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Effect of ultraviolet-c radiation processing on physical and microbial properties of horticulture produce

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Abstract

Ultraviolet irradiation is an emerging non-thermal processing technology that offers enhanced shelf life with better retention of organoleptic and nutritional properties. Ultraviolet-C irradiation is highly absorbed by DNA compared to UV-A and UV-B hence having a higher germicidal effect. This review paper presents brief information about UV-C irradiation and its effect physical (weight loss and firmness) and microbial properties of horticulture produce. Ultraviolet-C irradiation showed enhanced disease resistance and resistance against microbial growth. It also enhances plant defence by increasing the production of defence hormones such as phenylalanine ammonia-lyase and peroxidase. UV-C treatment maintained important parameters during storage such as firmness and weight loss. It can also reduce the activity of cell wall degrading enzymes and vary the genes related to them hence can maintain the texture of horticulture produce. Research efforts on the effect of intensity and harvest maturity in the efficacy of UV-C treatment are required to be explored. More studies are required to optimize the dose levels for different horticulture commodities is warranted.

Keywords: Ultraviolet, irradiation, horticulture, phenylalanine ammonia-lyase, peroxidase

Introduction

Agriculture was an important turnaround in human evolution. Soon after, humans started cultivation the need for preserving the produce was raised. Since then, we are adopting various processing techniques to preserve the quality and enhance the shelf life of the harvest. Most of the conventional processing techniques involve the use of heat either reduction in water content or use of chemicals etc. to preserve the product. The conventional processing technologies leads to detrimental effects on products' sensory as well as on nutritional quality^[1]. In the last decade, the consumer demand for nutritionally rich food with more natural aroma, along with minimum use of the chemical has increased. The increased market demand for minimally processed food has increased the focus of the scientific community and processing industries towards minimal processing techniques such as cold plasma, sonication, ozonation, pulsed electric fields, high hydrostatic pressure and irradiation. In contrast to conventional processing techniques, minimally processing techniques offer a commercial sterilized product with more natural quality, minimal effect on nutritional content and with no or slight use of chemicals^[2].

Ultraviolet irradiation is among the emerging minimal processing techniques which displayed promising results in enhancing the shelf life and maintaining the nutritional properties of the product without the use of any harsh processing condition. UV irradiation is a cost-effective physical technique that has a minimum effect on the environment^[3]. Ultraviolet light is non-ionizing germicidal radiation, which offers a vast opportunity for surface decontamination by controlling microbial growth in food processing with minimal effect on qualitative parameters^[4]. Among ultraviolet electromagnetic spectra, ultraviolet-c radiation is acknowledged for its germicidal property. UV-C radiation directly affects the DNA of the micro-organisms, which limits reproduction, and injured DNA leads to cell death^[5]. Apart from the direct germicidal effect, UV-C also induces a plant defense system by enhancing the production of stress-related enzymes^[6]. UV light has the limitation of low penetration depth, which restricts its use on the food surface, clear liquids and juices with turbulence systems & thin layers.

Several works of literature also report the increase in quality, shelf life and reduction in ethylene production of several fruits and vegetables. This review paper will highlight brief information about ultraviolet-C irradiation its effect on the physicochemical and microbial characteristics of horticulture products.

Ultraviolet Irradiation

Ultraviolet light and its types

UV is a part of enormous electromagnetic spectra with a wavelength between 10-400 nm, which lies between X-rays and visible electromagnetic spectra (Fig 1). UV spectra are divided into UV-A, UV-B and UV-C with wavelength 315-400nm, 280-315 and 200-280nm respectively. Vacuum UV has a wavelength of 100-200nm and exists in a vacuum only. Among UV radiation UV-C has the highest energy followed by UV-B and UV-A. UV-A doesn't have enough energy to achieve disinfection which is considered a major function of UV radiation [7, 8] reported that to inactivate *E. coli* using UV-A LED a fluency of 315 J cm⁻² and dose of 70 mW cm⁻². As the treatment dose and treatment time is too high it makes the use of UV-A alone for disinfection obsolete. However, UV-A doesn't affect the human body and exist in the atmosphere. UV-B has more energy compared to UV-A and is responsible for skin tanning. UV-B irradiation has the potential to improve the nutritional benefits of fruits and vegetables [9]. UV-B irradiation has shown potential in delaying yellowing, improving chlorophyll content, improving antioxidant qualities and maintaining sensory qualities [10-11]. However, UV-B has less potential in sterilizing the food products compared to UV-C [12]. The UV-C radiations are well known for their germicidal effect and are extensively used in the food industry for different purposes. Becquerel and Draper independently conducted a first-ever experiment showing photochemical changes caused by UV rays (340-400nm) on daguerreotype plates in 1842. In 1877, Downes and Blunt, who inactivated bacteria using the violet-blue spectrum of light, reported the germicidal effect of UV rays. The harmful effect of UV rays on human skin, likely skin burn, was first-ever reported by Widmark by experimenting on human skin. The use of UV-C for bacterial disinfection of water started as early as 1906. WHO also recommended its use for controlling tuberculosis [13]. In 2001, U.S.FDA permitted the use of UV in juice processing, which enhanced the commercial application of ultraviolet irradiation in the food industry [14]. Like UV-B, UV-C can also increase the nutritional composition along with added benefits of the significant reduction in fungal growth. [12] compared the effect of UV-B and UV-C on the phenolic content of table grapes. It was reported that the antioxidant activity and total phenol content was found highest in UV-C treated fruit compared to UV-B and combined (UV-C and UV-B) treatment. In another study, [15] investigated the effect of UV-B, C and combined (UV-B and

UV-C) treatment on decay and organoleptic properties of peach fruit. It was reported that UV-C treated fruits displayed improved physico-chemical and sensory properties compared to UV-B treated fruits. However, the combined treatments displayed the best result in reducing decay as well as maintaining the post-harvest qualities of fruit. Similarly, [16] also investigated the effect of UV-B, C and their combination on the phenolic compound content of fresh-cut carrots. It was reported that the UV-B treated samples displayed the highest phenolic content compared to other treatments. UV-C can significantly reduce fungal growth and has the potential in increasing the shelf life of food products. UV-C also has the potential in increasing the nutritional composition making it a prime attraction in food processing. The concerning point in the application of any processing technique is its effect on the sensorial and nutritional composition of the product. The current review focuses on the effect of UV-C treatment physical and microbial properties of horticulture produce.

Principle of ultraviolet irradiation

Light has both wave and particle nature which transfers energy in the form of small energy packets called photons. Ultraviolet is also a kind of light that commences a photochemical reaction by transferring energy while getting it absorbed by the molecule [17]. The escalating energy leads to the orbital jump of electrons in higher orbits, which is in a highly unstable excited state. The duration of the following process ranges from 10⁻¹⁰ to 10⁻⁸ seconds during which the molecules undergo photochemical reaction to achieve a stable state. To achieve the desired change, the UV light should have enough energy, second, it should get absorbed by the molecule [14].

Concerning food, ultraviolet irradiation works on two principles (1) Germicidal effect (2) Hormesis effect. The germicidal effect occurs because of the absorption of UV light by DNA, which results in crosslinking between neighbouring thymine and cytosine of the same DNA strand. The intra-cross linked mutated DNA base result in the breaking of hydrogen bond on the opposite strand, which disrupts the DNA structure ultimately affecting the transcription and replication, which is important for cellular functioning [18]. The roadblock in cellular functioning ultimately affect the reproduction process and leads to cell death [19]. UV-C has a wavelength range between 200-300 nm, which is highly absorbed by DNA thus makes a substantial effect on the microbial population such as protozoa, bacteria, moulds and viruses [20-17]. However, the following range of electromagnetic spectrum has the least effect on the proteins and doesn't produce any byproduct which can deteriorate the quality of the food. There are chances of photoreactivation of some organisms after UV-C irradiation when expose to visible light hence, the product should be stored under dark and refrigerated conditions after irradiation [21-22].

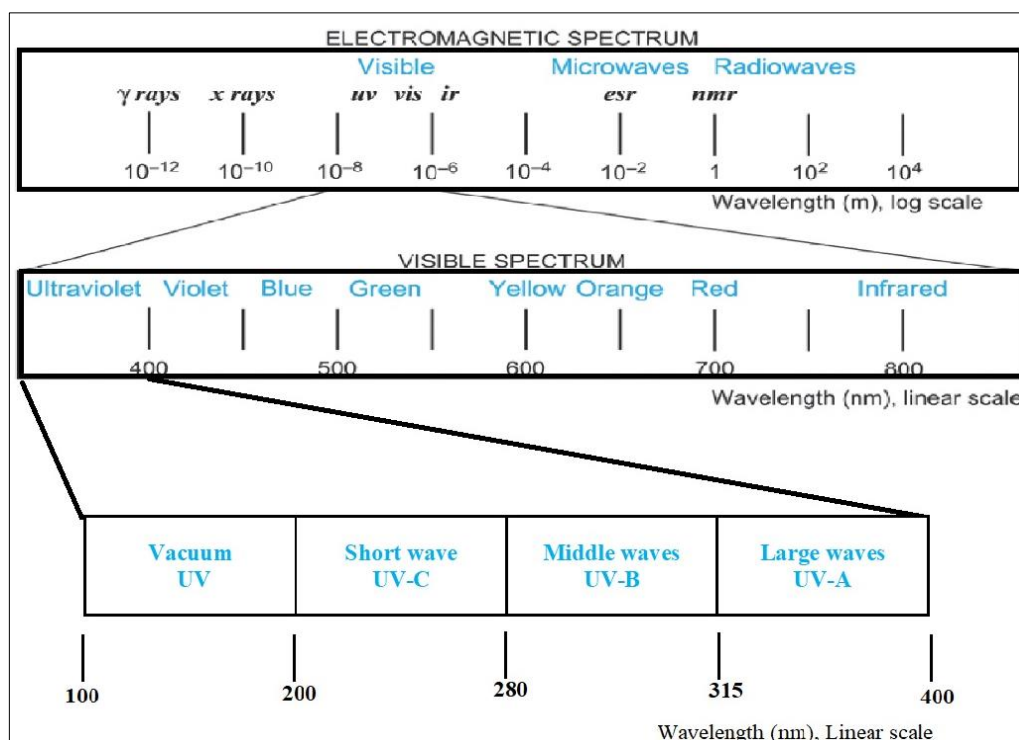


Fig 1: Electromagnetic spectrum and UV spectrum classification

Hormesis is defined as the stimulation by application of low doses of harmful agents [19, 23] stated that low doses of ultraviolet-c can generate a hormesis effect in the plant that can create positive effect homeostasis of a plant. UV-C light acts as hormetin to produce a beneficial hormesis effect that stimulates the production of phenylalanine ammonia-lyase (PAL). PAL stimulates the biosynthesis of phytoalexins (e.g., scopoletin and scoparone) and increases the resistance of plant tissue to spoilage causing factors [24, 3]. The hormesis effect is also known to enhance their biochemical parameter, which mimics the effects produced by abiotic stresses [25, 26]. reported that UV-C irradiation proved beneficial in delaying the chlorophyll change and yellowing in broccoli. The beneficial effect of UV-C induced antifungal compounds also involves delayed softening, delayed ripening and decline in chilling injury [27]. The major problem of several fruits is quick ripening due to excessive ethylene production. UV-C has shown potential in reducing the ethylene production rate, which delays ripening ultimately senescence and increase the shelf life of the product [28].

Effect of UV-C on physical properties

UV-C irradiation improves the quality of horticulture produce by reducing the microbial count and by enhancing the host resistance of the commodity by releasing certain biochemical compounds. Apart from shelf life and reduced chances of diseases, UV-C irradiation also leads to certain beneficial physical changes in fruits. Moisture loss in fruit and vegetables through metabolic activities such as respiration and evaporation losses directly affect their quality. UV irradiation showed a positive response in reducing weight loss [3] investigated the exposure of mangosteen with ultraviolet-c irradiation, they reported a significantly lower weight loss in the product during the storage period of 9 days. Similar results were reported in bitter melon when treated with a UV-C lamp of 25W with different exposure times of 20, 30, 40 minutes. The control sample faced the highest weight loss of

10.2% at the end of storage while only 6.2% (lowest) was observed in samples with treatment time of 40 minutes [29]. Tomatoes and strawberries also displayed a significant reduction in weight loss on UV-C irradiation. Higher UV-C intensity had a significant effect on the reduction of weight loss compared to the lower intensity with the same overall dose [30]. Similarly, broccoli florets displayed lower weight loss in treated samples (1.2 kJ m^{-2}) compared to control samples [31, 2] also reported the reduction of weight loss on UV-C application in shallots and fresh pistachio, respectively. [32] observed similar results in brussels sprouts and attributed the reduced weight loss to the crystalline modification, which leads to reduced permeability. The compaction of the wax structure was thought to result from reduced size and shorter wax structures. Similarly, [33] described that the reduced weight loss resulted from the change in the structure of epicuticular waxes. On exposure to UV-C irradiation, the outer layer of the fruit surface experienced reduced permeability of water vapour and volatile compound, due to changes in the microcrystalline structure of waxes. Firmness is an essential quality attribute in fruits and vegetables that determines the maturity of the agricultural commodity. The firmness reduces with the maturity of the fruit and rapidly with ripening. Firmness is directly related to the ripening, which ultimately affects the shelf life of the product [34]. UV-C irradiation results in higher firmness, thus ultimately resulting in a better quality of fruits and vegetables. [35] investigated the effect of UV-C (0, 1, 2, 3 and 6 kJm^{-2}) on metabolic features of sweet cherry fruit and reported an increase in skin resistance to penetration. A 15% increase in firmness was reported on irradiation with higher doses of 3 and 6 kJm^{-2} . Similar findings were observed in bitter melon irradiated with UV-C. As per nature, firmness reduced proportionally to the storage period but treated sample displayed significantly higher firmness in contrast to the untreated [29, 27] also reported a similar trend of UV-C irradiation on strawberries with the lowest 0.201N firmness in

untreated samples, while 0.9 kJ m^{-2} displayed the highest 0.688N firmness. A similar trend of maintaining firmness was reported by ^[36] in baby corn, ^[37] in persimmon fruit, ^[38] in Fresh-Cut Carambola, ^[30] in tomatoes and strawberry, ^[39] in peeled garlic and ^[40] in strawberry fruit ^[41] associated the increase in firmness with the enhanced level of polyamine, which functions in a similar mechanism to calcium. Polyamine limits the enzyme accessibility to the cell wall by forming cation cross-links with polysaccharides and pectic acid ^[40] attributed the higher firmness to a set of genes encoding for enzymes and protein involved in cell wall degradation ^[27] also reported the delayed firmness loss because of the UV-C effect on degrading enzyme activity. The cell membrane function and integrity were maintained because of inhibition of lipid peroxidation on UV-C application ^[42].

Effect of UV-C on the cell wall of horticulture produce

The quality of horticulture produce is judged based on attributes such as flavour, colour, textural, functional and nutritional characteristics. These attributes are dynamic in nature and vary during the development, ripening, senescence and storage. The cell wall and its component play an important role in deciding the texture of the fruits and vegetables. The major component involved in the structural changes in the cell wall during ripening and senescence is pectin, cellulose, hemicellulose and cell wall degrading enzymes ^[43] reported that UV-C treatment has the potential in inhibiting the expression of genes related to cell wall degradation, lipid metabolism and photosynthesis. The authors performed microarray analysis by using Affymetrix Tomato Gene Chip UV-C irradiated tomato to view the overall gene expression during 24 hours of treatment. It was reported that the genes related to the cell wall disassembly, lipid oxidation and photosynthesis were down regulated however the genes related to metabolism and defence response were upregulated. Pectin solubilisation is a common feature during ripening that decides the fruit texture. Pectin is the key factor in cell to cell adhesion which maintains the firmness of the cell wall. The major enzymes related to the solubilisation of pectin are pectin methylesterase (PME) and polygalacturonase (PG) which are also known as the major enzymes responsible for cell wall degradation ^[44, 45] investigated the effect of UV-C irradiation and calcium chloride on enzymatic activity and decay of tomato fruit during storage. It was reported that UV-C irradiation significantly reduced the activity of PG and PME. Similarly ^[46] reported a 40% reduction in PG activity of tomatoes on UV-C treatment. Likewise, ^[42] also reported a significant decrease in PG and PME activity of tomato fruit on UV-C irradiation ^[47] reported that UV-C treatment suppressed the height of peak PME activity in wounded fruit and suppressed the solubilisation of pectin ^[48] investigated the effect of postharvest UV-C treatment of tomato fruit on ethylene and cell wall degrading enzymes. The microscopic analysis revealed that after 15 days of storage control fruits began to shrink and the intercellular layer disappeared. In contrast to this UV-C treated fruit doesn't display adverse effects on the cell wall. Instead, the cell wall remained intact even after 30 days of storage whereas a cell wall with the scattered arrangement of filament was observed in the control fruit. The better retention of cell wall properties was attributed to the reduction in the cell wall degrading enzymes. In concomitance to the above discussion, the authors reported a

significant reduction in PG activity and a reduced peak of PME activity in treated fruits. It was also reported that UV-C altered the mRNA expression of four genes encrypting the cell wall degrading enzymes. Similarly, ^[49] reported that on UV-C treatment of strawberry, a radiation-induced modification in the expression of genes related to the enzyme activity and reduction in enzymes activity responsible for cell wall degradation.

Effect of UV-C on microbial properties

The germicidal effect of ultraviolet-c irradiation is well known and explained in detail earlier. Apart from the disinfection of food, it is also used to disinfect equipment and machines in the food processing industries ^[24] studied the effect of postharvest application of UV-C on the microbial quality of apricot. Apricot was treated with a fluency of 32.3W m^{-2} for different time duration ranging from 0 to 25 min. Initial exposure to UV-C decreased the total aerobic plate count (TAPC) from 6.1 to 3.10 log CFU g^{-1} . However, the maximum reduction was reported in 16 minutes treatment and there was no further change in the microbial count on increasing the dose. Similarly, ^[50] studied the response of persimmon fruit on UV-C treatment of fluency 0.6034 mW/cm^2 for different exposure times viz. 1, 3, 6, 12, or 24 h. It was reported that the killing rate was maximum with 6h exposure time after that there was no significant increase ^[51] researched the influence of combined and individual treatment of UV-C and freezing on date fruit. It was concluded that combined treatment of UV-C and freezing showed a minimum microbial count after 90 days compared to their treatments and control. It was reported that UV-C can significantly reduce yeast and mould count. Similarly exposure of fresh pistachio with an irradiation dose of 2.1 and 4.5 kJm^{-2} resulted in a decrease in total bacterial fungal count under detection limit from 3.42 and 1.66 CFU g^{-1} , respectively ^[2]. Besides their immediate effect in reducing microbial count, UV-C irradiated fresh-cut carambola showed a lower microbial count than control even after 14 days of storage ^[38]. Similar findings were investigated in peeled garlic on UV-C application. The samples treated with UV showed a lower aerobic microbial population compared to control ^[39, 52] also stated a significant decrease in enterobacteria, yeast and moulds count on the application of UV-C irradiation on watermelon cylinders with different cut intensities. The use of UV-C for liquid disinfection of water and liquid was started long before it was used for disinfection of solid foods. However, its use in the solid food industry is still limited compared to liquid foods ^[53] investigated the effect of UV-C with different exposure times (0-60 min) on tomato juice. A significant decrease in the microbial count was reported for 15-minute exposure however there was no significant difference between different UV-C treatments. The UV-C intensity along with UV-C dose had a significant effect on the microbial population ^[30] studied the response of UV-C irradiation intensity with the same dose on tomatoes and strawberries. The high intensity irradiated strawberry showed only 12% fungal growth, whereas low intensity and control showed 46% and 68% respectively after 5 days of storage ^[54] also stated significantly higher inactivation of pathogens on exposure to higher UV fluency ^[55] also established the use of higher intensity may cause the accumulation of phenolic and leads to better antimicrobial properties.

Effect of UV-C in disease prevention

The hormesis effect induced due to irradiation leads to the release of certain plant defense enzymes in response to the stresses developed. UV-C application increases the activity of plant defense enzymes like peroxidase (POD), PAL, β -1,3-glucanase, chitinase and also increases hydrogen peroxidase and total phenol content [56, 3] studied the effect of UV-C doses (0 - 40 kJ m⁻²) on mangosteen and concluded that can reduce the fruit rot disease. Though, the incidence of disease was minimum in 13 kJ m⁻² trailed by 26 kJ m⁻². No significant difference was found between higher dose (40 kJ m⁻²), a lower dose (6 kJ m⁻²) and control. UV treatments significantly increased PAL and POD activity. Similarly, [57] also reported that low UV-C doses significantly controlled the blackhead disease in 'Korla' fragrant pears. The persimmon fruit was also reported with enhanced disease resistance on UV-C irradiation [38, 44] stated a significant increase in PAL activity on UV-C application, which reached a maximum of 176% compared to control after 48 hours in yali pear. [58] also reported a two-fold increase in PAL activity in pear fruit on UV-C exposure [59] experimented to test the effect of UV-C irradiation in reducing the post-harvest decay of star ruby fruit. The decay percentage was measured based on the visual assessment of blue mold, green mold and brown rot. A significant reduction in decay development was reported in treated fruit compared to control. Moreover, there was no significant improvement in decay percentage in fruits at higher doses after 0.5 kJ m⁻². It was also concluded that the percentage of fruit damage (Treatment damage such as peel necrosis and rind browning) was greater in high doses. The treatment damage at higher doses reached 13.7% at 3 kJ m⁻². Although, it was only 3.2 to 4.2% for 1.5 kJ m⁻². The decrease in decay development was attributed to the accumulation of scoparone and scopoletin, which induces the production of phytoalexins and controls pathogens [60]. Mango on irradiation with UV-C doses of 2.46 and 4.93 kJ m⁻² showed enhancement in resistance to decay symptoms. Fruits treated with a higher dose had a slightly lower decay value compared with fruits irradiated with the lower dose. The overall appearance of treated samples was better because of the least decay. At storage end, control mango showed 100% fungal infection while treated fruit suffered 40-45% disorder. It was concluded that UV-C irradiation can decrease the fungal infection and maintain the quality of fruit. Apart from the reduced fungal infection, plant defense enzyme PAL activity also increased significantly on irradiation compared to control, which increases plant resistance to pathogenic attack [61]. Surface pitting occurs due to mechanical injury in the fleshy mesocarp, which is skin depression overlies necrotic lesions that result in disorder surface pitting [62, 35]. Conducted research to test the effect of UV-C irradiation on sweet cherry. The experiment revealed that UV significantly reduced surface pitting percentage and number of fruits without surface pitting compared to control.

Conclusion

Based on literature evidence UV-C irradiation had shown potential in the processing of horticulture produce. UV-C irradiation provides a product with better sensorial properties and a lesser microbial population. The major advantage of UV-C is its lethal effect on microbial DNA, which reduces the microbial load and provide a sound product free from disease-causing microorganisms. UV-C irradiation enhances

the hormesis effect in the horticulture produce, which leads to increases in plant defence enzymes. The increased activity of PAL and POD leads to a surge in disease resistance of the horticulture produce. The reduced microbial load and increased activity of plant defence enzymes enhance the shelf life of the product.

The literature evidence showed that UV-C has a positive impact on the physicochemical properties of horticulture produce. UV-C treatment reduced weight loss and firmness loss which is highly required during the storage of horticulture produce. UV-C treatment had shown potential in maintaining the cell wall integrity by reducing the activity of cell wall degrading enzymes. The UV-C radiation also has the potential in varying the genes expression related to cell wall degrading enzymes. Fewer literatures were available to discuss the response of produce towards UV-C intensity and product maturity stage at application time. Enhanced study in that area can turn beneficial in reducing the application time and deciding the correct maturity stage for treatment. The application of higher doses has shown negative impacts on commodity or doesn't show a marked difference from the following dose. More studies are required to optimize the doses for different commodities and different cultivars.

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