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REVIEW



Emerging chemical and physical disinfection technologies of fruits and vegetables: a comprehensive review

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ABSTRACT

With a growing demand for safe, nutritious, and fresh-like produce, a number of disinfection technologies have been developed. This review comprehensively examines the working principles and applications of several emerging disinfection technologies. The chemical treatments, including chlorine dioxide, ozone, electrolyzed water, essential oils, high-pressure carbon dioxide, and organic acids, have been improved as alternatives to traditional disinfection methods to meet current safety standards. Non-thermal physical treatments, such as UV-light, pulsed light, ionizing radiation, high hydrostatic pressure, cold plasma, and high-intensity ultrasound, have shown significant advantages in improving microbial safety and maintaining the desirable quality of produce. However, using these disinfection technologies alone may not meet the requirement of food safety and high product quality. Several hurdle technologies have been developed, which achieved synergistic effects to maximize lethality against microorganisms and minimize deterioration of produce quality. The review also identifies further research opportunities for the cost-effective commercialization of these technologies.

KEYWORDS

Fruits; vegetables; *E. coli*; *Salmonella*; disinfection; safety

Introduction

As income and urbanization have increased around the world, global food consumption has been changed to diets that are higher in protein, meats, refined sugars, fats, and oils (Gu et al. 2019; David and Michael 2014). Such global dietary, processing and production shifts are threatening human health, environmental sustainability, and biodiversity (Fischer 2018; Springmann et al. 2018). Fresh fruits and vegetables (F&V) are integral components of the human diet due to their health and nutritional benefits (Liu 2013). Increasing the consumption of F&V can provide substantial health benefits and reduce the risk for many diet-related chronic diseases, such as obesity, type II diabetes, coronary heart disease and some cancers (Willett et al. 2019; Clark, Hill, and Tilman 2018; Foley et al. 2011).

In recent years, the consumption of F&V in raw or minimally processed forms has increased continuously (Ramos et al. 2013). Consequently, the frequency of documented produce-related foodborne illness outbreaks has increased (Scallan et al. 2011; Scharff 2012; Center for Science in Public Interest (CSPI) 2015; Centers for Disease Control and Prevention (CDC) 2017; García and Heredia 2017). The economic cost related to foodborne illnesses in the US is

estimated to be higher than \$50 billion per year, involving more than 48 million of persons (Scharff 2012), 128,000 hospitalizations, and 3,000 deaths (CDC 2017). Therefore, the microbiological safety has become a challenging concern for the food industry and consumer.

F&V are easily contaminated by microorganisms at various points during production, handling, and packing process (García & Heredia 2017). The pathogenic microorganisms commonly involved in foodborne outbreaks resulting from contaminated produce consumption are *Escherichia coli*, *Salmonella*, *Shigella* spp., *Listeria monocytogenes*, *Bacillus cereus*, *Staphylococcus aureus*, etc. (Alwi & Ali 2014; Severino et al. 2014; Afari et al. 2016; García and Heredia 2017; Kang and Song 2018). Some common F&V susceptible to pathogen contamination are leafy greens (lettuce, spinach, cabbages, etc.; Bhargava et al. 2015; Agüero et al. 2016; Chen, Xue et al. 2018), tomato (Kwon, Chang, & Han 2017), pepper (Alwi & Ali 2014), grape (Botondi et al. 2015), blueberry (Bridges, Rane, & Wu 2018), and others. The high moisture content of fresh F&V provides an environment to support spoilage microorganisms (Sun et al. 2017). It has been reported that more than 35% fresh F&V are lost during harvesting, transportation and storage due to

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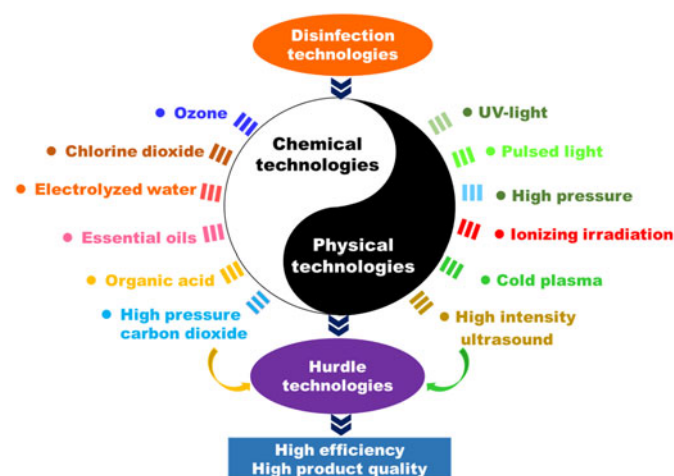


Figure 1. Emerging disinfection technologies of fruits and vegetables.

microbial spoilage, mechanical injury, improper storage temperature and relative humidity management (Shafiee-Jood & Cai 2016). Spoilage is a devastating food supply problem in developing countries, where as high as 75% of the food is lost on-farm, during transport and processing (Lung et al. 2015).

Therefore, eliminating microorganisms on F&V during post-harvest processing is vital to assure microbial safety and prolong shelf life of F&V. A number of disinfection methods are currently practiced. Chlorine is the most common antimicrobial sanitizer applied in the food industry (Gil et al. 2009; Olaimat and Holley 2012; Ramos et al. 2013). However, the reduction of microorganisms with chlorine is limited. Chlorine treatment usually results in only 1–2 log reduction of most bacteria on fresh produce (Mukhopadhyay et al. 2014). The overall contamination levels of the chlorine washed produce can still be relatively high and pathogens may still survive (Goodburn & Wallace 2013). Furthermore, chlorine treatment leads to the liberation of chlorine vapors and formation of potentially carcinogenic by-products—trihalomethanes and haloacetic acids (Praeger, Herppich, & Hassenberg 2018). They can trigger concerns for both human health and environmental pollution. Due to the safety and efficacy issues, the use of chlorine for sterilization of fresh-cut produce has been prohibited in several countries, such as the Netherlands, Switzerland, and Belgium (Rico et al. 2007).

Consumers are becoming increasingly aware of safety issues of F&V consumption and at the same time are demanding fresh-like high quality products (García & Heredia 2017). To satisfy consumers, alternative sanitation agents and novel disinfection technologies have been developed, as shown in Figure 1. These emerging technologies include chemical methods involving the use of chlorine dioxide (Lee, Beuchat et al. 2018), ozone (Bridges, Rane, & Wu 2018), electrolyzed water (EW, Rahman, Khan, and Oh 2016), essential oils (EO; Kwon, Chang, and Han 2017), high-pressure carbon dioxide (HPCD; Ferrentino, Komes, and Spilimbergo 2015), and organic acids (Chen, Zhang, and Zhong 2019; Chen et al. 2019a) as the sanitation agents, as well as non-thermal physical treatments, such as UV-light

(Beristaín-Bauza et al. 2018), pulsed light (PL; Keklik, Krishnamurthy, and Demirci 2012), ionizing radiation (Molnár et al. 2018), high hydrostatic pressure (HHP; Maresca and Ferrari 2017), cold plasma (CP; Min et al. 2017), high-intensity ultrasound (Millan-Sango et al. 2016), etc. To further to improve sterilization processes, a number of hurdle technologies have been developed (Ramos-Villarroel, Martín-Belloso, & Soliva-Fortuny 2015; Ngnitcho et al. 2017).

Therefore, this review aims to summarize the working principles and applications of various emerging disinfection technologies. The limitations and future research opportunities are also identified and discussed.

Chemical disinfection technologies

Over the past few decades, chlorine dioxide, ozone, EW, EOs, HPCD, and organic acids have gained growing interests by the public and the food industry. They have advantages of high biocidal efficacy, broad antimicrobial spectrum, and low or negligible toxicity of resulting by-products and residues.

Chlorine dioxide

Chlorine dioxide (ClO_2) is an oxidative gas and commonly produced by the reaction of acid and sodium chlorite, or sodium chlorite and chlorine gas (Ölmez & Kretzschmar 2009). ClO_2 can be used as a sanitizing agent with broad antimicrobial effects on bacteria, fungi and virus. It has 2.5 times higher oxidative capacity than chlorine in hypochlorous acid (HOCl), without forming carcinogenic by-products (Praeger, Herppich, & Hassenberg 2018). Hence, the sterilization efficiency and safety of ClO_2 are more advantageous than chlorine and ClO_2 has been adopted as an alternative to chlorine disinfection in recent decades.

Disinfection mechanisms

ClO_2 kills microorganisms by its destabilizing effects on cell membranes. ClO_2 can alter the structure of proteins and lipids in the outer membrane, thus enhances membrane permeability, induces the efflux of intracellular components, and eventually causes the cell death (Praeger, Herppich, & Hassenberg 2018). Ogata (2007) found that protein side chains (tryptophan and tyrosine residues) of yeast were covalently modified by ClO_2 , that caused the denaturation of protein. Meanwhile, ClO_2 induces the oxidative stress to microorganisms, which lead to the alteration of the microbial transcriptional and phenotypic profile. Pleitner et al. (2014) found that after exposed to ClO_2 , the *L. monocytogenes* cell genes involved in heat shock response, redox reactions, cell replication, and universal stress response were significantly differentially expressed.

Applications

Using ClO_2 for sterilizing fresh produce is permitted by the FDA (FDA 2017). ClO_2 can be used in aqueous and gaseous forms (Kim, Lee et al. 2017; Lee, Beuchat et al. 2018). The

Table 1. Gas-phase chemical disinfection technologies for fruits and vegetables.

Methods	Material	Microorganisms	Treatment conditions	Microbial reduction	Reference
Chlorine dioxide	Carrot	<i>E. coli</i> , <i>S. enterica</i> , and <i>L. monocytogenes</i>	0.07 mg ClO ₂ /g produce, 5 h	7.7, 4.8, and 2.5 log CFU/g, respectively.	Bridges, Rane, and Wu 2018
	Lettuce	<i>E. coli</i> O157:H7	1.0 mg/L, 10 min	1.91 log CFU /g.	Singh et al. 2002
	Blueberry	<i>E. coli</i> , <i>S. enterica</i> , and <i>L. monocytogenes</i>	0.12 mg ClO ₂ /g produce, 2.5 h	3.6, 1.6, and 2.1 log CFU/g, respectively.	Bridges, Rane, and Wu 2018
	Tomato	<i>E. coli</i> , <i>S. enterica</i> , and <i>L. monocytogenes</i>	0.12 mg ClO ₂ /g produce, 2.5 h	5.6, 5.0, and 6.0 log CFU/g, respectively.	Bridges, Rane, and Wu 2018
	Grape tomato	<i>S. Typhimurium</i>	0.85 mg, 58 min, 4–25 °C	3.95–7.37 log CFU/fruit.	Netramai et al. 2016
	Grape tomato	<i>E. coli</i> and <i>A. alternata</i>	2, 4, 6 and 8 ppm, 14 d, 20 °C	2.9–4.7 and 1.6–4.0 log CFU/g, respectively,	Sun et al. 2017
	Apple	<i>E. coli</i> O157:H7, <i>S. Typhimurium</i> , and <i>L. monocytogenes</i>	0 ppm, 5–15 min	1.39–3.95, 1.25–3.95 and 1.47–3.50 log CFU/cm ² , respectively.	Park and Kang 2017
Ozone	Carrot	<i>E. coli</i> , <i>S. enterica</i> , and <i>L. monocytogenes</i>	300 ppm, 5 h	1.2, 0.5, and 0.8 log CFU/g, respectively.	Bridges, Rane, and Wu 2018
	Baby carrot	<i>E. coli</i> O157:H7	7.6 mg/L, 15 min	2.64 log CFU/g.	Singh et al. 2002
	Grape	Molds and yeast	1.5 g/h, 18 h	Up to 50%.	Botondi et al. 2015
	Tomato	<i>E. coli</i> , <i>S. enterica</i> , and <i>L. monocytogenes</i>	300 ppm, 5 h	1.6, 1.1, and 1.1 log CFU/g, respectively.	Bridges, Rane, and Wu 2018
	Bell pepper	<i>E. coli</i> O157, <i>S. Typhimurium</i> and <i>L. monocytogenes</i>	9 ppm, 6 h	2.89, 2.56 and 3.06 log CFU/g, respectively.	Alwi and Ali 2014

applications of ClO₂ for effective disinfection of various microorganisms on F&V have been widely reported (Table 1). Lee, Beuchat et al. (2018) explored the feasibility of gaseous ClO₂ for sterilizing chili peppers. They observed that 6 h treatment reduced the population of *S. Typhimurium* by > 5.6 log CFU/g, and the naturally occurring aerobic bacteria was less than the detection limit (<0.7 log CFU/g). Kim, Lee et al. (2017) treated pepper with gaseous ClO₂ (0–200 µg/mL) and found that the *B. cereus* spores on pepper were decreased by 1.2–3.1 log CFU/sample. The *Salmonella*, *E. coli* O157:H7, and *L. monocytogenes* on fresh-cut cabbage and carrots were reduced by 3.13–4.42 and 5.15–5.88 log CFU/g after gaseous ClO₂ treatment (Sy et al. 2005). Gaseous ClO₂ treatment reduced the total bacteria and yeast & mold counts on grapes after storage for 90 d by 1.3 and 2.7 log CFU/g, respectively, and reduced decay incidence by 30% (Chen, Wang et al. 2018). Therefore, ClO₂ treatment can effectively reduce microbial loads on F&V. Furthermore, ClO₂ has high potential to preserve the quality of F&V. It was found that ClO₂ treatment alleviated browning in grapes (Chen, Wang et al. 2018) and longan (Chomkitichai et al. 2014) during storage. Grape tomatoes treated by ClO₂ were firmer and loss less weight compared to the untreated sample after storage for 14 days at 20 °C (Sun et al. 2017).

Factors influencing efficacy

The efficacy of ClO₂ depends on the application form, concentration, holding time, microorganisms, F&V types, temperature, and other factors. Gaseous ClO₂ has higher sterilizing efficacy than its aqueous solution, due to the higher penetration capability of gas into microorganism harboring sites at the produce surface (Praeger, Herppich, & Hassenberg 2018). The ClO₂ gas and solution treatment (0.3 mg/L, 10 min) reduced *L. monocytogenes* on green pepper surfaces by 3.05 and 1.87 log CFU/5g, respectively (Han et al. 2001). However, ClO₂ aqueous solutions are more commonly used due to the easy combination with the

existing washing lines. Usually, the reduction in the microbe population increased with increasing ClO₂ concentration and holding time. For instance, treating red chili with ClO₂ at 0, 50, 100, and 200 µg/mL resulted in decreases in the *Bacillus cereus* spore populations by 1.2, 2.3, 2.6, and 3.1 log CFU/chili, respectively (Kim, Oh et al. 2017). The populations of *E. coli* O157:H7 on tomatoes treated with ClO₂ gas for 5, 10, and 15 min were reduced by 1.33, 2.64, and 4.72 log CFU/cm², respectively (Park & Kang 2017). It has been reported that the Gram-negative bacteria were more sensitive to ClO₂ than Gram-positive bacteria, and yeasts had intermediate sensitivity, while the spores of mold and *B. cereus* had a strong resistance to ClO₂ (Praeger, Herppich, & Hassenberg 2018). Increasing temperature results in an increase of log reduction of microorganisms to some extent. Netramai et al. (2016) treated grape tomatoes with ClO₂ gas for 50 min at 4 and 25 °C and observed that *S. enterica* Typhimurium was reduced by 3.95 and 7.37 log CFU/fruit, respectively. Meanwhile, F&V species is also a non-negligible factor affects the antimicrobial efficiency of ClO₂. Gaseous ClO₂ treatment (0.06–0.15 mg/g, 2.5–5 h) decreased bacterial pathogens (*E. coli*, *S. enterica*, and *L. monocytogenes*) by 2.0–7.7, 0.9–3.7, and 3.4–8.8 log CFU/g on carrots, blueberries, and tomatoes, respectively (Bridges, Rane, & Wu 2018). The water quality (e.g. pH, temperature, turbidity, conductivity, and organic matter content) should be taken into consideration since they are related to the disinfection effectiveness of aqueous ClO₂ (López-Velasco et al. 2012).

Limitations

Usually, ClO₂ treatment requires long exposure time (10 min–2 h) to obtain the desired microbial loads reduction, which may limit industrial application (Singh et al. 2002; Goodburn and Wallace 2013). The degradation of produce color and aroma during ClO₂ treatment has been observed in some cases. For example, Lee, Beuchat et al. (2018) observed the redness and yellowness decrease in chili pepper, whereas Sy et al. (2005) revealed the browning effect in

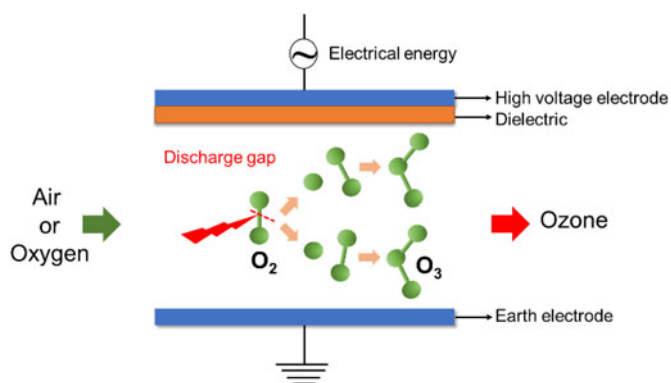


Figure 2. Ozone generation process by means of dielectric barrier discharge.

fresh-cut lettuce, cabbage, and peaches upon the treatment with ClO_2 . There are some other limitations of ClO_2 application. ClO_2 is thermally unstable and a powerful oxidizing agent which makes it prone to explosion by any source of thermal energy, such as sunlight and heat. It can decompose and explode under intense fire conditions and form corrosive and highly toxic chlorine and hydrogen chloride gases (ERCO 2012), causing a safety concern in the work place. In addition, it is toxic at high concentrations and the concentration of residual ClO_2 in produce must be lower than 3 ppm, hence the treated produce must be followed by a potable water rinse (Praeger, Herppich, & Hassenberg 2018). Moreover, ClO_2 treatment may not irreversibly damage microbes. For example, *B. cereus* and *Pseudomonas fluorescens* have been observed to survive and increase after ClO_2 treatment (Lindsay et al. 2002) and repeated treatments were required to curb the microbial growth in F&V during storage (Chen, Wang et al. 2018).

Ozone

Ozone (O_3) is a gas that occurs naturally in the atmosphere. It is a strong oxidative agent and powerful broad-spectrum microbicide that is active against bacteria, fungi, virus, and bacterial and fungal spores (Khadre, Yousef, & Kim 2001). At present, ozone is commonly generated by the passage of air, or oxygen gas through a high-voltage electrical discharge (Perry & Yousef 2011; Brodowska, Nowak, & Śmigielski 2018). As shown in Figure 2, air or oxygen flows through the gap between two electrodes separated by a dielectric material, a high energy discharge splits molecular oxygen into atomic oxygen radicals, which spontaneously combines with molecular oxygen to form ozone molecules (Brodowska, Nowak, & Śmigielski 2018). Ozone has the advantages of having high reactivity, oxidation potential, and penetration capacity (Kim, Yousef, & Dave 1999). Ozone has the additional food processing advantage that it spontaneously decomposes to oxygen, leaving no residues on treated produce. Ozone is generally recognized as safe (GRAS) for food contact applications (Botondi et al. 2015) and approved by the FDA in 2001 as a direct contact food sanitizing agent (Khadre, Yousef, & Kim 2001).

Disinfection mechanisms

Ozone has been proven to destroy microbe via the progressive oxidation of vital cellular constituents, including proteins and peptidoglycans in cell wall, enzymes, and nucleic acids in the cytoplasm, as well as unsaturated lipids in the cell membranes (Miller, Silva, & Brandão 2013). In detail, ozone can react with polysaccharides and break the glycosidic bonds; oxidize the sulfhydryl groups and amino acid of protein and peptides; and oxidize polyunsaturated fatty acids to acid peroxides (Khadre, Yousef, & Kim 2001; Brodowska, Nowak, & Śmigielski 2018). These modifications cause cell envelope disruption or disintegration, leading to leakage of the cell contents and finally results in cell lysis (Pascual, Llorca, & Canut 2007). Membrane disruption allows ozone to penetrate into the cell, reacts with DNA and RNA, especially thymine, guanine, and uracil, and damages to nucleic acids (Perry & Yousef 2011). Besides, the ozone decomposition in water creates free radicals, such as hydroperoxyl ($\bullet HO_2$), hydroxyl ($\bullet OH$), and superoxide ($\bullet O_2^-$) are also responsible for the antimicrobial activity (Perry & Yousef 2011; Brodowska, Nowak, & Śmigielski 2018).

Applications

Ozone has been extensively tested for use in inactivating microorganisms on F&V, such as tomato (Venta et al. 2010), bell pepper (Alwi & Ali 2014), broccoli (Severino et al. 2014), and grape (Botondi et al. 2015), as detailed in Table 1. Daş, Gürakan, and Bayındırlı (2006) observed that gaseous O_3 treatment (20 mg/L, 15 min) achieved more than 7 log CFU/fruit elimination of viable *Salmonella* cells on cherry tomatoes. The *E. coli* on tomatoes was reduced by 2–3 log CFU/fruit after treated with ozonated water at 0.5 mg/L for 15–30 min (Venta et al. 2010). Aqueous ozone treatment (1.4 mg/L) reduced the total bacteria on fresh-cut apple by 1.83–2.13 log CFU/g, and extended the shelf life up to 10 days (Liu et al. 2016). Therefore, ozone provides a promising alternative to conventional chemical sanitizers. The quality and nutrition of F&V were not significantly change by proper ozone treatment. No considerable changes in color and flavor of lettuce, sprouts, and spinach were observed after ozone treatment at 1.0 ppm for 360 min (Ngnitcho et al. 2017). Ethylene production, polyphenol oxidase and peroxidase activities, and malondialdehyde contents of fresh-cut apple were reduced by ozone treatment (Liu et al. 2016). In addition, higher extractability of phytochemicals was obtained after ozone treatment (100 g O_3/m^3 , 30 min), and the total phenolic content in ozone treated juniper berries (15.47 mg CE/g) was about twofold higher than that of the sample without ozone treatment (9.81 mg CE/g) (Brodowska et al. 2015). Ozone has also been shown to eliminate of off-flavor, mycotoxins and pesticide residues (Agriopoulou et al. 2016; Brodowska, Nowak, & Śmigielski 2018). Aflatoxin AFG2 and AFB2 were decreased by 42.1–53.9% and 17.4–29.6%, respectively, after gaseous ozone treatment at 8.5–40 ppm for 20 min (Agriopoulou et al. 2016).

Factors influencing efficacy

The efficacy of ozone to inactivate microorganisms depends on many factors, including form and concentration of ozone, exposure time, microbial species, and food property (Horvitz & Cantalejo 2014). Ozone can perform in both gaseous and aqueous forms (Perry & Yousef 2011). The sterilizing efficiency of aqueous ozone was found to be greater than the gaseous form (Pascual, Llorca, & Canut 2007). However, the decomposition rate of ozone in water is much higher than its gaseous form (Pascual, Llorca, & Canut 2007; Oner and Demirci 2016). In general, increasing ozone concentration or exposure time results in a higher reduction of the microbial population. The total mesophilic bacteria count on juniper berries treated with gaseous ozone at 100, 130, and 160 g O₃/m³ for 90 min were reduced by 0.8, 1.6, and 1.5 log CFU/g, respectively (Brodowska et al. 2015). Tomatoes washed by 0.5 mg/L ozonated water for 15 and 30 min achieved 2.03 and 2.89 log reductions of *E. coli* populations, respectively (Venta et al. 2010). The microorganism and F&V species are important factors related to efficacy. Usually, bacteria are more sensitive to ozone than yeasts and fungi, Gram-positive bacteria are more sensitive than Gram-negative organisms, and spores are more resistant than vegetative cells (Pascual, Llorca, & Canut 2007). Gaseous ozone treatment (9 ppm, 6 h) reduced *E. coli* O157, *S. Typhimurium* and *L. monocytogenes* populations on fresh-cut bell pepper by 2.89, 2.56, and 3.06 log CFU/g, respectively (Alwi & Ali 2014). The populations of *E. coli* on blueberry and tomato treated with ozone under the same condition (300 ppm, 5 h) were reduced by 0.5 and 1.6 log CFU/g, respectively (Bridges, Rane, & Wu 2018). There is a conflict with choice of treatment temperature, a rise in temperature increases the proportion of microbial destruction, but it decreases the stability and solubility of ozone (Pascual, Llorca, & Canut 2007). Additionally, water quality and environmental relative humidity also affect the efficiency of aqueous and gaseous ozone, respectively (Brodowska, Nowak, & Śmigielski 2018).

Limitations

High ozone concentration or long exposure time is needed to attain significant reductions of microorganisms. An excessive use of ozone may cause oxidation of some ingredients in produce (Kim, Yousef, & Dave 1999), resulting in discoloration, flavor deterioration, and phytochemicals degradation (Brodowska, Nowak, & Śmigielski 2018; Botondi et al. 2015). Noticeable aroma loss and bleaching were observed in ozone treated strawberries (Nadas, Olmo, & Garcia 2003), lettuce (Singh et al. 2002), tomato and carrot (Bridges, Rane, & Wu 2018). Gaseous ozone treatment decreased approximately 12, 47, and 22% of EO, phenolics and ferric-reducing antioxidant power in juniper berry, respectively, as compared to untreated samples (Brodowska et al. 2015). Meanwhile, the highly unstable and explosive property of ozone has also limited its application in postharvest processing of F&V (Brodowska, Nowak, & Śmigielski 2018). Ozone must be generated on-site as needed, and it is costly to maintain the concentration (Goodburn & Wallace 2013).

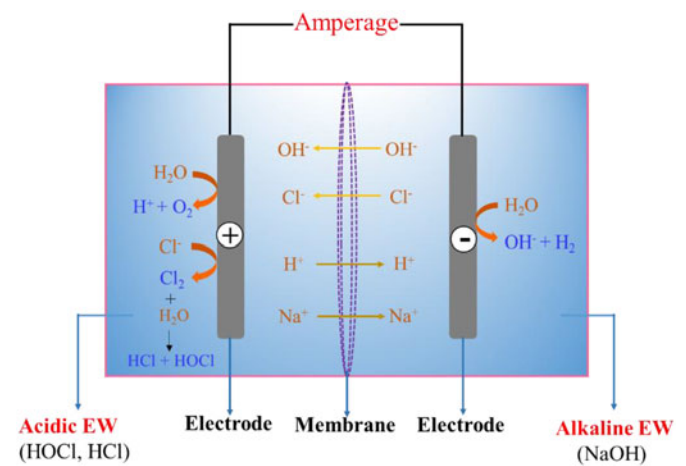


Figure 3. Schematic diagram of EW generation system (adapted from Hati et al. 2012).

Besides, excessive exposure to ozone (>1 ppm) may cause damage to the respiratory tract, lungs, and eyes of humans (Oner & Demirci 2016). Good air-handling and ozone destruction systems are needed to ensure the safety of workplace (Perry & Yousef 2011). Additionally, the capacity of ozone generator requires further enhancement to meet the demand of large-scale industrial application (Oner & Demirci 2016).

Electrolyzed water

Electrolyzed water (EW) is produced by electrolysis of a dilute salt solution (generally NaCl) passing through an electrolytic cell which contains positively and negatively inert charged platinum electrodes separated by a bipolar membrane (Ngnitcho et al. 2017). The principle of EW production is illustrated in Figure 3. NaCl solution dissociates into Na⁺ and Cl⁻ ions; negatively charged ions (OH⁻ and Cl⁻) move toward the anode where they are oxidized, generating hypochlorous acid, hypochlorite ion, hydrochloric acid, oxygen gas, and chlorine gas; positively charged ions (H⁺ and Na⁺) move to the cathode and reduced, resulting in formation of hydrogen and sodium hydroxide (Hati et al. 2012). When anode and cathode are separated by a membrane, acidic EW with low pH (2.3–2.7) and high oxidation-reduction potential (>1000 mV) is produced on anode side (Rahman, Khan, & Oh 2016), and alkaline EW with high pH (10.0–11.5) and low oxidation-reduction potential (–800 to –900 mV) is produced on the cathode side (Rahman, Khan, & Oh 2016). When anode and cathode are not separated, the process results in producing neutral EW (Rahman, Khan, & Oh 2016).

Disinfection mechanisms

EW has strong bactericidal effects on a variety of bacteria. The disinfection mechanisms of EW including the modification of metabolic fluxes and adenosine triphosphate production, increase of membrane penetrability, the release of intracellular components that lead to necrosis of bacteria (Hati et al. 2012; Rahman, Khan, and Oh 2016; Liu et al.

Table 2. Liquid-phase chemical disinfection technologies for fruits and vegetables.

Methods	Material	Microorganisms	Treatment conditions	Microbial reduction	Reference
Electrolyzed water	Lettuce	<i>E. coli</i> and <i>Salmonella</i> spp.	22.1 mg ACC/L, pH 5.6, 5 min	2.8 and 2.91 log CFU/g, respectively.	Issa-Zacharia et al. 2011
	Daikon sprouts	<i>E. coli</i> and <i>Salmonella</i> spp.	22.1 mg ACC/L, pH 5.6, 5 min	2.8 and 2.91 log CFU/g, respectively.	Issa-Zacharia et al. 2011
	Pear	<i>E. coli</i> , <i>S. enterica</i> and <i>Listeria</i> spp.	102 mg ACC/L, pH 8.2, 5 min	0.53–1.1 log CFU/g.	Graça et al. 2017
	Pear	<i>Cronobacter sakazakii</i>	103 ACC mg/L, pH 2.82, 5 min	1.3 log CFU/g.	Santo et al. 2016
	Melon	<i>Cronobacter sakazakii</i>	103 ACC mg/L, pH 2.82, 5 min	1.7 log CFU/g.	Santo et al. 2016
	Cilantro	Total aerobic bacteria, yeasts and molds	15–30 mg ACC/L, pH 5.76–6.05, 5 min	1.78–4.63 and 1.03–3.86 log CFU/g, respectively.	Zhang et al. 2016
Essential oils	Mango	<i>E. coli</i> and <i>C. sakazakii</i>	101 mg ACC/L, pH 7.95, 5 min	2.19 and 1.70 log CFU/g, respectively.	Santo et al. 2018
	Mature and baby spinach	<i>S. enterica</i>	Lemongrass oil: 0.1–0.5%, 1–2 min	0.5–2.5 log CFU/g.	Moore-Neibel et al. 2011
	Lettuce and spinach	<i>S. enterica</i>	Oregano oil: 0.1–0.5%, 1–2 min	0.5–4.9 log CFU/g.	Moore-Neibel et al. 2013
	Lettuce	<i>S. Typhimurium</i> and <i>E. coli</i> O157:H7	Clove extract: 10%, 10 min	4.04 and 3.68 log CFU/g, respectively.	Kim et al. 2011
	Baby carrot	<i>E. coli</i> O157:H7	Thyme oil—1.0 mL/L, 10 min	2.06 log CFU/g.	Singh et al. 2002
	Green bean	<i>L. innocua</i>	0.05% mandarin oil + 1% N-palmitoyl chitosan	Reductions of 1–3 log CFU/g during 14 d storage.	Donsì et al. 2015
	Broccoli florets	<i>L. monocytogenes</i>	0.05% carvacrol + 1.0% modified chitosan	1.46 log CFU/g after 6 d storage	Severino et al. 2014
	Tomato	<i>S. enterica</i>	3% of Oregano essential oil + 1% polyvinyl alcohol films containing	>2.7 log CFU/fruit during storage for 7 d.	Kwon, Chang, and Han 2017
Organic acid	Lettuce	<i>E. coli</i> O157:H7, <i>S. aureus</i> , <i>L. monocytogenes</i> and <i>Salmonella</i> spp.	Fumaric acid: 0.5%, pH 2.5, 3 min	Appropriately 2.0–2.5 log CFU/g.	Ngnitcho et al. 2017
	Lettuce	<i>E. coli</i> O157:H7 and <i>Salmonella</i>	Lactic acid: 2%, pH 1.5, 55 °C (spray)	2.7 and 2.3 log CFU/g, respectively.	Neal et al. 2012
	Sprout	<i>E. coli</i> O157:H7, <i>L. monocytogenes</i> and <i>Salmonella</i> spp.	Fumaric acid: 0.5%, pH 2.5, 3 min	1.65, 2.17 and 2.12 log CFU/g, respectively.	Ngnitcho et al. 2017
	Avocado, watermelon, mushroom	<i>L. innocua</i> and <i>E. coli</i>	Malic acid: 2%, 2 min	1.73–1.82 log CFU/g during 15 d storage.	Ramos-Villarroel, Martín-Belloso, and Soliva-Fortuny 2015
	French beans	Total aerobic mesophilic bacteria	Citric acid: 1.6%, 5 min	2 log CFU/g.	Gupta et al. 2012
	Apple	Total bacteria	Citric acid: 0.5%, 5 min	2.1 log CFU/g.	Chen et al. 2016

2018). The active chlorine (Cl_2 , HOCl , and OCl^-) in EW (as shown in Figure 3) contributes to the inactivation of microbial cells (Rahman, Khan, & Oh 2016). In detail, OCl^- ion can damage the outer membrane and inactivate key proteins in the plasma membrane, resulting in the disintegration of the cell wall and membrane. HOCl can penetrate into cells and damage the microbial cell walls and the organelles within the cell by inhibiting enzymes and breaking down DNA, and disrupting cell metabolic processes. Liu, Wu et al. (2017) and Liu et al. (2018) observed that the EW stress induced the intracellular ROS, decreased the ribose-5-phosphate, amino acids and glucose levels, inhibited phosphate acetyltransferase-acetate kinase, and increased the 3-hydroxybutyrate level of bacteria. These changes perturbed the general metabolic pathways of bacteria, including reduced nucleotide and amino acid biosynthesis, suppressed energy-associated metabolism, altered osmotic adjustment, and promoted fatty acid metabolism, and eventually inhibited the cell proliferation (Liu, Wu et al. 2017; Liu et al. 2018). Besides active chlorine, some reactive oxygen species are formed in EW can cause a significant inactivation of microorganisms (Jeong, Kim, & Yoon 2006). The $\cdot\text{OH}$ was

observed in the NEW (Zhang, Yang, & Chan 2018), and with the existence of CO_3^{2-} and HCO_3^- , $\cdot\text{OH}$ can be captured and produces carbonate radicals ($\bullet\text{CO}_3^-$) (Zhang, Lai, & Yang 2018). Hati et al. (2012) also showed that O_3 , H_2O_2 , and $\cdot\text{OH}$ can be generated during electrolysis, that also contribute to the antimicrobial efficacy of EW.

Applications

EW has been highlighted as an emerging alternative to sodium hypochlorite disinfection because of its ease of operation, low cost, safe and eco-friendly nature (Gómez-López, Gil, & Allende 2017). It is effective for the disinfection of most microorganisms and is widely used for F&V processing, such as lettuce (Hao et al. 2015; Zhao et al. 2019), daikon sprouts (Issa-Zacharia et al. 2011), and red cabbage (Chen, Xue et al. 2018), as shown in Table 2. The aerobic mesophilic counts and yeasts & molds on lettuce treated with EW (4 mg available chlorine content (ACC)/L) for 7 min were reduced by 5.70 and 6.22 log CFU/g, respectively (Zhao et al. 2019). Issa-Zacharia et al. (2011) showed that EW treatment (22.1 mg ACC/L, 5 min) reduced bacteria



Figure 4. Commercial electrolyzed water generator (Hoshizaki Electric Corporation).

loads (total aerobic mesophilic bacteria, *E. coli*, and *Salmonella* spp.) on Chinese celery, lettuce and daikon sprouts by 2.45–2.7, 2.7–2.8, and 2.87–2.91 log CFU/g, respectively, which were comparable to the reductions achieved by NaOCl solution (100 mg ACC/L). Zhang et al. (2017) showed that the neutral EW (pH 7.08, 40 mg ACC/L) produced by their developed portable electrolytic sanitizing unit achieved >6 log reduction in *E. coli* O157:H7 and *L. monocytogenes* BAA-839. Meanwhile, the color and firmness of lettuce were not significantly changed after EW treatment (Zhao et al. 2019). Similarly, Graça et al. (2017) observed that there were no significant differences in color, soluble solid content, and acidity of untreated and EW treated fresh-cut pear. Therefore, EW exhibits strong bactericidal ability without significant alteration of product quality. The commercial EW generator is available (Figure 4), and it has been applied in food processing industry.

Factors influencing efficacy

The main factors influence the EW efficiency against microbes includes ACC and pH of EW, exposure time, microorganism and F&V types, and temperature. A higher ACC, longer exposure time, and lower pH may result in greater reductions in microbial populations. For instance, as the ACC increased from 50 to 150 ppm, the pH decreased from 3.94 to 3.31 and the reductions of the aerobic bacteria, molds and yeasts, and *S. Typhimurium* DT104 on red cabbage increased from 2.33 to 4.72, 0.94 to 3.48, and 2.46 to

2.89 log CFU/g, respectively (Chen, Xue et al. 2018). The populations of *E. coli* O78 on fresh-cut cilantro were reduced by 0.57, 1.96, and 2.76 log CFU/g, after being treated by alkaline, slightly acidic and strongly acidic EW, with pH values of 11.65, 5.85, and 2.48, respectively (Hao et al. 2015). The surviving total aerobic bacteria on celery were significantly decreased from 7.61 to 2.98 log CFU/g, when the exposure for 7 min (Zhang et al. 2016). Meanwhile, Rahman, Ding, and Oh (2010) also showed the reduction in the populations of *L. monocytogenes* increased from 4.98 to 7.42 log CFU/mL when EW temperature was raised from 4 to 50 °C. However, Liu, Tan et al. (2017) reported that an increase in temperature (from 20 to 50 °C) of EW did not achieve a significant reduction in *E. coli* O157:H7 and *L. monocytogenes* on broccoli, that might be due to the partial loss of ACC occurred when the high temperature was used (Rahman, Khan, & Oh 2016).

Limitations

Significant negative effect on the nutrient content of F&V was observed after EW treatment. The cyanidin, pelargonidin, and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity of red cabbage treated with EW (ACC 100 ppm, 3 min) were significantly reduced by 18.5, 22.1, and 11.2%, respectively (Chen, Xue et al. 2018). Strong acid EW possesses a higher efficiency, but it has limited application due to its corrosive and unstable characteristics (Zhang, Yang, & Chan 2018). Slightly acidic and neutral EW has been increasingly used (Santo et al. 2018). Additionally, chlorine gas can be generated and released during the EW generation process (Rahman, Khan, & Oh 2016).

Essential oils

Essential oils (EOs) or volatile oils, refer to complex volatile compounds with strong sensorial characteristics (Patrignani et al. 2015). EOs are natural secondary metabolites found in plant flowers, buds, seeds, leaves, roots, wood, stems, twigs, fruits or bark (Patrignani et al. 2015). They play a fundamental role in defending against pathogens, herbivores, insects, and UV light (de Oliveira et al. 2017).

Disinfection mechanisms

EOs possess strong broad-spectrum of antibacterial properties (Kwon, Chang, & Han 2017). The mechanisms of the antimicrobial actions of EOs hasn't been completely understood. The antimicrobial activities of the EOs seem to be related to their composition, and the possible synergistic interactions among the components (Patrignani et al. 2015). The major antimicrobial components in EOs have been proven to be phenolics, terpenes, aliphatic alcohols, aldehydes, ketones, acids, and isoflavonoids (Tiwari 2014). These compounds can interact with the lipid of cell membrane, increase the fluidity and permeability of the cytoplasmic membrane, and cause the leaching out of vital cell components (Tiwari 2014), and bind to proteins, ultimately leading to enzyme inactivation (Ochoa-Velasco et al. 2018). These

modifications destroy the microbial metabolism, such as inhibiting electron transport for energy production, disrupting the proton motive force, protein translocation and synthesis of cellular components, that lead to cell lysis and death (Nazzaro et al. 2013). A number of components of the EOs, such as carvon, thymol, and carvacrol have been proven to induce the decrease of the intracellular ATP pool of *E. coli*, indicating the destruction occurred on the cytoplasmic membrane (Helander et al. 1998). An increased the expression of chaperone proteins, and overexpression of genes were observed for *Salmonella* Thompson cell in presence of thymol, which induced the destabilization of transcriptional control, mRNA levels, translational control, protein levels, and metabolic fluxes (Di Pasqua et al. 2013).

Applications

Due to their environmentally friendly and safe properties, EOs have been approved by the FDA as GRAS substances. Recent decades, EOs have been gaining increasing interest for the elimination of microorganisms on F&V (Kang & Song 2018; Bhargava et al. 2015). Numerous EOs, such as clove, oregano, thyme, and cinnamon have shown broad-spectrum antimicrobial activity, as shown in Table 2. The populations of *L. monocytogenes*, *S. Typhimurium*, and *E. coli* O157:H7 on lettuce were reduced by 3.03–3.57 log CFU/g after oregano oil treatment (0.05–0.1, 1 min) (Bhargava et al. 2015). The populations of *S. Newport* on romaine lettuce, iceberg lettuce, mature spinach, and baby spinach were reduced by up to 4.8, 4.8, 4.9, and 4.7 log CFU/g, respectively, after oregano oil treatment (Moore-Neibel et al. 2013). EOs also have low resistance-inducing effects in microorganisms (de Oliveira et al. 2017). Furthermore, it has been reported that EOs do not affect the firmness, color, weight loss, and sensory characteristics of products (Donsì et al. 2015; de Oliveira et al. 2017).

Factors influencing efficacy

The antimicrobial efficiency of EOs is closely related to chemical class and concentration, holding time, microorganism and F&V species, and other variables. After treatment with EOs from cinnamon-leaf, cinnamon-bark, and oregano at concentration of 0.05%, the reductions of *L. monocytogenes* inoculated on red mustard leaves were 0.62, 0.55, and 0.78 log CFU/g, respectively (Kang & Song 2018). In general, the microbial reduction increases with increasing EOs concentration and holding time. The reductions of *E. coli* O157:H7 on red mustard leaves were increased from 0.53 to 0.81 log CFU/g, as the cinnamon leaf oil concentration increased from 0.02 to 0.05% (Kang & Song 2018). The reduction in the population of *S. Newport* on romaine lettuce treated with 0.3% oregano oil for 2 min was 2.8 log higher than those treated for 1 min (Moore-Neibel et al. 2013). Gram-positive bacteria were more sensitive to EOs than Gram-negative bacteria, since the lipopolysaccharide in the outer membrane of Gram-negative bacteria could act as a diffusion barrier to phenolic compounds (Maisanaba et al. 2017). The *L. monocytogenes*, *S. Typhimurium*, and *E. coli*

O157:H7 populations on lettuce were reduced by 3.44, 2.31, and 3.05 log CFU/g, respectively, after being dipped in 0.05% oregano oil for 1 min (Bhargava et al. 2015). The reductions of *L. monocytogenes*, *S. Typhimurium*, and *E. coli* O157:H7 on lettuce treated by 10% clove extract for 10 min were >2, 3.68, and 4.04 log CFU/g, respectively (Kim et al. 2011).

Limitations

The practical application of EOs on food is limited due to their hydrophobic, sensory, volatile and unstable nature (Kang & Song 2018). Currently, the emulsification and microencapsulation techniques have usually been applied to enhance the solubility, stability and antimicrobial efficiency of EOs (Kwon, Chang, & Han 2017; Donsì et al. 2015; de Oliveira et al. 2017; Severino et al. 2014). Meanwhile, according to many studies, a higher concentration of EOs is needed in vivo applications to obtain the same microbial reduction in vitro test, but the sensory (taste or flavor) of produce can be severely altered (Donsì et al. 2015; Ochoa-Velasco et al. 2018). Moreover, the composition and efficacy of EOs are variable in geographic origin, agricultural techniques, harvest season, and extraction methods, etc. (Patrignani et al. 2015). Additionally, studies on the toxicity, safety and interaction of EOs in the human gut, as well as the antibacterial activity of minor components are still scarce and require further investigation (Prakash et al. 2018).

High-pressure carbon dioxide

High-pressure carbon dioxide (HPCD) is a relatively novel pasteurization technique. Foods are subject to sub- or super-critical (i.e., pressurized) CO₂ at relatively low temperature (20–50 °C) under moderate pressure (below 50 MPa) for 5–30 min (Ferrentino & Spilimbergo 2011). CO₂ is an inert, nontoxic, readily available, and inexpensive gas, therefore HPCD technology has been generally recognized as a safe (GRAS) and an environmentally friendly technique to sterilize foods (Ferrentino & Spilimbergo 2011).

Disinfection mechanisms

HPCD has been described as an effective method to inactivate microorganisms, and the related mechanisms are shown in Figure 5. The inactivation mechanism is primarily due to CO₂ solubilization in the external reaction medium where it can modify the cell membrane, increases the membrane permeability, decreases the intracellular pH and inhibits enzymes, as well as disorder the intracellular electrolyte balance and cellular metabolism (Garcia-Gonzalez et al. 2007).

Applications

Application of HPCD to F&V microbial inactivation has been reported. For instance, pears treated with HPCD at 10 MPa and 55 °C for 10 min resulted in up to 5 logs reductions of *S. cerevisiae* (Valverde, Marín-Iniesta, & Calvo

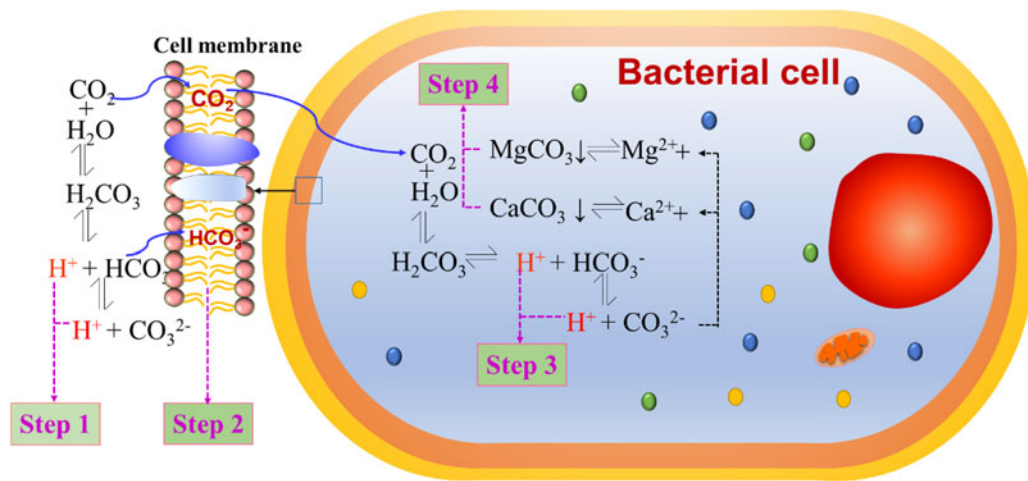


Figure 5. A schematic diagram of the lethal action of HPCD on bacteria (adapted from Garcia-Gonzalez et al. 2007).

2010). The application of HPCD at 12 MPa and 35 °C for 3 min inactivated yeasts & molds and total coliform bacteria on fresh-cut carrot to undetectable levels (about 5 log CFU/g reductions) (Spilimbergo et al. 2013). A 7 log CFU/g reduction of *E. coli* was obtained on fresh-cut carrot treated by HPCD at 12 MPa and 26 °C for 10 min (Ferrentino et al. 2014), and 4 log CFU/g reduction of *S. Typhimurium* on fresh-cut coconut was achieved by HPCD treatment at 12 MPa and 40 °C for 20 min (Ferrentino, Komes, & Spilimbergo 2015). HPCD treatment had no detrimental effects on flavor and nutrients (Zhou et al. 2015). The polyphenolics, carotenoids and antioxidant capacity of fresh-cut carrot were not changed after HPCD treatment (Spilimbergo et al. 2013). HPCD also has potential to inactivate enzymes. The residual activities of peroxidase, polyphenol oxidase, and pectin methylesterase of carrot were decreased to 75.8, 90.9, and 52.8%, respectively, after HPCD treatment at 5 MPa and 20 °C for 15 min (Bi et al. 2011). Therefore, HPCD can effectively destroy microorganisms without affecting the nutritional attributes of F&V.

Factors influencing efficacy

The efficiency of HPCD treatment is highly influenced by temperature, pressure, exposure time, microorganism and F&V species (Garcia-Gonzalez et al. 2007). Pressure and temperature are the principle factors for microbial inactivation of HPCD treatment. Density and solvation power of CO₂ are increased with pressure, the diffusivity, and cell membrane penetration of CO₂ are facilitated with increasing temperature (Ferrentino et al. 2014). Spilimbergo et al. (2013) showed that when the temperature was raised from 22 to 45 °C at 12 MPa for 15 min, the reduction of mesophilic microorganisms on fresh-cut carrot was increased from 2.5 to 3.5 log CFU/g. Yeasts and molds on fresh-cut carrot decreased to undetectable levels (about 5 log reduction) at 22 °C by HPCD treatment in 5 and 10 min at 12 and 8 MPa, respectively (Spilimbergo et al. 2013). Similarly, Ferrentino et al. (2014) observed that increasing pressure and temperature induced a significant reduction of the treatment time to achieve the same level of microbial reduction.

Besides, Bi et al. (2011) showed that HPCD treatment (5 MPa, 20 °C, 20 min) reduced aerobic bacteria and yeasts & molds on fresh-cut carrot slices by 1.86 and 1.25 log CFU/g, respectively.

Limitations

HPCD acidifies the medium and may alter the quality of products (Valverde, Marín-Iniesta, & Calvo 2010). For instance, the ascorbic acid content and texture of fresh-cut carrot decreased up to 40 and 90%, respectively, as compared to the untreated samples (Spilimbergo et al. 2013). HPCD has failed to control the microbial populations during produce storage. A fraction of microbes is able to maintain their metabolic activity, and regrowth can occur after HPCD treatment (Garcia-Gonzalez et al. 2007). Faster growth rates of both mesophilic microorganisms and lactic acid bacteria were observed on HPCD treated carrot compared to untreated controls during a 2 weeks storage (Spilimbergo et al. 2013). There was an 8 log CFU/g inactivation of *E. coli* after HPCD treatment for 10 min, but flow cytometry analysis showed that the inactivation of intact cells was only 2.0–2.5 log CFU/g (Tamburini et al. 2014). Additionally, the high costs of the equipment and operation are obstacles to the application of HPCD. Current research has focused on pure microbial cultures occurring in liquid foods (e.g. fruit juices, wine, and milk) (Ferrentino & Spilimbergo 2011). Therefore, more studies on the effects of the HPCD on sterilization and quality of F&V are required.

Organic acids

Organic acids have acceptable daily intake and do not have a significant adverse impact on human health (Park et al. 2011). Organic acids can rapidly inactivate a broad-spectrum of bacteria within a wide temperature range (Sagong et al. 2011). Some of the organic acids, that is, lactic acid, citric acid, acetic acid, and tartaric acid, have commonly been utilized as food additives and preservatives to extend the shelf life of food for centuries. In recent years, attention has turned to the use of organic acid as sanitizers for F&V.

Disinfection mechanisms

Organic acid kills microorganisms by reducing the environmental and cellular pH of microbial cells, which disturbs membrane transport and permeability (Rico et al. 2007). In detail, organic acids exist in the undissociated and dissociated forms. The undissociated molecules can penetrate the microbial membrane and enter microbial cells. And then the undissociated molecule encounters a near-neutral pH environment and dissociates into the charged anions and protons, resulting in acidification of cell interior, elimination of the electrochemical proton gradient across the cell membrane, inactivation of intracellular enzymes, and inhibition of metabolic reactions of cells (Zhou, Xu, & Liu 2010). Acid can induce oxidative stress to bacteria. Mols and Abee (2011) found acetic acid, lactic acid and sorbic acid exposure promoted the formation of reactive oxygen species, which might induce the cell death of *B. subtilis*. Ter Beek et al. (2008) also observed that sorbic acid-stressed *B. subtilis* cells experienced the strong depression of the CcpA, CodY, and Fur regulon and the induction of tricarboxylic acid cycle genes, and the upregulation of fatty acid biosynthesis genes. In addition, the protein profiles of *E. coli* exposure to levulinic acid were changed, and the fewer and fainter bands present after treatment, indicating the destruction of cellular proteins (Zhao et al. 2019).

Applications

Organic acids have been used as sanitizers for various F&V. Citric acid and acetic acid (0.3–1.4%) treatment reduced *S. aureus* on tabbouleh salad by 2.7–3.9 log CFU/g after storage for 7 days at 21 °C (Al-Rousan et al. 2018). Pathogens (*E. coli* O157:H7, *S. Typhimurium*, and *L. monocytogenes*) on apple treated with 2% lactic acid and citric acid for 10 min were reduced by more than 2.56 and 3.42 log CFU/fruit (Park et al. 2011). The color of apples and lettuce subjected to organic acid treatment was not significantly changed during storage (Park et al. 2011). Some salt or derivatives of organic acids have been reported with high germicidal activity. For example, Chen, Zhang, and Zhong (2019) and Chen et al. (2019a) washed cherry tomatoes using acidified sodium benzoate (0.3%, pH 2.0) for 3 min. They found that the *L. monocytogenes*, *E. coli* O157:H7, and *S. enterica* were reduced by 5.20, 5.96, and 4.84 log CFU/g, respectively, without marked modification of product quality. Hence, organic acids can decrease the microbial loads on F&V, and maintain the products' quality (Sagong et al. 2011).

Factors influencing efficacy

The disinfection efficacy of organic acid is related to the acid types and concentration, treatment time, microorganism and F&V species. When 2% propionic-, acetic-, lactic-, malic-, and citric-acid were applied on apples, and the holding time increased from 0.5 to 10 min, consequently, the corresponding reductions of *E. coli* O157:H7 were increased from 1.77 to 2.75, 1.12 to 2.69, 1.64 to >3.42, 1.33 to >3.42, and 1.99 to >3.42 log CFU/fruit (Park et al. 2011). Similar trends were also found by Sagong et al. (2011) where the

reductions of *S. Typhimurium* on lettuce increased from 1.62 to 4.50 log CFU/g, when malic acid concentrations increased from 0.3% to 2.0% for 30 min. They also observed that the *S. Typhimurium* reduction increased from 2.03 to 4.50 log CFU/g, when the exposure time of 2% propionic acid increased from 10 to 30 min. After 10 min of treatment, the *E. coli* O157:H7, *S. Typhimurium*, and *L. monocytogenes* were reduced by 1.04, 2.84, and 2.50 log CFU/fruit on apples, and by 1.52, 1.22, and 1.18 log CFU/g on lettuce, respectively (Park et al. 2011).

Limitations

Organic acids have a relatively low antimicrobial efficacy, low concentration or short duration time treatment was not successful in controlling microorganisms. Bermúdez-Aguirre, Wemlinger et al. (2013), and Bermúdez-Aguirre & Barbosa-Cánovas (2013) showed that the citric acid treatment (0.5–1.5%, 15 min) failed to reduce the *E. coli* ATCC 11775 on carrots, lettuce, and tomatoes. The reductions of *E. coli* O157:H7, *S. Typhimurium*, and *L. monocytogenes* on lettuce were 1.55–1.76, 1.30–1.74, and 1.35–1.46 log CFU/g, respectively, after 5 min of treatment with 2% malic acid, lactic acid and citric acid (Sagong et al. 2011). The aerobic bacteria in broccoli sprouts was reduced 0.77 log CFU/g after lactic acid treatment (2%, v/v) for 2 min (Chen, Zhang, & Zhong 2019; Chen et al. 2019b). A prolong treatment time (20 min) caused significant quality changes in color and general appearance of the product (Sagong et al. 2011). Moreover, organic acids used with high concentration also had disadvantages of altering flavor and odor of F&V and might have a corrosive effect to human tissue and treatment equipment (Khan et al. 2017).

In general, the emerging chemical disinfection technologies can effectively improve the safety of F&V under proper conditions. However, some problems caused by antimicrobial washes should be taken into consideration. On the one hand, large volumes of water are needed for sanitizer washing treatment, which can cause pathogen cross-contamination and environment pollution. The residual moisture after the washing treatment also creates an environment that promotes mold growth (Alwi & Ali 2014; Gil et al. 2009). Increasing sanitizer concentration or extending exposure time could increase the reductions of microbe populations, but the quality deterioration should not be ignored. Therefore, treatment parameters should be further optimized to achieve a significant reduction in microorganism populations and minimize the impact on the quality of F&V.

Physical disinfection technologies

Recently, interest in developing non-thermal physical technology to reduce microbial loads on F&V has emerged, since thermal treatments induce undesirable physiological deterioration and biochemical changes, such as color degradation, tissues softening, and nutrients loss. Heating approaches are not suitable for the disinfection of fresh F&V due to its fragile nature (Rico et al. 2007; Zhang et al.

Table 3. Radiation disinfection technologies for fruits and vegetables.

Method	Material	Microorganisms	Treatment conditions	Microbial reduction	Reference
UV-light	Tomato	<i>Salmonella</i> spp., <i>Staphylococcus aureus</i> , and <i>L. monocytogenes</i>	Dose of 5 kJ/m ²	3.08, 3.13, and 2.59 log CFU/g, respectively.	Sommers, Sites, and Musgrove 2010
	Pepper	<i>Salmonella</i> spp., <i>Staphylococcus aureus</i> , and <i>L. monocytogenes</i>	Dose of 5 kJ/m ²	3.02, 3.09, and 3.11 log CFU/g, respectively.	Sommers, Sites, and Musgrove 2010
	Apple	Total mesophilic bacteria, yeasts	Dose of 1.2–24 kJ/m ²	1–2 log CFU/g.	Manzocco et al. 2011
	Pear	<i>Cronobacter sakazakii</i>	Doses of 2.5–10 kJ/m ²	1.6–2.3 log CFU/g	Santo et al. 2016
	Apricot	<i>E. coli</i> O157:H7 and <i>S.</i> Typhimurium	Dose of 13.26 kJ/m ²	2.2 and 2.5 log CFU/fruit, respectively.	Yun et al. 2013
	Garlic	Total aerobic bacteria	Dose of 2 kJ/m ²	1.04 and 0.62 log CFU/g after 30 and 45 days storage at 0 °C, respectively.	Park and Kim 2015
Pulsed light	Mellon	<i>Cronobacter sakazakii</i>	Doses of 7.5–10 kJ/m ²	1.8–2.4 log CFU/g.	Santo et al. 2016
	Apple	<i>L. innocua</i> ATCC 33090 and <i>E.</i> <i>coli</i> ATCC 11229	3 pulses/s, 1.27 J/ cm ² , 100 s	1.70 and 2.25 log CFU/cm ² , respectively.	Gómez et al. 2012
	Spinach	<i>E. coli</i> O157:H7	3 pulses/s, 1.27 J/ cm ² , 1–30 s	1.7–3.4 log CFU/g.	Mukhopadhyay et al. 2019
	Spinach	<i>L. innocua</i> and <i>E. coli</i>	0.3 ms/pulse, 4 kJ/m ² , 2 pulses	1.85 and 1.72 log CFU/g, respectively.	Agüero et al. 2016
	Tomato	Aerobic mesophilic bacteria, mold and yeast	2.68 J/cm ² , 2 pulses	2.9 and 1.9 log CFU/g, respectively	Aguiló-Aguayo et al. 2013
	Mushroom	<i>L. innocua</i>	0.3 ms/pulse, 9 s	2.17 log CFU/g.	Ramos-Villarroel, Martín- Belloso, and Soliva- Fortuny 2015
Ionizing radiation	Baby spinach	<i>Salmonella</i> spp. and <i>Listeria</i> spp.	e-beam, 0.7 kGy	5.0 log CFU/g.	Gomes et al. 2011
	Tomato	Bacteria, yeast and molds	X-ray, 1.5 kGy	Under the detectable limit.	Mahmoud 2010
	Spinach	Bacteria, yeast and mold, <i>E.</i> <i>coli</i> O157: H7, <i>L.</i> <i>monocytogenes</i> , <i>S. enterica</i> , and <i>Shigella flexner</i>	X-ray, 2.0 kGy	Under the detectable limit.	Mahmoud, Bachman, and Linton 2010
	Mushroom	Mesophilic bacteria	γ-ray, 1.0–2.0 kGy	1.5–2.5 log CFU/g when stored at 4 °C for 20 d.	Jiang et al. 2010
	Cucumber	<i>S. Typhimurium</i> , <i>E. coli</i> , <i>Staphylococcus aureus</i> and <i>L. ivanovii</i>	γ-ray, 1 kGy	3.12, 1.94, 2.11, and 2.97 log CFU/g, respectively.	Lee et al. 2006
	Spinach	<i>S. Typhimurium</i> , <i>E. coli</i> , <i>Staphylococcus aureus</i> and <i>L. ivanovii</i>	γ-ray, 1 kGy	2.83, 2.53, 2.77, and 2.36 log CFU/g, respectively	Lee et al. 2006

2013). As a consequence, several novel non-thermal physical technologies, including UV-light, PL, ionizing radiation, high pressure, CP, and high-intensity ultrasound, have emerged as attractive alternatives to conventional thermal processing.

Ultraviolet light

Ultraviolet (UV) light is a portion of the electromagnetic spectrum with the wavelength in the range of 100–400 nm (Keklik, Krishnamurthy, & Demirci 2012). UV-light can be divided into UV-A (315–400 nm), UV-B (280–315 nm), UV-C (200–280 nm), and vacuum-UV (100–200 nm) (Keklik, Krishnamurthy, & Demirci 2012). UV light has advantages of having a broad-spectrum bactericidal effect, cost-efficiency, ease of use and being environmentally friendly (Gayán et al. 2014). UV light has been proposed as a potential alternative to chemical and thermal disinfection methods.

Disinfection mechanisms

Genetic damage caused by UV light is primarily responsible for microbial inactivation (Gayán et al. 2014). The UV-C

has the highest germicidal activity, since it can be absorbed by DNA, especially at around 254 nm where the maximum absorption is achieved (Sinha & Häder 2002). The UV-C absorption induces the photochemical changes of microbial DNA by forming thymine dimers (cyclobutane pyrimidine dimers and pyrimidine 6–4 pyrimidone). The thymine dimers can interrupt DNA replication and transcription, and eventually inactivate the microorganism (Keklik, Krishnamurthy, & Demirci 2012; Gayán et al. 2014).

Applications

UV light has been widely applied on surface disinfection of F&V, as shown in Table 3. The population of *S. Typhimurium* in coconut water was decreased by 3.8, 5.2, and 6 log CFU/mL after treatment with UV-C light for 3.5, 7, and 10.5 min, respectively (Beristáin-Bauza et al. 2018). Mukhopadhyay et al. (2014) used UV-C at doses of 0.6–6.0 kJ/m² to eliminate pathogens on grape tomato and found that the populations of *E. coli* O157:H7 and *Salmonella* were reduced by 2.3–3.5 and 2.15–3.1 log CFU/fruit, respectively. The firmness and color of grape tomato during storage were not affected by the UV-C treatment. UV light treatment at 1.6 mW/cm² for 60 min reduced the

E. coli on tomatoes by 2.8 logs (Bermúdez-Aguirre, Wemlinger et al. 2013; Bermúdez-Aguirre, Martínez-Niño et al. 2013). Moreover, UV treatment mitigated the softening, respiration and decay of fresh-cut pepper during its storage (Rodoni et al. 2015). The weight loss and browning of UV light treated apples was reduced during storage (Chen et al. 2016). The total phenolics (3%) and quercetin (25%) contents in garlic increased after being treated with UV and stored at 0°C for 15 days (Park & Kim 2015). Therefore, UV light can significantly reduce microbial loads and extend the shelf life of F&V.

Factors influencing efficacy

The effectiveness of UV light against microorganisms depends on many factors, such as wavelength and dose of the light, food property and microbial related features (Gayán et al. 2014; Mukhopadhyay et al. 2014). In general, a higher lethal efficacy is achieved at wavelengths closer to DNA's absorption peak or higher applied doses (Gayán et al. 2014). When grape tomatoes were exposed to UV-C light at dose of 1.2, 2.4, and 4.8 kJ/m², *E. coli* O157:H7 was reduced by 2.70, 3.05, and 3.44 log CFU/fruit, respectively (Mukhopadhyay et al. 2014). Significant differences in sensitivity to UV-light among microorganisms were previously observed, which depended on microbial types, structure, repair system, and pyrimidine amounts in DNA (Gayán et al. 2014). Ochoa-Velasco et al. (2018) showed that the reductions of 7.54–7.79, 5.96–7.32, and 3.12–4.46 log CFU/mL were obtained for *L. rhamnosus*, *S. Typhimurium*, and *S. cerevisiae* in coconut water after UV-C light treatment for 10 min, respectively. Usually, the efficacy of UV light was higher for F&V with smoother surfaces. *E. coli* O157:H7 was reduced by 3.05 and 2.59 log CFU/fruit on the surface and stem scar of grape tomato after treatment with UV-C light at a dose of 2.4 kJ/m², respectively (Mukhopadhyay et al. 2014). Apple, pear, and strawberry inoculated *E. coli* O157:H7 were treated with UV-C light at 0.92 kJ/m², and the microorganisms were reduced by 2.9, 2.1, and 0.9 log CFU/g, respectively (Adhikari et al. 2015). Whereas, *L. monocytogenes* was just reduced by 1.4, 1.2, and 0.7 log CFU/g at a higher dose of 3.65 kJ/m² for the corresponding fruits, respectively (Adhikari et al. 2015).

Limitations

The major limitations of the application of UV light are the low penetration of UV light and the shade effect from complex surface properties of foods (Gayán et al. 2014). A slight reduction (< 1 log CFU/g) in microbial populations was observed on fresh-cut peppers (Rodoni et al. 2015) and peeled garlic (Park & Kim 2015). Meanwhile, quality deterioration may occur during long exposure to UV light to achieve substantial microbial inactivation. For instance, more than 90 min of UV irradiation was needed to reduce the microbe on figs to satisfactory levels (Isman & Biyik 2009). UV-C light (>1.2 kJ/m²) treatment destroyed the surface cells, promoted dehydration and caused off-flavor of fresh-cut apple (Manzocco et al. 2011). It has been reported

that UV light can alter some “light sensitive” compounds of the sample, such as vitamins, carotenes, folic acid, tryptophan, and unsaturated fatty acid (Koutchma, Forney, & Moraru 2009). Moreover, some microorganisms can repair DNA damage induced by UV irradiation, especially at low dosage treatments which create a stress process, then the microbe can gradually recover during storage (Ochoa-Velasco et al. 2018).

Pulsed light

Pulsed light (PL) refers to short and intense pulses of broad spectrum radiation (200–1100 nm) ranging from UV to near infrared (Mahendran et al. 2019). It is generated by accumulating the electrical energy in a capacitor and releasing it over an inert gas (such as xenon) in the lamp within a short time, which greatly magnifies the power (Gómez-López et al. 2007). PL is also known as pulsed UV-light, because approximately 50% of energy is in the UV-C range (200–280 nm) (Gómez-López et al. 2007). PL has advantages of high efficacy as compared to the continuous irradiation form, due to its instantaneous delivery of more intense energy (Rowan 2019). The antimicrobial ability of PL is 4–6 folds higher than that of continuous UV light (Keklik, Krishnamurthy, & Demirci 2012; Pataro et al. 2015).

Disinfection mechanisms

The UV-C light is considered as the most important for microbial inactivation (Gómez-López et al. 2007). The antimicrobial effect of PL has been mainly attributed to photochemical damage to DNA caused by UV light, which induces the formation of pyrimidine dimers and inhibits the formation of new DNA chains during cell replication (Rowan, Valdramidis, & Gomez-Lopez 2015). Meanwhile, the disintegration of microbial cells could be induced by the photothermal and photophysical effect, which was partially associated with the temporary overheating and constant disturbance caused by the high-energy pulses (Keklik, Krishnamurthy, & Demirci 2012). An immediate shutdown of metabolic activity, inactivation of intracellular esterase or membrane disruption induced by PL have been demonstrated as an additional inactivation mechanism (Krishnamurthy et al. 2010).

Applications

PL has been approved by the FDA for food treatment with maximum fluence of 12 J/cm² (FDA 1996). Lately, PL has attracted increasing attention as a promising tool for food-surface disinfection. There are commercial PL systems for batch or continuous treatments for high throughput production, Figure 6 shows an equipment developed by Xenon Corporation. PL has been used as a surface disinfection method on a great variety of F&V, such as apple (Gómez et al. 2012), spinach (Mukhopadhyay et al. 2019), tomato (Aguiló-Aguayo et al. 2013), and mushroom (Ramos-Villarroel, Martín-Belloso, & Soliva-Fortuny 2015), etc., as shown in Table 3. PL treatment (59.2–72 J/cm²) reduced the



Figure 6. Pulsed light system for continuous conveyor surface sanitization (Xenon Corporation).

E. coli O157:H7 and *Salmonella* on raspberries by 3.9 and 3.4 log CFU/g, respectively (Bialka & Demirci 2008). The reductions of *E. coli* O157:H7 on blueberry were greater than 5.8 log CFU/g after PL treatment (1.27 J/cm^2) (Huang & Chen 2014). The shelf life of fresh-cut cantaloupes treated by PL (7.8 J/cm^2) was extended by 8 days at 4°C compared to the sample with no treatment (control) (Koh et al. 2016). Valdivia-Nájjar, Martín-Belloso, and Soliva-Fortuny (2018) observed that PL maintained the fresh-like quality of fresh-cut tomato for 10 days. In addition, PL has significant potential for enhancing the accumulation of bioactive components. The lycopene, total carotenoid, phenolic compounds and antioxidant activity of tomatoes treated by PL increased up to 6.2, 2.5, 1.3, and 1.5 times, respectively when compared with those of untreated samples (Pataro et al. 2015). Thus, PL can effectively improve microbial safety, maintain quality and extend shelf life of F&V.

Factors influencing efficacy

The effectiveness of PL against microorganisms depends on several factors, such as pulse fluence of PL, microorganism species, and food properties. In general, a higher pulse fluence lowers the microbial survival rate. The reductions of *E. coli* O157:H7 populations on blueberry calyx were increased from 2.1 to 4.2 log CFU/g when PL fluence increased from 5.0 to 56.1 J/cm^2 (Huang & Chen 2014). However, there was no significant differences on *E. coli* O157: H7 and naturally occurring aerobes inactivation when the fluence was greater than 5.25 and 7.80 J/cm^2 , respectively (Mukhopadhyay et al. 2019; Llano et al. 2016). This might be due to the plateauing of cell death at high fluence (Koh et al. 2016). The sensitivity to PL varies in different microorganism species. In general, Gram-positive bacteria are believed to be more resistant to PL than Gram-negative bacteria, fungi are more resistant than bacteria, and bacterial spores are more resistant than their vegetative cells (Gómez-López et al. 2007). The reductions of *E. coli* O157:H7 and *Salmonella* on strawberries treated by PL at the same condition were 3.3 and 4.3

log CFU/g, respectively (Bialka & Demirci 2008). Tomatoes processed by PL (5.36 J/cm^2) showed reductions of 2.9 and 1.9 log CFU/g of aerobic mesophilic bacteria and mold & yeast, respectively (Aguiló-Aguayo et al. 2013). In addition, food properties vary in different parts of a fruit and species. The loads of *L. innocua* on tomato slices and spinach leaves treated by PL at 8 J/cm^2 were reduced by 0.90 and 1.85 log CFU/g, respectively (Valdivia-Nájjar, Martín-Belloso, & Soliva-Fortuny 2018; Agüero et al. 2016). The amounts of *E. coli* O157:H7 on blueberry calyx and skin treated with PL at 1.27 J/cm^2 were reduced by 3.0 and >5.8 log CFU/g, respectively (Huang & Chen 2014). The microbiological stability was affected by food cut type, the inactivation of total plate count and yeast & mold for sphere (49.29–50.35%) were higher than those of cuboid (25.41–40.78%) and triangular prism (21.36–45.91%) (Koh et al. 2016). In addition, the germicidal efficiency of PL was greatly interfered by the shadow effect (Hwang, Cheigh, & Chung 2017).

Limitations

PL is a non-thermal technology only under short durations or low fluence, whereas prolonging exposure time would significantly increase the sample temperature. The product quality depends on the application of PL at proper fluences (Ignat et al. 2014). Though the high PL fluence allows greater microbial reductions, it promotes temperature increase and product quality deterioration (Gómez et al. 2012; Huang and Chen 2014). Bialka and Demirci (2008) reported that raspberry temperature reached up to 80°C at a fluence level of 72 J/cm^2 . It was observed that after being treated by PL for 60 s, the temperature of blueberries surface increased to 64.8°C , which induced a severe discoloration and burnt appearance to the blueberry (Huang & Chen 2014). In addition, PL treatment also induced softening and increased weight loss and wrinkles of tomatoes during storage (Aguiló-Aguayo et al. 2013). Ignat et al. (2014) verified that the cell damage, browning and cooked flavor occurred in fresh-cut apple after being treated by PL (157.5 kJ/m^2). Hence, to achieve industrial scale use of PL technology for food disinfection, more efforts are required to optimize the operating conditions, develop novel equipment to minimize shadow effect, and use cooling systems to reduce rising temperatures.

Ionizing radiation

Ionizing irradiation has been proposed as an alternative technique for decontaminating F&V. It has been approved by the US FDA to eliminate pathogens and undesirable spoilage microorganisms of foods (FDA 2016). There are three sources of radiation permitted to use on foods, including gamma rays (γ -ray) from cobalt 60 or cesium 137, electrons generated from machine sources (e-beam), and X-rays created when high energy electrons strike a metal plate (Yang et al. 2013). Ionizing irradiation has advantages of proven efficacy, being environmentally-friendly and time effective (Moosekian et al. 2012).

Disinfection mechanisms

Ionizing irradiation has great potential for inhibiting a variety of foodborne pathogens and spoilage microorganisms (Yang et al. 2013; Lung et al. 2015). Ionizing irradiation has significant effects on all components of organisms. It can cause the breakage of chemical and biomolecular bonds, resulting in the strand-break in DNA, and denaturation of enzymes and membrane proteins (Moosekian et al. 2012; Severino et al. 2014). Meanwhile, water molecules in microorganisms can be ionized, and generate free radicals (i.e. OH^\bullet , H^\bullet , and HO_2^\bullet), which can break chemical bonds and alter various molecules, leading to the destruction or deactivation of bacterial components (Lung et al. 2015).

Applications

A large number of studies have used ionizing irradiation to sterilize and prolong the shelf-life of F&V, as shown in Table 3. The γ -rays at dose of 2.0 kGy reduced the mesophilic, psychophilic, pseudomonad bacteria, and yeasts & molds on shiitake mushroom to below detection limits (Jiang et al. 2010). The populations of mesophilic bacteria, psychrotrophic bacteria, and yeast and mold counts on Roma tomatoes treated with X-ray (1.5 kGy) were reduced from initial values of 5.4, 4.7, and 4.4 log CFU/g to non-detectable limit (<1.0 log CFU/g). Meanwhile, the treatment prolonged the shelf life of tomatoes from 6 days to 20 days at 22 °C (Mahmoud 2010). The e-beam treatment reduced the *Salmonella* Poona on cucumber slices by 4.96 log CFU/g, and the quality (color, pH, and water activity) of fruits did not change during the 3 days of refrigerated storage (Joshi et al. 2018). The γ -ray irradiation (2 kGy) extended the shelf life for about 1 week, decreased the chilling injuries and water loss, and maintained the vitamin C and chlorophyll of cucumbers (Khalili et al. 2017). Additionally, γ -ray irradiation treatment has been found to prevent the browning of mushroom, and improve the product's appearance (Jiang et al. 2010). Therefore, ionizing irradiation shows considerably beneficial effects in reducing the microbial population, extending the shelf life, and maintaining the quality of F&V.

Factors influencing efficacy

The efficacy of ionizing irradiation on microorganisms is closely related to the irradiation source and dose, and microorganism and F&V species. The γ -ray has excellent penetration capacity (30–40 cm) and higher energy-efficiency than e-beam or X-ray (Niemira 2017). But the radiation source of γ -ray cannot be “turned off” readily, whereas e-beams can be completely deactivated or activated depending on the requirements (Niemira 2017), which offers e-beam a commercial advantage over γ -ray. When irradiation is applied at higher doses, sterilization efficacy can be enhanced. When the aerobic mesophilic organisms on paprika was treated with γ -ray at doses of 1, 5, and 10 kGy were decreased by 1.76, 2.13, and 3.93 log CFU/g, respectively (Molnár et al. 2018). Normally, Gram-positive bacteria show stronger resistance to γ -ray than Gram-negative

bacteria, whereas prokaryotic microorganisms are more resistant to γ -ray than eukaryotic microorganisms (Khan et al. 2017). Approximately 4.2, 2.3, 3.7, and 3.6 log CFU/fruit reductions of *E. coli* O157:H7, *L. monocytogenes*, *S. enterica*, and *S. flexneri* on tomato were achieved by X-ray treatment at 0.75 kGy, respectively (Mahmoud 2010). More than 5 log CFU/g reductions of pathogens (*E. coli* O157: H7, *L. monocytogenes*, *S. enterica* and *S. flexneri*) on spinach were achieved with 2.0 kGy X-ray exposure (Mahmoud, Bachman, & Linton 2010).

Limitations

For certain treatments, in order to achieve substantial reductions of microorganisms, a high-dose irradiation is required. However, quality deterioration of F&V occurred when treated by irradiation at high dose (>1 kGy) (Olaimat & Holley 2012). The firmness of irradiated (1.92 kGy) cucumber slices was 50% lower than that of untreated samples (control) (Joshi et al. 2018). The γ -ray irradiation decreased the total carotenoid and tocopherol contents of paprika by 54.8–62.7% and 23.1–39.8%, respectively, meanwhile, the volatile aroma and color of the product were also destroyed (Molnár et al. 2018). Though FDA determined that food irradiation can be used without posing human health risks, but the long-term consumption of irradiated produce remains a cause of concern to the general public (Moosekian et al. 2012). Additionally, ionizing irradiation requires high initial investment and maintenance costs (Goodburn & Wallace 2013).

High hydrostatic pressure

High hydrostatic pressure (HHP) involves treating foodstuff at high pressure of 100–800 MPa conveyed by liquids (typically water) for an appropriate period (Wang et al. 2016). HHP is recognized as one of the most promising pasteurization technologies, as it has high efficient sterilization performance with zero or a minimal negative impact on the sensory and nutritional properties of F&V (Rendueles et al. 2011).

Disinfection mechanisms

HHP treatment can effectively inactivate bacterial vegetative cells, yeasts, and molds via multiple mechanisms on cells, including inhibition of DNA synthesis, induction of protein denaturation and enzyme inactivation, and damage to cell membrane (Lou et al. 2015; Huang et al. 2017). The cytoplasmic membrane is the key target for HHP inactivation. HHP can alter membrane structure via increasing the order of lipid molecules on membrane, that causes a decrease in cell membrane fluidity, and followed by an increase in thickness. HHP induces the detachment and inactivation of membrane proteins. Eventually, resulting in the modification of cell permeability and functionality (Knorr et al. 2011). HHP also promotes a cytoplasm acidification and oxidative stress of cell, and alters the metabolism and energy gene expression program (Fernandes et al. 2004).

Table 4. Non-radiation physical disinfection technologies for fruits and vegetables.

Method	Material	Microorganisms	Treatment conditions	Microbial reduction	Reference
High hydrostatic pressure	Green bean	<i>L. innocua</i>	300 MPa, 5 min	1.7 Log CFU/g.	Donsì et al. 2015
	Lotus root	Total plate counts	500 MPa, 5 min	4.36 log CFU/g after 45 days of storage.	Dong et al. 2013
	Pumpkin	Total aerobic bacteria, yeast and mold	350 MPa, 0.5–30 min	1.06–3.55 and 0.22–2.33 log CFU/g, respectively.	Zhou et al. 2014
	Pumpkin	Total aerobic bacteria, yeast and mold	450 MPa, 0.5–5 min	1.08–3.42 and 0.31–2.34 log CFU/g, respectively.	Zhou et al. 2014
	Radish	Total aerobic bacteria, yeast and mold	550 MPa, 5 min	5.57 log CFU/mL and totally inactivated, respectively.	Bao et al. 2016
Cold plasma	Strawberry	<i>E. coli</i> , <i>Salmonella</i> and <i>L. monocytogenes</i>	DBD-ACP: max voltage 120 kV, frequency 50 Hz, 300 s	3.5, 3.8 and 4.2 log, respectively.	Ziuzina et al. 2014
	Blueberry	Yeast and mold	ACP: frequency 47 kHz, power 549 W, 90 s	1.70 log CFU/g after 7 days in 4 °C storage.	Lacombe et al. 2015
	Red chicory	<i>L. monocytogenes</i>	DBD-ACP: max voltage 15 kV, frequency 12.5 kHz, 30 min	2.2 log CFU/cm ² .	Pasquali et al. 2016
	Cabbage	<i>S. Typhimurium</i>	Microwave-powered CP: use N ₂ , 900 W, 10 min.	1.5 log CFU/g.	Lee et al. 2015
	Apple, cantaloupe, lettuce	<i>Salmonella</i>	DBD-ACP: voltage 9 kV, frequency 6 kHz, 1–5 min	>2 log CUF/sample after 1 min, >3 log CUF/sample after 3 and 5 min exposure.	Critzer et al. 2007
	Apple, cantaloupe, lettuce	<i>L. monocytogenes</i>	DBD-ACP: voltage 9 kV, frequency 6 kHz, 1–5 min	>3 and >5 log CUF/sample after 3 and 5 min exposure, respectively.	Critzer et al. 2007
High-intensity ultrasound	Alfalfa	<i>E. coli</i> and <i>S. enteritidis</i>	26 kHz, 200 W, 5 min	1.40 and 1.06 log CFU/g, respectively.	Millan-Sango et al. 2017
	Lettuce	<i>S. enterica</i>	26 kHz, 200 W, 5 min	2.23 log CFU/cm ² .	Millan-Sango et al. 2016
	Cherry tomato	<i>S. enterica</i>	45 kHz, 10–30 min	0.83–1.73 log CFU/cm ² .	São José and Vanetti 2012
	Tomato	<i>E. coli</i> O157:H7	20 kHz, 130–210 W, 5–15 min	2.88–4.22 log CFU/g.	Afari et al. 2016

Applications

HHP sterilization against a number of microorganisms on various F&V has been investigated (Table 4). For instance, as compared to untreated sample, HHP treatment (600 MPa, 5 min) effectively reduced the natural-occurring microorganisms on red bean powder by 7.18 log CFU/g, which was comparable to the values achieved by thermal treatment at 90 °C for 15 min (Lee, Ha et al. 2018). Both *S. enterica* and *E. coli* O157:H7 on green onions were reduced by over 5 log CFU/g after HHP treatment at 450–550 MPa and 20 °C for 2 min (Neetoo et al. 2011). The total plate counts in lotus root treated with HHP (400 MPa, 10 min) were reduced by 5.12 log CFU/g on 45th day's storage (Dong et al. 2013). HHP greatly reduced the microbial counts and maintained the organoleptic (appearance, color, and aroma) and nutritional quality of green onion (Neetoo et al. 2011). HHP is able to inhibit undesirable enzyme activity. For instance, the trypsin inhibitor activity of bean after HHP treatment (600 MPa, 5 min) was significantly reduced by 80.5%, which might help to preserve sensory and nutritional quality of produce (Lee, Ha et al. 2018). Meanwhile, total phenolic, flavonoid, proanthocyanidin, and antioxidant capacity of the treated samples were 38, 12, 26, and 15% higher than those of thermal-treated sample (90 °C, 15 min), respectively (Lee, Ha et al. 2018). Therefore, HHP treatment can effectively inactivate microorganisms and maintain the quality of F&V. The HHP sterilization technology have been commercialized, the commercial scale HHP treatment system is currently available (Figure 7).

**Figure 7.** Industrial high pressure processing equipment (Hiperbaric LLC., 525L capacity, over 3,000 kg/h throughputs).

Factors influencing efficacy

The germicidal efficiency of HHP depends on various parameters, such as pressure level, treatment duration, temperature, microorganism species, and food property. In general, microbial counts decreased with the increase of processing pressure, duration and temperature. For instance, the remaining number of natural microorganisms in lotus root was 1.76 log CFU/g at 500 MPa for 25 min, compared to 1.94 log CFU/g at 600 MPa for 5 min (Dong et al. 2013). Similarly, naturally occurring microbial loads in red bean powder were reduced from 8.23 to 1.83, 1.55, and 1.05 log CFU/g when the product was treated at 400, 500, and 600 MPa for 5 min, respectively (Lee, Ha et al. 2018). The reductions of the *L. lactis* increased from 1.4 to 4.7 log cycles when the pressure was raised from 200 to 400 MPa (Maresca & Ferrari 2017). The reductions of *S. enterica* on

green onion treated with HHP (300 MPa, 2 min) was increased from 3.0 to 4.2 log CFU/g, when the temperature increased from 4 to 40 °C (Neetoo et al. 2011). Structural properties and various components are various among microorganisms and their resistance to HHP differs too. Gram-positive bacteria showed higher resistant to pressure than Gram-negative ones, molds mycelia were more susceptible to HHP than spores, and microorganisms at logarithmic growth were more sensitive to HHP than other phases (Rendueles et al. 2011). After HHP treatment at the same pressure, *L. lactis* and *E. coli* were reduced by 1.4–4.7 and 0.8–3.4 log cycles, respectively (Maresca & Ferrari 2017). Meanwhile, the efficacy of HHP also depends on F&V properties. The *S. enterica* and *E. coli* O157:H7 in un-wetted, wetted and soaked green onions treated with HHP (350 MPa, 20 °C, 2 min) were reduced by 1.2 and 1.4, 3.1 and 3.0, and 3.8 and 3.3 log CFU/g, respectively (Neetoo et al. 2011).

Limitations

There are still several issues that hinder the application of HHP. Significant alteration of the texture and color of some products were observed after HHP treatment. Cell disruption and intense browning occurred on apples after HHP treatment (Dong et al. 2013). The L^* and b^* of green bean treated with HHP decreased by 19 and 23%, respectively (Donsì et al. 2015). The elasticity of radish tubers decreased up to 89% after HHP treatment (Rux et al. 2019). At the same time, high-cost equipment, and non-continuous nature of the process are unfavorable factors for the spread of this technology (Lou et al. 2015; Huang et al. 2017). Furthermore, significant reductions of spores are difficult to achieve due to their resistance to high pressure. Some cells can repair the sub-lethal damage induced by HPP, hence they can proliferate after the injury (Baptista et al. 2015). Hence, HHP treated products need to be stored and transported under refrigerated conditions. Therefore, HHP processing parameters must be optimized in order to assure safety and maintain the quality of products.

Cold plasma

Plasma can be characterized as quasi-neutral particle systems in gaseous mixture of photons, free electrons, positive and negative ions, metastables, atoms, and free radicals (Hertwig, Meneses, & Mathys 2018). Based on temperature, plasma can be classified into thermal or non-thermal plasmas. In the case of thermal plasmas, the temperatures of all the species are the same, which is a thermal equilibrium exists between electrons and other heavier species (Misra & Jo 2017). The non-thermal plasmas are further subdivided into quasi-equilibrium plasma and non-equilibrium plasma (Misra et al. 2016). CP refers to non-equilibrium plasma, which has high potential for inactivating microorganisms on food surface without increasing the temperature markedly (Niemira 2012; Kim, Oh et al. 2017). CP can be generated by means of electric discharge, using corona discharge,

dielectric barrier discharges (DBD), microwaves (MW), radio frequency waves, or gliding arc discharge, under either atmospheric or low pressure (Misra et al. 2016). At present, the DBD and plasma jet are the two commonly explored areas in food research because their simple construction and ease of control (Misra et al. 2016; Hertwig, Meneses, and Mathys 2018).

Disinfection mechanisms

The mechanisms responsible for microbial inactivation of CP could attribute to the lethal action of ions and highly reactive gaseous molecules interacting with biological materials, as shown in Figure 8 (Misra & Jo 2017; Hertwig, Meneses, and Mathys 2018). During CP treatment, reactive oxygen species (ROS) (e.g. H_2O_2 , O_3 , $O_2^{\bullet-}$, HO_2^{\bullet} , 1O_2 , $^{\bullet}OH$, $CO_3^{\bullet-}$, etc.), reactive nitrogen species (RNS) (e.g. $^{\bullet}NO$, $^{\bullet}NO_2$, $ONOO^-$, $ROONO$, etc.), and UV photons could be involved in the antimicrobial processes (Misra & Jo 2017; Hertwig, Meneses, and Mathys 2018). The reactive species (RNS and ROS) can directly interact with cellular macromolecules, including the oxidation of polysaccharides, lipids, proteins and DNA (Misra et al. 2011; Niemira 2012). DNA damage may retard cells' multiplication, whereas, cell membrane damage can cause the cell leakage and loss of cell functionality, peptidoglycan or lipopolysaccharides oxidation may modify the cell walls of Gram-positive and Gram-negative bacteria, respectively (Lee et al. 2015; Misra and Jo 2017). Meanwhile, the ROS/RNS can induce the generation and accumulation of intracellular oxidative stress, and cells generate the cytoplasmic and nuclear responses, lead to DNA damage, cell cycle modification and apoptosis (Arjunan, Sharma, & Ptasinska 2015). Sharma, Collins, and Pruden (2009) observed that most genes involved in SOS and oxidative stress response significantly up-regulated after CP exposure, and a vast majority of genes involved in energy metabolism and transport processes markedly down-regulated. After CP treatment, the bacteria proteins exhibited changes in abundance, and genes belong to the functional categories cell wall, motility and chemotaxis, coping with stress, prophages, sporulation and homeostasis were upregulated, while to nucleotides, nucleic acids and carbon metabolism were downregulated (Winter et al. 2011).

Applications

CP has been gained a rising attention during past decades, and both scientific research and industrial applications of CP for F&V disinfection have been widely reported (Table 4). Baier et al. (2013) demonstrated that CP treatment at 20 W for 1 min obtained 4 log CFU/cm² reductions of *E. coli* on corn salad leaves. Zhang et al. (2013) observed a 3 log reduction of *S. Typhimurium* LT2 on spinach after low-pressure oxygen plasma treatment at 0.34 W/cm³ for 600 s. Recently, DBD atmospheric CP (DACP) treatment has been developed as a method for the in-package disinfection of F&V and has gained great commercial interest from the food industry. Min et al. (2017) showed that DACP uniformly inhibited the *E. coli* O157:H7 on cut lettuce in

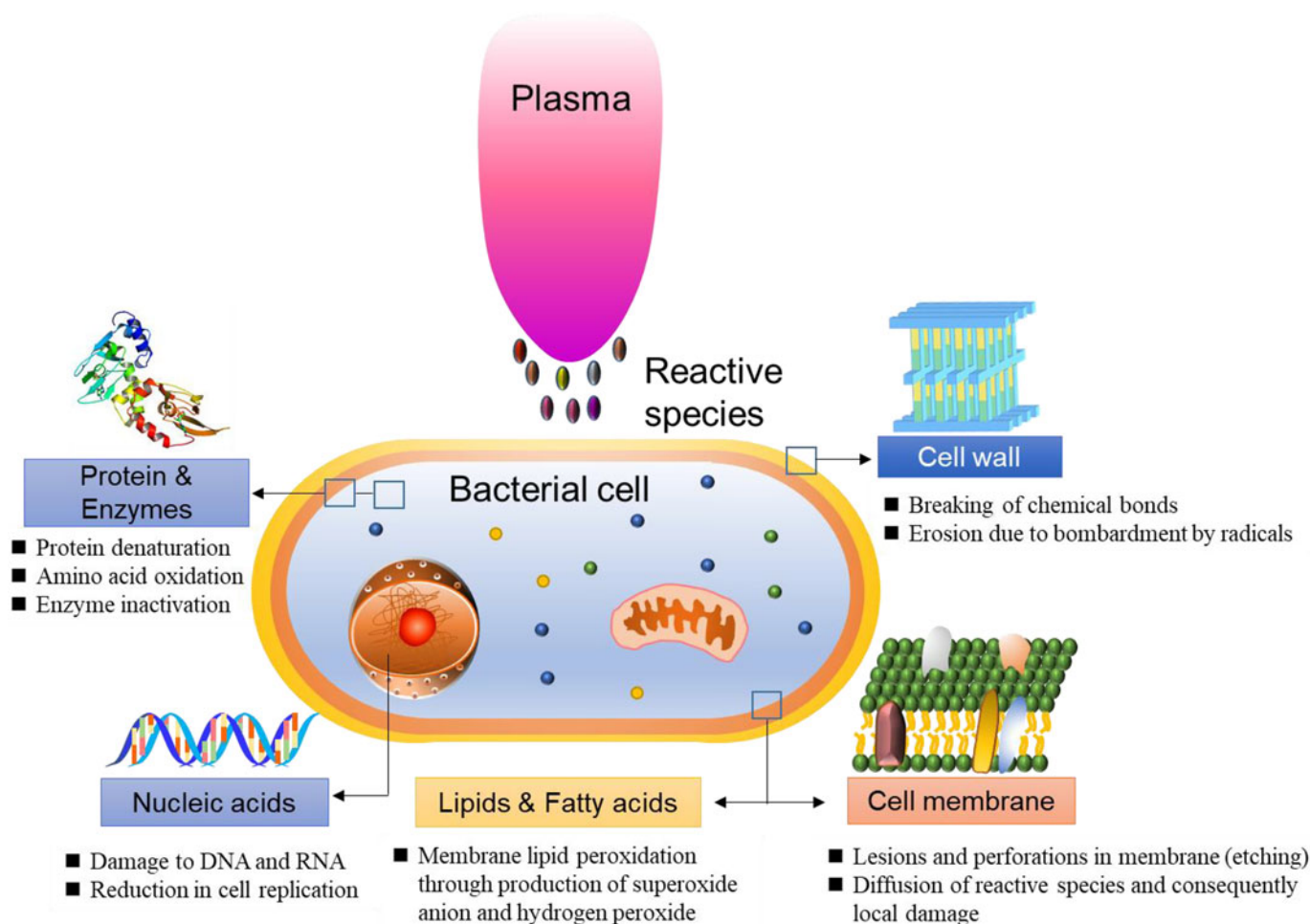


Figure 8. A schematic of the mechanics of cold plasma sterilization of bacteria (adapted from Misra and Jo 2017).

commercial containers. A treatment of packaged strawberry by DACP for 5 min reduced the total mesophiles and yeasts & molds by 2.4 and 3.3 log CFU/g, respectively (Misra et al. 2014). Therefore, CP has great potential to eliminate the microbial contamination of F&V. Additionally, CP treatment has no significant effect on the physical and biological properties (e.g. color, respiration rate, and weight loss) of F&V (Misra et al. 2014; Pasquali et al. 2016; Min et al. 2017).

Factors influencing efficacy

The bactericidal efficacy of CP depends on many factors, including plasma source, operating conditions, gas used, exposure time, microorganisms, and F&V species (Zhang et al. 2013; Lacombe et al. 2015). Misra and Jo (2017) showed that plasma operated at atmospheric pressure was more effective for bacterial inactivation than those operated at low pressure. They also found that pure noble gases provided abundant charged particles that caused bacterial cell surface etching. The pressure determines the total density of gas, and higher pressures increase collision frequencies (Bárdos & Baránková 2010). However, due to the high equipment cost, and short longevity of the charged He and Ar ions, the CP using air at atmospheric pressure has been widely explored (Misra & Jo 2017). Higher antimicrobial efficiency can be achieved at higher generator power and

longer duration time (Baier et al. 2013). High MW density-CP treatment (HMCPT) was more effective than low MW density-CP treatment (LMCPT) in inhibiting *B. cereus* spores (Kim, Oh et al. 2017). Increasing treatment time yielded a greater inhibition of *B. cereus* spores (Kim, Oh et al. 2017). A CP treatment of corn salad leaves inoculated with *E. coli* for 10 and 30 s resulted in inactivation of 1 and 2 log, respectively (Baier et al. 2013). HMCP (400 W, 0.7 kPa, 40 min) using helium gas reduced the numbers of *B. cereus*, *A. brasiliensis*, and *E. coli* O157:H7 in onion powder by 2.1 log spores/cm², 1.6 log spores/cm², and 1.9 log CFU/cm², respectively (Kim, Oh et al. 2017). Atmospheric CP (ACP) treatment for 45 s reduced the *E. coli* and *L. monocytogenes* on cherry tomatoes by 2.0 and 4.5 log CFU/fruit, respectively (Ziuzina et al. 2014). Meanwhile, higher antimicrobial efficiencies were also achieved at lower initial microbial counts (Baier et al. 2013). A significant higher reduction of *E. coli* on tomato and lettuce was observed at the initial microbial concentration of 10⁵ CFU/mL than at 10⁷ CFU/mL (Bermúdez-Aguirre, Wemlinger et al. 2013; Bermúdez-Aguirre, Martínez-Niño et al. 2013). F&V type is also an important factor. Usually, microbial populations can be easily inactivated on produce with clean and flat surfaces when compared to those with intricate and complex surfaces (Critzler et al. 2007). Ziuzina et al. (2014) showed that ACP treatments for 10 and 60 s reduced the populations of

Salmonella and *E. coli* on cherry tomatoes to undetectable levels, however, it took about 300 s for strawberries to achieve the similar results. Similarly, Fernández, Noriega, and Thompson (2013) observed that ACP treatment for 15 min achieved 2.72, 1.76, and 0.94 log reductions of *S. enterica* serovar Typhimurium on lettuce, strawberry, and potato, respectively.

Limitations

A higher power of CP resulted in noticeably higher temperature of produce, which caused thermal damage to F&V (Baier et al. 2013), and altered organoleptic profiles of the product (Kim, Oh et al. 2017). For instance, the temperature of onion was increased by 15.9, 19.9, 36.5, and 40.2 °C after CP treatment at 10, 20, 30, and 40 W, respectively (Kim, Oh et al. 2017). In addition, under atmospheric conditions, higher voltages are required to generate plasma (Lieberman & Lichtenberg 2005). Arcing usually occurs between the electrodes when high voltages applied, which may result in the burn damage of produce surface (Lee et al. 2015). Significant reduction in firmness, anthocyanins, and surface color values (L^* , a^* , and b^*) were observed for blueberry after ACP treatment duration for 60, 90 and 120 s, respectively (Lacombe et al. 2015). Nevertheless, CP still shows promise to replace conventional disinfection technologies. More research should be focused on the interactions between CP and food components. The operational parameters should be optimized to maintain the quality of the product. Meanwhile, investment and cost should be analyzed for large-scale applications.

High-intensity ultrasound

High-intensity ultrasound (HIUS) refers to ultrasound operating at high frequency (ranging from 20 to 100 kHz) and power (between 100 and 500 W/cm²). HIUS is generally considered safe, nontoxic, and environmentally friendly. Over the last decade, HIUS processing has attracted increasing interest in the field of food disinfection (Afari et al. 2016; Millan-Sango et al. 2016).

Disinfection mechanisms

HIUS can inactivate microorganisms via the disruption of the microbial cells' components by intracellular acoustic cavitations (Gómez, Welti-Chanes, & Alzamora 2011; Bilek and Turantaş 2013). The cavitation bubbles are generated when HIUS is applied. Small bubbles undergo regions of alternating compression and expansion in very short periods, which is accompanied with violent collapse and further break up into many smaller bubbles. Shock waves with very high energy densities can radiate from collapsing bubbles that are strong enough to shear and break cell walls and membrane structures, as well as depolymerize large molecules (Gómez, Welti-Chanes, & Alzamora 2011). Besides, the chemical effect of ultrasonication also contributes to the antimicrobial effect. The hydroxyl radicals can be formed due to rise of temperature at a localized position inside a collapsing

bubble. They can react with the DNA chain and break the double strand microbial DNA (Bilek & Turantaş 2013).

Applications

HIUS has been used as a surface cleaning technique for inactivating various microorganisms on F&V (Table 4). For instance, HIUS treatment (32–40 kHz) of cut iceberg lettuce achieved a 1.6 log CFU/g (about 97.9%) reduction of *S. Typhimurium* (Seymour et al. 2002). Similarly, HIUS treatment (26 kHz, 200 W) reduced the *S. enterica* on romaine lettuce by 1.68–2.23 log CFU/cm² (Millan-Sango et al. 2016). In another study, HIUS treatment of alfalfa and mung bean sprouts reduced the *Salmonella* by 1.40 and 1.89 log CFU/g and the *E. coli* by 1.06 and 1.23 log CFU/g, respectively (Millan-Sango et al. 2017). The published data indicate that the HIUS may not be adequate to effectively inactivate microorganisms and guarantee the microbial safety of F&V.

Factors influencing efficacy

During HIUS treatment, the microbial reductions are closely associated with the operation parameters, such as power, exposure time and temperature. Usually, increases in exposure time and ultrasound frequency could achieve higher reductions of microorganisms. When HIUS treatment time was increased from 1 to 10 min, the reductions of *E. coli* on lettuce were increased from ~1 to 2.61 log CFU/cm² (Huang et al. 2018). As ultrasound power was amplified from 130 to 210 W, the reductions of *E. coli* O157:H7 on romaine lettuce increased from 1.92 to 2.24 log CFU/g (Afari et al. 2016). Meanwhile, both the microorganism and F&V species significantly influence the sterilization effect. HIUS treatment (26 kHz, 200 W) reduced *Salmonella* and *E. coli* by 1.89 and 1.23 log CFU/g on mung bean sprouts, respectively (Millan-Sango et al. 2017). HIUS treatment for 10 min reduced the *E. coli*, *L. innocua*, and *P. fluorescens* inoculated lettuce leaves by 2.61, 2.23, and 1.10 log CFU/cm², respectively (Huang et al. 2018). Reductions of 1.40, 0.97, 0.49, and 0.63 logs of *E. coli* NCIMB 12497 were observed on cabbage, iceberg lettuce, parsley, and strawberry after treated with HIUS at 70 kHz for 10 min (Seymour et al. 2002).

Limitations

HIUS has limited antimicrobial effect, it reduces approx. 0.6–1.5 log CFU/g under typical conditions experiments (São José & Vanetti 2012; Bilek and Turantaş 2013; Lou et al. 2015). It needs to be combined with chemical sanitizers, such as EOs (Millan-Sango et al. 2016), ClO₂ (Millan-Sango et al. 2017), and chlorine (Huang et al. 2018) to enhance disinfection efficacy. Meanwhile, the shear forces generated by acoustic micro-streams in the ultrasonic field can lead to cell disruption and tissue damage (Ngnitcho et al. 2017). Prolonged exposure to ultrasound increased the electrolyte leakage rate and reduced the turgor of lettuce (Huang et al. 2018). Information on the commercial

application of ultrasound is scarce. The small radiation area of transducers is a vital shortcoming for industrial applications. Efforts are needed to develop robust large-scale equipment (Deng et al. 2019).

In summary, novel non-thermal physical technologies provide a gentle pasteurization mode that are effective reduction in microbial loads and minimizing impact on organoleptic and nutritional profiles of F&V. A number of physical disinfection technologies, such as UV light, PL, ionizing irradiation and HHP are currently under commercialization. However, many of them are expensive and technically difficult to be implemented compared to chemical sanitizer treatment (Bilek & Turantaş 2013). UV light system is affordable with a low initial investment and operating cost, which is estimated between \$10,000 and \$15,000 (Shah et al. 2016). While, PL facility is much more expensive than UV light, the high investment cost (€300,000–800,000) limits the applications to high value added (Pereira & Vicente 2010). Though the cost for fruits ionizing irradiation processing is reasonable (about \$0.10/kg), the initial investment for the facility is estimated between \$4 and \$10 million (Moreira & Castell-Perez 2012). The commercial scale HHP vessel usually costs between \$500,000 to \$2,500,000 depending upon equipment capacity and extent of automation (Tonello 2011). But the novel physical disinfection technologies have been proven to be energy-efficient, cost-effective, and environmentally friendly, which will help reduce cost and add value to the product, and make them economically advantageous and good payback on capital investment (Pereira & Vicente 2010). While, CP and HIUS have been explored in the lab with successful results, but it is still under development. More efforts are needed to develop large-scale inexpensive equipment for their application in the food industry.

Hurdle technologies

The emerging chemical and physical disinfection technologies are promising alternatives to traditional chlorine washing to address the microbial concerns of F&V. However, as summarized in Table 5, these technologies used alone are possibly not sufficient to assure the microbial safety, and may adversely affect the nutrition and organoleptic properties of produce. Due to these facts, hurdle technology, generally known as combined technologies, has been developed and adopted for improving the safety, and nutritious and sensory qualities of produce (Khan et al. 2017). Hurdle concept advocates the intelligent use of the combinations of different preservation factors or technologies, aiming to achieve multi-target, mild but reliable preservation effects (Leistner & Gorris 1995). These hurdles can be temperature, water activity (a_w), pH, redox potential or something else (Leistner & Gorris 1995).

Mechanisms

Usually, hurdle technology exhibits synergistic effects (Rahman, Khan, & Oh 2016). The hurdles may hit different

targets (e.g., cell membrane, DNA, enzyme systems, pH, a_w , etc.) within the microbial cells at the same time, and thus critically disturb the homeostasis of the cell (Rahman, Khan, & Oh 2016). Thereby, microorganisms will be more difficult to recover homeostasis and activate stress shock proteins (Leistner & Gorris 1995). The specific mechanisms are dependent on the technologies used. For example, HHP damages sublethal cells and increases the cellular permeabilization, which might enhance the sensitivity to antimicrobial activities and diffusion of sanitizer through the cell membrane (Ramos-Villarroel, Martín-Belloso, & Soliva-Fortuny 2015). Ultrasound helps aqueous sanitizers penetrate inaccessible sites, and then improves the effectiveness of sanitizers (Sagong et al. 2011; Bilek and Turantaş 2013). Usually, a number of efforts are required for a microorganism to overcome each hurdle (Leistner & Gorris 1995).

Applications

There are a number of successful combinations of disinfection technologies (Table 6). Garcia, Mount, and Davidson (2003) showed that the chlorine, ozone, and chlorine–ozone treatment reduced aerobic plate count up to 1.4, 1.1, and 2.5 log CFU/g, respectively, and achieved the shelf lives of the products of 16, 20, and 25 d, respectively. The results indicated that the chlorine–ozone combinations had beneficial effects on the shelf life and quality of lettuce. Severino et al. (2014) observed that the modified chitosan coating containing mandarin EO treatment alone failed to reduce the *L. monocytogenes* on broccoli florets, whereas, combining ozonated water washing with coating achieved 1.51 log CFU/g reduction, which was also higher than ozonated water washing alone (1.13 log CFU/g). Afari et al. (2016) found that while the NEO washing reduced the *E. coli* O157:H7 and *S. Typhimurium* DT 104 on lettuce by >2.5 log CFU/g, combining NEO water with HIUS reduced pathogens by >4 log CFU/g. Ramos-Villarroel, Martín-Belloso, and Soliva-Fortuny (2015) reported that the PL (180–1100 nm, 12 J/cm²) and malic acid (MA, 2%) treatments reduced *L. innocua* and *E. coli* on fresh-cut avocado, watermelon and mushroom by 0.91–1.1 and 1.92–2.97 log CFU/g, respectively. Nevertheless, both *L. innocua* and *E. coli* were reduced more than 5 log CFU/g when the PL and MA treatments were sequentially applied. The treatment of green beans by mandarin oil coating, HHP, and their combination reduced the population of *L. innocua* by 0.6, 1.7, and 5.0 log CFU/g, respectively (Donsì et al. 2015). Additionally, the combined treatment increased the firmness and maintained the color of green beans during refrigerated storage (Donsì et al. 2015). Gupta et al. (2012) found that the combined γ -ray and citric acid treatment significantly reduced microbial loads and extended shelf life by 1 week of French beans.

These findings suggest that hurdle technologies have higher sterilization efficacy than the individual treatments alone. Meanwhile, hurdle technologies can reduce the amounts of sanitizers used, lower the operation intensities (power, dosage or frequency), and decrease the duration time, thus minimize the impact on the quality of F&V. Therefore, hurdle technologies are promising treatments to

Table 5. Advantages and disadvantages of emerging disinfection technologies.

Technology	Advantages	Limitations	Reference
Chlorine dioxide	Can be applied in aqueous or gaseous forms; Higher oxidative capacity and antimicrobial efficacy than chlorine; Without forming carcinogenic by-products; Less corrosive than chlorine and ozone.	Low efficient at permitted concentration; Very short shelf life and requires on-site generation; Explosion and toxicity hazards at high concentrations; Requires water rinsing after treatment; Formation of by-products; Possibility of affecting products' quality; Possibility reversibly damage microbes.	Goodburn and Wallace 2013; Lee, Beuchat et al. 2018; Lindsay et al. 2002; Praeger, Herppich, and Hassenberg 2018; Singh et al. 2002.
Ozone	Can be applied in aqueous or gaseous forms; Environmentally friendly; Without residual compounds.	Oxidation of food components at prolong exposure or high concentration treatment; Safety issue of workers; High instability and requires on-site generation; Corrosive hazard; High management costs.	Botondi et al. 2015; Brodowska, Nowak, and Śmigielski 2018; Goodburn and Wallace 2013; Kim, Yousef, and Dave 1999; Oner and Demirci 2016.
Electrolyzed water	Low operational expenses; Easy operation; Safe and eco-friendly.	Very short shelf life and requires on-site generation; Corrosive hazard of strong acid EW; Cl ₂ production; Possibility negative effect on the nutrient.	Chen, Xue et al. 2018; Gómez-López, Gil, and Allende 2017; Rahman, Khan, and Oh 2016; Santo et al. 2018.
Essential oil	Natural antimicrobial agents; Environmentally friendly; Relative safe; Low resistance-inducing effects in microbe.	Hydrophobic, volatile and unstable nature; Alters the aroma of produce; Instability in composition and efficacy; Little knowledge about toxicity, safety and interaction of EOs in the human gut.	de Oliveira et al. 2017; Donsì et al. 2015; Kang and Song 2018; Patrignani et al. 2015; Prakash et al. 2018.
High pressure carbon dioxide	No toxicity; Relatively low operating costs compared to HHP; Without residual compounds.	High capital costs; Alters the quality of products; Fails to control microbe during produce storage.	Ferrentino and Spilimbergo 2011; Spilimbergo et al. 2013; Valverde, Marín-Iniesta, and Calvo 2010.
Organic acid	Easy to use; No toxicity.	Long contact time; Interferes with the organoleptic quality; Low antimicrobial efficacy; Corrosive hazard.	Bermúdez-Aguirre, Wemlinger et al. 2013 and Bermúdez-Aguirre, Martínez-Niño et al. 2013; Khan et al. 2017; Sagong et al. 2011.
UV light	Equipment of moderate to low cost; Easy to use; Little changes in product quality at low doses; Continuous process; Without residual compounds.	Low degree of penetration; Occurrence of shadow effects; Alters light sensitive compounds in produce; Some microbe can gradually recover.	Gayán et al. 2014; Ochoa-Velasco et al. 2018.
Pulsed light	High energetic and sterilization efficiency; Little changes in product quality at low doses; Suitable for packaging materials; Continuous process; Without of harmful residues.	Low degree of penetration; Occurrence of shadow effects; Thermal damage and quality deterioration of product at high doses.	Aguiló-Aguayo et al. 2013; Gómez et al. 2012; Pataro et al. 2015; Rowan 2019.
Ionizing radiation	High sterilization and energetic efficiency; Little changes to organoleptic properties of product; Suitable for packaging produce; Without of harmful residues.	Possibility of affecting products' quality; High capital investment; Strict safety standards; Public acceptance concern.	Goodburn and Wallace 2013; Moosekian et al. 2012; Olaimeat and Holley 2012.
High hydrostatic pressure	Uniformity of treatment throughout food; Effectively inactivates vegetative bacteria; Preserve product quality; Inactivate enzymes; In-packaging treatments; Without of residues.	Spores cannot be inactivated; Batch processing; High cost of equipment; Safety concerns on use of high pressures; Affects to certain product quality.	Baptista et al. 2015; Dong et al. 2013; Huang et al. 2017; Lee, Ha et al. 2018; Lou et al. 2015; Rendueles et al. 2011.
Cold plasma	High efficiency; In-package treatments; Without of harmful residues.	Thermal damage to produce at some certain condition; Types of foods treated may result in shadowing; Possibly alters organoleptic profiles of the product at intensive treatment; Technology in an early development stage.	Baier et al. 2013; Kim, Oh et al. 2017; Lacombe et al. 2015; Niemira 2012.
High-intensity ultrasound	Little change in quality of produce; Without of harmful residues.	Low antibacterial efficacy; Intensity of industrial-scale equipment limited; Undesirable cell rupture and quality changes at high doses.	Afari et al. 2016; Bilek and Turantaş 2013; Millan-Sango et al. 2016; São José and Vanetti 2012.

Table 6. Hurdle disinfection technologies for fruits and vegetables.

Material	Microorganisms	Treatment	Results	Reference
Lettuce and baby carrot	<i>E. coli</i> O157:H7	Sequential washing (SW): thyme oil suspension (1.0 mL/L for 5 min), followed by aqueous ClO ₂ (10.0 mg/L for 10 min), and ozonated water (9.7 mg/L for 10 min)	SW treatment reduced <i>E. coli</i> O157:H7 by 3.75–4.34 log CFU/g, which were greater than those treated by alone form (1.48–1.97 log CFU/g).	Singh et al. 2002
Apple	Total bacteria	Dipped in 0.5% citric acid (CA) for 5 min, then exposed to UV-C light for 5 min	The bacteria were reduced by 1.5, 2.1 and 2.6 log CFU/g after treatment with CA, UV-C and CA + UV-C, respectively; the CA + UV-C treatment retarded the microbial growth, and reduced weight loss and phenolic degradation during storage.	Chen et al. 2016
Cilantro	<i>E. Coli</i> O78	Dipped in acidic electrolyzed water (AcEW) (68.35 ACC mg/L, pH 2.48) for 5 min, and then in alkaline electrolyzed water (AIEW) (pH 11.65) for 5 min	AcEW + AIEW treatment reduced the bacterial counts to under detectable level (>7 log CFU/g), which much higher than those of single AcEW (2.67 CFU/g) and AIEW (0.57 CFU/g) treatments.	Hao e al. 2015
Cilantro	Total aerobic bacteria	Dipped in acidic electrolyzed water (AcEW) (68.35 ACC mg/L, pH 2.48) for 5 min, and then in alkaline electrolyzed water (AIEW) (pH 11.65) for 5 min	The AcEW + AIEW, AcEW and AIEW treatments reduced the bacterial counts from 6.89 to under detectable level, 4.41 and 6.02 log CFU/g, respectively.	Hao et al. 2015
Lettuce	<i>Salmonella</i>	Ultrasonic treatment (operating at 26 kHz and 200 W) for 5 min, and 0.018% v/v of EOO (carvacrol (75–85% v/v) and thymol (0.7–4% v/v)) for 5 min	Ultrasonic, EOO and the combination treatment reduced the <i>Salmonella</i> populations by 2.23, 0.75 and 3.08 log CFU/cm ² , respectively.	Millan-Sango et al. 2016
Spinach	<i>E. coli</i> O157:H7	PL (dose of 15.75 J/cm ²) and HEN sanitizer (3% hydrogen peroxide, 0.02 mM EDTA and 20 mg/ml Nisin, pH 6.6) washing 2 min	Reductions of 4.6 and >5 log CFU/g after Hen-PL and PL-HEN, respectively; they were much higher than those of PL (2.7 log CFU/g) and HEN (1.8 log CFU/g) alone.	Mukhopadhyay et al. 2019
Plum tomato	<i>S. enterica</i>	UV light (dose of 0.6 kJ/m ²), and HEN sanitizer washing for 2 min	UV, UV + HEN reduced the populations by 1.6 and 4.71 log CFU/fruit, respectively.	Mukhopadhyay, Ukuku, and Juneja 2015
Broccoli	<i>L. monocytogenes</i>	Combined EOs coating (1% Modified chitosan + 2.5% Mandarin oil nanoemulsion) and γ -ray (doses of 0.25 kGy)	The combined treatment reduced the <i>L. monocytogenes</i> by 2.5 log CFU/g after 13 days storage, which greater than EOs coating alone (1.11 log CFU/g).	Severino et al. 2014
Avocado, watermelon, and mushrooms	<i>L. innocua</i> and <i>E. coli</i>	Dipped in malic acid (MA) (2% w/v) for 2 min, then treated with PL (dose of 12 J/cm ²)	Reductions of 0.91–1.1, 1.92–2.97 and >5 log CFU/g after MA, PL, and MA + PL treatment, respectively.	Ramos-Villarroel, Martín-Belloso, and Soliva-Fortuny 2015

obtain the F&V that are safety and high quality and have long shelf life. They can be the key in providing solutions to future food safety.

Future trends

1. Wastewater and cross contamination occurrences during sanitizer washing should be given more careful attention. Aqueous sanitizers washing has gained more commercial use and interests due to easy-handling. However, the large volumes of water needed for treatment can cause microbial cross contamination and wastewater. Hence, gas treatments need to be developed as potential alternatives to aqueous antimicrobial washing, and good air handling and destruction systems are important to ensure safe processing environment and minimize negative environment impact.
2. The quality changes of F&V during storage and shelf life are frequently overlooked. Extending shelf-life is a key factor for making F&V products more profitable and commercially available for long periods of time of the best possible quality. Further research regarding the effects of these technologies on organoleptic and nutritional properties of F&V after the treatments and during storage, as well as the shelf life are needed.
3. Hurdle technologies should be specifically developed for future application to improve efficiency, minimize cost, and yield minimal quality changes.
4. The disinfection technologies of organic fresh produce should be further developed. Recently, there has been an increasing demand for organic produce, while most of chemical sanitizers are not allowed for organic food processing. The emerging physical disinfection technologies provide the opportunity to improve the microbial safety and quality of organic produce. In order to meet the requirements for industrial application, more efforts are required to improve the robustness and efficiency, and reduce the cost of these technologies.
5. Developing mathematical models for accurately assessing, predicting, and controlling the microbial

inactivation process is urgently needed. Mathematical model is important for establishing and optimizing process conditions to achieve stable and safe products as well as to minimize the undesirable changes of food quality. It is necessary for understanding the underlying antimicrobial mechanism using advanced micro-imaging and analytic tools, identifying the critical factors that influencing the microbial survival, and constructing predictive inactivation models for target strains for specified technology.

Conclusions

With the increase in safety awareness from consumers and demand for high quality F&V, several disinfection technologies have been developed for F&V. Chemical disinfection technologies, including chlorine dioxide, ozone, EW, EO, high HPCD, and organic acid, are promising alternatives to traditional disinfection technologies to meet the rigorous food safety and shelf life demands. Physical technologies, including UV-light, PL, ionizing radiation, HHP, CP, and high-intensity ultrasound technologies, provide a gentle pasteurization mode that are effective reduction of microbial loads with minimal effects on sensory and nutritional profiles. High treatment intensities are required while using an individual physical technique alone to achieve a substantial reduction of microorganisms. Therefore, hurdle technology has gained more attention and practical applications since it has shown promising results for improving the disinfection efficiency, overcoming the limitation of the specific technology, and helping to improve the quality of F&V.

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