



Review

Bio-Circular Perspective of Citrus Fruit Loss Caused by Pathogens: Occurrences, Active Ingredient Recovery and Applications

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Abstract: The Sustainable Development Goals (SDGs) contribute to the improvement of production and consumption systems, hence, assisting in the eradication of hunger and poverty. As a result, there is growing global interest in the direction of economic development to create a zero-waste economy or circular economy. Citrus fruits are a major fruit crop, with annual global production surpassing 100 million tons, while orange and tangerine production alone account for more than half of the overall production. During pre- and postharvest stages of citrus fruit production, it is estimated that more than 20% of fruit biomass is lost, due, primarily, to biotic stresses. This review emphasizes causes of fruit losses by pathogenic caused diseases and proposes a bio-circular perspective in the production of citrus fruits. Due to substantial changes in fruit characteristics and environmental conditions, some of the most economically significant pathogens infect fruits in the field during the growing season and remain dormant or inactive until they resume growth after harvest. Peel biomass is the most significant by-product in citrus fruit production. This biomass is enriched with the value-adding essential oils and polysaccharides. For the complete bio-circular economy, these active ingredients can be utilized as citrus postharvest coating materials based upon their functional properties. The overall outreach of the approach not only reduces the amount of agricultural byproducts and develops new applications for the pomology industry, it also promotes bio-circular green economic, which is in line with the SDGs for the citrus fruit industry.

Keywords: antifungal properties; by-products; citrus essential oils; fruit drop; fruit wax; greening; pectin



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1. Introduction

The Sustainable Development Goals (SDGs) of the United Nations aim at sustaining the well-being of the global population and preserving the environment due to the concerns of climate change and the scarcity of natural resources by 2030. The agenda advocates for a new paradigm of growth, in which economic and social development ensure sustainability [1]. Among all others, SDG 12 addresses sustainable consumption, the need to enhance

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resource use efficiency and reduce food loss by recycling and reusing [2]. This goal, as do other SDGs, such as Zero Hunger (SDG 2) and No Poverty (SDG 1), contributes to better production and consumption systems that contribute to the eradication of hunger and poverty [3]. As a result, scholars, policymakers, and practitioners throughout the world are increasingly interested in the path of economic development and enabling cyclical thinking toward developing a zero-waste economy or circular economy (CE) [4]. CE is an economic system in which the concept of end-of-life is substituted by reduce, reuse, recover, and recycle within the production line [5]. In recent years, the food industry has taken a number of steps to deal with issues, such as food waste and loss, food safety, production traceability, product quality, and environmental harm [6]. The reduction in food waste or agricultural biomass is one of the most critical sustainability issues for food and agricultural producers [7]. The value recovery process is an essential component in food supply chain circular movements [8]. In line with the CE principle, food biomass is now regarded as a resource for bioproducts, such as active ingredients, enzymes, and organic acids, along with energy and water through biorefinery approaches [8]. However, stakeholders across the value chain, from product design to production and distribution to waste disposal, must appreciate the advantages of using biomass [7,9].

Citrus production has exceeded 140 million tons per year globally, with orange and tangerine production alone accounting for more than half of the total volume [10–12]. Commercial citrus production has been recorded by the Food and Agriculture Organization [10] in over 100 different countries, across all regions in the tropical and subtropical areas that serve the demand, mainly for fresh consumption [10,13]. A tremendous amount of money is lost annually due to fruit drop, which occurs during the flowering stage and continues till harvesting. Fruit drop is caused by various biotic and abiotic factors [14,15]. Abscission is a physiological process that is active and involves the breakdown of cell walls at specific locations, called abscission zones, which are frequently related to stress (i.e., salinity and pathogens causes diseases) and senescence [16]. Three waves of abscission are known during fruit production. The first wave is generally at the blooming stage, causing high abscission in buds, flowers, and ovaries [17]. After fertilization fails, the second wave emerges, following competition for nutrition (both among fruitlets and between fruitlets and vegetative shoots) and failure of embryo development, and the third wave mainly contributes to losses in fruit drop occurrence [16,17]. More importantly, plant diseases, such as stem end rot and post-bloom fruit drop, are the primary biotic causes of loss in the orchard during pre-harvesting, with an estimated total loss of fruits of about 20% in the overall output yield [18–20]. While the effects of abiotic stresses (i.e., salt and heat stress) on fruit drop have been thoroughly explored [21-23], the influence of pathogen-caused disease has not been the subject of a collective review. After harvesting, fresh citrus fruits require standards of quality; therefore, "cosmetic thresholds", especially from pests and postharvest diseases, were devised [24]. Premature fruit drops are the major loss in orchards and farmers usually let them decompose naturally due to high management costs [25]. The accumulation of such waste is known to be the cause of disease pathogens in the orchard, which are difficult to eliminate and even cost more for maintenance. Postharvest losses due to fungal and microbial invasion account for up to 35% of total losses, with the most common diseases being caused by green mold, blue mold (Penicillium spp.), and sour rot (Geotrichum candidum) [26]. Moreover, fruit rot diseases are responsible for a variety of fungi, including Penicillium, Alternaria, Aspergillus, Colletotrichum, Botryodiplodia, and Phomopsis [27].

Being the main sources of pectin cellulose, hemicellulose, phenolic compounds, and citrus essential oils can be recovered from peel biomass [28–31]. Citrus essential oils have the capacity to suppress postharvest fungal infections, which is very useful in global fruit production [25,32,33]. Citrus polysaccharides, such as pectin, are used as hydrocolloids for food industries, with global demand reaching over USD 1 billion [34–36]. Considering the efforts for bio-circular green production and in line with the SDGs for sustainable development in the citrus industry, this review proposes a complete use of biomass from losses, particularly by pre-harvesting infectious diseases and suggests the applications of value-adding components from biomass during postharvest and handling. By minimizing the volume of agricultural by-

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products and creating novel applications in the pomology industry, this review can potentially be a significant step toward the Sustainable Development Goals (SDGs).

2. Citrus Pre-Harvest Losses

2.1. Gum Diseases of Citrus Trees Caused by Phytophthora spp. Infection

All commercial citrus scion cultivars are vulnerable to *Phytophthora* spp. Infection; however, when grafted onto certain rootstocks, they become moderately susceptible to bark infection [37]. These pathogens cause yield losses worldwide, especially in susceptible rootstocks in citrus plants, thereby causing considerable concern among growers [38,39]. Cankers and gum appear on the trunks and main branches of several citrus cultivars, indicating the presence of the disease (Table 1). These cankers girdled the tree's limbs and trunk, often resulting in the tree's demise. Cankers are visible in some situations, but only have minor external symptoms in others, and it is only after the outer bark is removed that significant necrotic areas are discovered. Cracks in the bark of affected trees frequently discharge a pale-yellow gum. Though a fungus complex has been linked to Rio Grande gummosis, its etiology is unknown [40]. Phytophthora spp. has been identified as harmful to citrus, causing a variety of diseases that affect the roots, trunk, branches, fruits, and shoots [41]. In Mediterranean regions, P. citrophthora causes gummosis and root rot and is the most common cause of brown rot. Phytophthora spp. is widely found in citrus soils, causing fibrous root deterioration in susceptible rootstocks, as well as lesions on structural roots and crown rot [42]. P. citrophthora attacks aerial plant parts more frequently than *P. parasitica* and also produces brown rot, a disease that affects fruits, causing a firm light-brown decay, and finally, fruit fall. The reason for this is because P. parasitica does not produce aerial sporangia but *P. citrophthora* and other species do. Therefore, a citrus tree is more susceptible to *P. citrophthora* than any *Phytophthora* spp. [43].

2.2. Citrus Greening Disease

Huanglongbing (HLB), also known as citrus greening disease, is the most serious citrus disease in areas where both the disease and its vector are present, primarily in Southeast Asia, India, and South Africa [44]. The term "Huanglongbing" is a Chinese term that translates as "yellow dragon disease", referring to the disease's symptoms, which include prominent yellow shoots. This disease is caused by a Gram-negative bacterium called *Candidatus* Liberibacter (*Ca.* L.) and is spread through natural vectors, such as the psyllids, *Trioza erytrea*, and *Diaphorina citri* [45,46]. This disease has been classified into three subtypes. The first type is Asiatic and it is associated with Ca. L. *asiaticus* [47]. Second, there is an African form associated with Ca. L. *africanus* [41]. Finally, there is an American form, associated with Ca. L. *americanus* [45,46,48]. The three strains of Ca. L. exhibit distinct temperature responses. The asiaticus is a thermo-tolerant species that can tolerate temperatures above 30 °C, whereas the africanus is thermolabile and prefers temperatures between 22 and 25 °C [46]. For the americanus, it, nonetheless, prefers lower temperatures between 17 and 27 °C [49].

Symptoms include asymmetrical mottling of the leaves and frequently yellowed midribs. Sectors of the canopy decline and dieback first, followed by the canopy as a whole declining and dying. Once the tree is nearly completely infected, yellow shoots will appear. Symptomatic fruits are lobbed, frequently contain aborted seeds, and have an unpleasant flavor. Fruit production is decreased, symptomatic fruits are small, and symptomatic fruits frequently drop prematurely. Over a period of two to three years, the tree deteriorates and eventually dies [45]. The nature of the disease occurs as a result of pathogens penetrating the phloem and attacking the vascular system, clogging the veins and significantly impairing water and nutrient transport [46]. Current management strategies are aimed at eradicating vectors, preventing infection spread, and managing infected trees. Individual or combined approaches will have varying degrees of success, depending on the severity of the infestation. The most frequently used practices are preventing infection spread through tree removal, protecting grove edges through intensive monitoring, pesticide use, and

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biological control of the vector [50]. According to Lee [45], to assure the production of healthy plants and the prevention of the spread of contaminated nursery stock, HLB control involves quarantine, clean stock, and certification procedures. Regular surveys are used to identify early signs on trees, which are subsequently removed; control of the psyllid vector through survey and pesticide application; and replanting with clean plant material are all beneficial in places where HLB has not yet been established. Depending on the percentage of the canopy impacted, yield decline can range from 30% to 100% and is primarily caused by early abortion of fruits from afflicted branches [51].

2.3. Citrus Bacterial Disease

2.3.1. Citrus Canker Disease

Currently, citrus canker has been detected in over thirty countries, including Southeast Asia, South America, the islands of the Pacific and Indian Oceans. Due to its high susceptibility, among commercial citrus types and rootstocks, Asiatic citrus canker is particularly harmful to grapefruits (C. paradisi), limes (C. aurantifolia and C. limettioides), trifoliate oranges (Poncirus trifoliata), and their hybrids. Citrus fruits that are infected during the early stages of growth fracture or have malformations and the seriously infected fruits fall immaturely, even though only sporadic canker lesions may appear on the surface of fruits in later growth stages. Light infection renders fresh fruits unfit for commercial distribution. Fruit infections typically have a similar severity as foliage infections. In sensitive citrus trees that have already experienced severe foliage infection, fruit infection of 80% to 90% is not unusual. Such extreme defoliation, leaving only bare twigs, frequently results from such high foliage infection [52]. One of the biggest and most significant families of bacterial phytopathogens is the Xanthomonadaceae, which includes Xanthomonas citri subsp. citri. Citrus canker disease is caused by the pathogen X. citri subsp. citri, which has been extensively studied in terms of epidemiology and disease control as a pathogen of a worldwide significant fruit crop [53]. Conspicuous elevated necrotic lesions that form on leaves, twigs, and fruits identify infected plants as having them. By running the fingertips over the surface of affected tissues, lesions can be found. On leaves, they initially show as 2-10 mm round, oily-looking patches, typically on the abaxial surface (reflecting stomatal entry following rain dispersal). Lesions frequently have similar sizes. Later, tissue hyperplasia brought on by the infection may cause both epidermal surfaces to break. Circular lesions on leaves, stems, thorns, and fruits develop into elevated, blister-like lesions that eventually turn into white or yellow spongy pustules. These pustules eventually thicken and darken to become a corky, rough-to-the-touch canker that ranges in color from pale tan to brown. With transmitted light, it is simple to see the water-soaked border that frequently forms surrounding the necrotic tissue. Pustules on stems may group together to divide the epidermis throughout the length of the stem and, occasionally, immature stems may girdle. Older lesions on leaves and fruit typically have rising margins, a sunken center, and, occasionally, a yellow chlorotic halo around the edges (which may vanish as canker lesions age) [54]. The bacterium (Xanthomonas) is rod shaped, Gram negative, and has a single polar flagellum. It measures $1.5-2.0 \times 0.5-0.75$ mm. Growth requires aerobic activity. Because xanthomonadin pigment is produced, colonies on culture media are typically yellow. The use of resistant cultivars in combination with integrated systems of suitable cultural practices and phytosanitary measures, including quarantine and regulatory programs, results in the most successful management of canker. The fundamental tenets of the specific approaches include avoiding, excluding, or eradicating the pathogen, minimizing the spread of the pathogen, reducing the amount of inoculum available for infection, and protecting susceptible tissue against infection [55]. There have been reports of copper resistance in X. citri subsp. citri and X. alfalfae subsp. citrumelonis strains. The usage of copper-based bactericides, which are crucial substances for the management of Xanthomonas-related diseases on citrus, is seriously impacted by copper resistance [56].

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2.3.2. Bacterial Blast Disease

More than 180 plant species, including *Citrus* spp., are infected by *Pseudomonas syringae* pv. *syringae*, which also produces black pits in orange fruits and bacterial blast in orange (*C. sinensis*) and mandarin (*C. rediculate*) fruits [57,58]. This disease has been reported in Iran, Montenegro, Tunisia, Turkey, and the USA [59]. Black spots on the petiole wings and water-soaked lesions were the first noticeable symptoms on leaves. Later lesions spread to the twigs surrounding the base of the petiole as well as to the midvein of the leaves. The leaves soon withered, rolled while remaining securely attached, and eventually fell to the ground without petioles. Twigs' necrotic regions became larger and after 20 to 30 days, the twigs succumbed to death. A 50-hectare citrus plantation in Antalya suffered significant damage, with a disease prevalence of about 100%. The most popular therapy for a number of bacterial illnesses of fruit trees was the use of copper compound sprays (mostly the Bordeaux mixture) during the fall and winter before the infection *P. syringae* pv. *syringae* appears in late winter and spring [60]. The selection of copper-resistant *P. syringae* pv. *syringae* strains in mango orchards, where heavy copper spraying was utilized for disease management, may be the cause of their frequently reduced efficacy [61].

Table 1. Causes of losses and types of by-products during the production of citrus fruits.

Citrus Fruit Diseases	Disease Characteristics	Types of by-Products	Citations
Citrus gumnosis	Pathogen: Phytophthora spp. Symptoms: Cankers and gum or citrus trunks and branches. The infection causes damage to fibror roots in vulnerable rootstocks an crown rot, thereby causing fruit fall.	branches, shoots die	[40–42]
Citrus greening	Pathogen: Candidatus Liberibact Symptoms: Pathogens infiltrate the phloem and assault the vascular system, blocking water and nutrient transport. Yellow shoots and mottled leaves with yellow midribs may occur causir die-back. Infected fruits are lobbed, have aborted seeds, and prematurely drop off.	Fruit drop	[62,63]
Black rot	Pathogen: Alternaria spp. Symptoms: On fruits, symptom range from light brown, depresse patches to dark brown circles. Young shoot apexes are defoliate in severely affected trees. Young infected fruits and leaves fall, an mature lesions-covered fruits ar unmarketable. Alternaria spp. ca cause black rot by causing laten infections on the calyx and disc then entering the columella as th fruit grows.	d G Die shoots and d fruit drops e in t	[64–66]

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Table 1. Cont.

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Citrus Fruit Diseases	Disea	nse Characteristics	Types of by-Products	Citations
Brown rot		Pathogen: Phytophthora citrophthora and Phytophthora nicotianae Symptom: light brown, leathery decay. This disease is associated with citrus gummosis or citrus foot rot.	Fruit drops	[67–70]
Anthracnose		Pathogen: Colletotrichum spp. Symptoms: The disease caused necrotic petals and fruit lesions during pre-harvesting. Infected twigs and lesions generally had black fructifications. During postharvest, the infected fruits had brown-to-black tear stains that turned silver-gray, 1.5 mm or bigger lesions.	Branch die-off, fruit drops during pre-harvesting stage and fruit loss during postharvesting	[71,72]
Green and blue mold	Green mold	Pathogen: Penicillium digitatum and P. italicum Symptoms: during postharvest, the fruit peel is soft and decolored and soaky. During disease development, the fruit surface is covered with aerial white mycelium turns to olive with spore development.	Fruit loss	[73,74]
	Blue mold			
Sour rot		Pathogen: Geotrichum citri-aurantii Symptoms: Storage fruits in the humid condition cause the fungus growth with a light brown to yellow tint. The symptom appears extensive water-soaked lesions, and artroconidia and mycelia on the fruit surface.	Fruit loss	[75–77]

3. Citrus Postharvest Disease

Filamentous fungi are the most common cause of citrus fruit postharvest diseases. Because of major changes in fruit features and environmental conditions, they are some of the most economically important pathogens that infect fruits in the field during the growing season and stay latent or quiescent until they resume growth after harvest. *Alternaria* sp. Ellis & N. Pierce in N. Pierce causes alternaria rot or black rot; *Colletotrichum* sp. (Penz.)

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Penz. & Sacc. in Penz. causes anthracnose; or *Phytophthora* spp. causes brown rot in Penz. Other economically significant diseases infect the fruits by rind wounds or damage sustained during harvest, transportation, or postharvest handling. Green and blue molds are caused by *Penicillium digitatum* (Pers.: Fr.) Sacc. and *P. italicum* Wehmer, respectively; sour rot is caused by *Geotrichum citri-aurantii* Ferraris E.E. Butler [78].

3.1. Black Rot Caused by Alternaria spp.

Alternaria spp. is responsible for a variety of citrus diseases, including alternaria brown spot in tangerines (Citrus reticulata Blanco) and black rot in a variety of Citrus spp. fruits [79-81]. The disease is a significant postharvest problem that may appear in the field prior to harvesting. In fruits, symptoms include light brown, slightly depressed spots to circular and dark brown areas on the external surface (Figure 1A). Young infected fruits and leaves frequently fall and mature fruits with lesions are unmarketable, resulting in significant economic losses [66]. During postharvest handling, extreme weather conditions, such as hot, dry summers and cool, moist winters, favor the disease. Citrus black rot begins as a core rot, in which the pathogen colonizes the fruit's columella. The disease occurs most commonly on navel oranges and on tangerines and their hybrids during storage [82]. The fungus invades and colonizes the columella in black rot, which is usually a core rot. Except for the fact that the fruits are frequently more vividly colored than healthy fruit, there is usually little visible sign of infection. Infection can enter the fruits through wounds or natural apertures in the stylar end. The blossoms that have been affected by the fungus may wither before opening or drop off right after fruit set. Alternaria spp. can also cause black rot by forming latent infections on the calyx and disc, then invading the columella as the fruit matures [64]. Young leaves show irregular brown necrotic patches with characteristic yellow halos as symptoms [83]. In severely afflicted trees, the apex of young shoots is entirely defoliated. Citrus black rot disease can be caused by a variety of Alternaria species, including those that are not *A. alternata* citri [64].

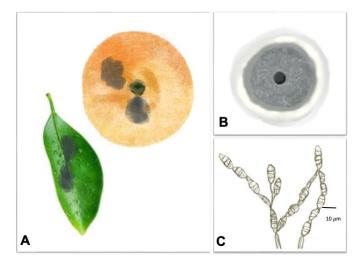


Figure 1. Black rot of citrus fruit causes by *Alternaria alternata*, (A) = the symptom on fruit and leaf; (B) = colony and (C) = conidia morphology.

Morphologically, typical *A. alternata* colonies on potato dextrose agar media (PDA) are lettuce green to olive green in color, with a conspicuous (2–5 mm) white border and spread to a size larger than 70 mm in diameter after 7–10 days (Figure 1B). Based on the sporulation behaviors of single-spored colonies, *A. alternata* is distinguished by the formation of conidial chains six to fourteen conidia in length and the growth of several secondary, and rarely tertiary, chains, two to eight conidia in length. Through the extension of secondary conidiophores from distal terminal conidial cells and subsequent conidium production, chain branching happens in a sympodial way. Conidia are tiny (20.0–50.0 μm) length and

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oval in shape, with transverse and vertical walls dividing them and minimal apical extension growth (Figure 1C). The septate hyphae and conidiophores are light brown [65].

3.2. Brown Rot Caused by Phytophthora citrophthora

The fungi Phytophthora citrophthora (R. E. Sm. & E. H. Sm.) Leonian and P. nicotianae Breda de Haan (P. parasitica Dastur) are the most commonly connected with citrus foot rot, also known as gummosis or root rot [67]. It is commonly known as brown rot in storage fruits. Infected fruits when entering the packing house are frequently the major cause of postharvest deterioration issues in the entire batch. The infectious symptoms are leathery, light-brown lesions (Figure 2A) [70]. The morphological characteristics of *Phytophthora* spp. have been described [68,69]. Mycelium is thick and cotton like [84]. The colony is finely radiated with stellate and flame-like growth, growing at optimum temperatures of 24–28 °C (Figure 2B). Sporangiophores branch irregularly, with swelling at the point of branching. Some sporangia grow single or in loose sympodia, with a swelling at the branching point. Sporangia are noncaducous and generally papillate, with two apices that are often divergent. P. citrophthora makes sporangia in a variety of forms, including spherical, ovoid, obpyriform, obturbinate, and ellipsoid. Multiple papillae and offset pedicel attachments can cause distorted sporangial forms in water. Sporangia are $18.0-60.0 \times 23.0-90.0$ µm (average 30.0×45.0 µm). The length x breadth ratio is less than 1.6 (Figure 2C). Chlamydospores are rarely found on citrus isolates in culture, although they do form in the roots. Chlamydospores are 25.0–35.0 μm in diameter (average 28 μm). Hyphae are smooth and coarse and 3.0–7.0 μm in diameter [68,69].

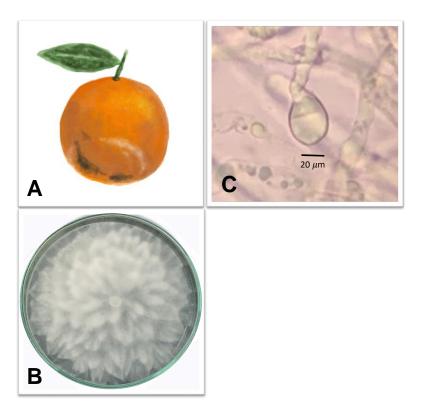


Figure 2. Brown rot of citrus fruit causes by *Phytophthora* spp., (A) = the symptom on fruit; (B) = colony and (C) = morphology of sporangia.

3.3. Anthracnose Caused by Colletotrichum spp.

Colletotrichum spp. causes citrus anthracnose, which is a serious disease in many citrusgrowing regions across the world. This disease has been reported in Algeria, Bermuda, Brazil, California, Italy, Mexico, Morocco, Pakistan, Portugal, Tunisia, and Turkey [72,85–88]. Many citrus species, including Mexican lime (*C. aurantifolia*), sweet orange (*C. sinensis*), and

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grapefruit (*C. paradisi*), have petal necrosis and necrotic lesions on their fruits. On mature fruits, brown to black streaks (tear stain) appeared, sometimes turning silver gray (Figure 3A). These lesions were 1.5 mm in diameter or greater. On the surface of the lesions and the dry extremities of diseased branches, dark-colored fructifications were found in general [71].

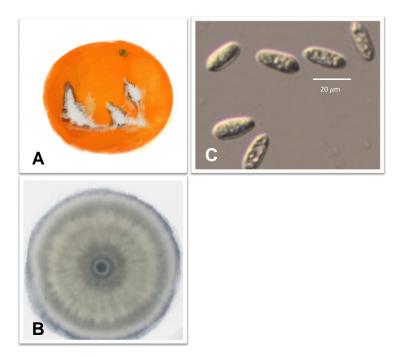


Figure 3. Black rot of citrus fruit causes by *Colletotrichum gloeosporioides*, (\mathbf{A}) = the symptom on fruit; (\mathbf{B}) = colony; and (\mathbf{C}) = conidia morphology.

Morphologically, the mycelium in the colonies was creamy, white to pale gray, sparse, and more or less cottony, with profuse bright-orange conidiomata (acervuli) and setae (Figure 3B). Conidiophores within the conidiomata were branching with hyaline conidiogenous cells that were sub-cylindrical and more or less straight. Septate, brown, and slightly pale at the apex, setae were found (Figure 3C). Conidia ($15.0 \times 5.0 \, \mu m$) mounted in lactic acid from actively growing colonies were hyaline and sub-cylindrical with bluntly rounded ends [71,72].

3.4. Green and Blue Molds Caused, Respectively, by Penicillium digitatum Sacc. and P. italicum

Green and blue molds have been discovered in all commercial citrus species and cultivars, including oranges (C. sinensis L.), mandarins or tangerines and their hybrids, clementines (C. clementina hort. ex Tanaka), satsumas (C. unshiu Marcow.), lemons (C. limon Burm. f.), limes (C. aurantiifolia (Christm.) Swingle), and grapefruits (C. paradisi Macfad.) [82]. P. digitatum is a fungal species belonging to the Ascomycota division that is important to both the environment and the food industry. This species, followed by P. italicum, is the most common cause of citrus fruit postharvest rot in the world [89]. Green mold disease contributes to enormous losses all around the world and may be responsible for up to 90% of citrus industry postharvest losses [90]. After about three days of incubation at room temperature, incipient *P. digitatum* and *P. italicum* infections are normally visible to the human eye [90]. The fungus can enter and infect the fruits through wounds caused by wind, hail, and insects, as well as during the harvesting and handling processes. If suitable temperature and conditions are available, the infection area on the fruit peel appears water soaked, soft, and decolorized and can be easily penetrated with the finger (sometimes referred to as clear rot), and a white mycelium grows on it that later turns a blue-green color with spore production [73]. During disease development, the fruit surface is completely covered with an aerial mycelium that produces spores and then begins to shrink, resulting in a sunken mummified shape in the case of green mold and a sticky mass in the case of blue

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mold (Figure 4A,E) [91]. Both fungi produce hydrolytic enzymes, primarily polygalacturonases and cellulases, as necrotrophic pathogens, which appear to be responsible for tissue maceration throughout disease progression [92]. In contrast to green mold, which seldom contaminates surrounding fruits, blue mold can spread more quickly and directly in healthy fruits in storage boxes. Although it is not uncommon to detect indications of both diseases in the same fruits in packing facilities or marketplaces, with combined infections on fruits stored at room temperature, green mold frequently outgrows blue mold [73,74]. A considerable number of thiabendazole (TBZ)- (84%) and imazalil (IMZ)-resistant (77%) strains of *P. digitatum* were found when the sensitivity of 75 distinct *P. digitatum* strains to seven different fungicides was assessed. These fungicides are the two most frequently employed in citrus postharvest [93].

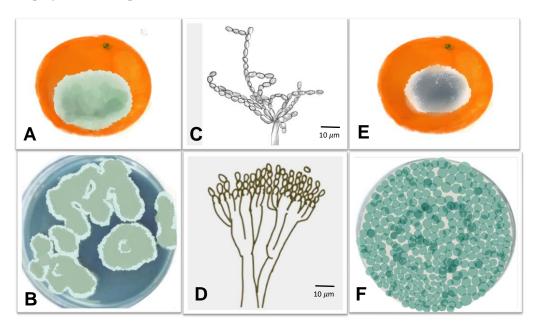


Figure 4. Green and blue molds caused by *Penicillium* spp., (A,E) = the symptom on fruit from *P. digitatum* and *P. italicum*; (B,F) = colony from *P. digitatum* and *P. italicum* and (C,D) = conidia morphology from *P. italicum* and *P. digitatum*.

The colonies of *P. digitatum* are planar and olive green on one side and colorless to cream yellow or mild dull brown on the reverse (Figure 4B). The texture of the colony is velutinous, with no exudate droplets. The fungus can germinate in PDA at 5 °C and generate colonies up to 3.0 mm in diameter, but no growth is identified at 37 °C [94]. The conidial apparatus is extremely weak and it tends to disintegrate into several cellular components. Conidiophores are terverticillate, borne from subsurface or aerial hyphae, irregularly branched, and made up of short stipes with few metulae and branches that end in whorls of three to six phialides, which are sometimes solitary, cylindrical, and have a short neck. Conidia are smooth walled, ellipsoidal to cylindrical, variable in size, roughly $3.5-8.0 \times 3.0-4.0 \mu m$ (Figure 4C) [92]. P. italicum colonies are flat, sporing heavily, blue or gray green in color, and granular due to the presence of conidiophore bundles and conidial heads. The reverse of the Petri dish is uncolored or gray to yellow brown, although it can turn to brownish orange or red brown. Asymmetric penicilli containing tangled strands of conidia makes up the conidial apparatus (Figure 4F). Conidiophores are terverticillate, hyaline, usually with the branches appressed, with $100.0-250.0 \times 3.5-5.0 \,\mu m$ stipes and smooth-walled metulae containing three to six phialides each and they originate from the substratum or occasionally from superficial hyphae [74]. The phialides are cylindrical in shape and have small but noticeable necks. Conidia are cylindrical at first, then elliptical or subglobose. They are smooth, $4.0-5.0 \times 2.5-3.5 \mu m$ in diameter, greenish, and smooth

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walled. Fresh isolates have occasionally revealed colorless to light-brown sclerotia with a diameter of 200–500 m (Figure 4D).

3.5. Sour Rot Caused by Geotrichum citri-aurantii

The disease has been reported from most areas of the world where citrus is grown and with evidence of infection in tangerines, oranges, grapefruit, and lemons. Citrus sour rot is one of the most serious citrus diseases caused by a heterothallic fungus G. citri-aurantii. After ten days in 85–90% relative humidity (RH), the fungus' major characteristics in pathogenicity testing were light brown to yellow color and large water-soaked lesions with significant quantities of artroconidia and mycelia on the fruit surface (Figure 5A) [75]. Guazatine is the only chemical fungicide that efficiently controls sour rot, in addition. In several Chinese citrus-producing regions, sour rot has become more common in recent years. At the same time, G. citri-aurantii resistance has risen yearly in citrus-growing regions [95]. Colonies on PDA were usually dull white, but some were dazzling white, and they grew at a daily rate of 8.8–16.0 mm for 5 days at 25 °C. All isolates on PDA showed dichotomous branching of mycelium at the colony's edge, a hallmark of G. citri-auranti (Figure 5B). All isolates had chains of arthrospores that originated from hyphal segmentation. Arthrospores were generally oval, with a few cylindrical ones thrown in for good measure. Spores are cylindrical at first, then barrel shaped, ellipsoidal, or subglobose, measuring $2.0-8.0 \times 3.0-50.0 \mu m$, but most typically $3.0-6.0 \times 6.0-12.0 \mu m$. Each spore has one to four nuclei, most commonly two (Figure 5C) [76,77].

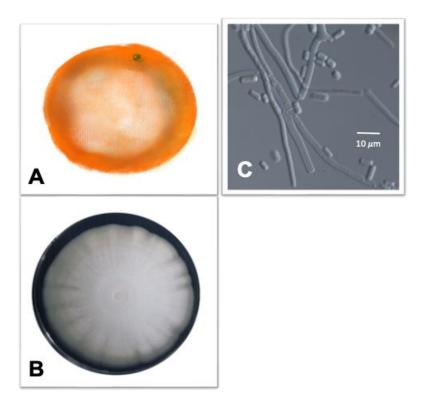


Figure 5. Sour rot caused by *Geotrichum citri-aurantii*, (\mathbf{A}) = the symptom on fruit; (\mathbf{B}) = colony and (\mathbf{C}) = conidia morphology.

4. High-Value Component Recovery

4.1. Citrus Essential Oils

Essential oils are by-products of plant defense and pollinator attraction among other ecological functions. As other secondary metabolite groups, they illustrate biological activities that make them able to be used as herbicides, pesticides, and anticancer compounds [96,97]. They are also utilized as in the food and pharmaceutical industries due to their therapeutic, antimicrobial, and antioxidant activities [98,99]. There are more than

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200 components present in the essential oils, both volatile (90.0-95.0%) and non-volatile (1.0–10.0%) [100]. Normally, the volatile compounds are those of phenylpropanic derivatives or terpenes [101,102]. Citrus spp. fruits are susceptible to a variety of fungal, viral, and bacterial infections from the nursery through the postharvesting and bearing phases, resulting in enormous losses to the plantation and its output. Khamsaw et al. [25] described that fruit drops were the most significant pre-harvest loss and the volume increased over time till harvesting. Given the volume and practicality of biomass recovery in the citrus industry, ongoing efforts are being made to investigate new applications, especially when bio-circular green production is a core concern [103-106]. Essential oils, especially from the citrus species, are the most important raw materials in the fragrance and pharmaceutical industry [107]. Citrus peel is the most familiar and rich source of essential oils. The oil gland is localized in the exocarp of citrus fruits. The outer colored peel is often referred to as the flavedo [108,109] (Figure 6). The amounts of citrus oil range between 2.0 and 5.0%, depending on the methods of extraction [110,111]. Fruit beverages, confectioneries, soft drinks, eau de cologne, soaps, cosmetics, and household items are all flavored using these oils [112,113]. They are also employed as immune stimulants and anti-inflammatory drugs in medicinal therapies.

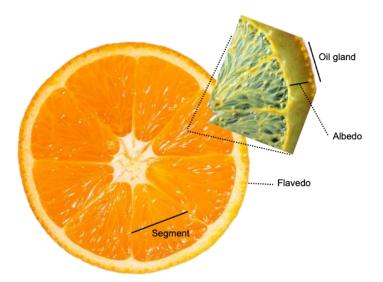


Figure 6. Localization of oil gland in orange peel (flavedo).

In total, 29 compounds were found in the *C. reticulata* Blanco (Ponkan) essential oil, 27 of which account for 99.8% (w/w) of the total oils. The main components in the essential oil were monoterpene hydrocarbons ($C_{10}H_{16}$), which included principally D-limonene (60.8%), γ -terpinene (10.0%), and β -myrcene (7.43%) [32], while the limonene is highly sought for commercial applications as a food additive and in pharmaceuticals [114,115]. Although the volatile profile was consistent throughout the period of fruit growth, it was also discovered that peel biomass obtained from the early stage of fruit development included larger amounts of limonene (1.2%) and β -pinene (0.02%) in the essential oil [25].

Globally, citrus oil sales make up roughly 20–25% of the whole market for essential oils in terms of value. Persistence market research described that the citrus oil market has a current value of about USD 3.3 billion in 2022 and is anticipated to grow at a compound annual growth rate (CAGR) of 5% to reach USD 5.3 billion by 2031. East Asia and South Asia are markets rising at significant CAGRs, according to this thorough market assessment. As of 2021, more than 65% of the total market volume for citrus oils is held by North America and Europe due to the growing popularity of aromatherapy products; these regions are seeing an increase in demand for essential oil [116]. More importantly, citrus essential oils are approved by the Food and Drug Administration (FDA) as additives in certain

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types of foods, with the capacity to delay the onset of food deterioration and enhance the organoleptic aspects [117].

4.2. Citrus Polysaccharides

Citrus peel biomass is the major source of polysaccharides that are commercially needed for the pharmaceutical, nutraceutical, food, and cosmetic industries [118]. In the 1920s and 1930s, many companies began producing pectin from citrus pulp biomass from the juice and wine industries [119]. Various polysaccharide types are recovered from the citrus peel, such as soluble sugars, starches, and fibers, including celluloses, hemicelluloses, lignins, and pectins. At present, commercial pectins are almost exclusively derived from citrus peel, apple pomace, and sugar beet pulp [120]. Pectins are acidic heteropolysaccharides, which are classified into three main groups and are widely used due to their gelling properties [121]. It is also advised that the emulsifying activity of citrus peel pectin is higher than that of pectin from other sources [122]. These gelation properties strongly affect their structure, especially on the degree of methyl-esterification [123]. The citrus peel pectin, in particular, has a high commercial need as a gelling agent in jams, confectionary, and bakery fillings, as well as a stabilizer in yoghurts and milk beverages. Other relevant applications are in the cosmetics, personal care (paints, toothpaste, and shampoos), and pharmaceutical (gel caps, detoxifying agents, and drug carriers) industries, as well as the emerging use as a nutraceutical ingredient [124,125]. The peels of lemon and orange are good sources of pectin, which can be extracted using alcohol precipitation [28,126]. Common orange peel contains 6.0% pectin, whereas lemon peel yields 8.0%. The pectin yield recovered from pomelo (*C. maxima* or *C. grandis*) peels was measured to be as high as 23% [47]. Additionally, with the highest content obtained among the neutral sugars, arabinose was the main component in the pectin side chains, followed by galactose, which suggested the presence of rhamnogalacturonan I (RG I). The pectin yield of sour orange (C. aurantium L.) peels is 28.0%. Structurally, the backbones of the homogalacturonan (HG) and rhamnogalacturonan I and II (RG I and II) regions are composed of galacturonic acids (GalA). Neutral sugars, such as galactose [106], rhamnose [71], arabinose, xylose, and fructose, are the main constituents of pectin side chains. Further, the galacturonic acid and glucose contents in the pectin of this type were 65% and 0.4%, illustrating a high purity [122]. The cellulose content from citrus peels ranges from 13% to 14% and hemicellulose is 5% to 6%, respectively [127].

Citrus polysaccharides can be recovered using various techniques, including enzymatic, physical, and chemical methods, with the latter being the most prevalent in industry [128]. The process involves enzymatic hydrolysis and approximately 70.0% of the biomass can be converted to ethanol [129]. Furthermore, citrus peel cellulose was mixed with zinc nano-composite and the defense mechanism against microbial attack and healing due to antioxidative property, therefore, can be exploited in wound dressing [130].

5. Bio-Circular Approaches

Synthetic fungicides, particularly imazalil, thiabendazole, sodium ortho-phenyl phenate, fludioxonil, pyrimethanil, or combinations of these compounds are currently the principal means of preventing postharvest infections in citrus fruits. However, continuous use of these fungicides has resulted in the emergence of isolates of fungi with multiple fungicide resistances, complicating disease management (especially penicillium rots). The current challenge is to provide safer and greener alternatives to existing management methods for citrus postharvest infections that pose less harm to both human health and the environment [27]. The use of plant-derived chemicals has taken precedence due to their antibacterial and antifungal capabilities. These compounds have gained popularity and scientific attention because of their antifungal action, nonphytotoxicity, systemicity, and biodegradability; natural plant products are an appealing alternative or complementary control strategy [27,127].

After harvested, mandarin utilizes the nutrients in itself through a respiration process, leading to further senescence and pathogen infection. Natural compounds, such as

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essential oil, have been demonstrated to be useful in controlling these diseases by lowering the physiological activities of fruits during storage while also reducing overall qualitative and quantitative losses [131]; consequently, they have gained considerable attention. For example, a study from Yang et al. [89] found that eugenol, carvacrol, and cinnamaldehyde were encapsulated in an oil-in-water nanoemulsion using a high-pressure microfluidizer. The antifungal impact of these produced nanoemulsions against *P. digitatum* was found to be significant, with minimum inhibitory concentration (MIC) at 0.125 mg/mL and minimum fungicidal concentration (MFC) at 0.25 mg/mL values. Furthermore, the essential oil of C. reticulata Blanco inhibited P. italicum and P. digitatum dose dependently. Citronellol, octanal, citral, decanal, nonanal, b-pinene, linalool, and c-terpinene were identified as antifungal components in the oils against P. italicum, whereas octanal, decanal, nonanal, limonene, citral, c-terpinene, linalool, and a-terpineol were identified as antifungal to *P. digitatum* [32]. Moreover, the antifungal properties in essential oils from four *Thymus* spp. were studied against P. digitatum, P. italicum, and G. citri-aurantii. Essential oils of wild thymes, Thymus leptobotrys and T. riatarum, at 100 μL/mL, completely inhibited the three pathogens in an in vitro mycelial growth experiment. For the three pathogens, the essential oil of T. leptobotrys showed the lowest at 500 μL/mL MIC value [94]. In another study, Fusarium sarcochroum and P. digitatum, were inhibited at the highest concentration of tangerine oil (256 μL/mL). The MIC of 'Sai-Namphaung' was at 64 μL/mL for C. gloeosporioides. The essential oil of 'Fremont' citrus was clearly effective in the inhibition of C. gloeosporioides at an MIC as small as $16 \mu L/mL$ [25]. In fact, all other essential oil types illustrated the same pattern. The MIC of the 'Fremont' essential oil was 128 μ L/mL for G. candidum, F. sarcochroum, and P. digitatum. G. candidum had MICs of 64 µL/mL, F. sarcochroum and P. digitatum had MICs of 16 μL/mL using commercial citrus oil.

In addition, to delay these onsets of postharvest damage, it is crucial to maintain fruit respiration and dehydration as minimum. Coating has been a common postharvest practice that, in addition to extending the shelf life of fruits, the appearance can also be improved. Coatings are employed as passive and inactive barriers to preserve the quality of citrus fruits and they may also help to reduce the negative effects of chemical and mechanical stresses. Coating can control the release of essential oil and prevent physical damage, such as mechanical or burning, due to the exposal of the essential oils. It protects the fruit to disclose directly to the essential oils and the sticky surface of coating can prevent mechanical damage during transportation [132,133]. Synthetic waxes and/or chemical fungicides were used in traditional coatings, which could be harmful to customers' health and pollute the environment [134]. Essential oils are used in edible active coatings. This technology has developed as a viable and environmentally acceptable alternative to traditional non-edible coatings [135], with the capacity to maintain the quality, stability, and safety of citrus fruits, while reducing the harmful effects of chemicals on consumers and the environment [133,136]. Polysaccharides have been widely used as a coating material in recent years, owing to their inexpensive cost and availability, as well as their increased solubility, stability, safety, nontoxicity, lack of allergens, lack of added taste and odor, and capacity to form clear coatings [137]. Among the various polysaccharides, cellulose derivatives and pectin are two of the most common substances used in edible coatings [138]. Cellulose, the most prevalent component in plant cell walls, has a high number of intra-molecular hydrogen bonds, resulting in water insolubility and a crystalline structure [139]. Because of its linear structure, cellulose is durable, flexible, transparent, and resistant to fats and oils, making it a great coating material with outstanding mechanical and structural capabilities [138]. Commercially available cellulose derivatives, such as methylcellulose (MC), carboxymethyl cellulose (CMC), hydroxypropyl cellulose (HPC), and hydroxypropylmethyl cellulose (HPMC), have been employed as edible coatings for a range of citrus fruits [140]. They act as moisture, oxygen, and carbon dioxide barriers, as well as improving coating formulation adhesion to the product surface [133,136]. Pectin, plant cell walls' major ingredient, is located in the middle lamella of plant cells. They are D-galacturonic acid-based complex heteropolymers, with a wide range of content, structure, and molecular weight. Nontoxic, biodegradable, biocompatible, transparent, and oil- and fat-resistant, Horticulturae 2022, 8, 748 15 of 21

pectin-based coatings have selective gas permeability and low mechanical characteristics [141]. Due to their hydrophilic nature, they have a high water vapor transmission rate [142] and maintain the sensory characteristics and quality of citrus fruits [143].

Mandarin coating agent is usually in the form of water-wax emulsion, such as carnauba wax, shellac, and resin, with or without pesticide [144]. The coating components include primarily proteins or polysaccharides and lipids. At present, there is an increasing interest in the use of polysaccharides, especially from agricultural biomass as a coating agent in combination with natural antifungal components, such as essential oil plant extracts, food additives, low-toxicity compounds generally recognized as safe (GRAS), and microbial antagonists as biocontrol agents [145-148]. When appropriately blended, these ingredients create a thin coating over the fruits, forming a semi-permeable barrier against gases and water vapor that helps coated fruits keep their weight, firmness, and other quality features during storage [149,150]. Along with notably active substances, such as flavonoids, citrus peel biomass contains a considerable amount of both soluble (i.e., pectin) and non-soluble (i.e., cellulose) polysaccharides [151,152]. Figure 7 illustrates the bio-circular approach that makes use of the major biomass during the pre-harvesting process of citrus fruits. Peels from fruit drops have the potential to be utilized as raw materials for antimicrobial citrus essential oil. After the extraction, several types of polysaccharides may, indeed, be recovered from the biomass, which can be formed as a coating agent by integrating the citrus essential oils.

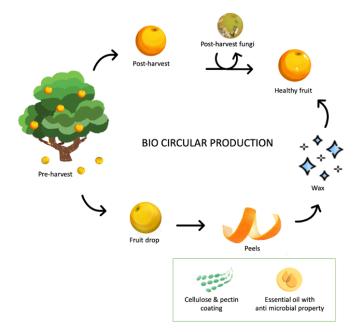


Figure 7. A proposed bio-circular approach for citrus fruit production.

6. Conclusions

During the pre- and postharvest stages of citrus fruit production, more than a quarter of production volume is considered as biomass, mainly from fruit drop and fruit loss. Several pathogens that cause citrus diseases during pre- and post-production are responsible for the fruit losses. Peel biomass is the major by-product from citrus fruit production. It can be utilized for valuable components, such as essential oils and polysaccharides. Citrus postharvest loss due to pathogen attack has been the major challenge for the industry. Edible films and coatings have attracted considerable attention in this respect due to their capacity to stop food items from spoiling during handling, shipping, and storage. Based on the functional properties of the possible value-adding ingredients, they can be bio-circularly incorporated as coating materials for citrus postharvest. This perspective can not only reduce agricultural biomass, it also supports sustainable development in the pomology industry on a global scale.

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