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Review

Fruit Stem-End Rot

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Abstract: After harvest, the fruit ripens and stem-end rot (SER) starts to develop, leading to significant fruit losses. SER is caused by diverse pathogenic fungi that endophytically colonize the stem during fruit development in the orchard or field and remain quiescent until the onset of fruit ripening. During the endophytic-like stage, the pathogenic fungus colonizes the phloem and xylem of the fruit stem-end; after fruit ripening, the fungus converts to a necrotrophic lifestyle, while colonizing the fruit parenchyma, and causes SER. The fruit stem-end is colonized not only by pathogenic fungi, but also by various nonpathogenic endophytic microorganisms, including fungi, yeast and bacteria. However, little is known about the fruit stem-end endophytic microbiome, which could contain new and existing biocontrol agents. To control fruit SER, treatments such as ripening inhibition, harvesting with the stem, application of chemical or biological fungicides, or physical control such as heat treatments, cold storage, or exposure to light have been suggested. This review focuses on the characterization of SER pathogens, the stem-end microbiome, and different pre- and postharvest practices that could control fruit SER.

Keywords: stem-end rot; *Botryosphaeria*; fruit; fungicide; ripening; microbiome; biological control; physical control

1. Characterization of Stem-End Rot Causing Pathogens and Their Lifestyle

In recent years, there has been a rising demand for ripe and ready-to-eat fruit. However, as the fruit ripens, it becomes susceptible to various postharvest diseases [1]. Among them is the emergence of stem-end rot (SER) disease. SER occurs in various fruit, and particularly in tropical and subtropical fruit, including mango, avocado, citrus, mangosteen, carambola, and others. In mangoes, for example, SER is considered to be the second most severe disease worldwide, after anthracnose, caused by *Colletotrichum gloeosporioides* [2], while in dry areas, SER is the major postharvest pathogen. For example, in Israel, SER caused 30–40% loss of harvested mango fruit during 2014 (Diskin et al., in press).

SER-causing pathogens penetrate to the stem through natural openings and wounds, mainly during inflorescence and flowering stages [3–5]. Those fungal pathogens live endophytically, mainly in the phloem but also in the xylem, and exist asymptomatically in the stem tissue until fruit ripening (Figure 1) [2,5–7]. Unripe fruits are resistant to SER [7]. This resistance is compromised when fruit ripening initiates during fruit storage. During ripening, fruits undergo dramatic biochemical and physiological changes including ethylene emission in climacteric fruit and other phytohormone changes, accumulation of soluble sugar, cell wall loosening, a decrease in phytoanticipin and phytoalexin levels, a decline in inducible plant defense mechanisms, and changes in ambient host pH [1].

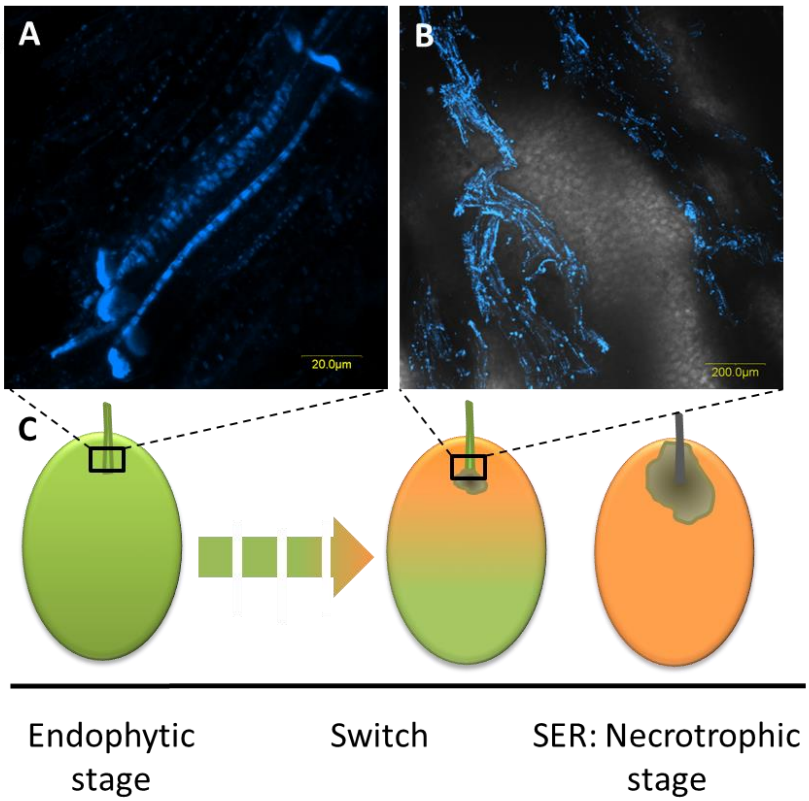

Figure 1. Stem-end rot development during the fruit ripening process. Confocal images of mango stem-end stained with aniline blue show (**A**) endophytic colonization of the phloem, and (**B**) necrotrophic colonization of ripe fruit. (**C**) Illustration of the development of stem-end rot during fruit ripening (adapted from Diskin et al. [6] with permission).

The endophytic pathogens probably sense the changes during fruit ripening and respond to them by switching from an endophytic, asymptomatic lifestyle termed 'quiescent' or 'latent' stage, to an aggressive necrotrophic stage, causing SER [5–7]. These physiological alterations modify the endophytic microorganism's environment in the fruit and consequently influence the fruit's susceptibility to SER [6]. At the early ripening stage, SER symptoms appear as a small dark-brown to black spot at the fruit stem-end. In advanced stages of ripening, SER progresses to decay, resulting in fruit discoloration, brown flesh, and fruit softening [8,9]. A positive correlation was found between length of ripening time and severity of several postharvest diseases, including SER, in avocado fruit [10]. Indeed, fruits that ripen faster have less SER than fruits that are slower to ripen [11], and therefore the longer ripening time in avocado increases the time available for fungal colonization and the opportunity for SER symptoms to develop [12].

SER occurs in various fruits, but it has mainly been studied in mango and avocado. Interestingly, similar pathogens cause SER in both mango and avocado fruit. The major pathogens causing SER in mango are illustrated in Figure 2 and including mainly *Botryosphaeria*-related species such as: *Dothiorella dominicana*, *Dothiorella mangiferae*, *Lasiodiplodia theobromae*, *Neofusicoccum* spp., *Phomopsis mangiferae*, *Cytosphaera mangiferae* and *Pestalotiopsis* sp. [5,13], and *Alternaria alternata* as well as *Colletotrichum gloeosporioides* [2,5]. Similarly, SER-causing pathogens of avocado include: *Colletotrichum gloeosporioides*, *Alternaria alternata* and various species of the *Botryosphaeria* family as described for mango [14]. The genus *Lasiodiplodia* is an emerging pathogen, associated with SER worldwide. In recent years, there has been a rise in reports of this pathogen causing heavy losses to the fruit industry in Brazil [15], China [16], Peru [17], and India [18]. As *Lasiodiplodia* prefer higher temperatures and attack during plant stress, this rise might be connected with global climate warming and has to be further explored.

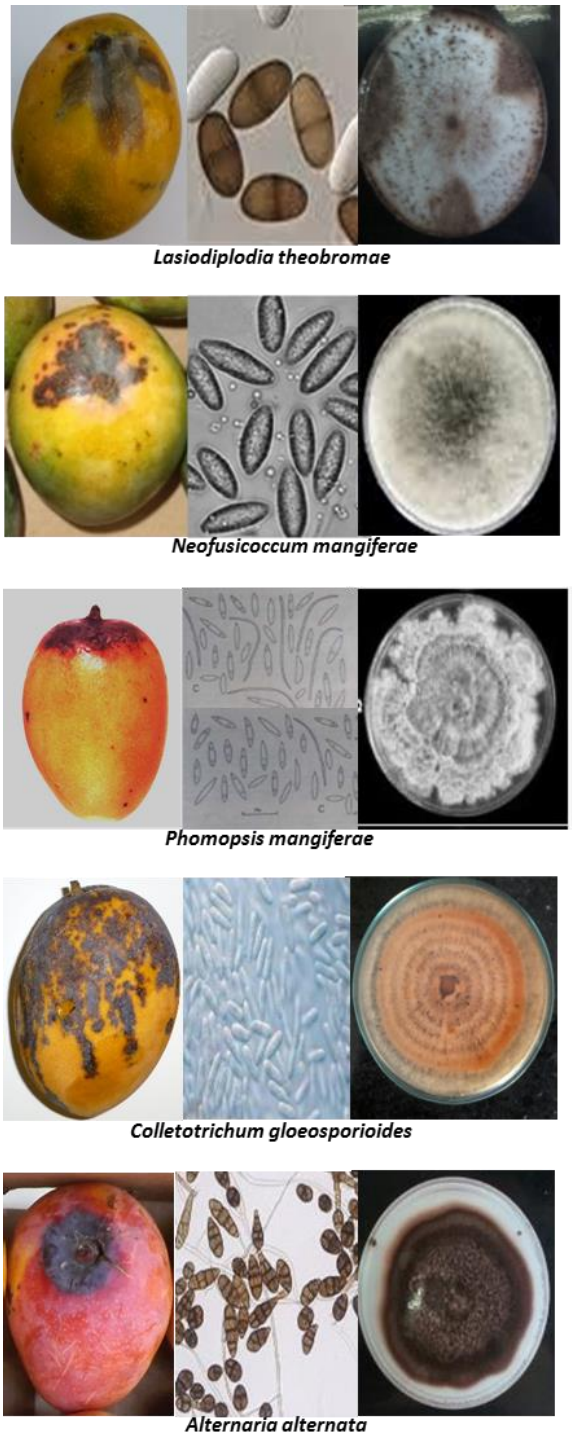

Figure 2. Characterization of fungal pathogens that cause stem-end rot in mango fruit. Left column: typical disease symptoms. Middle column: typical conidia. Right column: fungal growth on PDA media.

2. Endophytic Community in Fruit Stem-End

Not all fungi present in the stem-end make the transition from endophytic to necrotrophic lifestyle and become pathogenic during fruit ripening. The plant stem is populated with various species of microorganisms, including fungi, yeast and bacteria, most of which are not pathogenic [6,19]. These microorganisms can live in symbiosis or mutualism with the plant. They are termed 'endophytes' if they colonize the plant tissue internally. Endophytes persist in the plant tissue without causing any

apparent symptoms or damage [20]. Diverse endophytic communities are considered to be important in maintaining a healthy plant biosystem. Little is known about the endophytic community of the fruit stem-end. However, endophytic microbiomes have been well studied in other plant organs, such as seeds, bark, foliage and roots [20,21]. Fungal endophytes are found in most plant families. Tropical trees represent hotspots of fungal species diversity, containing numerous species that have not been recovered [22]. The tree bark of Indian Bael trees was shown to have more endophytic fungi than its leaves or roots [23]. Thus, the fruit stem-end microbiome probably contains various microorganisms that should be further studied.

Recent advances in DNA sequencing and "omics" technologies have enabled evaluating the diversity and understanding the function of microbial communities existing within the plant tissue. In recent years, a few publications have also begun exploring the fruit stem-end microbiome in apple [19,24] and mango [6]. During storage and fruit ripening, the microbial community changes and the abundance of pathogenic fungi increases dramatically, along with increasing incidence of SER. These findings highlight the existence of an inherent mechanism by which pathogens have a quiescent endophytic stage and become active and cause disease when the composition of the microbiota changes in response to fruit ripening and storage [6].

3. Factors and Treatments Affecting SER

Postharvest disease management has the goal of preserving fruit quality without disease until consumption. Thus, management approaches are aimed at preventing, suppressing or delaying disease symptoms during storage [25]. Postharvest disease management in general, and SER in particular, can be achieved by several main approaches, such as chemical, biological and physical treatments that directly inhibit fungal pathogens on the one hand, or regulate fruit resistance on the other (Table 1).

3.1. Inhibition of Fruit Ripening

Ethylene is the main phytohormone controlling most of the events associated with the climacteric fruit-ripening process. Other phytohormones, such as auxin and abscisic acid, are also closely associated with fruit ripening [26]. Since there is a positive correlation between fruit ripening and postharvest decay, several studies have evaluated the potential application of phytohormones to prevent postharvest decay. Indeed, application of the auxin derivative 2,4-dichlorophenoxyacetic acid (2,4-D) reduced fruit ripening and prevented abscission of the stem-end in citrus and mango fruit, which reduced SER [27,28]. Other studies assessed the effect of postharvest application of ethylene receptor inhibitor, 1-MCP, which delays fruit senescence and prolongs storage [29,30], on the inhibition of postharvest decay. However, because ethylene plays a dual role, in fruit ripening and in the fruit defense response [1], those studies yielded conflicting findings. In Indian jujube fruit [31,32] and avocado fruit [33], 1-MCP treatment reduced fungal pathogen rot and SER. However, other studies in citrus [34], mango [35], and avocado [36] showed that 1-MCP promotes fruit susceptibility to SER pathogens. It seems that 1-MCP could affect fruits susceptibility in a concentration and timely dependent manner [37–39]. Thus, small amounts of ethylene are probably necessary to maintain fruit resistance to pathogens [1], and a high concentration of 1-MCP probably both delays the ripening process and hampers the fruit's natural defense.

3.2. Harvesting with Stem

One of the most intriguing ways to reduce SER is derived from a simple harvest practice, i.e., harvesting fruit with short stems (pedicel) using secateurs as opposed to the common practice of detaching the fruit, which leaves no stem at all. Surprisingly, this minor change in harvesting practice had a major and significant impact on reducing SER incidence in mango and avocado fruit [40]. Similarly, harvesting mango with long pedicels reduced SER in comparison to harvesting with short pedicels [41]. Interestingly, in the sap, there are some compounds with antimicrobial properties, which could cause the difference in SER incidence. For example, in mango sap, the alk(en)ylresorcinols

(5-n-heptadecenylresorcinol and 5-n-pentadecylresorcinol) have antimicrobial and antifungal activities, especially against *Alternaria alternata* [42,43]. Furthermore, 'Kensington Pride' mango fruit stored with 2- to 3-cm long stems had significantly more resorcinol in their peel and smaller anthracnose lesions than de-sapped fruit [44]. Similarly, mango cultivar with higher sap flow had less incidence of anthracnose [45]. Therefore, it seems that when harvesting fruit with stems, more sap that contains antifungal compounds is left in the fruit stem and peel, leading to decreased postharvest side decay and SER.

3.3. Chemical Treatments

Fungicides are generally the most traditional and effective strategy for controlling postharvest diseases [46]. Fungicide type and timing of application depend on the target pathogen and its lifecycle. A variety of pre- and postharvest fungicidal treatments were suggested to reduce or delay the onset of SER.

3.3.1. Preharvest Chemical Control

Preharvest application of fungicide is efficient in reducing SER, while the fungicide residue decline with time. Preharvest sprays with Benlate applied to 'Hamlin' orange trees was found to eliminate SER in orange fruit harvested a week later, and efficiently reduced green mold (*Penicillium digitatum*) in fruit harvested six weeks after the spray application [47]. Preharvest sprays with various chemicals applied to 'Fuerte' avocado fruit were efficient in controlling SER and anthracnose [48]. Difolatan was found to be the most effective product closely followed by Cu-hydroxide and Baycor. Good control was also achieved with Aliette, Benlate and Cu-oxychloride [48].

Since SER-causing pathogens penetrate mainly during flowering and colonize the fruit stem before harvest [5], targeting the flowering stage during fungal penetration could reduce SER. Preharvest spray application of copper oxychloride, combined with mancozeb, from flowering until harvest, controls most mango postharvest diseases [49]. Diskin, Feygenberg, Maurer and Alkan [4] recently showed that fungicide application of Luna Tranquility (fluopyram and pyrimethanil) or Switch (fludioxonil and cyprodinil) during flowering, as *Lasiodiplodia* penetrates, significantly reduces the incidence and severity of postharvest SER and side decay in mango fruits. These results suggest that fungicide application during flowering reduced the penetration and initial colonization of pathogenic fungi in the fruit stem-end, which shifted the fruit stem-end microbiome toward a more diverse and less pathogenic community, leading to a reduction in SER incidence [4].

3.3.2. Postharvest Chemical Control

Postharvest fungicidal treatments are more common for controlling SER and can be applied by dipping or spraying, or in waxes or coatings. Prochloraz, a nonsystemic imidazole, is a well-recognized fungicide that is used commercially for controlling postharvest diseases in avocado and mango fruit [50–52]. However, application of prochloraz has been reported to be more effective against side decay (anthracnose), and less effective against SER. In general, benzimidazole fungicides, including benomyl and thiabendazole, have the advantage of also being effective against SER caused by *Lasiodiplodia theobromae* on mango, whereas imidazoles such as prochloraz and imazalil are not effective for SER control [53]. Similarly, Plan, et al. [54] found that benomyl is more effective than prochloraz and pyrimethanil for controlling mango SER caused by *Botryosphaeria*. Fludioxonil was more effective against mango fruit SER, whereas prochloraz was more effective against anthracnose [55]. The combination of prochloraz and fludioxonil was most effective at controlling both postharvest diseases—anthracnose and SER—in 'Kent' mango fruit [56]. In a comparative study, the efficacy of six fungicides—carbendazim, azoxystrobin, tebuconazole + trifloxystrobin, difenoconazole, thiabendazole and propiconazole—was assayed on mango artificially inoculated with *Lasiodiplodia theobromae* inoculum. They showed that carbendazim, followed by thiabendazole were highly effective at inhibiting the mycelial growth of *Lasiodiplodia theobromae*. They also reported that tebuconazole

+ trifloxystrobin, azoxystrobin and carbendazim significantly reduce SER disease severity on mango fruit [57].

A comparative study of azoxystrobin, fludioxonil, pyrimethanil, imazalil and thiabendazole against *Diplodia* SER in citrus fruit showed highest effectiveness for thiabendazole, imazalil and fludioxonil [58]. In artificially inoculated lemons, Phomopsis stem-end rot caused by *Diaporthe citri* was effectively controlled by low toxicity salts as potassium sorbate and potassium phosphite at 20 °C, although Diplodia stem-end rot caused by *Lasiodiplodia theobromae* was partially controlled only by potassium sorbate [59]. Recently, the conventional fungicides imazalil and thiabendazole were found to be effective at controlling Diplodia SER caused by *Lasiodiplodia theobromae.* The best control of Diplodia SER was achieved by immersion in thiabendazole at pH 5 and 20 °C. It was concluded that thiabendazole application for lemon treatment is the best alternative to controlling SER and should replace carbendazim, which is, however, not allowed in the European Union [60].

3.4. Biological Control

Postharvest applications of chemical fungicides are probably the best means of controlling postharvest decay. However, there is an increase in public concern over the use of chemical fungicides due to their negative effects on the environment and consumer health. In addition, repeated use of fungicide could lead to the development of resistant strains of pathogens. Therefore, there is a need for alternative approaches for postharvest disease management [61]. Biological control, which use microbial antagonists such as bacteria, yeast and fungi against postharvest pathogens, is an efficient strategy for controlling SER [62,63]. While most of the postharvest microbial antagonist research has focused on controlling side decay of fruit, some of those microbial antagonists have biological control activity against SER-causing pathogens. Timing of application is of crucial importance in biological control programs and significantly influence on control efficiency. Thus antagonists can be applied pre and post-harvest.

3.4.1. Preharvest Biological Control

Several preharvest treatments were studied in order to reduce SER disease. Preharvest application of *Bacillus subtilis* were found to be effective in controlling several postharvest decays as anthracnose, SER, and *Dothiorella–Colletotrichum* complex in avocado fruits [64]. To control SER disease using biological control strategy it could be important to apply the antagonist during inflorescence and flowering, when SER-causing pathogens penetrate. Interestingly, *B. subtilis* was found to attach and colonize avocado flowers and interfere with SER-causing pathogen penetration and initial colonization, by attach the conidia and hyphae of SER-causing pathogens and cause cell degradation [65]. Indeed, application of *B. subtilis* during mango flowering reduced pathogenic fungal colonization in the stem-end and reduced mango SER [4].

3.4.2. Postharvest Biological Control

Postharvest application of *Bacillus licheniformis* reduced mango anthracnose and SER [66]. In banana, anthracnose and crown rots caused by *Colletotrichum* is controlled by the biological agents *Burkholderia cepacia*, *Pichia anomala*, *Pseudomonas* sp. and *Candida oleophila* [67,68]. In addition, *Trichoderma harzianum* was found to reduce SER caused by *Lasiodiplodia theobromae* in Rambutan fruit [69], and *Trichoderma viride* was reported to control SER caused by *L. theobromae* in mango fruit [70]. Thus, a variety of microbial agents have been found effective at controlling fruit SER-causing pathogens. The antagonistic mechanism of the various microorganisms could include competition for nutrients, production of antibiotics, direct parasitism or induction of fruit resistance [63].

3.5. Plant Extracts

Another approach for SER management is plant extracts, which proved to be a safe alternative to control postharvest diseases and are listed as food additives by the U.S. FDA [71,72]. The efficiency of

plant volatiles has been demonstrated for reducing postharvest disease incidence, including prolonged shelf life and improved fruit quality in mangoes by hexanal [73] and in citrus by citral [74,75].

Many studies have revealed the antifungal potential of plant extracts against a range of fungal pathogens [76–78]. For example, *Moringa oleifera*, *Syzygium aromaticum* and *Cinnamomum zeylanicum* showed significant antifungal activity against mycelial growth of fungal pathogens that cause SER, and a reduction in SER development in mango fruit [79]. Similarly, a comparative analysis of plant extracts showed that extracts of *Datura stramonium* and *Eucalyptus camaldulensis* efficiently reduce the radial growth of *Lasiodiplodia* isolates in vitro [80]. Another comparative study showed that thyme oil vapours, and clove and cinnamon oil completely inhibited *Colletotrichum gloeosporioides* and *Lasiodiplodia theobromae* growth in vitro. They also showed that thyme oil significantly inhibited the postharvest pathogens on mango fruits after storage at 25 °C for six days [81].

Plant extracts have also been reported as inducers of the defense response in fruit against potential pathogens [82,83]. Obianom and Sivakumar [84], recently showed that a combination of prochloraz and 0.1% (*v*/*v*) thyme oil significantly reduces anthracnose and SER in the 'Fuerte' avocado. They also reported that the combined treatment induces activity of defense enzymes in 'Fuerte' avocados inoculated with *Lasiodiplodia theobromae* and *Colletotrichum gloeosporioides*. In another study, dipping treatment with combination of bacterial antagonists, *Bacillus subtilis* and hexanal induced systemic resistance of mango fruits against *Lasiodiplodia theobromae* by inducing several defense-related enzymes in mango [85].

3.6. Physical Control

Physical technologies, including heat treatment, irradiation and cold-temperature storage, are also common and safe approaches for SER control. Cold storage is one of the best ways to delay fruit ripening and therefore decrease postharvest decay. The effects of temperature on endophytes have been poorly characterized [86]. However, each fungus has an optimal temperature and a temperature that limits their hyphal growth, conidial germination and pathogenicity [87]. On the other hand, each fruit has an optimal storage temperature. Storage below this temperature leads to chilling injuries, and storage above this temperature leads to faster ripening. Indeed, storage of the 'Hass' avocado, for example, at a temperature higher than 6 °C increased fruit ripening and the occurrence of SER [11], while storage at suboptimal temperature (lower than 5 °C) also increased the occurrence of SER [88].

Gamma irradiation can kill microorganisms by damaging their DNA [89], and can even be used to extend the shelf life of foods [90]. However, gamma irradiation did not reduce mango or citrus SER [91,92], or only mildly reduced SER [93], whereas a combination of hot water with gamma irradiation significantly reduced mango SER and anthracnose [92,93]. UV-C was found to control various fungal pathogens, including SER-causing pathogens [94,95], by inducing fruit resistance [96], and can also be considered for the organic market. Similarly, red mango fruit that was exposed to sunlight in the orchard accumulated anthocyanin and was more resistant to SER than fruit that developed within the tree canopy [6]. Therefore, pruning and exposure of fruit to sunlight could be a good method for reducing postharvest SER.

Heat treatment can induce the fruit's natural resistance, remove the unattached pathogens and cause spreading of the fruit's waxy covering, leading to a reduction in postharvest diseases, including SER, reduction in chilling injury and improvement of shelf life [97,98]. Different heat-treatment approaches include hot-water dipping and rinsing; hot vapor and dry-air treatments have been suggested to reduce postharvest diseases via induction of a defense response [99]. Hot-water immersion also reduced SER of papaya and mango [92,93,100,101], albeit with less efficiency than its effect on anthracnose. Therefore, various studies have integrated hot-water treatments in their postharvest treatment protocol.

Table 1. Summarize the technologies used for controlling fruit SER.

	Treatment	SER-Causing Target Pathogen	Fruit Host	References
Preharvest chemical control	Benlate	*Lasiodiplodia theobromae*	Citrus	[47]
	Copper-based chemicals	*Lasiodiplodia* spp.	Mango	[49]
	Fluopyram + pyrimethanil or fludioxonil + cyprodinil	*Lasiodiplodia theobromae*	Mango	[4]
Postharvest chemical control	Benzimidazole: benomyl or thiabendazole	*Lasiodiplodia theobromae*	Mango	[53]
	Prochloraz and fludioxonil	*Lasiodiplodia theobromae*	Mango	[56]
	Tebuconazole + trifloxystrobin, azoxystrobin or carbendazim	*Lasiodiplodia theobromae*	Mango	[57]
	Thiabendazole, imazalil and fludioxonil	*Diplodia natalensis*	Mango	[58]
	Salts: potassium sorbate and potassium phosphite	*Diaporthe citri*, *Lasiodiplodia theobromae*	Lemon	[59]
	Thiabendazole	*Lasiodiplodia theobromae*	Lemon	[60]
Preharvest biological control	*Bacillus subtilis*	*Lasiodiplodia theobromae*	Avocado	[64,65]
	Bacillus subtilis	*Lasiodiplodia theobromae*	Mango	[4]
Postharvest biological control	*Bacillus licheniformis*	*Botryosphaeria* spp. and *L. theobromae*	Mango	[66]
	Trichoderma harzianum	*Lasiodiplodia theobromae*	Rambutan	[69]
	Trichoderma viride	*Lasiodiplodia theobromae*	Mango	[70]
Plant extracts	*Thyme oil* vapors	*Lasiodiplodia theobromae* and *Colletotrichum gloeosporioides*	Mango	[81]
	Combined prochloraz and *thyme oil*	*Lasiodiplodia theobromae* and *Colletotrichum gloeosporioides*	Avocado	[84]
	Moringa oleifera, *Syzygium aromaticum* and *Cinnamomum zeylanicum*	*Lasiodiplodia theobromae*, *Phomopsis mangiferae*, *Colletotrichum gloeosporioides*	Mango	[79]
Fruit Ripening inhibition	1-MCP	*Lasiodiplodia theobromae*	Jujube fruit	[31,32]
	1-MCP	*Lasiodiplodia theobromae*	Avocado	[33]
	2,4-D	*Phomopsis* spp. and *Lasiodiplodia* spp.	Citrus and mango	[27,28]
Agrotechnical methods	Harvesting with short stems (pedicel)	*Lasiodiplodia theobromae*	Mango	[40,41]
	Pruning, exposure to sunlight	*Lasiodiplodia theobromae*	Mango	[6]
Physical treatment	Hot-water or combined hot water and gamma irradiation	*Lasiodiplodia theobromae*	Papya and Mango	[92,93,101]

Table 1. *Cont.*

	Treatment	SER-Causing Target Pathogen	Fruit Host	References
Combined	Benomyl dip in hot water	*Dothiorella dominicana* and *Lasiodiplodia theobromae*	Mango	[102–104]
	Combined HWB along with prochloraz followed by 2,4-D	*Phomopsis* spp. and *Lasiodiplodia* spp.	Mango	[28]
	Hot-water treatment with benomyl followed by a prochloraz	*Dothiorella dominicana*	Mango	[104]
	Combined *Bacillus subtilis* and hexanal	*Lasiodiplodia theobromae*	Mango	[85]

3.7. Combined Treatments

With the emergence of various fungicide-resistant isolates, and the specificity of each fungicide on the one hand, and difficulty achieving complete protection against postharvest decay using only physical or natural control on the other, one treatment alone cannot generally provide complete protection against all postharvest diseases. Thus, a combination of strategies must be applied to enhance the efficiency of coping with various postharvest diseases.

Integrating heat treatment with some chemical compounds resulted in a synergistic increase in control effectiveness leading to a significant decline in the chemical concentration needed to control postharvest decay. In several countries, mangoes were treated by immersing the fruits for 5 min at 52 °C combined with benomyl [76]. Indeed, benomyl dip in hot water was reported to efficiently control SER caused by both *Dothiorella dominicana* and *Lasiodiplodia theobromae* [102–104]. Carbendazim can also be applied with hot water (52 °C) for SER and anthracnose control [105]. Hot thiabendazole is generally effective at controlling SER, but provides poor control of anthracnose [102].

In mango, a combination of hot water brushing (HWB) along with prochloraz followed by 2,4-D significantly reduced SER and side decay by 50–70% and improved mango fruit quality during prolonged storage [28]. This combination reduced the incidence of SER from 86 to 10% in 'Tommy Atkins' mangoes. One of the common treatments for mangoes includes the combination of chlorine sterilization followed by HWB, then acidic prochloraz application followed by waxing [106]. Another combination offered in Australia is hot-water treatment with benomyl followed by a prochloraz spray, which provides effective control of anthracnose, SER and alternaria rot in mangoes [104].

Efficient control of postharvest disease, including SER, should therefore integrate several approaches. These can include preharvest chemical or biological application, harvesting with short stems, removing the sap, surface sterilization, hot-water treatment, chemical or biological fungicide treatment, waxing or coating, ripening inhibition, and cold storage. However, not all of these treatments are necessary if the disease rate is low or if the storage period is relatively short. In addition, each approach costs money. Thus, each packing house must customize their own protocols to control postharvest diseases and prolong fruit quality during storage.

4. Fruit Stem-End Microbiome and Modern Molecular Tools

The last decade was accompanied with major advantages in high-throughput sequencing of DNA and RNA and computational mapping. Those methods could be applied to fast sequencing of fungal genomes, fruit-pathogen interaction in the transcriptional level and study the dynamics of microorganism in the fruit stem-end. Today, the genomes of most of the hosts and SER causing pathogens are available, which will enable transcriptome analysis that could open new insights for better understanding the host effective defense response and the switch of pathogenic fungi from endophytic to pathogenic stage. This transcriptome analysis could lead to the development of new control methods.

The microbiome consists of all of the microorganisms (fungi and bacteria) inhabiting the plant tissue. The advances in high-throughput sequencing of DNA have been applied to study the plant microbiome. In apples, variety and rootstock modulate the endophytic microbiome, suggesting coevolution of a specific genotype with its microbiome [19]. When these methods were used to study the mango stem-end microbiome during postharvest storage and disease development, healthier stem-ends (with a lower incidence of SER) showed more diverse microbial communities [6]. In contrast, an increase in SER incidence was correlated with a reduction in microbiome diversity and an expansion of one or several fungal pathogen families, such as *Pleosporaceae* and *Botryosphaeriaceae*. In addition, the increase in fungal abundance and SER was correlated with an increase in the chitin-degrading *Chitinophagaceae* bacteria and a reduction in biocontrol agents [6]. This implies that the stem-end microbiota is a dynamic system that can be modified to control SER-causing pathogens.

5. Summary and Future Directions

Several fungal pathogens can cause SER. Their conidia or spores are carried by wind or water and penetrate through natural openings in the fruit. The penetrated fungi endophytically colonize the phloem or xylem of the pedicel and localize in the tissue connecting the stem-end to the fruit body (Figure 1). During ripening, the fruit becomes susceptible to various fungal pathogens (Figure 2) that switch from endophytic stage to necrotrophic stage and cause SER (Figure 1). Although SER disease leads to significant fruit loss, the basic science of fruit SER is largely unknown. Nevertheless, studies have shown that different treatments can decrease the incidence of SER by directly inhibiting the fungal growth or indirectly inducing host resistance (Table 1), or by indirectly changing the stem-end microbiome to a more diverse and less pathogenic community. With the increased availability of new tools such as deep sequencing, new studies are expected to emerge, leading to a better understanding of the host–pathogen interaction and the stem-end holobiont, which could lead to the development of new means to reduce fruit SER.

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