

INSIGHTS INTO OZONE AND ELECTROLYZED WATER HURDLE SYSTEMS CONFERRING DECONTAMINATION IN FRESH PRODUCE

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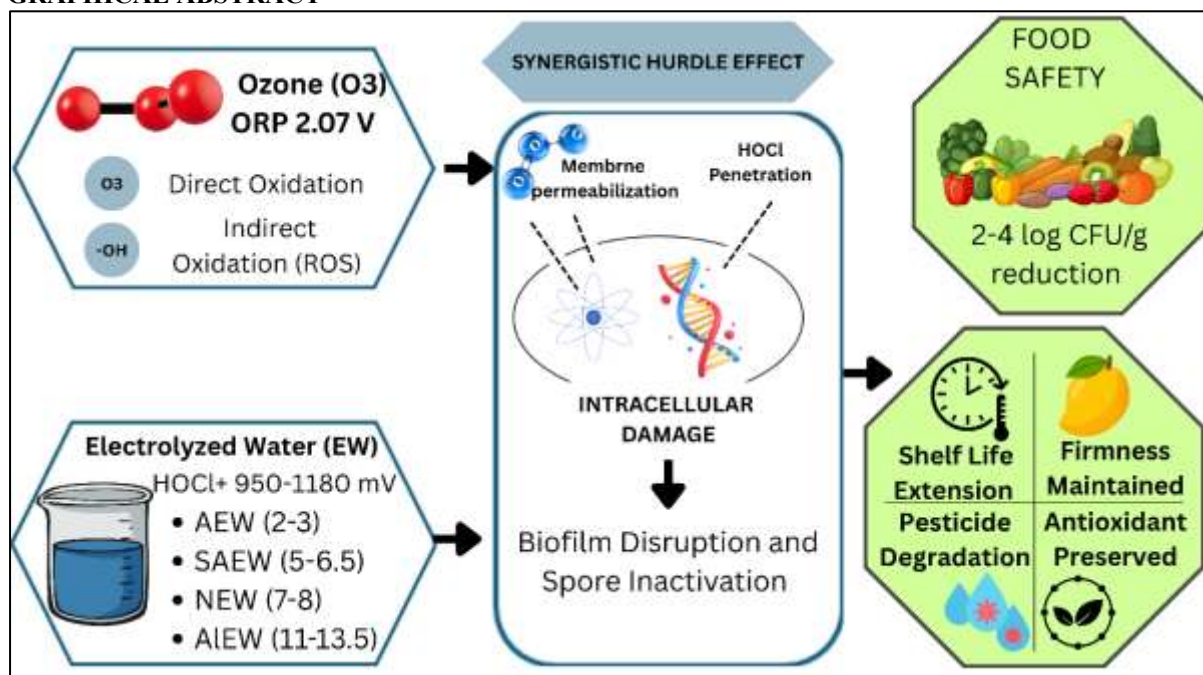
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ABSTRACT

Fresh produce and processed food are highly susceptible to microbial contamination during production, transportation, and storage, leading to significant economic losses and health impacts. Though traditional thermal technologies effectively control pathogens, they often compromise the food's nutritional and sensory qualities. As a result, non-thermal alternatives such as ozone (O₃) and electrolyzed water (EW) have emerged as promising sanitization hurdles for food-contact surfaces and raw materials. This review evaluates the synergistic application of ozone and electrolyzed water in food safety management. Both technologies rely on high oxidation-reduction potential (ORP) to inactivate the microbial contamination. EW is categorised by pH levels into acidic (AEW), slightly acidic (SAEW), neutral (NEW) and alkaline (ALEW). It works on a complementary mechanism, where ozone enhances cell permeability for EW's HOCl penetration, achieving superior pathogen inactivation, biofilms and spores compared to individual treatments. By using lower dosages, this synergistic method reduces individual drawbacks such as metal corrosion and off-flavour, and produces bleaching, while minimizing chemical residue and maintaining quality attributes such as firmness, texture, colour, and antioxidants in food products. The review discusses underlying mechanisms, physicochemical properties, and practical application of this synergy, including pesticide removal and shelf-life extension, as well as key constraints such as organic load interference. Overall, ozone-electrolyzed water treatment represents a sustainable alternative to chlorine-based sanitization with strong potential for food safety applications and industrial scale-up.

GRAPHICAL ABSTRACT



KEYWORDS: Ozone (O₃), Electrolyzed water (EW), Synergistic sanitization, Non-thermal processing, food safety

1. INTRODUCTION

Freshly harvested fruits and vegetables are highly perishable and are prone to spoilage during production, transportation, and storage, resulting in substantial losses and food safety risks (Jiang et al., 2001). The global burden of foodborne diseases remains significant, with bacterial contamination of food and food contact surfaces identified as the primary cause of outbreaks and economic losses in the food industry (Nowshad et al., 2021; Sehgal et al., 2024). Food safety must be ensured at each postharvest processing step, including handling, washing of raw materials, cleaning of utensils and pipelines, and packaging (Al-Haq et al., 2005). During processing and storage, raw food materials come in contact with the moist surface of equipment and utensils, creating an ideal environment for bacterial biofilm formation (Sharma et al., 2022).

Conventional thermal sanitization methods, while effective against a broad spectrum of pathogens, cause detrimental effects on heat-sensitive food components, leading to protein denaturation, nutrient loss, and sensory quality deterioration that fail to meet consumer demand for minimally processed foods (Sun et al., 2022). These limitations have driven the rapid development of non-thermal sanitization technologies as alternatives that preserve food quality while ensuring microbial safety. As a part of the circular economy approach, the implementation of non-thermal sanitization techniques in food processing has the potential of sustainable development (Adal et al., 2024). Among emerging non-thermal technologies, electrolyzed water (EW) has gained attention as a sanitizer for the food industry (Rico et al., 2008). EW is generated by passing an electric current through a diluted sodium chloride solution (NaCl) across an anode and a cathode compartment separated by a polyester membrane. The cathode area produces alkaline reducing water, while the anode area produces acidic oxidizing water (Lee et al., 2007). The electrolysis process yields different EW types depending on pH: acidic EW (AEW, pH 2-3), slightly acidic EW (SAEW, pH 5.0-6.5), neutral EW (NEW, pH 7.0-8.0), and alkaline EW (ALEW, pH 10-13). AEW inactivates bacteria, including *Vibrio parahaemolyticus*, *Shewanella putrefaciens*, *Escherichia coli*, and *Listeria monocytogenes*. It also eradicates resistant microorganisms like *Aspergillus sydowii*, *Rhizopus stolonifer*, and *Bacillus subtilis* (Xu et al., 2025). EW offers on-site production, cost effectiveness, environment friendly, effective sanitizing agent, ease of application. It provides safety to humans and the environment (Zhao et al., 2017).

While EW is effectively used as a surfactant, other oxidants like ozone (O_3) are also being explored. The ozone (O_3) is a potent non-thermal sanitizer recognized as a safe food disinfectant authorized in many countries for direct contact with food (Food & Administration, 1997). This triatomic molecule possesses a high oxidation-reduction potential (2.07 V) and degrades the cell membranes, unsaturated lipids, proteins, DNA and intracellular enzymes of micro-organisms (Epelle et al., 2022). It was observed that O_3 gas or aqueous ozone lowered decay during cold storage of papaya fruits (Ali et al., 2014), persimmon leaves (Ikeura et al., 2013), lowbush blueberries (Crowe et al., 2012), actinidia deliciosa (Barboni et al., 2010), spinach (Klockow & Keener, 2009), and peanuts (Chen et al., 2014). Ozone exhibits a broad spectrum antimicrobial activity against Gram-positive and Gram-negative bacteria, as well as spores and vegetative cells, with bacteria being more sensitive to O_3 than yeasts and fungi (Joshi et al., 2013; Pascual et al., 2007). Despite the well-documented efficacy of EW and ozone as separate sanitizers, most published reviews have examined each technology in isolation. Comprehensive reviews on EW applications in food postharvest and processing are available (Adal et al., 2024; Rahman et al., 2016), as do reviews on ozone for produce decontamination (Xue et al., 2023). The complementary mechanisms of these two sanitizers- where ozone alters cell membranes and enhances permeability, enabling deeper penetration of EW-derived hypochlorous acid (HOCl)- offer a promising hurdle strategy that may achieve superior microbial inactivation at lower individual doses, thus minimizing the drawbacks of each treatment alone, such as ozone-induced bleaching, EW corrosiveness, and off-flavour development. Accordingly, this review discusses the individual and combined roles of ozone and electrolyzed water in food sanitation, with emphasis on their mechanism of action, application in fresh and processed foods, impact on product quality, and future prospects for industrial implementation.

2. METHODOLOGY

This systematic review adopted a systematic methodology aligned with PRISMA 2020 guidelines to ensure transparent and reproducible evidence synthesis while systematically addressing the lack of a unified evidence base on the synergistic application of ozone and electrolyzed water (Sohrabi et al., 2021). The methodology adopted for this systematic review is outlined below.

2.1. LITERATURE SEARCH

A systematic literature search was conducted to identify relevant studies on the application of ozone (O_3), electrolyzed water (EW) and their synergistic use in food systems. The search was conducted across several reputable databases, including Scopus, ScienceDirect, Web of Science, PubMed, MDPI, and Google Scholar, to ensure comprehensive coverage of the available literature. The literature search was conducted between January 2026 and February 2026 using primary search terms 'ozone', 'ozonated water', 'electrolyzed water', and 'acidic electrolyzed water (AEW)'. Additional keywords included 'food safety', 'food sanitation', 'non-thermal processing', and 'food contact surfaces'. Boolean operators were used to refine the search strategy. The operator AND was employed to combine major concepts, while OR was employed to include synonymous terms. A representative search string was ("ozone" OR "ozonated water" OR "gaseous ozone") AND ("electrolyzed water"

OR "acidic electrolyzed water"). To ensure the retrieved studies were relevant, records related to medical ozone therapy, wastewater treatment, pharmaceutical use, and clinical disinfection were excluded using the AND NOT operators where applicable. The review included peer-reviewed original research articles, short communications and conference papers published in the English language. Studies published between 2000 and 2025 were included to capture recent advances in ozone and electrolyzed water technologies. Studies focus solely on non-food applications, such as human and veterinary medicine, and industrial chemical processing, were excluded from the search parameters.

2.2. SELECTION OF SUITABLE RECORDS

The citations extracted from the literature search were meticulously curated using EndNote, the citation management tool (<http://www.endnote.com/>). Each recorded was appropriately tagged against its respective database of origin to facilitate tracking and management. Duplicate records identified across databases were removed automatically using the software's duplicate detection function, followed by manual verification to ensure data integrity.

A total of 553 records were identified through database searching, of which 197 duplicate records were removed, resulting in 356 unique records for title and abstract screening. The screened records were evaluated for relevance to the objective of this review, which focused on the synergistic application of ozone (O₃) and electrolyzed water (EW) in the postharvest management of fresh horticultural produce. Studies unrelated to food applications, including medical, pharmaceutical, wastewater treatment, and industrial chemical processing, were excluded according to the predefined eligibility criteria. In addition, the reference lists of relevant publications were manually examined to identify additional eligible studies. Following the screening and eligibility assessment, 77 studies were included in the qualitative synthesis. The literature selection process is summarized in the PRISMA 2020 flow diagram (Figure 1).

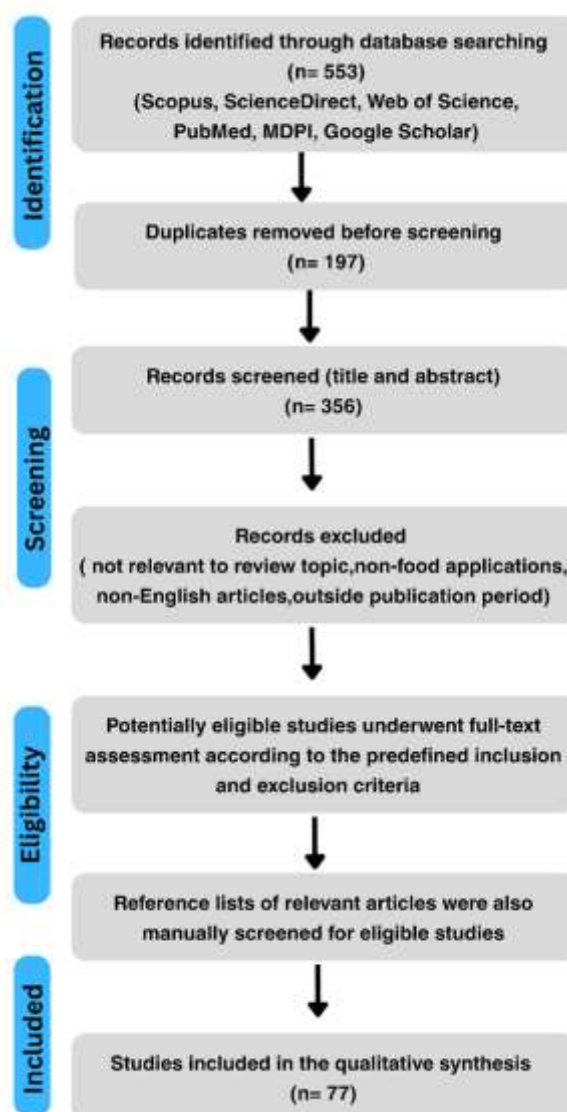


Figure 1 PRISMA 2020 Flow Diagram of Literature Selection

2.3. RESULTS AND DISCUSSION

The systematic literature search identified 553 records from six electronic databases namely Scopus, followed by ScienceDirect, Web of Science, PubMed, MDPI, and Google Scholar, ensuring comprehensive coverage of the available literature on ozone and electrolyzed water applications in postharvest management. After removing 197 duplicates, 356 unique records were screened by title and abstract. Following eligibility assessment, 77 studies met all inclusion criteria and were retained for qualitative synthesis. The removal of approximately 35.6% of the initially identified records as duplicates highlights the importance of multi-database searching. Each database indexes partially overlapping but distinct journal portfolios, and a single database approach would have risked omission of relevant studies, a concern frequently noted in methodological guidance on comprehensive literature searching. The subsequent reduction from 356 to 77 included studies reflects the stringent application of predefined eligibility criteria. A large proportion of excluded articles focused on non-food applications (e.g., water disinfection, medical sterilization) or failed to report extractable quantitative outcome data, a pattern commonly observed in systematic reviews in food science.

The final corpus of 77 studies encompassed a broad range of food commodities (fruits, vegetables, sprouts, muscle foods, processed products) and food-contact surfaces (stainless steel, polyethylene). Treatment protocols varied widely with respect to sanitizer type, concentration, contact time, and whether agents were applied individually or in combination. This heterogeneity, while precluding a formal meta-analysis, supports a robust narrative synthesis organized by food category, treatment type, and outcome measure. Notably, a smaller proportion of the included studies specifically examined the synergistic or combined use of ozone and electrolyzed water; most investigations focused on individual treatments alone. This observation suggests that while the theoretical basis for hurdle-based synergy is well established, the empirical evidence for combined application remains developing, highlighting a clear direction for future standardized research.

3. PHYSICO-CHEMICAL PROPERTIES AND GENERATION OF SANITIZING AGENT

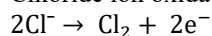
The antimicrobial effect of ozone (O₃) and electrolyzed water (EW) is governed by their physicochemical properties and parameters of on-site generation (Epelle et al., 2023; Lee et al., 2007). Although both sanitizers act through high oxidation reduction potential (ORP), they differ in reaction kinetics, active species, and environmental stability (Lee et al., 2007; Xue et al., 2023). Because the performance of the combined system depends critically on the concentration and speciation of these oxidants at the point of application, a clear understanding of their generation chemistry, stability constraints, and key operating parameters is required for the food sanitation process.

3.1. ELECTROLYZED WATER (EW)

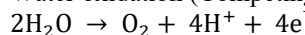
Electrolyzed water is used as an antimicrobial substitute for chlorine-based sanitizers (Rebezov et al., 2022). It is generated by passing an electric current through a dilute salt solution in an electrolytic cell containing a positively charged anode and a negatively charged cathode, typically separated by a diaphragm (Youssef & Hussien, 2020). At the anode, chloride ions undergo oxidation to chlorine gas, which hydrolyzes to form hypochlorous acid (HOCl) and hydrochloric acid. Simultaneously, water oxidation occurs at the anode, while reduction reactions proceed at the cathode. The primary reaction occurs at the anode and cathode poles can be summarised as follows:

At anode:

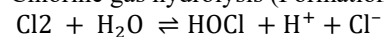
Chloride ion oxidation (Primary chlorine evolution)



Water oxidation (Competing oxygen evolution)

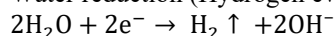


Chlorine gas hydrolysis (Formation of sanitizing species)

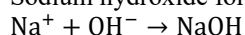


At cathode:

Water reduction (Hydrogen evolution)



Sodium hydroxide formation (Alkaline stream solution)



Resulting EW is classified into four types based on different pH: acidic electrolyzed water (AEW) (pH 2-3), slightly acidic electrolyzed water (SAEW) (pH 5.0-6.5), neutral electrolyzed water (NEW) pH 7-8, and alkaline electrolyzed water (ALEW) (pH 11-13.5). **Error! Reference source not found.** illustrates the pH of various electrolyzed water. Figure 2 Classification of electrolyzed water according to pH and oxidation-reduction potential (ORP), showing acidic, neutral, and alkaline types, along with their corresponding subcategories and indicative ORP ranges relevant to sanitation applications in food

Table 1 summarizes the four types of electrolyzed water.

3.1.1. ACIDIC ELECTROLYZED WATER (AEW)

While migrating to anode Cl^- ions undergone in oxidation process to form chlorine gas (Cl_2). These will react with water to form hypochlorous (HOCl) and hydrochloric acid (HCl). Anode has low pH, high ORP, and free-chlorine concentration values ranging from 30 to 90 ppm (Chen et al., 2016). Due to the low pH (2-3), strong ORP (+950 to +1180 mV), and high HOCl has a strong bactericidal effect (Rahman et al., 2016). While the antimicrobial activity of AEW is desired to have a sanitization effect, some concern related to food quality and nutritional values arises when applied below optimum pH values. Due to the pH values (2-3) of ACEW increase sensitivity of corrosion against the metal surface (Chen et al., 2020; Rahman et al., 2010).

3.1.2. SLIGHTLY ACIDIC ELECTROLYZED WATER (SAEW)

Only tap water and salt (NaCl) is required for generating SAEW in a non-membrane electrolytic chamber (Akther et al., 2023). SAEW has pH range of 5.0-6.5 and ORP values 800-900 mV. The main constitute of SAEW is HOCl which has more disinfection effect. SAEW can be produced quickly; being environment friendly, used in the food industry, and has an effect on a wide range of pathogens (Huang et al., 2021; Yan et al., 2020). SAEW is effective against *Escherichia coli*, *Listeria monocytogenes*, and *Salmonella enterica* (Kim et al., 2019).

3.1.3. NEUTRAL ELECTROLYZED WATER (NEW):

Neutral electrolyzed water (NEW) is generated in a single cell chamber without a diaphragm by partially mixing hydroxyl ions from the anode. It has a pH range of 7-8 and an ORP of +800 to +900 mV, having less corrosive effect on metals or food compared to AEW (Dowidar et al., 2024). Several studies stated that NEW is effective against pathogens like mesophilic anaerobic bacteria (MAB), yeast, and mold (Yamaner, 2021).

3.1.4. ALKALINE ELECTROLYZED WATER (ALEW):

Alkaline electrolyzed water (ALEW) is generated at the cathode side with Na^+ , OH^- and NaOH ions characterized by high pH (11.0 to 13.5) and low ORP (-800 to -900 mV) (Belay et al., 2025). ALEW shows detergent and anti-bacterial properties, but does not create a strong effect like AEW (Athayde et al., 2018). (Tomasello et al., 2021) stated that ALEW is highly effective against Gram-negative bacteria like *Salmonella* spp., *E.coli*. *Staphylococcus aureus* and *Listeria* spp.

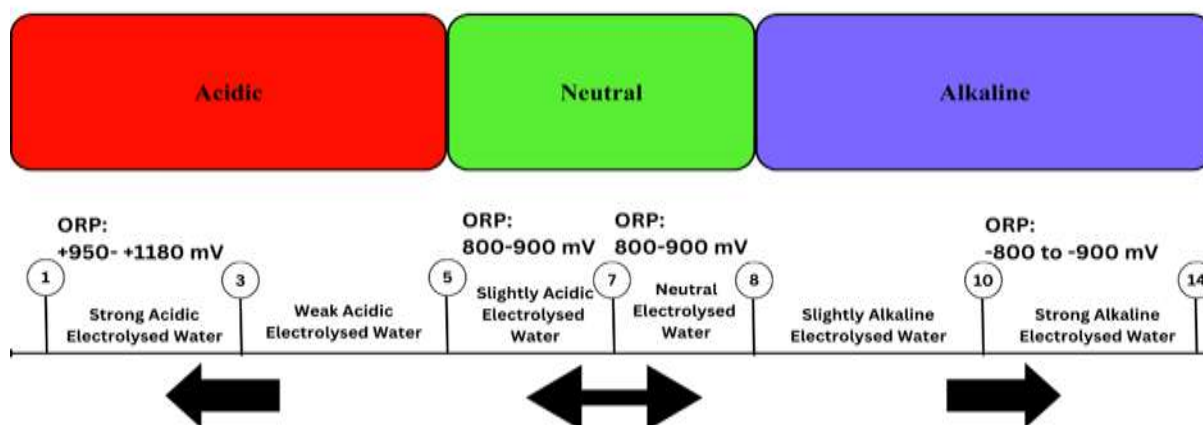


Figure 2 Classification of electrolyzed water according to pH and oxidation-reduction potential (ORP), showing acidic, neutral, and alkaline types, along with their corresponding subcategories and indicative ORP ranges relevant to sanitation applications in food

Table 1 Standardized Electrolyzed Water Classification Table

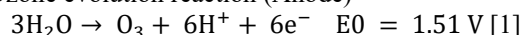
EW Type	pH Range	Unified ORP Range (mV)	Dominant Chemical Species / Properties	Reference
Acidic Electrolyzed Water (AEW)	2-3	+950 to +1180	HOCl ; dissolved Cl_2 , H^+ ions; high corrosion risk	(Rahman et al., 2016)
Slightly Acidic Electrolyzed Water (SAEW)	5.0 – 6.5	+800 to +900	High purity HOCl ; non-corrosive	(Huang et al., 2021)
Neutral Electrolyzed Water (NEW)	7.0 – 8.0	+800 to +900	Mixed HOCl and OCl^- ; stable and metal safe	(Dowidar et al., 2024)

Alkaline Electrolyzed Water (ALEW)	11.0 – 13.5	-800 to -900	NaOH, OH ⁻ ; strong surfactant/detergent properties	(Belay et al., 2025)
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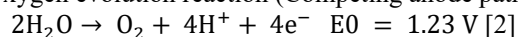
3.2. OZONE (O₃)

Ozone is a highly reactive triatomic oxygen molecule (O₃) with a high oxidation potential of 2.07 V, making it one of the most powerful food-grade sanitizers approved for direct contact with food (**Britton et al., 2020**). It decomposes to oxygen more rapidly, which makes it effective against the major microorganisms of the food industry without leaving toxic residual effects on food or equipment, such as *E.coli*, *Staphylococcus aureus*, *Listeria monocytogenes*, and *Pseudomonas fluorescens* (**de Mendonça Silva & Gonçalves, 2017**). Ozone is generated on-site by corona discharge (electrical discharge) in purified oxygen or dry air, which that passes through a high-voltage electric field, splitting O₂ into atomic oxygen, which subsequently recombines to form O₃. (**de Mendonça Silva & Gonçalves, 2017**). (**da Silva et al., 2023**) reported that the alternative option is electrocatalytic ozone generation technology that will oxidize water to ozone at the anode, as shown in (equation [1]). At acidic condition it is produced by the oxidation of oxygen (equation [2]).

Ozone evolution reaction (Anode)



Oxygen evolution reaction (Competing anode pathway)



The antimicrobial activity of ozonated water depends on ozone solubility in water and reaction stability, both of which are sensitive to operational parameters. Solubility is favoured by low temperature, particularly at (4°C) and under acidic pH, whereas higher temperatures, alkaline conditions, and organic matter accelerate ozone decay and reduce the effective oxidant concentration. Excessive usage of ozone can cause oxidative spoilage, discolouration of the surface, and unfavourable odour (**Bridges et al., 2018**).

Together, the generation chemistry, pH-dependent reactivity, and stability constraints of ozone and electrolyzed water establish the operational framework for their synergistic application. The complementary nature of these sanitizers one pH-tunable and water-derived, the other transient and gas-derived, establishes the mechanistic foundation for the hurdle approach discussed in section 4.

4. MECHANISMS OF ACTION

The antimicrobial activity of ozone and EW system arises from two complementary oxidative pathways that damage microbial cell structure and function. Both sanitizers act through high oxidation-reduction potential (ORP), but they differ in their primary targets and kinetics, which provide the basis for their synergistic use as a hurdle technology (**Kovač et al., 2023**).

4.1. ANTIMICROBIAL ACTION OF OZONE - MECHANISM

Ozone inactivates microorganisms through direct oxidation of the membrane and indirectly by generating reactive oxygen species (ROS) (**Patil et al., 2011**). Direct oxidation involves electrophilic attack of molecular O₃ (oxidation potential 2.07eV) on unsaturated lipids, structural protein, DNA and RNA in the cell membrane (**Casas et al., 2021**). Simultaneously, indirect oxidation occurs via ozone decomposition into ROS such as hydroxyl radicals, superoxide, and hydrogen peroxide which propagate intracellular oxidative cascades. Due to the combined effect of ozone and the free radicals, several effects like protein oxidation, enzyme disruption, and disruption of the cell membrane occur, ultimately causing cell lysis (**Rangel et al., 2021**). Figure 2 indicates the factors affecting the efficiency of microbial inactivation during aqueous and gaseous ozonation.

Ozone decontamination effectiveness is dependent on various other factors, categorised into two: extrinsic (concentration of ozone and the presence of organic materials that consume ozone in the decontamination environment) and intrinsic (growth stage, cell envelope, and the efficiency of the microorganism's repair mechanisms) (**Drazic et al., 2013**). Microorganisms form biofilms to protect themselves from any adverse conditions and are more resistant than in their planktonic state. In the food industry, bacterial biofilms are mostly in contact with, or without, food and may cause a source of contamination of foodstuff by spoilage (**González-Rivas et al., 2018**). In recent years, action against biofilm-embedded microorganisms has also been confirmed. In practice, aqueous ozone often acts more quickly on wet surfaces, whereas gaseous ozone may require higher concentration or longer contact time to achieve similar microbial reduction (**Panebianco et al., 2022**).

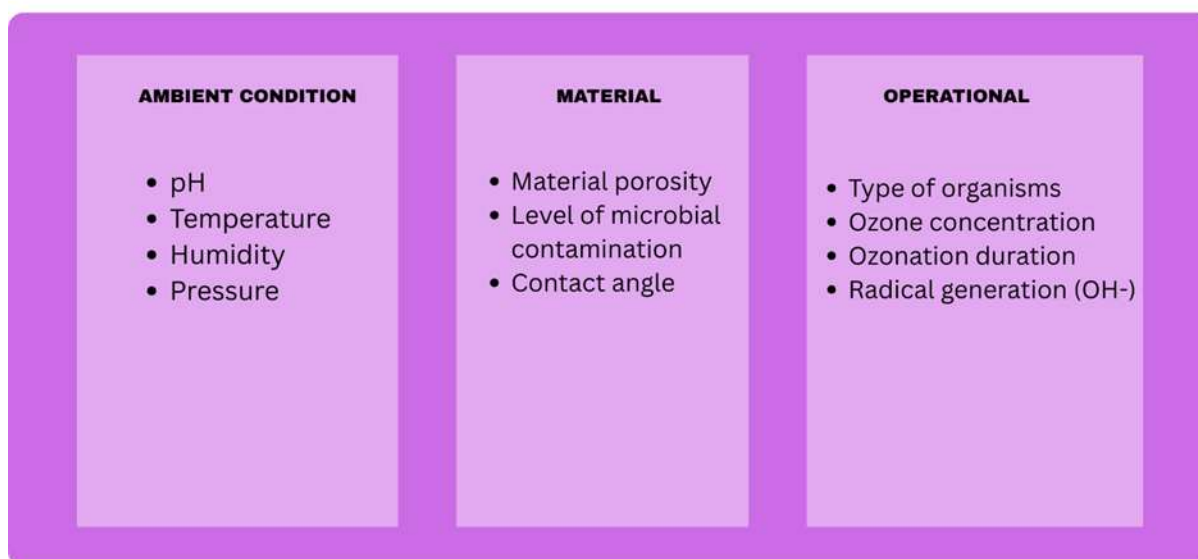


Figure 2 Schematic representation of the major environmental, material-related, and operational variables that affect the antimicrobial performance of ozone treatment in food sanitation applications.

4.2. ANTIMICROBIAL ACTION OF ELECTROLYZED WATER - MECHANISM

Electrolyzed water is a potent antimicrobial agent characterized by high oxidation-reduction potential (ORP), a low pH, and free chlorine, primarily hypochlorous acid (HOCl). (Ogunniyi et al., 2019) stated that HOCl and reactive oxygen species (OH, O₃, H₂O₂) are effective against microorganisms. This HOCl is more effective than ClO⁻ for microbial cell wall penetration and oxidize protein, particularly cystine and methionine-containing enzymes, leading to rapid loss of function and cell death (Wang et al., 2023). Electrolyzed water (EW), particularly acidic electrolyzed water (AEW; pH 2-3, ORP 950-1180 mV), has also been shown to damage biofilms by disrupting extracellular polymeric substances (EPS) and reducing embedded cell population. However, its activity can be reduced by organic load and variations in pH, chlorine concentration, and exposure time (Han et al., 2017). Therefore, the true efficacy of EW depends not only on formulation but also on the contamination environment and the nature of the target microorganism.

4.3. HURDLE EFFECT AND SYNERGY

The hurdle effect refers to the combination of several techniques to ensure microbial safety and stability, thereby maintaining food safety and quality while using a lower dosage of individual treatments. The synergistic effect is achieved by combining the ozone and electrolyzed water from their complementary mechanisms. Combining aqueous ozone (O₃) and electrolyzed water (EW) creates a multi-layered defence system that is more effective than using either agent alone. Ozone may weaken the cell surface first, increasing permeability and making cells more vulnerable to HOCl or other oxidative agents in EW (Hong et al., 2024; Jia et al., 2023). Likewise, combining EW with UV or ozone with chlorine has been reported to improve log reduction, especially against fungal spores and surface contaminants.

For example, combined ozone-chlorine treatment achieved higher spore reduction for *Aspergillus niger* and *Trichoderma harzianum* than individual treatments (Liang et al., 2021), and slightly acidic electrolyzed water combined with UV showed stronger activity against *Staphylococcus aureus* than single treatments (Zhang et al., 2023). (Panebianco et al., 2024) evaluates the sanitization effect on food-contacting surfaces, for example, stainless steel and other utensils. Furthermore, combined treatments of electrolyzed water, ozone and ultraviolet (UV) help to maintain the overall quality of produce (Jemni et al., 2014b). The advancement of hurdle technology by EW and ozone, along with some other non-thermal technique represent advancement in food safety and prolongs the shelf life of fresh produce and other food products (Rahman et al., 2022). However, the comparative value of these combinations is difficult to generalize because studies vary widely in target organism, contact time, surface type, and contamination level. More standardized, produce-specific validation studies are still needed before firm commercial recommendations can be made. Figure 3 illustrates the antimicrobial mechanisms of ozone and electrolyzed water

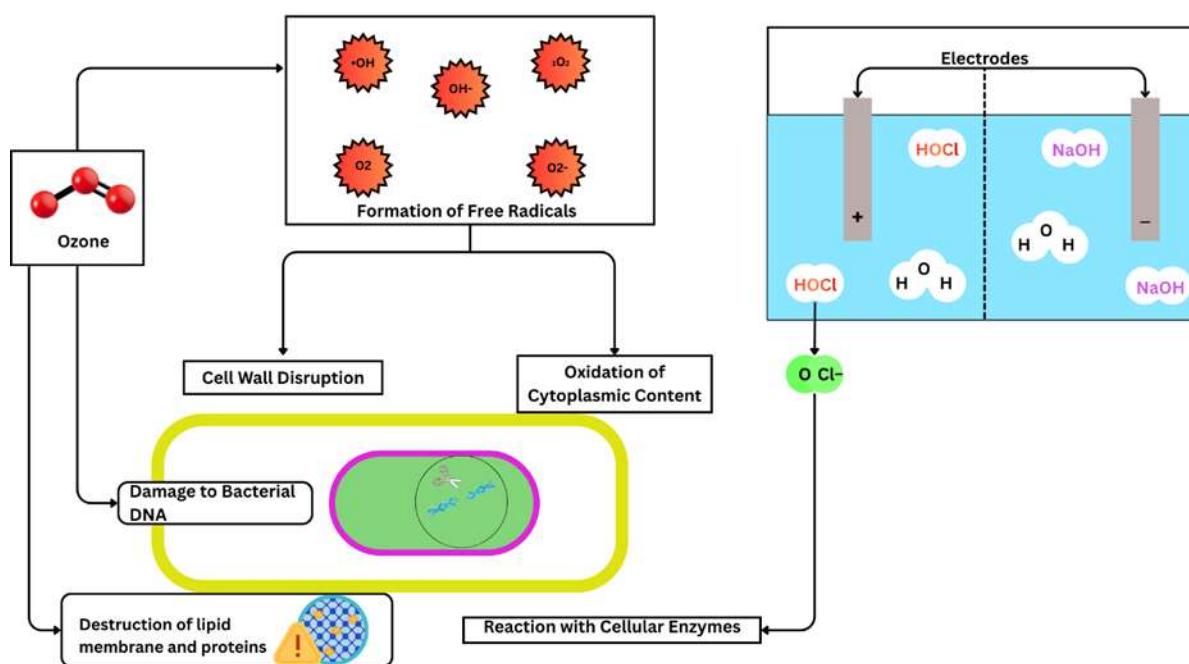


Figure 3 Proposed antimicrobial mechanisms of ozone and electrolyzed water in food sanitation, including free-radical formation, cell wall disruption, oxidation of cytoplasmic components, damage to bacterial DNA, lipid membrane and protein destruction, and reaction with cellular enzymes. The figure also illustrates the generation of hypochlorous acid and sodium hydroxide during the production of electrolyzed water.

5. APPLICATION IN FRESH HORTICULTURAL PRODUCE

Fresh horticultural produce differs in surface morphology, natural microflora, and organic load; the performance of ozone (O_3) and electrolyzed water (EW) should be evaluated as commodity-specific sanitizers rather than treated as one-size-fits-all solutions. Across the literature, sequential or combined O_3 -EW treatments often achieve 2-4 log CFU/g reduction of foodborne pathogens while maintaining postharvest quality (Koseki et al., 2001; Pang & Hung, 2016).

Table 2 summarizes the antimicrobial efficacy of ozone and electrolyzed water against foodborne pathogens and spoilage organisms. However, the magnitude of benefit varies with commodity, treatment order, contact time, and storage conditions, so the evidence supports a context-dependent rather than universal advantage.

Leafy vegetables provide some of the clearest evidence for the use of combined O_3 -EW treatments because their uneven surfaces make it easier for microbes to attach. Sequential application of slightly acidic electrolyzed water (SAEW) and ozonated water achieved approximately 3-log CFU/g reductions of *Salmonella enterica* on iceberg lettuce, exceeding the efficacy of either sanitizer used alone (Dev Kumar & Ravishankar, 2019). Comparable improvements were reported on fresh-cut cilantro, where acidic electrolyzed water and aqueous ozone lowered microbial population while still preserving sensory quality during refrigerated storage (Wang et al., 2004). These findings suggest that combined oxidative treatments are more effective than individual sanitizers for leafy vegetables, although direct comparisons across studies are limited by variation in sanitizer concentration and exposure time.

In contrast, more complex and heterogeneous matrices like alfalfa seeds and sprouts present protective microenvironments that hinder antimicrobial efficacy. Transitioning from SAEW to Acidic Electrolyzed Water (AEW) sequenced after aqueous ozone is mandatory to match comparable or greater reductions (≥ 3.0 log CFU/g) against *Salmonella* and Shiga toxin-producing *Escherichia coli* (STEC) (Mohammad et al., 2020). The higher oxidation-reduction potential of AEW (ORP $> +950$ mV) enables more effective disruption of organic matter and microbial shielding within these intricate surface structures (Rahman et al., 2016). Similarly, blueberries exhibited better microbial control when electrolyzed water and ozone were integrated with ultraviolet treatment, resulting in about a 3.5 log reduction of *E. coli* O157:H7. This suggests that commodities possessing protected microbial niches may require multi-hurdle approaches rather than oxidative sanitizers alone (Kim & Hung, 2012). Beyond microbial inactivation, preserving quality is essential for commercial use. Research on grapes, dates, cherry tomatoes, and clementines shows that well-optimized O_3 -EW treatments can help maintain firmness, soluble solids, colour, and vitamin C while still limiting microbial growth (Islam et al., 2018; Jemni et al., 2014a; Laureano et al., 2016; Yaseen et al., 2016). More recently, applying electrolyzed water followed by ozone was also found to speed up the breakdown of mancozeb residues on grapes more than either treatment alone, suggesting that combined oxidative treatments may improve both food safety and pesticide residue control (Wen et al., 2024). Overall, the evidence suggests that sequential ozone-electrolyzed water treatment can provide wider antimicrobial activity than either method used alone, while also helping preserve produce quality. However, the

evidence is only moderate because studies differ in ozone dose, chlorine level, treatment time, inoculum load, storage conditions, and how quality is assessed. For this reason, well-standardized comparative trials across a range of horticultural commodities are still needed before broad industrial recommendations can be made.

Table 2 Antimicrobial efficacy of ozone and electrolyzed water applied individually or in combination against foodborne pathogens and spoilage organisms (Al-Khalili et al., 2025)

SUBSTRATE	TREATMENT	TARGET ORGANIS	ANTIMICROBIAL OUTCOME	REFERENCE
Fungal Spores (in vitro)	Aqueous O ₃ + Active Chlorine contact time: 10 mins	Aspergillus niger and Trichoderma harzianum spores	0.47-0.55 log reduction	(Liang et al., 2021)
Alfalfa Seeds and sprouts	Sequential O ₃ then AEW	Salmonella and E. coli (STEC)	>3.0 log CFU/g	(Mohammad et al., 2020)
Blueberry calyx	Triple hudle O ₃ + EW + UV light	Escherichia coli O157:H7	~3.5 log CFU/g	(Kim & Hung, 2012)
Iceberg Lettuce	SAEW followed by Ozonated Water	Salmonella enterica ser. Newport	~ 3.0 log CFU/g	(Dev Kumar & Ravishankar, 2019)

6. IMPACT ON FOOD QUALITY AND SENSORY ATTRIBUTES

The commercial viability of sanitization technologies depends not only on antimicrobial efficacy but also on the preservation quality. Although ozone and electrolyzed water can each cause quality defects, such as oxidative bleaching, tissue softening, or off-colour development, their combined use often achieves equal or greater microbial reduction at lower individual doses, thereby minimizing these side effects (Roobab et al., 2023). Across commodities, the O₃-EW combination has shown promising effects on texture, colour, nutritional quality, residue removal, and sensory attributes, although the magnitude of benefit varies with produce type, treatment sequence, and exposure intensity. The effect of ozone and EW application on fresh produce quality is mentioned in Table 3.

Texture retention is one of the most consistent benefits reported for combined treatments. Grapes exposed to continuous ozone followed by electrolyzed water showed greater skin firmness without affecting fruit integrity (Laureano et al., 2016), while dates treated sequentially with UV-C, ozone, and EW maintained firmness better during storage than untreated fruit (Jemni et al., 2014a). Together, these findings indicate that lowering the severity of individual oxidative treatments while still taking advantage of their synergistic antimicrobial effects can help limit tissue softening. However, variations in cultivar, storage temperature, and firmness measurement techniques still make it difficult to compare results quantitatively across commodities. Colour stability and nutrient retention are also affected by how intense the treatment. In cherry tomatoes, ozonated water slowed respiration, lowered ethylene production, and helped preserve vitamin C during storage (Islam et al., 2018), while combined ozone and EW helped clementines retain soluble solids and maintain their external appearance (Yaseen et al., 2016). Similar results in dates suggest that, when properly optimized, oxidative treatments can improve microbial safety without causing undesirable bleaching or discolouration. Even so, using standardized colour metrics such as L*, a*, and b* values or consistent nutritional indicators makes cross-study comparison difficult. Combined O₃-EW treatment can offer advantages beyond microbial reduction. In grapes, applying electrolyzed water before ozone was reported to speed up the breakdown of mancozeb residues more than either treatment used alone (Wen et al., 2024), suggesting that this approach may help improve both microbial safety and pesticide residue control at the same time. Even so, pesticide degradation has been studied far less than microbial inactivation, and only a few studies assess residue removal alongside sensory quality, nutritional value, and storage behaviour.

Overall, the evidence suggests that sequential ozone-electrolyzed water treatment can achieve a good balance between microbial safety and postharvest quality. Even so, the strength of the evidence is still moderate because studies differ widely in sanitizer dose, available chlorine level, ozone production method, contact time, storage conditions, and commodity type. Future work should use standardized protocols that measure microbial reduction, shelf-life extension, colour, firmness, nutritional quality, sensory acceptability, and economic feasibility together so that different horticultural commodities can be compared more meaningfully.

Table 3 Impact of ozone and electrolyzed water treatments on the shelf life, microbial quality, and physicochemical attributes of fresh produce and food products

PRODUCE	TREATMENT METHOD	MICROBIAL REDUCTION	QUALITY OUTCOME	REFERENCE
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Dates	UV-C + Ozonated Water or EW	lowest counts for coliforms and total mesophilic bacteria	Highest firmness and overall quality	(Jemni et al., 2014a)
Cherry tomatoes	Ozonated Water (O ₃)	Reduced microbial load	Higher vitamin C; delayed ripening	(Islam et al., 2018)
Cilantro	Aqueous O ₃ followed by Acidic EW (AEW)	Effective initial microbial reduction	maintained low microbial growth during 14-day storage at 0°C	(Wang et al., 2004)
Clementines	Electrolyzed Water + Ozone exposure	Reduced microbial contamination	Maintained soluble solid content (SSC) during storage at 4°C	(Yaseen et al., 2016)
Grapes	EW + Ozone (gas exposure and immersion)	Significant reduction	Increased skin firmness; anthocyanins retained	(Wen et al., 2024) (Laureano et al., 2016)
Iceberg lettuce	(SAEW) followed by Ozonated Water	~ 3 log CFU	Good texture retained	(Dev Kumar & Ravishankar, 2019)
Blueberries	O ₃ + EW + UV	~ 3.5 log CFU	Acceptable quality	(Kim & Hung, 2012)

7. CHALLENGES AND FUTURE PROSPECTS

The combined use of ozone (O₃) and electrolyzed water (EW) has emerged as an effective non-thermal sanitation strategy for fresh horticultural produce. However, current evidence suggests that its efficacy is highly dependent on commodity characteristics and processing parameters rather than representing a universally applicable treatment. Comparative studies indicate that leafy vegetables such as iceberg lettuce and cilantro typically achieve microbial reductions of approximately 2-4 log CFU/g while maintaining acceptable sensory quality following sequential ozone-EW treatments **(Pang & Hung, 2016; Wang et al., 2004)**. In contrast, commodities with more complex surface morphologies, including alfalfa sprouts and blueberries, generally require greater oxidant exposure or integration with additional hurdle technologies, such as ultraviolet treatment, to achieve comparable microbial reduction **(Kim & Hung, 2012; Mohammad et al., 2020)**. These observations highlight the critical role of surface morphology, natural wax layers, and organic load in determining sanitizer performance and should form the basis for commodity-specific sanitation strategies rather than generalized applications.

A key limitation in the existing literature is the substantial variability in treatment conditions across studies. Ozone concentration, available chlorine content, type of electrolyzed water (AEW, SAEW, or NEW), oxidation-reduction potential (ORP), treatment sequence, exposure time, microbial inoculum, storage temperature, and quality assessment methods differ considerably **(Adal et al., 2024; Rahman et al., 2016)**. Consequently, although many studies reports improved microbial inactivation and quality preservation, the absence of standardized experimental protocols limits evidence synthesis and prevents the establishment of optimized treatment recommendations for different horticultural produce. Future research should therefore prioritize harmonized experimental designs that simultaneously evaluate microbial inactivation, shelf-life extension, physicochemical and nutritional quality, sensory acceptance, and economic feasibility under comparable conditions.

The proposed synergistic antimicrobial mechanisms of O₃-EW treatment also require further experimental validation. Current evidence suggests that ozone disrupts microbial cell membranes, thereby facilitating the penetration of hypochlorous acid (HOCl) produced by electrolyzed water, thereby intensifying oxidative damage compared to individual treatments **(Hong et al., 2024; Wang et al., 2023)**. However, this hypothesis is largely supported by indirect microbiological evidence. Future research should employ advanced analytical approaches including confocal laser scanning microscopy, electron microscopy, atomic force microscopy, fluorescent membrane probes, and proteomic analyses, to verify membrane disruption, intracellular oxidative damage, and biofilm removal under practical postharvest conditions. Another significant challenge is to rapid depletion of oxidative sanitizers in the presence of organic matter. Organic residues released during washing can consume ozone and available chlorine, thereby reducing antimicrobial and increasing sanitizer demand in commercial systems **(Han et al., 2017; Panebianco et al., 2024)**. This effect may partially explain the reduced performance observed in produce with rough or waxy surfaces compared to leafy vegetables. Future studies should investigate sanitizer dynamics under realistic wash-water conditions and develop adaptive control systems to maintain effective oxidant levels during continuous processing. Food safety consideration should extend beyond microbial inactivation to include the potential formation of disinfection by-products and environmental impacts. While ozone and electrolyzed water individually generate minimal residues, their combined use may promote the formation of reactive byproducts such as bromate and trihalomethanes under specific water chemistry conditions **(Liang et al., 2021)**. Comprehensive residue monitoring, toxicological assessment, and wash-water management strategies are therefore essential to ensure consumer safety and environmental sustainability. Additionally, improved approaches to water reuse, oxidant stability, and water management to enhance the sustainability of these green technologies.

Regulatory challenges also pose barriers to the widespread adoption of ozone-EW systems. Although ozone is classified as Generally Recognized As Safe (GRAS) for food application in the United States, regulatory frameworks governing ozone, electrolyzed water, residual oxidant, and by-products vary significantly across regions (**European Food Safety Authority (EFSA), 2023; Rice & Graham, 2001**). The lack of harmonized international standards complicates commercialization and may limit global acceptance. Generating standardized efficacy and safety data under commercial conditions will be critical to support regulatory approval and facilitate global adoption. Economic and engineering consideration remain equally important for large-scale implementation. Although ozone and electrolyzed water can reduce dependence on conventional sanitizer and minimize chemical residues, the capital investment, equipment maintenance, energy consumption, and process integration, which may hinder adoption, particularly among small-and medium-scale enterprises (**Artés et al., 2009; da Silva et al., 2023**). Future techno-economic studies should compare ozone-EW systems with conventional sanitation methods, including assessments of operational costs, return on investment, water consumption, energy efficiency, and environmental impacts through life-cycle analysis.

Future advancements should focus on integrating ozone-EW treatments with complementary non-thermal approaches, including ultraviolet light, ultrasound, cold plasma, and automated Clean-in-Place (CIP) systems. Studies on blueberries and dates fruits suggest that multi-hurdle approaches can achieve superior microbial control while preserving product quality compared to single treatments (**Jemni et al., 2014a; Kim & Hung, 2012**). In addition, the incorporation of Artificial Intelligence, computational fluid dynamics, and real-time monitoring of ORP, pH, available chlorine, and microbial load could enable dynamic optimization of sanitizer dosage according to commodity characteristics and processing conditions, thereby enhancing efficiency and reducing chemical usage (**National Institute of Food and Agriculture, 2021**). In summary, ozone- electrolyzed water treatment represents a promising and environmentally sustainable strategy for postharvest sanitation of fresh produce. However, its successful commercialization will depend on commodity-specific optimization, standardized validation protocols, mechanistic confirmation, comprehensive safety assessment, economic feasibility, and regulatory harmonization. Addressing these knowledge gaps will support the development of robust, evidence-based sanitation systems capable of improving food safety, maintaining postharvest quality, and advancing sustainable horticultural production.

CONCLUSION

The combined use of ozone and electrolyzed water represents a promising non-thermal and environmentally friendly sanitization technique for fresh foods and food processing environments. By using complementary oxidative mechanisms, this hurdle approach enhances microbial inactivation, including in biofilm-forming systems, while allowing lower treatment intensities that help reduce ozone-induced bleaching and the corrosive off-flavour effects associated with some electrolyzed water formulations. This strategy has shown strong potential for improving pathogen control, extending shelf life, and removing chemical residue from a wide range of products and food-contact surfaces.

Its advantages, including on-site generation, low chemical residue, and compatibility with sustainable processing systems, make it an attractive and reliable alternative to conventional sanitization techniques. However, broader industrial adoption will require further research to address process stability, organic load inference, scale-up challenge, and equipment compatibility. Overall, the ozone-electrolyzed water combination offers a compelling green sanitation option with significant promise for safer food, reduced environmental hazard, and improved post-harvest and processing efficiency.

ETHICS DECLARATION

This article is a review and did not involve experiments on humans or animals, so ethical approval was not required.

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CONFLICT OF INTEREST

The authors declare that they have no conflicts of interest.

AUTHOR CONTRIBUTIONS

Author contributions: Divyakshi Sawant, Keisar Lourdasamy: Writing of original draft and conceptualization. Divyakshi Sawant, Keisar Lourdasamy, C. Indu Rani: Revision of draft, inclusion of tables and figures, proofreading.

Divyakshi Sawant, Keisar Lourdasamy, C. Indu Rani, K.B Sujatha, K. Gurusamy: Revision, formatting and Supervision. All the authors read and approved the final version of the manuscript.

DATA AVAILABILITY STATEMENT

Data supporting this study are available within the manuscript.

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