

Review Article

Light: An Alternative Method for Physical Control of Postharvest Rotting Caused by Fungi of Citrus Fruit

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Solar light has fundamental roles in vast chemical, biochemical, and physical process in biosphere and hence been declared as “source of life.” Solar light is further classified into a broad range of electromagnetic waves, and each region in the solar spectrum bears its unique actions in the universe or biosphere. Since centuries, solar light is believed as a potent source of killing pathogens causing postharvest losses on food products as well as human skin diseases. Citrus fruit crops are widely produced and consumed across the world, but due to their higher juicy contents, *Penicillium italicum* (blue mold) and *Penicillium digitatum* (green mold) make their entry to decay fruits and cause approximately 80% and 30% fruit losses, respectively. Agrochemicals or synthetic fungicides are highly efficient to control these postharvest fungal pathogens but have certain health concerns due to toxic environmental residues. Therefore, the scientific community is ever looking for some physical ways to eradicate such postharvest fungal pathogens and reduce the yield losses along with maintaining the public health concerns. This review article presents and discusses existing available information about the positive and negative impacts of different spectrums of solar light exposure on the postharvest storage of citrus fruits, especially to check citrus postharvest rotting caused by *Penicillium italicum* (blue mold) and *Penicillium digitatum* (green mold). Moreover, a special focus shall be paid to blue light (390–500 nm), which efficiently reduces the decay of fruits, while keeping the host tissues/cells healthy with no known cytotoxicity, killing the fungal pathogen probably by ferroptosis, but indepth knowledge is scanty. The study defines how to develop commercial applications of light in the postharvest citrus industry.

1. Introduction

Citrus (family, Rutaceae) fruits are famous all across the world and have a commercial production in over 137 countries [1]. The total production area of citrus (oranges, grapefruits, pomelos, lemons, limes, tangerines, mandarins, clementines, and satsumas) on the world was about 9.7 million ha, where about 138.5 million tones of fruits were harvested in 2018 [2]. The contribution of the citrus industry

to the world economy is enormous, and it provides jobs to millions of people around the world in harvesting, handling, transportation, storage, and marketing operations. The importance of citrus fruit is attributed to its diversified use, which is widely consumed either as fresh fruit, as juice or in confectionaries. Citrus fruits have pleasant flavors, attractive colors, and aroma and are well-preferred by the consumers. It also has high nutritional concentration and health-promoting bioactive compounds, including ascorbic acid

(vitamin C), phenolic acids, flavonoids, carotenoids, pectin, and other compounds [3, 4]. Due to their higher water content and nutrient composition, citrus fruit is very susceptible to infection by microbial pathogens during the period between harvest, transportation, and consumption [5]. Postharvest decays of citrus fruit can also originate from latent infections occurring in the orchard such as black rot caused by *Alternaria alternata* pv. *citri*, brown rot caused by *Phytophthora citrophthora*, and anthracnose caused by *Colletotrichum gloeosporioides* [4]. In developing and non-developed countries, high losses occurred due to poor storing, inadequate transportation, handling, and postharvest storage [6, 7]. Citrus fruits are highly susceptible to a variety of postharvest diseases causing huge losses during postharvest phase [8]. Postharvest losses, not only for citrus but also for all food products, are gaining more attention due to the global hunger [9], and prevention of postharvest losses is thought to be the common global goal of humanity for the issue of global achieving sustainable food security [10].

Most important postharvest losses of citrus, as of many other fruits, are caused by weight loss and pathogenic decay (mainly caused by *Penicillium italicum* Wehmer “blue mold” and *P. digitatum* Sacc. “green mold”). Total losses caused by these two pathogens were noted to reach up to 80% and 30%, respectively [11]. Both of them are known to be reproduced asexually by airborne spores and generally infect the fruits through wounds. The spores are greatly produced by rotten fruits and easily contaminate the surrounding fruits. The severity of the pathogen damages mainly depends on the amount of spores and optimal temperature which is about 20–25°C [12].

Chemical fungicides are the primary preferences of growers/packing houses for the postharvest control of pathogens [13]. However the excessive or misuse of agrochemicals was reported to result with resistant fungi strains which reduce the effectiveness of the fungicides [14] and associated with several environmental and health-base issues [15]. Furthermore, agrochemical residues in food have been the topic of numerous public discussions in the world [16], which reduced the acceptability of chemicals in agriculture [17]. Moreover, supermarkets, citrus export companies, and countries began to adopt strict policies regarding pesticide residues [4]. Therefore, an important need arises to develop alternative postharvest decay control measures to agrochemicals and it has been an important topic for the scientific world [18]. Since then, numerous studies have been conducted to develop/investigate alternatives to the chemical fungicides for the control of blue and green molds and came up with promising results where they suggested that biocontrol agents, i.e., some strains of yeasts and bacteria [19], hot water treatment (HWT) [20], hot air treatment [21], salts [22], second metabolites of plants [23], plant extracts [24], nanomaterials [25] and light irradiation [26] are effective.

The sun’s light is accepted by scientists as the source of life on the earth. The light radiation of the sun flows into space, warms our planet, and has fundamental roles in many chemical, biochemical, and physical processes, including the most known photosynthesis. It also possesses an important

role in water and nutrient cycles which are the basis of ecological balance on the earth [27]. Based on the well-known definition of Paracelsus, the “dose” is the determinant of toxicity, and it is obvious that the earth’s position in the solar system is optimal for preventing us from burning but sustaining the life by supplying enough radiation. The entire range of the light is named as the electromagnetic spectrum which ranges from long radio waves to gamma (γ) rays [28]. The energy and the wavelength are known to have a reverse relationship, where the energy increases while the wavelength decreases. The visible spectrum of the light by the human eye makes up only a small fraction. The right side of the visible spectrum is generally known as nondangerous due to its low energy, where the left side of the visible spectrum (ultraviolet “UV” rays, X-rays, and gamma rays) is classified as harmful to many living organisms, due to their extremely high energies. However, most of the spectrums are absorbed by the atmosphere (primarily CO₂, H₂O, and O₃) with the exceptions of visible light, microwaves, and radio light, as well as a little of infrared and ultraviolet light [27].

The role of different spectrums of light in food preservation has been well studied and reported. Here, an important technology comes to forefront importance with the name of light-emitting diode (LED). It is a semiconductor diode capable of producing light. The LED technology is capable of producing monochromatic light, consisting of a narrow bandwidth of wavelengths. Thus, there are UV LEDs, IR LEDs, and LED blue lights available in food preservation technologies [29]. The efficacy of irradiation on the control of pathogens is reported to be mainly influenced by its type, penetration ability, and duration exposed [30]. The types of irradiation can be grouped into two categories as nonionizing irradiations (UV-C, UV-B, UV-A, and blue light) and ionizing irradiations (gamma (γ) and X-rays). Previous studies suggested that the mechanism of action varies depending upon the type of irradiation and fresh product, and the general mechanisms are as follows: inhibition of spore germination, increase in PAL activity and cell wall thickness, production of reactive oxygen species (ROS) in fungal cells, and enhancement of phytoalexin synthesis [26].

Light irradiation has different roles in plant and fruit bodies. Herein, it is necessary to mention about photooxidative stress/damage. Molecular oxygen in the living cells is known to be relatively unreactive in its ground state form. However, environmental stress conditions such as drought, ozone, salinity, cold, heat, changes in atmospheric composition, and light irradiation may cause a rise to various toxic reactive forms, called free radicals. Generally, the reactive oxygen species (ROS) result by transfer of one, two, or three electrons to molecular oxygen (O₂) to form superoxide radical (O₂^{•-}), hydrogen peroxide (H₂O₂), or hydroxyl radical (•OH), respectively. ROS are normally produced in every aerobic organism as by-products of several metabolic pathways (i.e., photosynthesis, respiration, and photorespiration). ROS play a significant role in plant and fruit growth, development, and homeostasis at normal concentrations by signalling mediators for different cellular responses. However, a rapid rise in the ROS concentration due to stress conditions in living cells may cause damage to

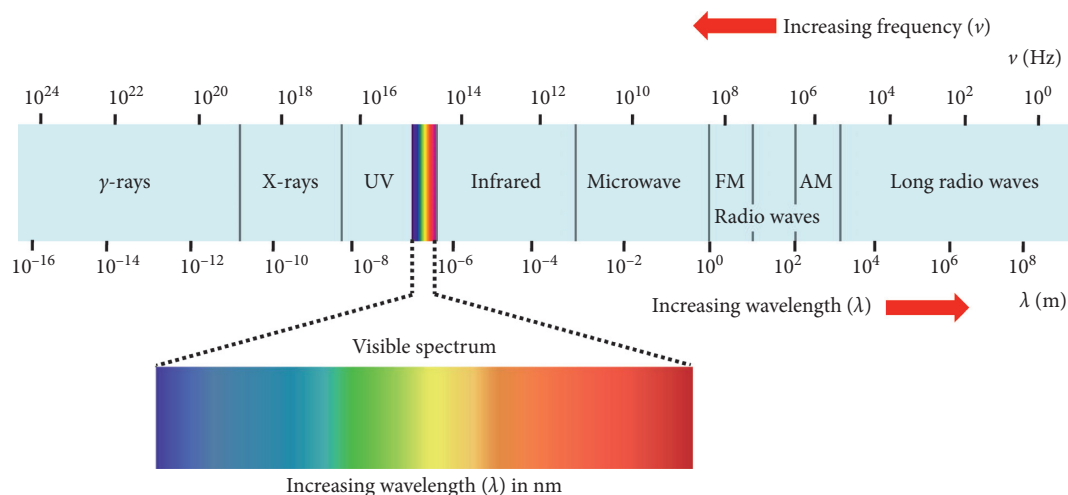


FIGURE 1: The solar electromagnetic spectrum.

proteins, lipids, carbohydrates, and DNA, which results in oxidative stress in the cells [31]. Among these stress conditions, light irradiation induced ROS generation is known as photooxidative stress/damage in plants [32]. Living plant cells have several defence mechanisms including nonenzymatic and enzymatic antioxidants to either prevent the formation or eliminate the ROS. The nonenzymatic antioxidant defence mechanism includes ascorbic acid, vitamin A, vitamin E, ascorbate, carotenoids, and phenols which directly react with ROS. Moreover, the enzymatic defence mechanism includes some enzymes, i.e., superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX), peroxidase (POD), and glutathione reductase (GR), which help in scavenging specific reactive oxygen species. Oxidative stress induced by light irradiation may stimulate the biosynthesis of those enzymatic and nonenzymatic components as a defence mechanism in plants and in harvest fresh product [31, 33, 34].

Plants and harvested fresh fruits and vegetables also produce ROS as a defence mechanism when they are attacked by pathogens as a result of the activation of defence genes. Those antimicrobial compounds have low molecular weight and are named as phytoalexins. They have special roles in the defence system and enable plants/foods to control pathogens [35]. Phytoalexin synthesis can be modified with the effect of several factors, such as light irradiation, temperature, and humidity [36, 37]. Phytoalexins are known to inhibit the mycelial growth of fungi, rupture the plasma membrane, and inhibit the elongation of the germ tube [38, 39]. One of the most important phytoalexins which is produced by the citrus species as a defence mechanism to pathogens is the scoparone (6,7-dimethoxy coumarin). It was firstly noted by Riov [40] that the accumulation of scopoletin, scopolin, and scoparone increases in the grapefruit peel under irradiated conditions (at 1–4 kGy). Researchers noted that the nonirradiated fruits do not have scoparone, whereas scopoletin and scopolin are found in very low concentrations. Scoparone was then isolated from the peel of grapefruit which infected with *P. italicum* [41]. 4-(3-Methyl-2-butenoxy) isonitroso acetophenone is another

irradiation-induced stress metabolite with antifungal activity noted from the irradiated peel of “Valencia” orange fruits [42].

In line with this information, this review attempts to discuss the possible mechanism of each irradiation type on the control of postharvest citrus rotting caused by *Penicillium italicum* (blue mold) and *Penicillium digitatum* (green mold).

2. Nonionizing Irradiations

Nonionizing radiation refers to any types of irradiation in the electromagnetic spectrum (from ultraviolet “including” to the right in Figure 1) where they do not carry sufficient energy to cause ionization, removing electrons from atoms and molecules. Among the nonionizing irradiations, ultraviolet (UV: 100–400 nm) irradiation and blue light (400–500 nm) were previously tested for postharvest control of pathogens. The FDA (the US Food and Drug Administration) has given permission to the application of UV for pathogen control in food storage [43]. The UV is divided into 3 groups as follows: UV-C (100–280 nm), UV-B (280–315 nm), and UV-A (315–400 nm). The treatment of UV generally took place by placing the fruits underneath the UV lamps. The duration of UV treatment may vary from a few seconds to a few hours, and this is the main determinant affecting the impact of treatment. The UV irradiation was known to sense by fruit tissues through photoreceptors, and it regulates some metabolic reactions in cells [44]. The positive effects of UV irradiation types were separately discussed in following sections.

2.1. UV-C Irradiation. Among the types of UV, the extensive works were previously conducted on UV-C and numerous studies reported success on the control of postharvest rotting caused by *P. digitatum* and *P. italicum*, whereas high intensities were reported to cause significant damages on the citrus fruit flavedo [45] (Table 1.). Gündüz and Pazır [48] carried out *in vivo* research, after an *in vitro* test, and studied the effects of UV-C treatment on the control of *P. digitatum*

TABLE 1: Effects of UV-C irradiation on the postharvest rotting caused by *P. italicum* and *P. digitatum*.

Citrus species	Fungi species	Treatment/intensity	Mechanism of action	References
Orange	<i>P. digitatum</i>	Fruits were placed 65 cm below the UV-C source (0.1 W·m ²)	Reduced growth of pathogen, attributed to increase in flavonoids content	[46]
Orange	<i>P. digitatum</i>	20 cm above the fruits with 3.6 W and 1.5 KJ·m ²	Reduction in fungi population which was attributed to the accumulation of phytoalexin scoparone in the fruits	[47]
Orange	<i>P. italicum</i> and <i>P. digitatum</i>	Irradiation (7.92 kJm ²)	Spore inactivation on the fruit surface	[48]
Orange	<i>P. italicum</i> and <i>P. digitatum</i>	UV-C light (254 nm) with intensity (2.64 mW cm ²) and doses from 0.26 to 15.84 kJ·m ² was applied with lamps over 10 cm of the fruits.	Reduction in the fungi spores by the germicidal effects of UV-C	[49]
<i>In vitro</i> at potato dextrose agar (PDA)	<i>P. digitatum</i>	Complete inhibition from 396 mws·cm ² UV-C exposure	Reported to have some type of a hedonal impact on reducing the reproduction of <i>P. digitatum</i>	[50]
Grapefruit	<i>P. digitatum</i>	Fruits were placed 10 cm away from the UV-C 254 nm lamps with 2.7 W·m ² fluency rate	Enhanced resistance to <i>P. digitatum</i> which was attributed to the accumulation of a chitinase and 1,3-endoglucanase proteins at the fruits' peel	[51]
Grapefruit and tangerine	<i>P. digitatum</i>	UV-C dose of 1.3 kJ·m ² d	Reducing the incidence of pathogen by induced resistance at the fruits	[52]
Tangerines	<i>P. digitatum</i>	UV-C dose of 1.3 kJ·m ² for 1.75 min (fruits were placed 10 cm away and rotated 4 times)	Reducing the incidence of pathogen due to the control of latent infection	[53]
Tangerines	<i>P. digitatum</i>	UV-C dose of 1.3 kJ·m ² for 1.75 min	Reducing the incidence of pathogen by induced host resistance to postharvest decay	[54]
Kumquats	<i>P. digitatum</i>	UV-C at 0.2 × 10 ³ to 1.5 × 10 ³ J·m ²	Induction of resistance to <i>P. digitatum</i> was previously attributed to phytoalexin and scoparone accumulation	[45]

and *P. italicum* spores inoculated at the orange fruits. Researchers placed oranges 10 cm below the UV-C lamps and treated for 5 min (7.92 kJ·m⁻²). Authors reported that the UV-C treatment is effective for reducing the percentages of infected fruits to about 3-fold as compared to control fruits, but they also suggested that the complete inactivation was not successful due to the low penetration ability of UV-C light. In a closely related previous study by Gündüz et al. [49], UV-C (0.26–15.84 kJ·m⁻²) was applied to the navel oranges with lamps located 10 cm upward of the fruits (Table 1). Prior to UV-C application (254 nm with an intensity of 2.64 mW·cm⁻²), spores of the two causes of citrus rotting, *P. digitatum* and *P. italicum*, were inoculated to the fruits with a rate of 4.00 and 4.50 log cfu fruit⁻¹, respectively. According to the results of the mentioned study, the 3.17 kJ·m⁻² UV-C dose was found to provide better control of *P. digitatum* and of 4.75 kJ·m⁻² UV-C dose was noted to provide maximum control of *P. italicum*. The UV-C treatment was noted to provide significant control over two fungi for both spot and wound inoculation methods. The effects of different doses of UV-C treatment on the *P. digitatum* at orange fruits were previously studied by Fernandez and Hall [50]. UV-C (254 nm with an output of 0.66 mws·cm⁻²) lamps were used in their study with exposure times varying from 10 s to 10 min. As a result, they reported that the complete inhibition of *P. digitatum* was only from 396 Mws·cm⁻² UV-C exposure, where almost complete, partial, slight, and no visible inhibition of myclial

growth was noted from 198, 99, 39.6, and 26.4 Mws·cm⁻² UV-C exposure, respectively. In a more detailed study about the effects of different doses of UV-C on the inhibition of *P. digitatum*, valuable results were noted for science by Trivittayasil et al. [55]. It was suggested by the researchers that the *P. digitatum* has a resistance to low doses of UV-C application and the survival curve of the pathogen is nearly linear (adjusted $R^2 = 0.95$). Thus, suggesting that the increase in the dose of UV-C provides better inhibition of the pathogen, as suggested by the Fernandez and Hall [50]. Rather than oranges, some studies were also conducted with tangerines. In one of these studies, Stevens et al. [53] tested the influence of UV-C (254 nm) alone or in combination with *Debaryomyces hansenii* on the control of *P. digitatum*. In this study, UV-C was applied at a dose of 1.3 kJ·m⁻² for 1.75 min, and during treatments, fruits were placed 10 cm away and rotated 4 times. Researchers reported that the UV-C treatment alone reduced the incidence of *P. digitatum*, but higher influence was reported for the combination of UV-C and *D. hansenii*. Similar results were also reported for both tangerines and grapefruits by Stevens et al. [52].

In an early study by Stevens et al. [54], the same dose of UV-C (254 nm at 1.3 kJ·m⁻²) was applied through the stem end in a stationary position without rotation and it was compared with the conventional procedure where the fruits were rotated 4 times. Furthermore, researchers noted that the treatment through the stem end provides better control of *P. digitatum* as compared with the rotation method.

Similar result for *P. digitatum* was previously reported by Arcas et al. [46], and their study went deeper to determine the changes in flavonoid levels. In their research studies, they placed fruits 65 cm below the UV-C source ($0.1 \text{ W}\cdot\text{m}^{-2}$). Researchers suggested that the UV-C treatment reduced the naringin content while promoted an increase in tangeretin contents. In a different study by D'hallewin et al. [47], the UV-C (254 nm) irradiation treatment (20 cm above the fruits with 3.6 W and $1.5 \text{ kJ}\cdot\text{m}^{-2}$) was tested against *P. digitatum* at orange fruits. Approximately 75% reduction was noted for the fungi population and this success was associated with the phytoalexin scoparone accumulation in the fruits. Induction of resistance to *P. digitatum* was previously attributed to phytoalexin and scoparone accumulation in kumquat fruits too [45].

In a different study, Papoutsis et al. [56] tested the effects of UV-C application (4, 19, 80, and $185 \text{ kJ}\cdot\text{m}^{-2}$) on the total phenolic content and antioxidant activity of dried powders of lemon pomace. No specific tests were performed on the growth and development of blue or green mold, but the results were meaningful in which the phenolics and antioxidant activities are known to enhance fruits resistance to pathogens [46]. According to the results of Papoutsis et al. [56], UV-C treatment was found to significantly affect the antioxidant activity, phenolic content, and flavonoid concentration of the dried melon pomace powders. The changes in the phenolic contents and flavonoid contents were found to be differently influenced by the UV-C doses, where the highest phenolic content was recorded from $19 \text{ kJ}\cdot\text{m}^{-2}$ UV-C irradiation and the highest flavonoid content was noted from $180 \text{ kJ}\cdot\text{m}^{-2}$ UV-C irradiation treatments. The results of this study highlight the importance of dose and imply that specific studies are required for maximum control of pathogens on different fruits. In an earlier study by Porat et al. [51], it was also suggested that the UV-C application (254 nm and $2.7 \text{ W}\cdot\text{m}^{-2}$ fluency rate) enhanced the resistance of grapefruits against *P. digitatum* and this was attributed to be a result of the accumulation of a 25 kDa chitinase and 39 kDa 1,3-endoglucanase proteins at the fruits' peel. Numerous studies have been conducted to identify the key factors and genome sequencing of some important flavonoid biosynthesis in different fruits [57, 58]. These studies showed that the R_2R_3 MYB transcription factors directly influence the expression of genes related with the flavonoid biosynthesis [59, 60]. Studies with fruits other than citrus, i.e., apple, strawberry, grape, and litchi, reported that R_2R_3 MYB transcription factors are inducible by light [61–64]. Moreover, it is well known that the biosynthesis of flavonoids is an important tool for the control of citrus rotting caused by *P. digitatum* and *P. italicum* [65].

2.2. UV-B Irradiation. Similar to the UV-C light, there are numerous studies in the published literature about the antifungal activities of UV-B, in which as a general, UV-B irradiation was reported to be less harmful for the fruits and less effective [66]. In a previous study by Ruiz et al. [67], the UV-B irradiation was reported to induce metabolic changes in the lemon fruit peel which then suggested contributing

fruit protection against *P. digitatum* (Table 2.). The UV-B treatment was found to change the respiratory profiles and increased the concentration of the phenolic compounds (flavones, flavonols, and anthocyanins). Thus, the antioxidant activity of the lemon fruits was also increased. In another study by Ruiz et al. [44], it was also noted that the UV-B treatment increases the ROS and membrane permeability of the lemon fruits. The UV-B exposure was also found to cause changes in the soluble carbohydrate metabolism and secondary metabolite accumulation of lemon fruits. It was suggested that the concentration of the secondary metabolites depends on the dose and duration of the UV-B, and the durations over 3 min ($0.43 \text{ W}\cdot\text{m}^{-2} = 22 \text{ kJ}\cdot\text{m}^{-2}\cdot\text{d}^{-1}$ UV-BBE) were noted to not cause any significant changes in the concentration of secondary metabolites. By contrast, the soluble sugars' concentration accumulation in the peel was found to increase with the durations over 3 min [69]. This result is in agreement with the notes discussed above for the UV-C and findings of Trivittayasil et al. [55] and Fernandez and Hall [50], where they reported that the intensity and duration of irradiation are highly important for the success of the treatment. Successful results for UV-B were also noted for mandarins. In one of these studies by Yamaga et al. [68], it was noted that the UV-B (15, 30, 60, and $120 \text{ kJ}\cdot\text{m}^{-2}$) treatment significantly reduces the germination of *P. italicum* spores on the "satsuma" mandarins.

2.3. UV-A Irradiation. The UV-A photons in the UV spectrum have less energy as compared with UV-C and UV-B, but they still have energy which can be absorbed by biological molecules and cause biochemical changes in the living tissues (Table 3). In a previous study, two phototoxins were reported to be activated by UV-A (334 nm), the a-terthienyl (a-T), and 8-methoxypsoralen (8-MOP), and these were noted to be effective in inactivation of some fungi, i.e., *P. italicum* [70]. It has been reported that harmol (1-methyl-9H-pyrido[3,4-b]indol-7-ol) is an active β -carboline against *P. digitatum* [72], and the UV-A treatment (at 365 nm and $8 \text{ W}\cdot\text{m}^{-2}$) was then reported to improve the harmol activity and improve antimicrobial activity [71]. Due to the low energy charge of UV-A, as compared with UV-B and UV-C, the studies are limited with UV-A.

2.4. Blue Light. As compared with the UV irradiation, blue light is a part of the visible spectrum and is known to be normally absorbed by some plant tissues and to have roles in some metabolic reactions in the living tissues [73]. Blue light (390–500 nm) is reported to reduce decay of foods caused by fungal pathogens *P. digitatum* and *P. italicum* [74–76] (Table 4.) and promote the growth of plants, and its receptors participate in pathogenesis response signalling [79]. Blue light activates endogenous photoactive porphyrins within the bacterial or fungal cells [80]. It was noted that the blue light treatment (450 nm at quantum fluxes between 60 and $630 \mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) causes an increase in scoparone concentration at the flavedo of sweet oranges and this improves the resistance of fruits to the *P. digitatum* [75].

TABLE 2: Effects of UV-B irradiation on the postharvest rotting caused by *P. italicum* and *P. digitatum*.

Citrus species	Fungi species	Treatment/intensity	Mechanism of action	References
Lemon	<i>P. digitatum</i>	UV-B (22,000 J·m ⁻² ·d ⁻¹) lamps placed 50 cm above the fruit	Increase in cell wall thickness and concentration of phenolics in the flavedo	[67]
Lemon	<i>P. digitatum</i>	UV-B (22,000 J·m ⁻² ·d ⁻¹) lamps placed 50 cm above the fruit	Increase in ROS and membrane permeability, which results in improved antifungal activity	[44]
Mandarin	<i>P. italicum</i>	In <i>in vitro</i> studies, fruits were placed 15 cm below UV-B lamps at 15, 30, 60, and 120 kJ·m ⁻²	Inhibit the germination of fungi spores by directly inactivating fungi and inducing an antifungal response in fruits	[68]

TABLE 3: Effects of UV-A irradiation on the postharvest rotting caused by *P. italicum* and *P. digitatum*.

Citrus species	Fungal species	Treatment/intensity	Mechanism of action	References
<i>In vitro</i> at PDA	<i>P. italicum</i>	At a fluence rate of 40–43 J·m ⁻² ·sec ⁻¹	Inactivation of some fungi which associated with the activation of two phototoxins: the a-terthienyl (a-T) and 8-methoxypsoralen (8-MOP)	[70]
<i>In vitro</i> at PDA	<i>P. italicum</i>	Irradiation at 365 nm and 8 W·m ⁻²	Improvement in antimicrobial activity associated with increasing harmol activity	[71]

TABLE 4: Effects of blue light irradiation on the postharvest rotting caused by *P. italicum* and *P. digitatum*.

Citrus species	Fungi species	Treatment/intensity	Mechanism of action	References
Orange	<i>P. digitatum</i>	Blue light (450 nm) at quantum fluxes between 210 and 630 μmol·m ⁻² ·s ⁻¹	Increase in scoparone at fruit flavedo	[75]
Mandarin	<i>P. italicum</i>	Low-intensity blue LED (465 nm) with a fluency of 8 μmol·m ⁻² ·s ⁻¹	Suppression of fungi spore development	[76]
Mandarin	<i>P. digitatum</i> and <i>P. italicum</i>	Blue light at 465 nm with a photon flux of 80 μmol·m ⁻² ·s ⁻¹	Prevention of microbial growth by increasing the phytoalexin scoparone concentration	[77]
Tangerine	<i>P. digitatum</i>	Exposure of fruits to 410–540 nm blue light at a fluency of 40 μmol·m ⁻² ·s ⁻¹	Induction in PLA2 gene expression which resulted with reduced pathogen infection	[74]
Tangerine	<i>P. digitatum</i> and <i>P. italicum</i>	Blue light at the intensity of 40 μmol·m ⁻² ·s ⁻¹	Induction of the octanal production and reducing the activities of <i>P. digitatum</i> and <i>P. italicum</i>	[78]

Antifungal effect of blue light irradiation (465 nm with a photon flux of 80 μmol·m⁻²·s⁻¹) on the blue mold (*P. italicum*) and on satsuma mandarin fruits was also studied in both before and after fungal inoculation with no effect on the quality of fruits except for the citric acid concentration and moisture loss [81]. After the inoculation of the blue mold fungal pathogen, blue light reduces the sporulation and mycelium of *P. italicum*, while the 6-day irradiation period before the inoculation with pathogen showed less symptom development [81]. Another study claims that when blue light was exposed on the wounded satsuma mandarin fruits under storage conditions, it enhanced the production of phytoalexin scoparone and the blue light treated fruits had only 13.3% decayed fruits, where the nontreated fruits had 51.1% fruit decay caused by *P. digitatum* and *P. italicum* [77]. Direct exposure of blue light in *in vitro* conditions reduces the citrus decay rates caused by the *P. italicum* and *P. citri*. Blue light posed its effects mainly by changing citrus phospholipase A2 at transcriptional levels in citrus peels and hence lowered the decay process by decreasing the growth of *P. digitatum* [74, 82]. The basic mechanism in this blue light-mediated fungal growth inhibition is the involvement of the lipid-derived pathways [82]. These studies further highlighted that blue light treatment has no effects on the rind

color nor affects fruit quality parameters (soluble solid content, titratable acidity, specific gravity, and percentage of flesh) [77]. The citrus peels are rich in oils which contain lipid-derived constituents having antifungal activity [83]. Furthermore, it can easily be concluded from these studies that the blue light induces scoparone production, thus reducing the fruit decay caused by *P. digitatum* and *P. italicum*, but correct combination of intensity and duration is highly important for maximum control. In a different study with bayberries, the blue light treatment was noted to increase the anthocyanin concentration of the fruits [84], which might be a subject for the further studies with citrus fruits.

The blue light treatment (exposure of fruits to 410–540 nm blue light at a fluency of 40 μmol·m⁻²·s⁻¹) was also noted to induce PLA2 gene expression in tangerine fruits which results with a reduced pathogen infection [74]. Exposure of fruits to low-intensity blue light (at 465 nm) with a fluency of 8 μmol·m⁻²·s⁻¹ was also noted to suppress the development of *P. italicum* spores at mandarin fruits [76]. Additionally, blue light at the intensity of 40 μmol·m⁻²·s⁻¹ was found to reduce the cell wall digest enzyme activity of *P. digitatum* and *P. italicum* by inducing the octanal production in the tangerine fruits [78]. The findings of Liao et al. [78] are meaningful in which the

TABLE 5: Effects of X-ray irradiation on the postharvest rotting caused by *P. italicum* and *P. digitatum*.

Citrus species	Fungi species	Treatment/intensity	Mechanism of action	References
Mandarins	<i>P. digitatum</i>	Irradiation at 510 gray (Gy)	Delays the pathogen development	[88]
Mandarins	<i>P. digitatum</i> and <i>P. italicum</i>	Irradiation at 510 and 875 gray (Gy)	Inhibition of sporulation	[89]

increase in exposure duration to the blue light was found to have an increasing influence on the prevention of the citrus pathogens. Numerous previous studies suggested that the combination of more than one measure would provide higher efficacy for the food preservation, i.e., combination of modified atmosphere packaging and lemongrass oil [85]. Similarly, combination of blue light and erythrosine was found to provide better control of *P. digitatum* [86].

It is evident from this discussion that utilizing different approaches and techniques to eradicate the citrus blue mold and green mold is an important alternative for agrochemicals, and blue light has proven itself as one of the superior techniques to eradicate blue mold disease. Blue light is environmental friendly and cost effective, and it keeps the citrus skin color as well as fruit quality parameters. Moreover, with the passage of time, most pathogenic fungal strains may resist the traditional fungicides, and hence, demand for a superior and novel antifungal agent is demanding ever. To sum up, although the general mechanism of action of blue light treatment has not been fully understood up to now, there are two main hypotheses: one is the occurrence of endogenous photosensitizers within the pathogen cells which causes the inactivation of the pathogens and the other is the induction of secondary metabolites in fruit tissues which increases the resistance of the products to the pathogens [75, 87].

3. Ionizing Irradiations

Ionizing irradiation is a form of energy in the electromagnetic spectrum (from ultraviolet “excluding” to the left in Figure 1) where they carry sufficient energy to cause ionization, removing electrons from atoms and molecules, including air, water, and living tissues.

3.1. X-Ray Irradiation. X-ray is one of the two ionizing irradiation forms of energy in the electromagnetic spectrum knowing to carry high energy. It was reported to be a promising technology for the disinfection of foods due to its antipathogenic characteristic (Table 5). X-ray irradiation was reported to disinfest *Ceratitidis capitata* at the citrus fruits, and treating mandarins with X-ray irradiation was found to not adversely affect the fruits’ quality [90]. Although it has potential to prevent postharvest quality of foods, its acceptability by the consumers is in question due to its known ionizing characteristic [91]. Ionizing characteristics of X-ray irradiation cause oxidative stress in fruits, which can directly influence the bioactive compounds located in fruit tissues and could improve the resistance of those fruits to the pathogens [92]. The ionizing in the food products is thought to generate free radicals which may stimulate unwanted reactions in the living tissues [93]. However, there are some studies in the published literature which have been reported

that the low doses of X-rays are effective in controlling citrus rotting caused by blue and green mold [88, 89]. Moreover, the X-ray irradiation (510 and 875 Gy) of mandarin fruits was found to increase phytoalexin scoparone and scopoletin concentrations in the fruit rind and reduce the *P. digitatum* infections [88]. Studies with the positive and negative effects of X-ray irradiation are limited with citrus fruits, and further studies are obligated to do any recommendations for their commercial use in the postharvest citrus handling.

3.2. Gamma (γ)-Ray Irradiation. Gamma (γ)-rays is the other type of irradiation in the electromagnetic spectrum which has the highest energy, and the low doses of γ -ray irradiation were also proven to regulate activities of some enzymes involved in scavenging of free radicals [94] and inhibit the development of fungal pathogens [30]. However, it was strongly highlighted by previous studies that the potential negative effects of γ -ray irradiated foods must be studied before their use [95]. In one of these studies, Jeong et al. [30] tested the *in vitro* activity of γ -rays (1.0 kGy) on the *P. digitatum* at “satsuma” mandarins and found out that it inhibits spore germination, mycelia growth of pathogen, and elongation of germ tube. However, the *in vivo* studies showed that this or higher doses cause severe damages on the fruit tissues. To eliminate the damages on the fruit tissue, researchers suggested that the incorporation of γ -rays at lower intensity (0.4 kGy) with dichloro-striazinetrione (NaDCC, 10 ppm) provides similar effect on the pathogen and less damage on the fruit tissue. The impact of the combined treatment was suggested to be due to the synergistical damage on the pathogens’ membrane. Moreover, these findings [30] are promising for future studies and development of appropriate technologies for the quality preservation of foods without significant damages on the fruit tissues, because it is a well-known phenomenon that higher doses of gamma radiation cause severe damages and mutation breeding in fruit tissues. In a study with *Citrus sinensis*, it was found that the doses more than 20 Gy γ -ray irradiation cause significant increase in peroxidase activity and decrease in chlorophyll content [96]. A study by Mahmoud et al. [97] noted that the application of gamma rays at doses of 0.4, 0.8, 1.6, and 3.2 kGy to the grapefruits helps to control postharvest decay. It was then noted by another study that the 150 and 200 krad gamma radiation suspends the spore germination of *P. expansum* [98]. However, even the less intensity of γ -ray irradiation was noted to cause significant changes in the phenolic compounds and enzymatic activities of citrus fruits. In one of these studies, 0.3 kGy γ -ray irradiation was noted to induce the biosynthesis of total phenolic compounds and also increase the phenylalanine ammonia-lyase (PAL) activity of *Citrus clementina* Hort. ex. Tanaka. Among the phenolic

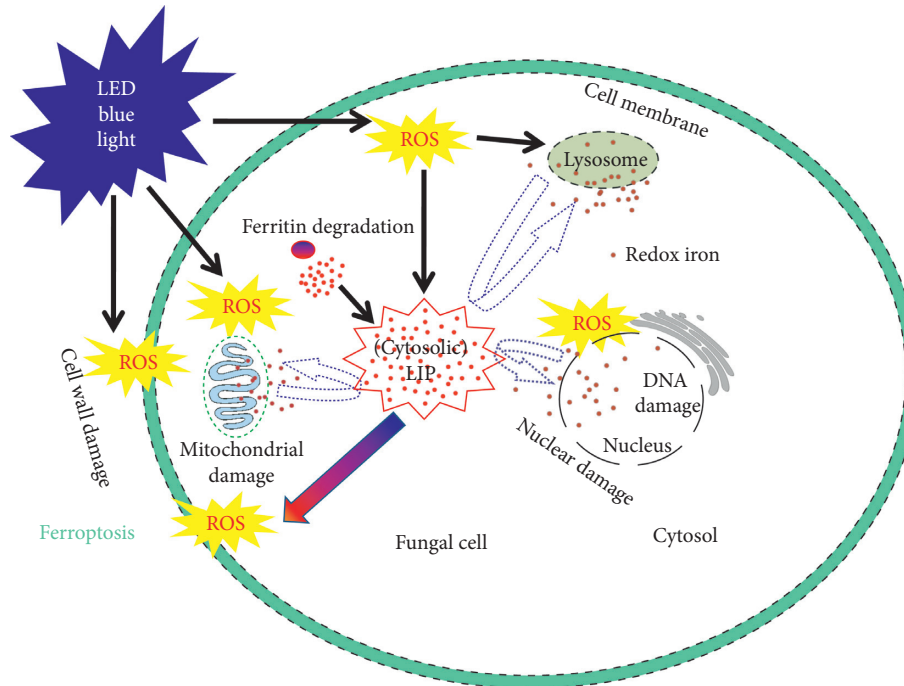


FIGURE 2: Blue light-induced ferroptosis in fungal cell.

compounds, the hesperidin, *p*-coumaric acid, and flavonoids were the compounds which highly influence by γ -ray irradiation [92]. Therefore, further studies are obligated to recommend any specific γ -ray irradiation treatment for the effective control of citrus rotting without any damages on the fruit quality and human health.

4. Light Radiation and Ferroptosis in Fungal Cells

Most studies concerning the application of either synthetic or the natural fungicides have been focused on the damage of the fungal hyphal cell walls leading to leakage of the cellular inclusions and finally the death of the fungal cells [99]. Recently, a phenomenon “ferroptosis” is introduced which is a regulated, nonapoptotic type of iron-dependent cell death reported in mammalian [100, 101] and plant cells [102, 103], and the whole phenomenon is portrayed in Figure 2. Herein, the light irradiation, especially the blue LED, induces the development of ROS in fungal cells, which then damages the cell wall and lysosome of the fungus and finally causes damage on the DNA. Ferroptotic cell death is distinct from apoptosis, necrosis, and autophagy [104]. Ferroptotic cell death requires iron and ROS accumulation [100, 101, 105]. Ferroptosis is initiated by inactivating glutathione-linked antioxidant defence and iron-mediated efflux of toxic lipid hydroperoxides along with numerous other reactive oxygen species (ROS) [106]. On the other hand, this can also be locked by iron chelators and antioxidants [101, 105]. Ferroptotic cells show different morphological and biochemical characteristics as compared with apoptosis. It includes cell rounding which is followed by rupturing of plasma membrane [105, 107]. As a result of the accumulation of ROS,

lipid peroxidation also occurs in ferroptotic cells [108]. It is further elucidated that free cellular iron along ROS and lipid hydroperoxides are directly involved in ferroptotic cell deaths [101]. At cellular level, iron behaves a strong redox catalyst [109], but it plays a great role in cell signalling and cell fate. In incompatible plant-pathogen interactions, a quick increase in ROS, iron, and α -glutamylcysteine synthetase is thought as an important marker for ferroptotic cell death [110, 111]. On the other hand, iron is essentially required by both the host and the pathogen for their normal growth, especially as a cofactor in certain metabolic processes. In many microbes, iron linked with heme is needed for the growth and virulence *in vitro*. However, the role of porphyrin-associated iron and iron-withholding proteins in the citrus rind and *P. italicum* is not described yet. Moreover, how iron loads get activated in blue light exposed citrus rind and *P. italicum* hyphae and the activation of ferroptosis in fungal hyphae is not elucidated yet. However, recent scientific information suggests that fungi may undergo iron-dependent ferroptosis and are noted to be resulted by the loss of membrane integrity as a consequence of lipid peroxidation [112]. On the other hand, some studies suggest that the fungi may encode some inhibitors of programmed cell death (i.e., conserved BIR “baculovirus inhibitor of apoptosis repeat”) [113]. Further studies are required to clarify the ferroptosis mechanism in fungus.

5. Conclusions

In conclusion, light irradiation (both nonionizing and ionizing irradiation) is a valuable physical technique for not only citrus but for other food products, by having a significant potential for controlling postharvest pathogens.

Even though the full mechanism of the action is not well understood or varying among the specific varieties/conditions, it is well known that the light irradiation, in general, has two types of mechanisms for controlling the citrus pathogens. One of these mechanisms is the inducement of the biosynthesis of specific secondary plant metabolites, which improves the fruit/tissue resistance against pathogens, and the other mechanism is the direct prevention of the pathogen development and/or spore inactivation. Therefore, further studies about the improvement of the understanding about the mechanism could potentially lead to the development of commercial applications in postharvest handling. Moreover, UVA, UVB, and blue-light are involved in the production of the ROS, which possibly leads to the fungal cell deaths by ferroptosis, a novel mechanism which has hardly been studied before for fungal cells. The increments in the LED industry and technology have been a precious potential for the development and use of quick sanitation technologies in the citrus industry. Here, the UV LEDs, especially UV-C, are believed to be powerful tools for the citrus postharvest industry. Further studies, with more specified subjects about the negative influences on fruit quality, seem to be most important requirements for the development of large-scale technologies, which would have an important role in the improvement of consumer acceptability. Needless to say, optimum dosage (type, intensity, and duration) is also a must to determine for each variety. Finally, it is important to remind that the ionizing irradiations (X-rays and γ -rays) would have negative effects on citrus fruit quality and on human health. Therefore, special attention should be paid on this subject.

Data Availability

No data were used to support this study.

Conflicts of Interest

The authors declare that there are no conflicts of interest regarding the publication of this paper.

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