalew, 2008). In the Eastern Caribbean, where anthracnose and fruit fly infestation are the main issues, the development of a sustainable mango sector has been severely hampered by strict quarantine and quality regulations in importing nations. Mango cv. Julie postharvest illness is not efficiently controlled by the globally recognized hot water treatment suggestion of 46.1°C for 65 minutes to meet fruit fly quarantine standards. A variety of hot water treatment temperatures, ranging from 45°C to 55°C, were examined in studies conducted in Dominica for periods ranging from 5 to 65 minutes. The findings showed that anthracnose (*Colletotrichum gloeosporioides*) and fruit fly (*Anastrepha obliqua*) may be controlled in Julie mangoes by raising the temperature over 46°C for brief periods (less than 65 minutes). In essence, it involves immersing the fruit in a hot water treatment

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that is between 50 – 55 °C for two - five minutes, whether or not fungicide is used. As soon as possible after harvest, but no later than two days after harvest, the therapy should be applied. Fruit damage can be avoided by carefully regulating the water bath's temperature to within 0.5 °C.

**Fungicides based (chemical) control:**

Chemical-based anthracnose management has been the primary method for preventing damage in fields and commercial packinghouses. Synthetic fungicides effectively reduce *Colletotrichum* inoculum in the field and fungus growth during storage (Siddiqui, 2018). In addition to additional drugs like prochloraz, imazalil, and chlorothalonils, the products that have historically been used to combat *Colletotrichum* spp. comprises various compounds such as triazole, benzimidazole, dithiocarbonates, strobilurins, and cupric products (Bordoh et al., 2020b). However, their inappropriate use and subsequent therapies based on chemicals with similar modes of action have been linked to the development of resistant strains and reductions in pathogen sensitivity (Martínez-Blay et al., 2020; Sanders et al., 2000; INADA et al., 2008; Zhang et al., 2020). For example, isolates of *C. gloeosporioides* sensu lato from avocado and mango fruits have been shown to have reduced susceptibility to prochloraz and benzimidazole (Sanders et al., 2000). Furthermore, because fungicides leave chemical residues on fruits and have polluting effects on soil, water, and creatures that are not their intended targets, their use in the agri-food industry poses a significant danger to both human health and the environment (Yuan et al., 2016; Ncama et al., 2019). Several countries have implemented regulations to reduce the adverse impacts of agricultural practices and promote safer methods. European Directive 2009/128/CE, aimed at reducing chemical residues on final goods, has led to the prohibition of certain fungicides used to control *Colletotrichum* species (Lokare et al., 2021).

Numerous countries have varying regulations on the types of active compounds and the permissible MLRs (Fiankor et al., 2021). Exporting farmers from countries with higher standards face significant financial burdens to adapt their production methods, including expensive inputs and skilled labor. Many

manufacturers are forced to leave the market due to their inability to pay for the costs of this remodeling procedure (Fiankor et al., 2021; Fatima et al., 2024; Javed et al., 2024). These considerations make it abundantly evident how vital it is to transition to innovative, workable, and sustainable practices; in fact, several attempts have been undertaken worldwide over the years to find substitute tools that would mitigate the impacts of fungicides. There have been occasions when research has focused on antifungal active agents with low fludioxonil toxicity, as, even if the majority of scientific studies have focused on the creation of chemical-free management strategies (Diskin et al., 2019; Shimshoni et al., 2020; Naeem et al., 2024).

**Innovative Approaches for Sustainability**

**Biological control**

Biocontrol-based methods can be a valid way to increase agricultural sustainability since they are based on the antagonistic behavior of fungal, bacterial, or yeast species that can be used to make non-polluting commercial formulations. They employ several strategies, including parasitism, the generation of harmful metabolites, and competition for resources and/or space (Carmona-Hernandez et al., 2019). Researchers investigated postharvest biological management of mango anthracnose using a *Bacillus* sp. strain. They found that in vivo disease control was achieved when fruit was inoculated with the bacterium 24 hours before the fungus, suggesting the antagonist did not affect the fungus's quiescent phase (Carmona-Hernandez et al., 2019; Fiankor et al., 2021).

*Bacillus subtilis* bioagent significantly reduced anthracnose entry into ripening fruits by preventing early fruit infection, significantly lower than traditional post-harvest treatments (Senghor et al., 2007). Kefalew and Ayalew (2008) discovered that bacterial isolates and yeast antagonists significantly reduced anthracnose on intentionally infected fruit, suggesting the use of biological control-based post-harvest management techniques to prevent disease spread (Droby et al., 2016; Zhang et al., 2018; Shi et al., 2021). Recent research has identified various biological agents or antifungal compounds as potential treatments for *Colletotrichum* spp. infection or development in sensitive fruits (Table 1).

**Table 1. List of biological agents for post-harvest control of *Colletotrichum* spp.**

Biological Agent (Species/Strain)	<i>Colletotrichum</i> Species	Host Species	Reference
<i>Bacillus subtilis</i> (S-16)	<i>C. acutatum</i> (complex species)	<i>Malus pumila</i>	(Lee et al., 2012)
<i>Streptomyces philanthi</i> (RM-1-138)	<i>C. gloeosporioides</i> (complex species)	<i>Capsicum annuum</i>	(Boukaew et al., 2018)
<i>Bacillus subtilis</i> (EA-CB0015)	<i>C. acutatum</i> (complex species)	<i>Cyphomandra betacea</i>	(Arroyave-Toro et al., 2017)
<i>Trichoderma harzianum</i> (T-39) or <i>Trichoderma hamatum</i> (T-105)	<i>C. acutatum</i> sensu lato	<i>Fragaria × ananassa</i>	(Freeman et al., 2004)

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<i>M. pulcherrima</i> T5-A2	<i>C. acutatum</i> sensu lato	<i>M. domestica</i>	(Janisiewicz et al., 2003)
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Research has largely focused on *C. acutatum*, *C. truncatum* as well as *C. gloeosporioides* species in singular or complex form because other species are occasionally or regularly harmful (Shi et al., 2021; Arroyave-Toro et al., 2017). Certain studies have shown that bacterial species, such as strains of *Paenibacillus polymyxa* and *Bacillus subtilis*, as well as their derivatives, are very effective in reducing the severity of post-harvest lesions and the incidence of anthracnose caused by the *C. acutatum* and *C. gloeosporioides* species complex (Shi et al., 2021; Arroyave-Toro et al., 2017; Lee et al., 2012). Because they can dramatically reduce the incidence (from 76% to 83%) and severity (from 65% to 85%) of anthracnose, *Bacillus* species, in particular, are promising candidates for the biological control of these pathogen species in a range of host species (Arroyave-Toro et al., 2017; Lee et al., 2012). Several strains of *Pseudomonas aeruginosa* and *Burkholderia* spp. shown a great effectiveness in managing *C. truncatum* in Chile, resulting in a 75% to 100% reduction in the occurrence of anthracnose (Sandani et al., 2019). Several yeast species, such as *Metchnikowia pulcherrima* and *Pichia kluyveri*, are highly effective *Colletotrichum* antagonists in treating complex infections in apple fruits, surpassing other tested bacterial or fungal species (Conway et al., 2005; Janisiewicz et al., 2003; Mewa-Ngongang et al., 2019). Yeast species effectively reduced anthracnose occurrence and were found to be more effective against *Colletotrichum* infections in field trials, with *Rhodotorula minuta* suspension being more successful than insecticides in suppressing anthracnose disease in a mango orchard. Despite scientific data, only two microorganism-based biofungicides are currently available against *Colletotrichum* spp. contain *Bacillus* bacterial strains (Shi et al., 2021). Two products, Double Nickel 55 (marketed by Certis and Bayer) and Serenade ASO (constructed by *B. amyloliquefaciens* QST713, formerly *B. subtilis* QST713), are available. Double Nickel 55 is not yet commercially accessible in the EU, while Serenade ASO has been registered in the EU, but commercialization has been approved in Canada and the USA. Highlight that other microbes effectively control anthracnose, suggesting that more work is needed to provide farmers with more biological tools (Shi et al., 2021).

#### Plant Derivates

Plant derivatives having direct antifungal activity such as secondary metabolites implicated in plant immune mechanisms have frequently been the focus

of research into environmentally friendly ways to treat anthracnose disease. Use of plant extracts: Due to their apparent safety, widespread consumer acceptability, and potential for several applications, plant extracts and essential oils from various plant genera are attracting attention. The significance of several plant species as a potential natural source of fungicidal compounds is demonstrated by the fungitoxic effects of crude extracts of various plant species. According to Gottlieb et al. (2002), they include complex combinations of secondary metabolites that are physiologically active and possess antibacterial, allelopathic, antioxidant, and bioregulatory qualities. According to several studies, extracts from various plant species have shown the ability to reduce the growth of anthracnose on mango fruit. This suggests that products produced from biologically active plants may be important for crop protection measures (Alemu et al., 2014a).

Research organizations in nations where anthracnose poses a serious economic concern have investigated the potential of several plant-based compounds against *Colletotrichum* spp. throughout the years. Nevertheless, a lot of research has only looked at the inhibitory effect that some unrefined vegetable extracts have on cultivated fungus, not their ability to prevent anthracnose on fruits, for example (Johnny et al., 2011; Bordoh et al., 2020a). Thus, nothing is presently known about their potential use in post-harvest management techniques. Furthermore, these compounds must be thoroughly examined to determine their antifungal potential as well as to rule out any unfavorable side effects from usage. Research on papaya fruits suggests refined extracts from *Vitex mollis* pulp may be more effective in managing pathogens than crude ones (López-Velázquez et al., 2021). Essential oils (EOs) from a variety of fragrant plants form a class of pure extracts that are gaining popularity as fungicide substitutes (Raveau et al., 2020). Terpenoids and phenylpropanoids, two classes of lipid-active chemicals that may interact with hydrophobic elements of the pathogen membrane, are the primary cause of EOs' potent antifungal action (Freiesleben and Jäger, 2014; Gutiérrez-del-Río et al., 2018; da Costa Gonçalves et al., 2021). EOs have been extensively studied in recent years to see if they are suitable to substitute pesticides in agriculture, and their antibacterial properties date back to ancient times. Several of these compounds have been thoroughly investigated and proven to be useful for managing anthracnose after harvest for various fruits (Table 2).

**Table 2. List of essential oils for managing *Colletotrichum* spp. in post-harvest**

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Essential Oil	Colletotrichum Species	Host Species	Reference
Ginger oil	<i>C. gloeosporioides</i> (complex species)	<i>C. papaya</i> L.	(Ali et al., 2016)
Lemongrass oil	<i>C. gloeosporioides</i> (complex species)	<i>Carica papaya</i> L.	(Ali et al., 2015)
Savory oil	<i>C. gloeosporioides</i> (complex species)	<i>C. papaya</i> L.	(Sarkhosh et al., 2018)
Cinnamon bark oil	<i>C. acutatum</i> (complex species)	<i>Fragaria</i> x <i>ananassa</i>	[104]
Thyme oil	<i>C. gloeosporioides</i> (complex species)	<i>Mangifera indica</i> L.	[103]

Research on papaya fruits revealed that various essential oils (EOs), including thyme, ginger, lemongrass, and savory oils, influence the development of the *C. gloeosporioides* species complex (Ali et al., 2015; Ali et al., 2016; Sarkhosh et al., 2018). Of these, the latter was found to be the most effective in lowering the incidence of anthracnose (-26.5%) (Sarkhosh et al., 2018). Thyme oils are effective in controlling *C. gloeosporioides* species in avocado fruits, reducing the incidence and severity of anthracnose disease in strawberry fruits. Thyme and cinnamon bark essential oils also decreased the incidence of anthracnose disease in strawberry fruits. On the other hand, essential oils from *Ocimum gratissimum* and *Ocimum basilicum* resulted in less severe anthracnose on banana fruits. However, the information regarding the negative effects of essential oils (EOs) is basic and limited, particularly for this chemical family. For instance, papaya fruits treated with lemongrass oils showed dose-dependent phytotoxicity (Ali et al., 2015). The suitability of various formulations to provide EOs, which have high volatility and hydrophobicity, stability, and durability, has, nonetheless, received a lot of attention. Nanotechnology-based methods for stabilizing formulations have been suggested, including encapsulating essential oils, integrating them into edible or biodegradable coatings, and producing microemulsions (Talón et al., 2019; Weisany et al., 2019; Wińska et al., 2019).

Several algae derivatives have been discovered to function as plant resistance inducers against biotic and abiotic stresses, making them one of the plant

active substances investigated as alternative crop protection options. Commercial formulations based on seaweed have been created for foliar (high/low pressure) spraying treatments or immersions of early seedlings. Furthermore, in vitro research employing cultivated fungus has shown that these compounds can also have inhibitory effects on the conidial germination and/or mycelial proliferation of pathogenic pathogens (Righini et al., 2018); proof of the effectiveness of some algal extracts has also been obtained against *Colletotrichum* spp. However, to proceed from laboratory testing to use in anthracnose control regimens, in vivo investigations are required for plant derivatives and essential oils. For instance, "Ulvan," a water-soluble polysaccharidic extract made from *Ulva* spp., has been demonstrated to be a highly successful resistance inducer in bean plants, consistently reducing the severity of *C. indemuthianum* anthracnose by up to 60% (de Freitas and Stadnik, 2012; Paulert et al., 2009). Instead, foliar spraying or infiltrating alfalfa specimens with an ethanolic fraction of the same species was linked to higher defense gene marker expression and, thus, improved resistance to *C. trifolii* infections (Cluzet et al., 2004).

In a similar vein, Kim noted that applying a *Chlorella fusca* solution topically to cucumber plants effectively activated the plants' natural defenses, resulting in a less severe case of anthracnose brought on by *C. orbiculare* (Kim et al., 2018) Lastly, spray treatments with a bio fungicide based on *Ascophyllum nodosum* (Aguado et al., 2012) consistently reduced the spread of anthracnose on strawberry leaves caused by the *C. acutatum* species complex (Table 3).

**Table 3. List of seaweed derivatives for management of *Colletotrichum* spp.**

Seaweed	Colletotrichum Species	Host Species	Reference
Ethanolic fraction- <i>Ulva</i> spp.	<i>C. trifolii</i>	<i>Medicago truncatula</i>	(Cluzet et al., 2004)
"Ulvan"- <i>Ulva</i> spp.	<i>C. lindemuthianum</i>	<i>Phaseolus vulgaris</i> L.	(de Freitas and Stadnik, 2012)
"Ulvan"- <i>Ulva fasciata</i>	<i>C. lindemuthianum</i>	<i>P. vulgaris</i> L.	(Paulert et al., 2009)
Algal suspension- <i>Chlorella fusca</i>	<i>C. orbiculare</i>	<i>Cucumis sativus</i>	(Kim et al., 2018)
Seaweed-based biofungicide- <i>Ascophyllum nodosum</i>	<i>C. acutatum</i> species complex	<i>Fragaria</i> x <i>ananassa</i>	(Aguado et al., 2012)

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Generally speaking, in addition to the absence of side effects and the creation of suitable delivery methods, several requirements must be met before plant-based chemicals may be used in crop defense measures. A detailed dossier with stability data which is currently unavailable for the bulk of these compounds must be created to receive approval from the regulatory bodies (da Costa Gonçalves *et al.*, 2021; Pavela and Benelli, 2016). Furthermore, it appears that these compounds' action is not sufficiently repeatable, mostly due to the great degree of heterogeneity in their chemical profile (Jugreet *et al.*, 2020). Therefore, it appears that the release of these chemicals onto the market is still a long way off, even with the encouraging findings obtained.

### Strategies Based on Biotechnology

Advanced Biotechnology-based approaches to managing pests and diseases have gained attention recently as a means of developing novel compounds that can replace pesticides or enhance plant resilience to infections. One such tactic is genome editing, which makes it possible to create novel resistant kinds. By causing breaks in specific regions of the plant genomic DNA, which are subsequently fixed by different cell-repairing mechanisms, single mutations (insertions, deletions, or substitutions) may be introduced into the target locus (Borrelli *et al.*, 2018). Meganucleases (MNs), zinc finger nucleases (ZFNs), transcription activator-like effector nucleases (TALENs), and clustered regularly interspaced short palindrome repeats protein 9 (CRISPR-Cas9) are the four types of nucleases that can be used in genome editing to create DNA breaks at particular locations.

This method, which is part of the so-called "New Breeding Technologies" (NBT), has a significant benefit due to its precise and targeted genetic changes. To boost the plant immune response, they may target the genetic characteristics of plants linked to disease vulnerability. These strategies significantly cut down on the amount of time needed to generate new kinds using conventional breeding technology. However the lack of knowledge about the genetic processes controlling plant response to pathogens which are unclear in the case of *Colletotrichum* may limit the use of this tactic. Mishra and associates created CRISPR-Cas9 T-DNA-free homozygous pepper plants (*Capsicum annum*) with a desired mutation in the CaERF28 gene (Mishra *et al.*, 2021) is the sole scientific proof to date of the effectiveness of NBT administration against this disease. The endogenous defensive systems are downregulated when this gene is expressed (Mishra *et al.*, 2018a; Mishra *et al.*, 2018b), and the altered plants were much less vulnerable to *C. truncatum*.

As potential next-generation tools for sustainable crop protection, RNA interference-based techniques have been studied more thoroughly than genome editing among biotech-based technologies. RNA interference is a post-transcriptional gene silencing regulatory mechanism that is widely conserved throughout Eucaryotes and contributes to host defense, pathogen pathogenicity, and host–pathogen communication (Wang *et al.*, 2016; Cai *et al.*, 2018; Gebremichael *et al.*, 2021). In addition to pathogenicity, RNA interference (RNAi) has a variety of roles in fungi, including controlling transposable elements, regulating endogenous gene transcription, forming heterochromatin, maintaining genomic integrity, and adapting to stressful situations (Lax *et al.*, 2020). By downregulating endogenous genes implicated in the scripts, the RNAi pathway helps plants defend against biotic stresses by allowing the RISC complex to cleave the genes and stop transcription (Senthil-Kumar and Mysore, 2010; Nunes and Dean, 2012; Capriotti *et al.*, 2020). Vulnerability of the host (Gebremichael *et al.*, 2021). Furthermore, it has been discovered that plant sRNAs may be transmitted to pathogens during the infection process to inhibit genes necessary for their development or virulence; similarly, pathogens can enhance their pathogenicity by transferring to sRNA that targets genes involved in host defense (Gebremichael *et al.*, 2021).

Numerous research papers have demonstrated the efficacy of HIGS technology in various crops to suppress a range of fungal species (Nowara *et al.*, 2010; Niu *et al.*, 2021; Hu *et al.*, 2020). Of them, Mahto et al. provided the first proof of anthracnose control in tomatoes and chillies in 2020 (Mahto *et al.*, 2020). To regulate the *C. gloeosporioides* species complex, the gene encoding the Conidial Morphology 1 protein (COM1) was selected as the target. This gene is homologous to a Magnaporthe oryzae gene that has been shown to play a critical role in the conidium morphology and appressorium development (Yang *et al.*, 2010).

*Anthracnose* symptoms in both leaves and fruit were significantly reduced when tomato and chilli plants were transformed by an agrobacterium using a cassette that expressed *C. gloeosporioides* CgCOM1 sRNA. Microscopy investigation supported these findings by showing that fungal growth was inhibited and that the generation of functional appressoria was hampered. Notwithstanding the encouraging outcomes, creating a HIGS approach necessitates significant time and financial commitments to produce plants that have undergone stable transformation. Furthermore, HIGS suggests the production of genetically modified plants, the use of which is restricted by national laws and societal considerations. As previously mentioned, SIGS crop protection techniques can also take use of the

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ambient RNAi process, which suggests that pathogens are capable of external RNA absorption. However, as fungal cells were unable to absorb fluorescein-labelled dsRNA even 20 hours after the treatments, SIGS proved ineffective as an environmental RNAi technique to manage the *C. gloeosporioides* species complex (Qiao *et al.*, 2021). The absence of dsRNA absorption ability in this species is supported by the fact that topical administration of dsRNA targeting the DCL gene of this species consistently did not lessen the development of symptoms in sensitive plant tissues (Wang and Jin, 2017).

### Final Thoughts

Research into creative crop protection methods in place of harmful pesticides was prompted by growing public concerns about food safety and the effects of the agri-food system on the environment, as well as impending limits on the use of phytosanitary agents. The most promising methods being researched to manage anthracnose in postharvest disease control are detailed in this review. For its use in large-scale production systems, several problems need to be resolved. Regarding biocontrol agents, for example, several studies revealed that a wide range of microorganisms may be useful for controlling anthracnose; however, in most cases, the antagonistic activity was only investigated on a small number of fruit systems, and further research is required to develop commercial preparations. Furthermore, special consideration should be given to their stability (i.e., capacity to adhere and colonize plant tissues, survival in harsh environments, and broad range of action), safety, and lack of harmful effects.

Testing of plant derivatives' antifungal efficacy was mostly conducted on cultivated fungi; however, there is no testing conducted in pertinent application environments. Several EOs shown good performance in controlling anthracnose; nevertheless, more study is required to rule out phytotoxic effects on the product, meet authorization criteria, and create suitable delivery methods. Considering the economic and environmental sustainability of delivery formulations, nanotechnology is now the more promising approach to improve their durability and bioavailability in the environment. Biotechnology-based methods, such as those that take advantage of the RNA interference mechanism, have also been proposed as new ways of managing *Colletotrichum* spp.

Furthermore, it is challenging to produce sRNA-expressing plants on a wide scale since rapid and affordable transformation procedures are yet unavailable for many vulnerable crops. However, when these restrictions are removed, this method can offer a reliable defense. Permitting a significant reduction in the use of harmful agrochemicals, is

very beneficial, particularly for fruit growers in underdeveloped nations where anthracnose frequently has a major impact on yields and postharvest methods are not developed. Because the SIGS approach applies interfering RNAs exogenously, it is unaffected by GMO-linked restrictions. More research is necessary to fully understand how RNA interference (RNAi) and siRNA absorption processes by *Colletotrichum* species work, especially in light of the disparate behaviors that have been documented thus far for various species. These factors become especially crucial for the postharvest management of fruit crops, where consumer concerns about fruit safety and limitations on the use of agrochemicals create special challenges. Both RNAi-based strategies have excellent selectivity toward target pathogens due to the sequence recognition mechanism; nevertheless, their lack of multi-spectrum effectiveness may be a limiting issue, particularly when it comes to protecting crops that are vulnerable to many *Colletotrichum* species. Because of this, the procedure of choosing target genes or sequences should concentrate on finding areas that effectively silence distinct, related pathogen species.

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#### Declaration

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#### Ethics Approval and Consent to Participate

Not applicable.

#### Consent for Publication

The study was approved by authors.

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All authors contributed equally.

**Conflict of interest**

There is no conflict of interest among the authors of the manuscript.



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