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Applications of water activated by ozone, electrolysis, or gas plasma for microbial decontamination of raw and processed meat

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A raw or processed meat product can be a breeding ground for spoilage bacteria (*Enterobacteriaceae*, *Lactobacillus* spp., *Pseudomonas* spp., etc.). Failure of decontamination results in food quality loss and foodborne illnesses caused by pathogens such as *Salmonella*, *Escherichia coli*, *Staphylococcus aureus*, and *Listeria monocytogenes*. Often, meat processors decontaminate the carcass using cheap chemicals or artificial antimicrobial agents not listed on the ingredient list, which is discouraged by health-conscious consumers. Foods with clean labels became more popular during the COVID-19 pandemic, which led consumers to choose healthier ingredients. Novel methods of controlling or improving meat safety are constantly being discovered. This review focuses on novel means of electrochemically activate water that is being investigated as a sanitizing agent for carcasses and processing area decontamination during production or at the end. Water can be activated by using non-thermal techniques such as ozonation, electrolysis, and cold plasma technologies. Recent studies showed that these activated liquids are powerful tools for reducing microbial activity in raw and processed meat. For instance, plasma-activated water can be used to enhance microbiological safety and avoid the negative effects of direct gaseous plasma on the organoleptic aspects of food products. In addition, electrolyzed water technology offers hurdle enhancement by combining with non-thermal strategies that have great potential. Ozonation is another way of activating water which provides a very convenient way to control microbiological safety and finds several recent applications as aqueous ozone for meat decontamination. These solutions are highly reactive and convenient for non-conventional applications in the meat industry related to food safety because of their antimicrobial or antiviral impact. The present review highlights the efficacy of activated-water decontamination of raw and processed meat via non-thermal solutions.

KEYWORDS

ozone, electrolyzed water, cold plasma, meat quality and safety, *Salmonella*, *Escherichia coli*, *Staphylococcus aureus*, *Listeria*

1. Introduction

Food safety and demand for cleaner labels continue to be an ongoing concern with consumers. Raw and minimally processed meat contains common foodborne pathogens such as *Listeria monocytogenes* (Zhang et al., 2020), *Salmonella Enteritidis* (Garrido-Maestu et al., 2019), *Escherichia coli* O157: H7 (de Assis et al., 2021), *Campylobacter* (Thomas et al., 2020) and *Staphylococcus aureus* (Ribeiro et al., 2018), which contributes to foodborne illnesses. The risk of microbial spoilage in raw, minimally processed, and ready-to-eat meat products is associated with temperature variations and extended distances of distribution. The solution is to apply an appropriate technology to tackle these issues or add some synthetic chemicals or artificial antimicrobial agents such as butylated and hydroxyanisole, butylated hydroxytoluene, tert-butyl hydroquinone, propyl gallate, phosphate, nitrate, and nitrite which are always discouraged by health-conscious consumers (Roobab et al., 2021). However, it is technically impossible to produce a bacteria-free, fresh-meat supply.

The meat industry continues tirelessly researching new strategies that reduce bacteria to as close to zero as possible. Traditional meat processing line usually involves cleaning and disinfection steps to reduce pathogens (Smith et al., 2015). Some of the most common conventional mild processing technologies used for the decontamination of commercial meat products include physical treatments, i.e., washing (Sá Júnior et al., 2021), steaming (Dixon et al., 2019), chemical treatments, i.e., organic acids (Heir et al., 2022), chlorinated chemicals (Kocharunchitt et al., 2020), hydrogen peroxide (Walsh et al., 2018), phosphate-based compounds (Sallam L. et al., 2020). Sodium hypochlorite is one of the most used, efficient, available, and low-cost disinfectants. Moreover, chlorine dioxide and lactic acid have broad antibacterial effects (Burfoot et al., 2015). In the meat industry, chlorinated water (0.5–1 ppm) is used to reduce microorganisms, prevent cross-contamination, and ensure carcass safety. Several research works revealed that chlorine efficacy is dependent on its concentration; however, its by-products such as trihalomethanes, haloacetic acids, and chloramines have carcinogenic potential (Gil et al., 2016). Similar is the case with chemical sanitizers which produce toxic by-products that are harmful to human health and the environment. Moreover, secondary rinsing with potable water may be needed to remove these acids when higher concentrations are used. This could be a challenge where potable water is scarce (Van Schalkwyk and Hoffman 2016). Besides, several pathogens such as *Salmonella* are becoming resistant to these traditional disinfectants (Youn et al., 2017; Cadena et al., 2019). Furthermore, the efficiency of chemical sanitizers depends on free chlorine availability, organic material load, microbial load, washing water quality, etc. (Murray et al., 2017). Nowadays, acetic, peroxyacetic acid, and lactic acid is more common for treating meat as a tool to improve meat safety (Nkosi et al., 2021).

Expanding applications for the existing decontamination technologies continue to be constantly studied and investigated by industry, technology suppliers, and academia. Currently, non-thermal interventions are being considered by the meat and poultry industries as alternative technologies. For instance, functionalized water as an environmentally clean label technology is a less expensive and safer technique as compared to traditional chemical sanitizers. The functionally activated water produced by using electricity

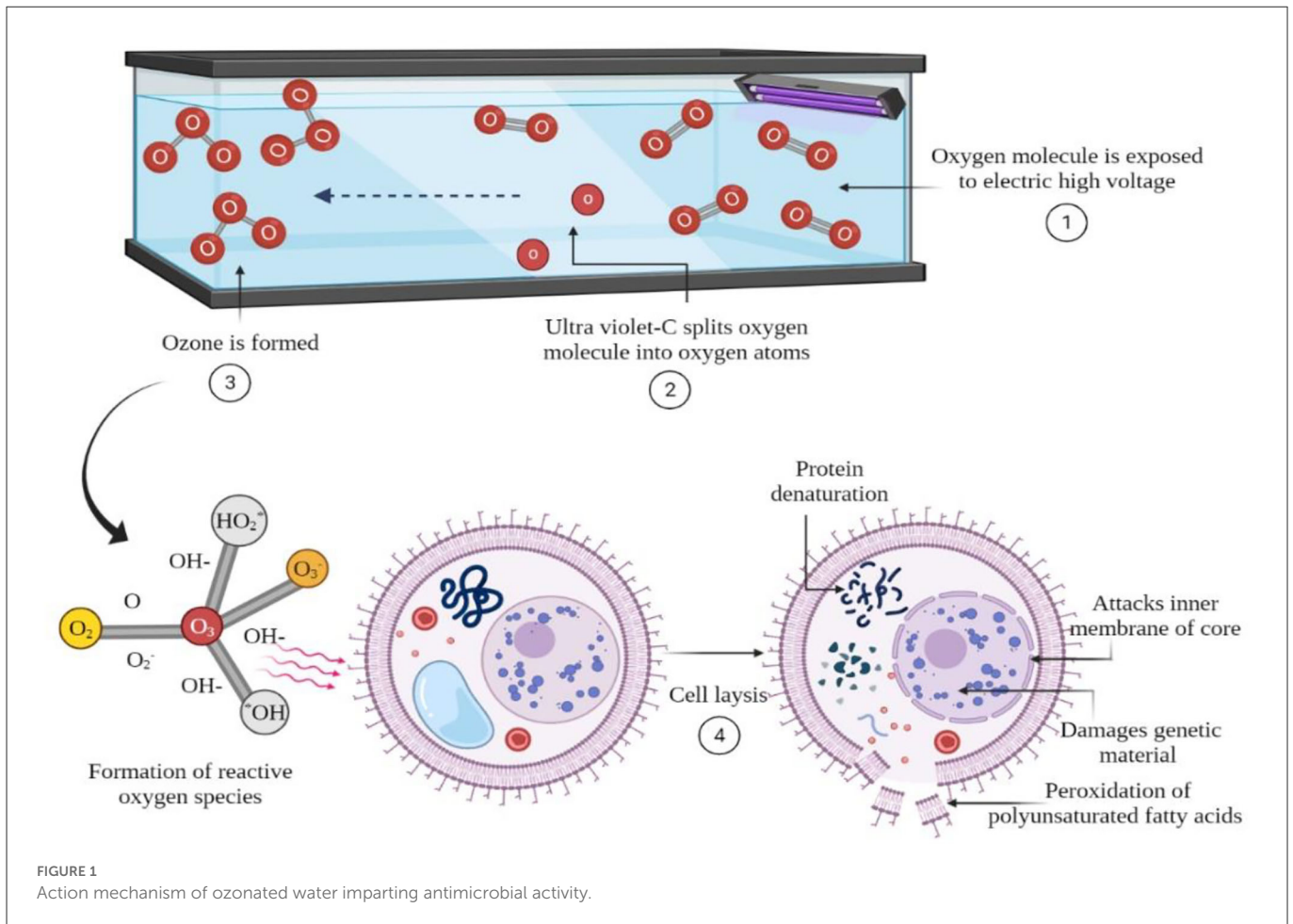
(Du et al., 2016; Iram et al., 2021; Moghassem Hamidi et al., 2021) ozone, (Fathul Karamah and Wajdi, 2018; Kanaan, 2018; Kalchayanand et al., 2019), and cold plasma technology (Liao et al., 2018; Yong et al., 2018; Inguglia et al., 2020; Zhao et al., 2020; Wang et al., 2021) have become a popular research topic. The antimicrobial effectiveness of electrochemically activated water is not only depending on its constituents but also on its oxidative power which accepts the electrons because of the highly deficient electron clusters. As explained by Aider et al. (2012) anode/solution interface produced anolytes (oxidizing agents), which contain a mixture of free radicals (Cl^* , O^* , OH^* , HO_2^*) having strong antimicrobial activities. However, the electrochemical activation of the anode reduced the surface tension of the electro-activated medium, increased electric conductivity, and modified the water structure. Water has been utilized for large-scale washing applications, which can be electrolyzed or chemically functionalized to be used for meat safety purposes. Considering the popularity of ozonated water, electrolyzed water, and cold plasma water, their recent applications for meat safety are reviewed. The research on novel non-thermal applications is ongoing, and the potential of electrochemically activated water for ensuring meat safety is being greatly explored. This review covers different non-thermal sources employed for water activation and the advancement of activated electrochemical solutions as the sanitizing agent in various meat products.

2. Activated water as enhancing microbiological safety

2.1. Ozonated water

Ozone is a Generally Recognized as Safe (GRAS) food processing aid, which has a powerful disinfectant and a strong sanitizing ability as compared to other chemical sanitizers. Current usage of ozone has proven effective antimicrobial agent against microbial populations that leaves no toxic residues on food or equipment, making it a greener environment-friendly technology (Pandiselvam et al., 2020; Roobab et al., 2022). Generally, low temperature (4°C) and pH of aqueous medium increased the solubility of ozone and enhanced its effectiveness. Hence, cool, damp, and refrigerated storage conditions for meat products facilitate the use of ozone in meat processing industries. However, excessive usage can produce oxidative spoilage, undesirable odors, and surface discoloration (Bridges et al., 2018). Moreover, its antimicrobial efficacy depends on the solubility in water and the reaction stability with organic and inorganic compounds. Other critical parameters affecting the antimicrobial efficiency of ozone include pH, temperature, and organic residues in food (Bridges et al., 2018). Ozonation is another way of activating water, accomplished by passing gas through ultraviolet irradiations (188 nm wavelength) and corona discharge that excites the oxygen electrons (Figure 1).

The electrochemical process in water generates high energy (around 6–7 eV) that breaks O–O bond and form ozone. The unstable molecule of ozone can decompose into superperoxide, hydroxyl, and hydroperoxyl radicals which have a strong oxidizing ability (2.07 mV oxidizing potential) (Gonçalves 2009). These active radicals can drive a series of chemical reactions when they encounter the bacterial cell. Ozone (2.07 V) causes peroxidation of phospholipids (present in the



bacterial cell wall), leading to the cell wall decomposition and leakage of intracellular components (Pandiselvam et al., 2017; Rudolphi-Skórska et al., 2017). The continuous degradative reactions alter the cellular integrity which could be a reason for cell death. However, the ozone efficacy responds differently to Gram-positive and Gram-negative bacteria due to the difference in the cell wall structure such as wavy or smooth cell wall, thick or thin peptidoglycan layer, presence, and absence of outer membrane and periplasmic space (Dizengremel et al., 2009; Sheng L. et al., 2018). Besides, ozone modified the primary structure of proteins and enzymes, thus inactivating several cytosolic enzymes (Dizengremel et al., 2009). Furthermore, secondary reactive species modified purines and pyrimidines (the building blocks of DNA) and damaged the genetic material of the host cell (Ito et al., 2005). However, it also altered the structure of prokaryotic plasmid DNA (Asfahl and Savin, 2012).

2.1.1. Decontamination of meat

Kalchayanand et al. (2019) evaluated a liquid ozone spray chill technique (at 4.6–5.6°C) to inactivate *E. coli* O157:H7 on surfaces of fresh beef as an alternative to traditional water sprays chill. Results showed 1.46 logs of *E. coli* O157:H7 reduction and 0.99 logs of aerobic bacteria reduction. Ozonated spray treatment caused sublethal injury to the target bacterial cells, which retarded their growth under highly stressed conditions such as low-temperature storage. Similarly, the

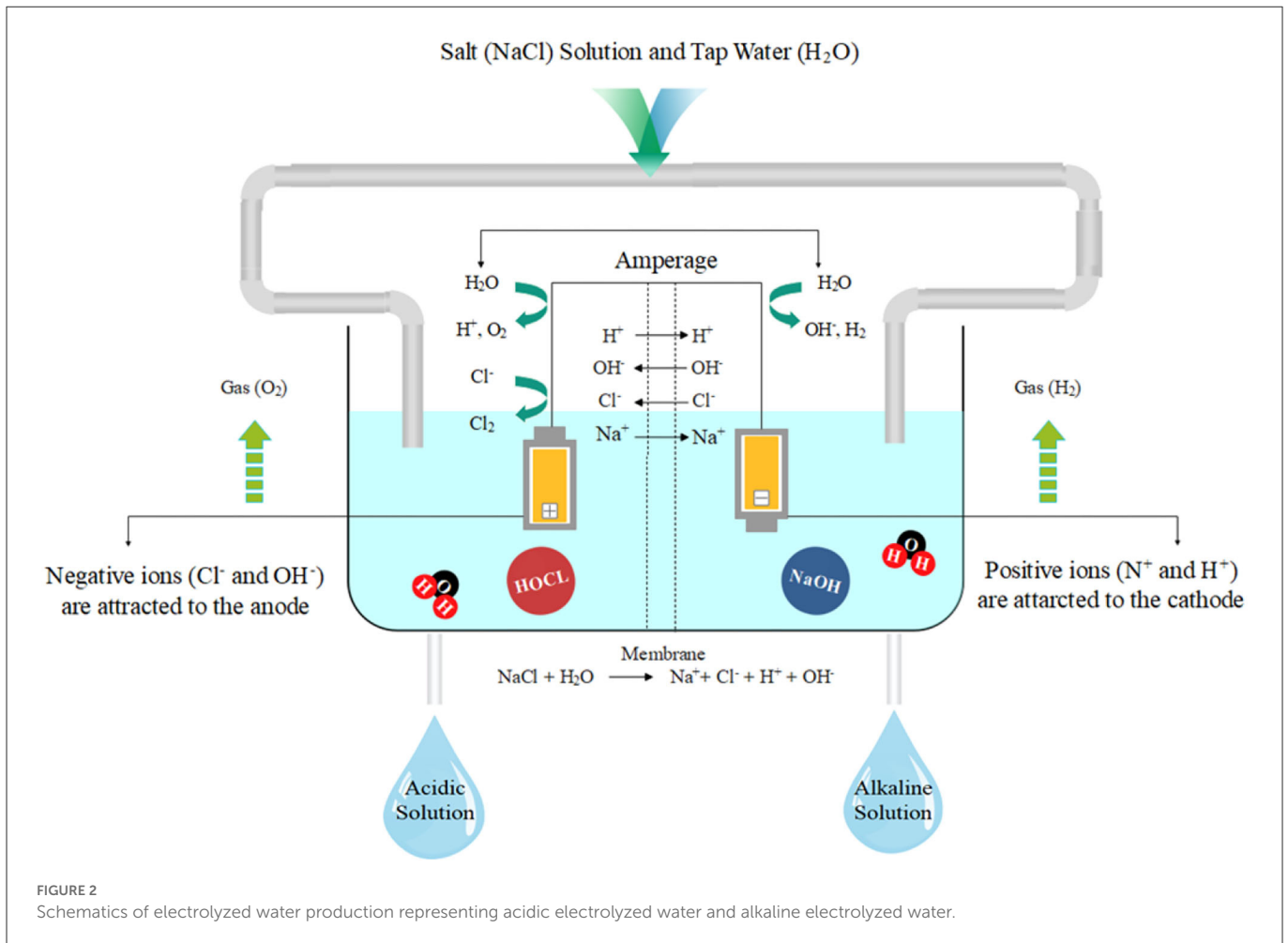
treatments of aqueous ozone (0.5 ppm for 45 min) on *S. aureus* in fresh and frozen chicken meat exhibited 2–4 logs CFU/mL reduction (Kanaan, 2018). The underlying mechanism has been explained by Aponte et al. (2018) who observed the modifications in cell membrane permeability as ozone may attack glycoproteins, glycolipids, and other amino acids (structural components of cells) which inhibit the enzymatic reactions and interrupt the growth and functionality of bacterial cells (Sheng X. et al., 2018). Table 1 lists the parameters used in ozonation to effectively decontaminate raw and processed meat.

The impact of ozonated water is not only limited to fresh carcasses but also effective for processed meat products. Botta et al. (2018) investigated the complexity and dynamics of aqueous ozone treated (6 mg/L for 90 s at 4°C) beefsteaks microbiota using RNA-based amplicon sequencing. *Pseudomonas fragi* were prominent in treated and control samples. While other microorganisms such as *Lactobacillus sakei*, *Leuconostoc gasicomitatum*, and *Lactococcus piscium* became active during chilled vacuum-packaged storage due to the unavailability of oxygen. Initially, aqueous ozone treatments were unable to modify the microbiota composition, dynamics, and the related volatilome; however, the efficacy to reduce microbial count increased during refrigerated storage. Because the injured bacterial cells cannot proliferate under stressed conditions of low-temperature storage and usually subsequently die under those conditions (Kalchayanand et al., 2019).

TABLE 1 A summary of studies reporting the decontamination of meat using ozonated water.

Sample	Specification	Microbes	Highlights	References
Chicken meat	0.5 ppm at 4°C for 15–45 min	<i>Staph. aureus</i>	2–4 logs CFU/mL after 45 min	Kanaan (2018)
Chicken meat	0.21 and 0.38 mg/L at 3–37°C for 40–120 min	TAMB	1 log CFU/g reduction at 0.38 mg/L for 120 min at 3°C. The longer the contact time, the more bacteria disinfected in the sample. The more frequently ozonated water is replaced, the more bacteria are disinfected in the sample	Fathul Karamah and Wajdi (2018)
Chicken meat	Immersed in 10 ppm + MAP with or without oxygen scavenger	TAMB, total aerobic psychrophilic bacteria, LAB, Enterobacteriaceae	MAP + Ozone = of 1.48–1.61 logs unite for TAMB count reduction. MAP + Ozone + oxygen scavenger = 1.30–1.46 for total aerobic psychrophilic bacteria counts, 0.94–1.17 for LAB counts, and 1.67–2.09 for Enterobacteriaceae counts on Day 4	Ünal (2017)
Chicken drumsticks	8 ppm soaking and spraying at 10–12°C for 4 min	<i>Salmonella</i>	The average killing capacity of aqueous O ₃ /cycle on the skin surface was 1.2–1.6 log/cm ² and on subcutaneous was 0.9–1.1 log/cm ² . The addition of lactic acid (0.3%) increased the microbial killing capacity by 0.3 and 0.2 log/cm ² on the skin and subcutaneous, respectively	Megahed et al. (2020)
Beef (vacuum packaged)	9 mg/L for 35 s at 4°C combined with 5 mL sodium citrate solution (1%)	TVC	The combined treatment showed better inhibiting the TVC, decreased TVBN, and decreased the deteriorated effect than the individual treatments	Zhang et al. (2020)
Beef	12 ppm at 4.6–5.68°C for 90 s of spray every 30 min for 12 h	<i>E. coli</i> O157:H7 and aerobic bacteria	1.46 and 0.99 logs reduction, respectively. ≤90% inactivation of <i>E. coli</i>	Kalchayanand et al. (2019)
Beef (raw)	0.5 ppm for 15, 30, and 45 min at 5, 15, and 20°C	<i>Aeromonas hydrophilia</i> , <i>Listeria monocytogene</i> and <i>Yersinia enterocolitica</i>	Rinsing for 45 min achieved 3 log ¹⁰ CFU/mL ⁻¹ reductions. The bacterial count diminished with increased exposure time to ozonated water at the same concentration	Ali et al. (2022)
Beef (head, heart, and liver)	Spray 1.5–2.3 ppm for 18 s at 10–24°C	Total aerobic bacteria and <i>E. coli</i>	Total aerobic bacteria counts were reduced on average by 1.66, 0.52, and 1.20 Log CFU/sample, while <i>E. coli</i> counts were reduced on average by 0.75, 0.62, and 1.25 Log CFU/sample, in the head, heart, and liver, respectively	Vargas et al. (2021)
Cattle meat	1/2 ppm for up to 30 min at 3–7°C	Multidrug-resistance <i>Staph. aureus</i>	Reduction 2–3 log ¹⁰ (CFU/mL) after 30 min	Hadi et al. (2021)
Beefsteaks (vacuum pack storage at 4°C for 15 days)	6 mg/L spray for 90 s	Total bacterial counts, LAB, coliforms and yeast	Non-significant reductions even after 15 days of storage. <i>Pseudomonas fragi</i> was the dominant species before and after the treatments	Botta et al. (2018)

O₃, Ozone; MAP, modified atmosphere packaging; TAMB, total aerobic mesophilic bacteria; LAB, lactic acid bacteria; TVC, total viable count; TVBN, total volatile basic nitrogen.



2.2. Electrolyzed water

Electrolyzed water (EW) was first developed in Japan as a medical product in the mid-1980s. EW has strong antimicrobial properties because of its pH, oxidation-reduction potential (ORP), and available chlorine content (ACC: 50 mg/L). EW is produced by passing a dilute salt solution through an electrolytic chamber containing anode and cathode electrodes with a bipolar membrane separation (Figure 2).

The salt electrolysis dissociates sodium and chloride into positive (Na⁺) and negative (Cl⁻) ions (Degala et al., 2020). The resulting solution can be collected separately from the anode or cathode regions and categorized into acidic (AEW) and alkaline electrolyzed water (AIEW), respectively (Hernández-Pimentel et al., 2020). The resultant solution contains active oxygen and other oxidants, including hydrogen peroxide, ozone, free chlorine, and chlorine dioxide. AEW (pH < 2.7, ORP > 1,000 mV) contains the hypochlorous acid (HOCl), which induced destructive oxidation in membrane-bound complexes and disturbed cellular electrical charge. HOCl as a sanitizer has about 80 times stronger antimicrobial capacity against *E. coli* compared to an equivalent concentration of the hypochlorite ion (ClO⁻). EW modified the meat protein secondary structure and effectively inactivates pathogens more than conventional chlorine compounds (sodium hypochlorite), hence its applications in meat products would be the second topic of interest in the future (Iram et al., 2021). However, AEW has great potential

for use as an antimicrobial agent in the meat industry. For instance, AEW-treated beef, chevon, and pork samples exhibited the highest inactivation of *E. coli* K12 by 1.16 logs in 4 min, 1.22 logs in 12 min, and 1.30 logs in 10 min, respectively; however, AIEW treatments (pH 10–11.5, ORP: 800–900 mV) reduced 1.61, 0.96, and 1.52 logs in 12 min, respectively (Arya et al., 2018). Several other types of EW are slightly acidic electrolyzed oxidizing water (SAEW), and neutral electrolyzed water (NEW). SAEW (pH 5.0–6.5, ORP 600 mV) is produced by the electrolysis of hydrochloric acid in a chamber without a membrane (Arya et al., 2018). However, NEW is generated by mixing the AEW and AIEW or using an electrolysis chamber without a membrane. NEW has solved the problems related to the storage and corrosion effect of AEW. Recently, the research focus has been shifted toward SAEW applications alone or with ultrasound and UV radiation (Iram et al., 2021).

The effect of EW on microorganisms is complex; however, some researchers linked it with active species produced during the process such as hydroxyl ions and hypochlorite (Wang et al., 2019). The electrolysis phenomenon (produced by immersed electrodes) is based on electrochemical reactions, which involve electron excitation and generates a variety of oxidants. The action of electrons (donors/acceptors) could induce irreversible modifications in the bacterial transmembrane potential. The resultant powerful electro-osmotic reactions disturb the active transport of substances into the cell and diffuse water against oxidation-reduction gradients. It

induces an electric charge in the bacterial membrane, following membrane rupture and an outflow of intracellular components (Athayde et al., 2017). Besides, excess anions (present in the anolyte solution) interrupt the functionality of the electrically charged bacterial cell membrane with modifications in solution transport or availability. However, solute transport is depending on the electrostatic interactions and small charged molecules. Thus, any significant change in the ORP of the immediate medium of the bacterial cells can negatively affect the electrochemical gradient (Aider et al., 2012).

2.2.1. Decontamination of meat

EW decontamination involves the disruption of bacterial membrane integrity and induces cell necrosis and apoptosis (Liao et al., 2017). Previously published studies on the effect of EW on raw and processed meat are listed in Table 2.

EW spraying is more convenient in mobile chicken slaughter lines as compared to dip and immersion methods. Additionally, spraying treatments reduce water waste and the formation of disinfectants by-products. Duan et al. (2017) compared the impact of traditional disinfectant sprays [sodium hypochlorite (50 and 100 mg/L), chlorine dioxide (50–100 mg/L), lactic acid (1–2%)] with AEW and SAEW on chicken carcasses. According to the results, 2% lactic acid, AEW, and SAEW reduced 0.47–0.83 log (CFU/cm²) total viable counts and 0.49–0.96 log (MPN/cm²) total coliforms. Moreover, AEW and SAEW exhibited a 2-day extension of microbial shelf life compared to other treatments. Although sodium hypochlorite and chlorine dioxide treatments were ineffective to reduce microorganisms, sodium hypochlorite promotes trihalomethanes formation when reacted with the organic matter. Besides, lactic acid exceeded the 2 mg/kg limit on the 8th day of storage, which led to high thiobarbituric acid reactive substances values and odors and flavors deterioration.

Similarly, Wang et al. (2018) reduced 1.0 logs total viable counts and total coliform in AEW-or SAEW-treated chicken carcasses by spraying with a nozzle at 0.3 MPa pressure for 15 s. Despite all advantages, AEW is becoming less popular because of corrosion and instability. Besides, SAEW (a pH of 5.0–6.5, an ORP of 750–850 mV, and an ACC of 10–30 mg/L) showed significant bactericidal effects with more stability and less corrosion as compared to AEW (Duan et al., 2017). Although 30 mg/L of SAEW has the same antimicrobial effect as 60 mg/L of AEW (Wang et al., 2018). SAEW is allowed to control the pH of the water used in the US meat industries (USDA, 2022). The total viable counts in the SAEW-treated beef meat decreased to 2.28 logs from 3.06 logs (Sheng L. et al., 2018). HOCl (present in SAEW) reduced microbial growth *via* modifying bacterial electron transfer mechanism (redox potential), which promoted cellular protein oxidation, and DNA damage. Furthermore, SEAW achieved high antimicrobial efficiency with a low available chlorine concentration (Xuan et al., 2016). According to Liao et al. (2020), SAEW thawing caused a 0.83–1.76 log reduction of total bacteria, fungi, and yeast without negatively affecting the texture, pH, and color of the beef carcass. Moreover, SAEW thawing prevented oxidation and degradation of lipid/protein as compared to traditional thawing treatments (air thawing, water thawing, and microwave thawing). Because it did not induce detrimental effects due to protein oxidation or changes in muscle microstructure (Liao et al., 2020).

NEW is another form of EW, approved in the US for poultry processing at doses ≤ 50 mg/L (USDA, 2022). NEW (ORP 750–900 mV) exhibited 10 times higher antimicrobial efficacy against *Salmonella* in pure culture than sodium hypochlorite treatments *via* bacterial cell enlargement and membrane structural modifications. In this study, the resistant *Salmonella* pure cultures mixture (adjusted to 6 log CFU/mL) was inoculated, allowing it to drain through the walls of the gastrointestinal cavity of the previously eviscerated carcasses and then air dried for 10 min in a laminar flow cabinet. Furthermore, broiler carcasses were immersed in NEW or NaClO at 50 mg/L of total available chlorine, and the control using distilled water at 3°C for 1.5 h. According to Hernández-Pimentel et al. (2020), NEW at 14 mg/L (total available chlorine) completely inactivated (>6 logs CFU/mL) the *Salmonella* pure cultures mixture after 1 min contact time. Moreover, NEW effectively reduced total viable counts and coliforms on broiler chicken carcasses, without color and pH modification. Though Hawkins et al. (2016) achieved 0.56 log CFU/g reduction of *Salmonella* by Near-NEW spraying (60 s) in raw chicken thigh meat. The active agents of NEW include HOCl and other components (~5%) such as hypochlorite ions and chlorine.

When compared to AEW, NEW has limited human health and safety concerns from chlorine dioxide off-gassing, reduces corrosion of surfaces, and limits phototoxic side effects while maximizing the application of hypochlorous acid species (Moghassem Hamidi et al., 2021). Besides, NEW produces less corrosion compared to other disinfectants such as AEW because of its neutral pH. NEW has comparable antimicrobial efficacy to the other types of sanitizers. For instance, NEW spray (free chlorine value of 58 ppm) and sodium hypochlorite (35 ppm) reduced 0.64 logs of *Listeria monocytogenes* and 0.3 log of *Salmonella Typhi* in highly contaminated pork chops (10⁶ CFU/mL) compared to the traditional saline solution (Torres-Rosales et al., 2020). NEW resulted in slight color modifications as compared to sodium hypochlorite or saline solution; however, it decreased lactic acid production and total volatile basic nitrogen during storage at 4°C for 19 days. Similarly, significant log reductions were observed in *E. coli* O157:H7 and *Salmonella* cultures when pork chops were treated with 6% NEW, while *Yersinia enterocolitica* required about 15% NEW for 2.5 logs reduction (Han et al., 2018). On the other hand, the color changes caused by NEW treatments were minor compared to sterilize deionized water on treated pork chops. Furthermore, high organic matter on the pork chops renders antimicrobial functioning of NEW and generates chloramines (Han et al., 2018). However, the application of NEW was more effective when supplemented with other physical and chemical treatment methods (Rahman et al., 2016; Oh et al., 2019) such as peroxyacetic acid (Moghassem Hamidi et al., 2021), nisin (Arevalos-Sánchez et al., 2012), UV-C radiation (Jemni et al., 2014), UV-A radiation (Jee and Ha, 2021), AEW (Jiménez-Pichardo et al., 2016), AIEW (Jadeja and Hung, 2014), ultrasound (Afari et al., 2016), modified atmosphere packaging (Posada-Izquierdo et al., 2014) and so on.

2.3. Plasma activated water

Several studies have reported the direct impact of gaseous plasma on food stuff for enhancing microbiological safety; however, it produced some negative effects on organoleptic characteristics

TABLE 2 A summary of studies reporting the decontamination of meat using electrolyzed water.

Meat type	EW	Experimental conditions		Target microorganism	Reduction	References
		Application	Exposure time			
Chicken	NEW (pH 6.5, ORP of 1,123 mV)	Immersion at 3°C	1.5 h	<i>Salmonella</i> , TVC and total coliform	Completely inhibited (>6 logs CFU/mL) of <i>Salmonella</i> with 14 mg/L TAC after 1 min and with 5 mg/L TAC after 5 min. 1.2 log CFU/mL reduction of TVC and total coliform after 10 days of refrigerated storage	Hernández-Pimentel et al. (2020)
Chicken breast	SAEW (pH 6.0, 5 ppm chlorine, ORP 800–850 mV) + ultrasound (25–130 kHz)	Immersion at 10°C	10 min	Psychrotrophic bacteria, LAB and mesophilic bacteria	76, 0.81, and 0.98 log CFU/g, respectively	Cichoski et al. (2019)
Chicken breast	Near-NEW (pH 6.8, ORP 830 mV, ACC 800 mg/mL) + Peroxyacetic acid (200 µg/mL)	Immersion	10 min	Aerobic, psychrophilic, <i>Enterobacteriaceae</i> , LAB and <i>Pseudomonas</i>	1.33, 1.40, 1.45, 1.01, and 1.45 log CFU/g, respectively on the 6 th day of storage	Moghassem Hamidi et al. (2021)
Chicken carcasses	AEW (pH 2.55, ORP 1150 mV, ACC 60 mg/L) or SAEW (pH 6.00, ORP 845 mV, ACC 30 mg/L)	Spray	15 s	TVC and total coliform	1 log CFU/cm ² or MPN/cm ²	Wang et al. (2018)
Chicken carcasses	AEW (pH 2.46, ORP 1126 mV, ACC 58 mg/L) or SAEW (pH 5.98, ORP 865 mV, ACC 30 mg/L)	Spray	15 s	TVC and total coliform	0.47–0.83 logs CFU/cm ² and 0.49–0.96 logs MPN/cm ² in TVC and total coliforms, respectively	Duan et al. (2017)
Chicken thigh meat	Near-NEW (pH 6.2–6.5, ORP 760–770 mV, ACC 10 mg/L)	Spray	60 s	<i>S. typhimurium</i>	0.56 log CFU/g	Hawkins et al. (2016)
Chicken (giblets)	NEW (pH 6.7, ORP 1030 mV, ACC 10 mg/L)	Submersion (50–200 ppm)	15 and 30 min	<i>E. coli</i> , <i>Staph. aureus</i>	A maximum microbial load reduction of 1.97 logs CFU/g for <i>E. coli</i> and 1.76 logs CFU/g for <i>Staph. aureus</i>	Sierra et al. (2022)
Goat meat	AEW (pH 2.73, ORP 831 mV, ACC 34.30 ppm) ALEW (pH 11.03, ORP–421 mV, ACC 0.06 ppm) ozonated water (pH 6.80, ORP 562 mV, ozone 0.68 mg/L)	Dip in 200 mL	2–12 min	<i>E. coli</i>	AEW = 0.86 CFU/mL in 10 min, ALEW = 0.74 CFU/mL in 12 min, ozonated water = 0.53 CFU/mL in 10 min, AEW + ozonated water = 0.84 CFU/mL in 10 min, ALEW + ozonated water = 1.03 CFU/mL in 8 min	Degala et al. (2020)
Beef	SAEW (pH 6.3, ORP 867.4 mV, ACC 30 mg/L)	Thawing at 20°C	Until the core temperature reached 0°C	TVC, fungi and yeast	sAEW reduced TVC by 0.83 log CFU/g and fungi and yeast by 1.16 logs CFU/g	Liao et al. (2020)
Beef	SAEW (pH 6.29, ORP 870–900 mV, ACC 40 ppm)	Dip at 23°C	5 min	TVC	Reduced to 2.28 logs CFU/g from 3.06 logs CFU/g. Maintained 14–16 days shelf life	Sheng X. et al. (2018)
Beef carcasses and frankfurters	EW (pH 6.5, ACC 250 ppm)	Spray	30 s	<i>L. monocytogenes</i> , <i>E. coli</i> O157:H7, and <i>Salmonella</i> spp.	Little or no reduction in controls was observed. >6-log reduction in 2 min was achieved when EW was directly applied to multi-strain cocktails. EW at 25, 50, and 100 ppm was effective in reducing <i>L. monocytogenes</i> by 1.67, 3.72, and 7.36 logs (CFU/mL), respectively	Veasey and Muriana (2016)

(Continued)

TABLE 2 (Continued)

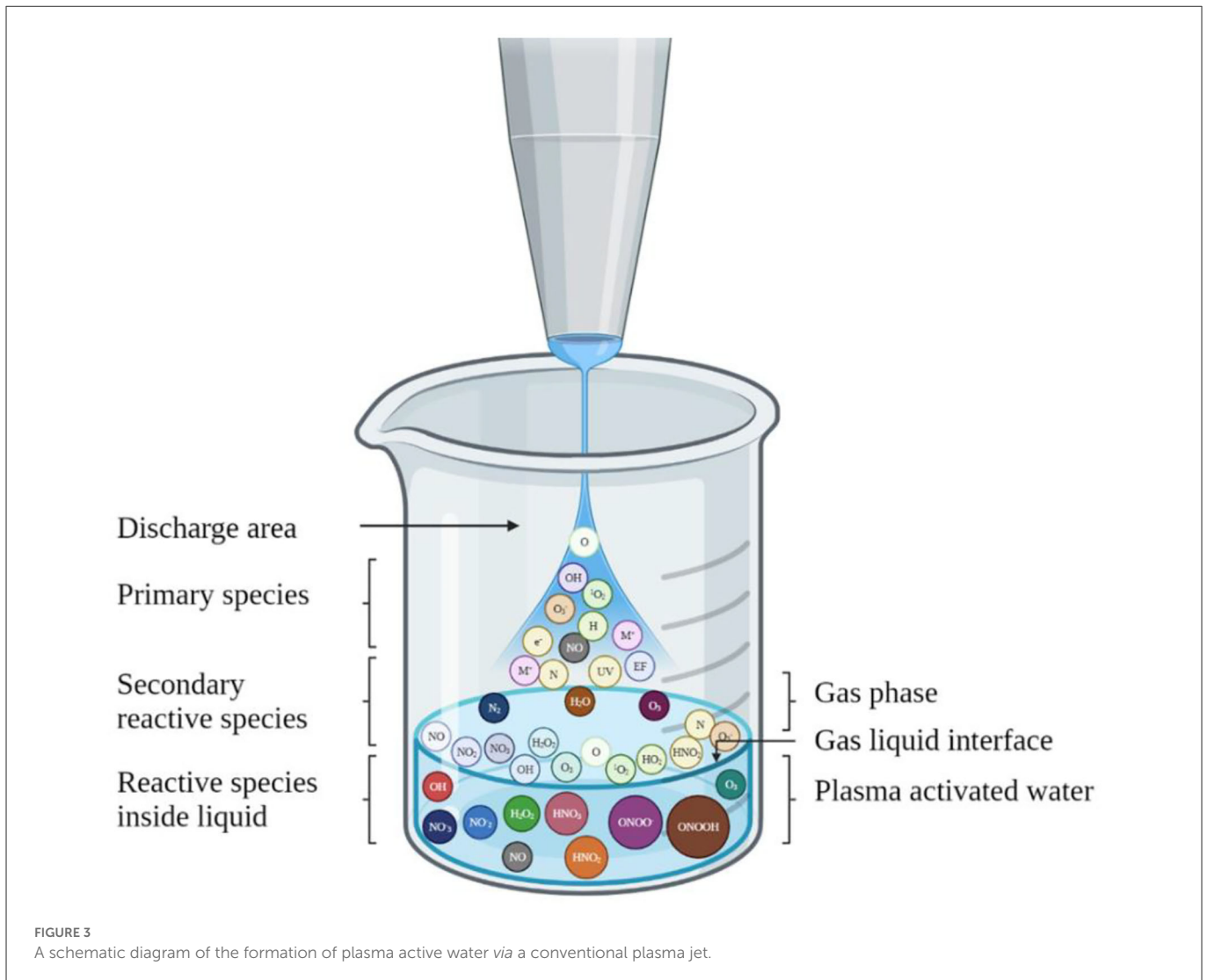
Meat type	EW	Experimental conditions		Target microorganism	Reduction	References
		Application	Exposure time			
Beef, chevon, and pork surfaces	AEW (pH 3.03, ORP 759.9 mV, ACC 34.3 mg/L), AIEW (pH 10.73, ORP -372.4 mV, ACC 0.06 mg/L)	Spray	2–12 min	<i>E. coli</i> K12	AEW-treated beef, chevon, and pork samples resulted in the highest log reductions of ~1.16 (4 min), 1.22 (12 min), and 1.30 logs CFU/mL (10 min), respectively; and AIEW resulted in 1.61, 0.96, and 1.52 logs CFU/mL reductions at 12 min, respectively	Arya et al. (2018)
Beef	SAEW (pH 6.51, ORP 655 mV, ACC 30 mg/L)	Immersion	1–5 min	<i>S. enteritidis</i>	The SAEW-tea polyphenols treatment resulted in an ~3.37 log ¹⁰ CFU/g reduction in <i>S. enteritidis</i> .	Bing et al. (2022)
Pork chops	NEW (pH 6.92, ORP 820 mV, ACC 58 ppm)	Spray	60 s	<i>L. monocytogenes</i> and <i>S. Typhi</i>	0.67 log CFU/g reduction in <i>L. monocytogenes</i> and 0.47 log CFU/g reduction in <i>Salmonella Typhi</i> compared to 0.65–0.3 log CFU/g reduction by NaClO, respectively	Torres-Rosales et al. (2020)
Pork (chops and skin)	NEW (pH 7.64, ORP 818 mV, ACC 74 mg/L)	Immersion (1–15% NEW)	2–10 min	<i>E. coli</i> O157:H7, <i>S. Enteritidis</i> and <i>Yersinia enterocolitica</i>	Skin samples: <i>E. coli</i> = 2.12–2.59 logs CFU/cm ² reductions. <i>Salmonella</i> = 2.22–2.37 logs CFU/cm ² reductions. <i>Yersinia</i> = 1.74–1.81 logs CFU/cm ² reductions. Pork chops: <i>E. coli</i> = 0.29–0.32 log CFU/cm ² reduction. <i>Yersinia</i> = 0.17–0.15 log CFU/cm ² reduction	Han et al. (2018)
Pork meat	AEW (pH 2.60, ORP 1185 mV) SAEW (pH 6.5, ORP 940 mV), AIEW (pH 11.40, ORP -826 mV)	Spray at 18 and 30°C	40 s	LAB, mesophilic bacteria and psychrotrophic bacteria	AEW alone or with AIEW decreased the microbial counts shortly after spraying. The combination of AIEW + AEW (30 psi) reduced the mesophilic and psychrotrophic bacteria counts throughout the refrigerated storage	Athayde et al. (2017)

AEW, acidic electrolyzed water; SAEW, Slightly acidic electrolyzed water; AIEW, alkaline electrolyzed water; NEW, neutral electrolyzed water; ACC, available chlorine concentration; ORP, oxidation reduction potential; TVC, total viable counts; TAC, total available chlorine; LAB, lactic acid bacteria.

such as color, firmness, etc. (Asghar et al., 2022; Roobab et al., 2022). To solve this problem, plasma-activated water (PAW) could be an alternative way to efficiently inactivate microorganisms and enhance food safety (Liao et al., 2020; Wang et al., 2021). PAW—a product of cold atmospheric plasma reacting with water—is a potential antimicrobial agent which has a rich diversity of highly reactive oxygen and nitrogen species. Chemical reactivity and energy transferred from gaseous plasmas to water without using any chemicals make it a clean-label alternative to conventional chemical disinfectants (Ostrikov et al., 2020). The working principle of PAW is based on the direct or indirect application of electrical discharge, which ionizes neutral gases in water (helium, argon, neon, nitrogen, oxygen, air) and forms excited/ionized atoms, ultraviolet rays, electric fields, and abundant reactive oxygen and nitrogen species (Figure 3). Based on the working principle, several

plasma devices have been used such as plasma jets and dielectric barrier discharge.

Due to the advantages of environmental friendliness and green aspects PAW has gained attention for replacing conventional chemical disinfectant treatments, such as chlorine (Thirumdas et al., 2018). PAW has several reactive species, including hydrogen peroxide, peroxyxynitrite, nitric oxide, nitrates, and nitrite ions, which have strong antimicrobial efficiency against a wide range of microorganisms. These active species can destroy bacterial cell membranes and disrupt genetic material (DNA and protein) inducing damage to cell macromolecules (Liao et al., 2020). According to Royintarat et al. (2019), reactive species such as •OH promote peroxidation of the bacterial cell membrane, induce oxidative stress, alter molecular structures, disrupt pH homeostasis, and cause cell death (Figure 3).



2.3.1. Decontamination of meat

PAW from underwater plasma jet treatments has been used on tap water to produce reactive species for the decontamination of chicken. According to Royintarat et al. (2018), PAW tap water treatments reduced *Salmonella Typhimurium* by 0.54 and 0.13 log CFU/mL in muscle and skin samples, respectively. The bactericidal efficiency of PAW was reduced in chicken muscle and skin due to high polyunsaturated fatty acids (omega 6 and omega 3), which inhibited reactive species (Ambrozova et al., 2010). However, the efficiency could be improved by increasing plasma intensity to generate more reactive species. Besides, PAW treatments reduced 0.46 log CFU/mL of *E. coli* K12 and 0.33 logs CFU/mL of *Staph. aureus*; however, the combined treatments with ultrasound inactivated *E. coli* K12 by 1.33 logs CFU/mL and *Staph. aureus* by 0.83 logs CFU/mL in chicken meat. The synergistic interaction of combined PAW–ultrasound enhanced the porosity of muscle structure with a dramatic difference in quality characteristics. According to Royintarat et al. (2019), ultrasonication was the reason behind the increased penetration of PAW into the sample. Ultrasonication induced lipid oxidation of bacterial cell membranes and speeded the reaction between cell membrane unsaturated lipids and free radicals in PAW. Moreover, chicken muscle bacteria were

more physically damaged as compared to chicken skin bacteria, indicating more organic material on the skin inhibits the formation of reactive species. The antimicrobial potential of PAW depends on the process parameters such as exposure time, targeted surface, microbial strain, reactive species, and so on (Perinban et al., 2019). Additional studies with different microorganisms' inactivation in several meat products with plasma-activated liquids have been mentioned in Table 3.

According to Zhao et al. (2020), 24 h PAW treatment achieved 3.1 logs reduction of surface bacteria and extended the shelf life of fresh beef for 4–6 days. A 30 min plasma activation contains 116 mg/L of hydrogen peroxide concentration in deionized water. However, the bactericidal impact of PAW decreased with the increase of the treatment interval time as microbial count increased during storage. Moreover, the treatment interval of more than 48 h was ineffective for controlling microbial growth in beef. Besides, excessive treatments could negatively affect the beef's physiochemical quality such as damage to the structural integrity. PAW thawing could be an effective approach to ensure microbial safety in meat.

Plasma-activated brine (plasma beam system at 20 kHz using air or nitrogen gas) was used to protect the jerky beef from *L. innocua*. Beef slices were cured in brine solutions [sodium nitrite

TABLE 3 A summary of studies reporting the decontamination of meat using plasma-activated water.

Sample	Specification	Microbes	Highlights	References
Chicken	Plasma-activated tap water	<i>S. typhimurium</i>	Reduced 0.54 logs in muscle and 0.13 logs in the skin	Royintarat et al. (2018)
Chicken breast	Sterile distilled water was activated by plasma for 30–90 s. Samples dip in PAW for 3–12 min	<i>Pseudomonas deceptionensis</i> CM2	Reduced by 1.05 logs CFU/g at PAW ₆₀ for 12 min	Kang et al. (2019)
Chicken breast (cooked)	Distilled water was activated by plasma for 5–20 min. Samples dip in PAW for 0–20 min	MRSA and MSSA.	Reduced by 2.09 and 2.29 logs CFU/g for MRSA and MSSA at PAW ₂₀ for 20 min	Wang et al. (2021)
Chicken	Soaked in PAW (1.5 kHz, 6.8 kV) and ultrasonicated (40 Hz, 220 W) at 4–40°C for 30–60 min	<i>E. coli</i> and <i>Staph. aureus</i>	Inactivated up to 1.33 logs CFU/mL of <i>E. coli</i> K12 and 0.83 log CFU/mL of <i>Staph. aureus</i>	Royintarat et al. (2020)
Chicken breasts (frozen)	PAW thawing; sample was thawed by being immersed in the thawing medium in a weight ratio of 1:4. All thawing methods were performed until the central temperature of the chicken was about 0°C	Total viable bacterial count and <i>Salmonella</i>	Showed reduction of the bacterium for 0.62–1.17 log CFU/g	Qian et al. (2019)
Chicken (myofibrillar protein gel)	The myofibrillar protein solution was incubated with PAW for 0–100 s	<i>S. Enteritidis</i> and <i>Staph. aureus</i>	The gels prepared with plasma-activated water showed obvious antibacterial activity	Qian et al. (2019)
Beef	1 mL PAW was sprayed on each piece of beef (30 × 30 × 10 mm). PAW treatment interval times (6–192 h)	Total bacteria	24 h PAW treatment achieved 3.1. log reduction of surface bacteria. 4–6 days extended shelf life	Zhao et al. (2020)
Beef	PAW thawing, distilled water was activated by plasma for 1 min. The sample thawing procedure continued until the core temperature reached around 0°C	Total bacteria, fungi and yeast	Showed the highest antimicrobial ability (1.62 logs reduction in total bacteria and 1.76 logs in fungi and yeast) compared with electrolyzed water and microwave thawing	Liao et al. (2020)
Beef	PAW was poured over samples at 0.57 mLPAW/g beef samples and allowed to air dry for 20 min before analysis	<i>S. Typhimurium</i> (NCTC74) and <i>E. coli</i> 0157:H7 (ATCC700728TM)	Showed 5.9 log reduction in <i>S. Typhimurium</i> population and a 4–log reduction in <i>E. coli</i> population shown after exposure to PAW for up to 240 and 300 s respectively	Astorga et al. (2022)
Beef	Distilled water containing lactic acid (0.05–0.20%) was activated by plasma jet for 40–100 s. UV-treated fresh beef slices were spot-inoculated with <i>S. enteritidis</i> (5.67 log CFU/g) and immersed in PAW solutions for 20 s	<i>S. enteritidis</i>	Reduction ranging from 1.24 to 3.52 logs CFU/g. The antibacterial efficacy of PAW was enhanced by 0.77–4.58 log CFU/mL with the addition of lactic acid	Qian et al. (2019)
Beef jerky	The brine solution was activated by plasma for 10 min. Beef slices (6 × ~ 10 g) were cured in 200 mL of brine solution for 18–20 h at 4°C, removed from the solution, and placed in a hot air-drying oven, at 70°C for 90 min	<i>L. innocua</i>	0.5 log inactivation in the brine. 0.85 log CFU/g reduction in beef jerky	Inguglia et al. (2020)
Loin ham	Distilled water containing 1% sodium pyrophosphate was treated with plasma for 2 h. PAW-treated brine solution was injected into pork lion, tumbled for 48 h at 4°C, and smoked until the internal temperature of loin ham reached 70°C	Total aerobic bacteria	The initial number of total aerobic bacteria cells in PAW-treated ham was 0.33 log CFU/g lower in comparison with sodium nitrite-treated samples	Yong et al. (2018)

PAW, plasma-activated water; MRSA, methicillin-resistant *Staph. Aureus*; MSSA, methicillin-susceptible *Staph. Aureus*.

(0–150 ppm) + sodium chloride (150 ppm) + sugar (100 ppm)] for about 18 h at 4°C then subjected to plasma treatment. Results showed a significant reduction in *L. innocua* population of about 0.5 log CFU/mL in brine and 0.8 CFU/mL when treated with plasma-activated brine (Inguglia et al., 2020). According to the authors, plasma technology could be used as an alternative nitrite source for meat curing with minimal impact on product quality characteristics. However, further optimization of plasma technology is required to ensure the microbiological safety of meat products.

2.4. Synergistic applications of meat decontamination

Ozonated water treatments can be combined with other clean-label ingredients such as lactic acid (Megahed et al., 2020) and sodium citrate (Zhang et al., 2020). Megahed et al. (2020) investigated the microbial killing capacity of ozonated water and ozone–lactic acid blend on *Salmonella*-contaminated chicken drumsticks using a multi-sequential application. The results showed that six consecutive soaking and spraying cycles with 8 ppm aqueous ozone reduced 7 logs *Salmonella*. However, the combination with 0.3% lactic acid increased the decontamination power of aqueous ozone with average differences of 0.3- and 0.2-log¹⁰/cm² on the skin surface using soaking and spraying approaches, respectively. Lactic acid reduced the pH of ozonated water, which intensified the antimicrobial effect. According to the authors, spraying or soaking in low pH ozonated water (pH range from 2 to 3) caused cytoplasmic acidification. The resultant aqueous medium has stable ozone, which disrupted the energy and regulatory parameters of the host bacterial cell. Moreover, the accumulated free acid anions inactivate microorganisms through malfunctioning of microbial growth and survival. However, the antimicrobial impact of ozone also depends on the initial bacterial load on the chicken carcass.

Zhang et al. (2020) have found that sodium citrate and aqueous ozone combination with vacuum-packaged beef inhibited microbial growth more effectively as compared to high temperature, alkali, or salt stress. Moreover, the treated beef meat had the longest shelf life (21 days) compared to individual treatments. However, aqueous ozonation negatively impacted the water-holding capacity (purge loss) and tenderness. The oxidative modifications of tenderizing enzymes strengthen muscle fibers; however, the reduced proteolytic activity caused cross-linking of muscle proteins. Furthermore, myoglobin oxidation (met myoglobin) produced darker and redder beef steaks compared to the control. However, the addition of sodium citrate (antioxidant) exhibited a higher *L** value (lightness), lower *a** (redness) value, and lower metmyoglobin of ozonated beef steaks. High dosages and prolonged contact time used for the antimicrobial purpose could negatively affect the quality parameters of meat. Adequate precautions should be taken while applying high ozone doses to inactivate microorganisms.

Several other non-thermal combinations have been introduced for meat decontamination such as Moghassem Hamidi et al. (2021) combined NEW (100 mg/mL) with peroxyacetic acid (200 mg/mL) to reduce aerobic plate counts, psychrophilic plate count, *Enterobacteriaceae*, LAB, and *Pseudomonas* counts of chicken breast meat. Peroxyacetic acid with high-oxidizing potential affects bacterial cell integrity and modifies protein synthesis. Furthermore, it acidifies the meat surface and enhances the diffusion of the undissociated acids

into the bacterial cell, which enhanced the antimicrobial efficacy of NEW. Veasey and Muriana (2016) suggested using EW for surface sanitizing of meat processing equipment (i.e., slicing blades) rather than a raw carcass. According to the author, the surface of raw meat (beef carcass) and ready-to-eat meat (frankfurters) contain high levels of organic material, which make HOCl ineffective as an antimicrobial. Although higher intensities of EW could inactivate microorganisms, they may produce undesirable sensory characteristics. Alternatively, a combined approach could be used to lower individual treatment intensities (Liao et al., 2020).

Similarly, the combination of ozonated water AEW, and AIEW showed promising antimicrobial potential against *E. coli* K12 in goat meat. According to Degala et al. (2020) the synergistic approach reduced 1.03 log CFU/mL in 8 min as compared to individual treatments of AEW, AIEW, and ozonated water, which caused 0.86; 0.74 and 0.53 log CFU/mL reduction in 10–12 min, respectively. In the same vein, Athayde et al. (2017) sprayed AEW, SAEW, and AIEW for 40 s with a pressure of 30 and 45 psi from 10 cm distance on pork loin with rotation at a 360° angle during application. The individual application of AEW and SAEW (compared to AIEW) increased carbonyl groups, shortly after application due to the molecular species (HCl, Cl₂, HOCl), which produced carbonylated proteins. However, the addition of AIEW exerted an antioxidant effect and protected the meat protein from the adverse effects of AEW and SAEW. Moreover, the combination AIEW + AEW exhibited a strong antimicrobial effect against mesophilic and psychotropic bacteria without producing carbonyl compounds and oxidating thiol groups. However, the production of acidic compounds by lactic acid bacteria stabilized the pH values throughout the refrigerated storage (Athayde et al., 2017).

Other non-thermal combinations have been used against different microorganisms on various types of meat. The ultrasound + SAEW treatments significantly reduced enterobacteria, mesophilic bacteria, lactic acid bacteria, and psychotropic bacteria without affecting lipid and protein oxidation, shear force, anaerobic glycolysis, and muscle structure of chicken breast (Cichoski et al., 2019). However, the thick peptidoglycan layer of *Staphylococcus* spp. cells renders the antimicrobial potential of SAEW by preventing entry into the microbial cell. According to the author, large cavitation bubbles produced by ultrasonication generated much energy to rupture bacterial cytoplasmic membranes. Hence, ultrasonic cavitation helped SAEW to enter the cell. Furthermore, SAEW before and after immersion did not show mesophilic and psychotropic bacteria growth, hence, could be reused in the cooling process (Joyce et al., 2011). Furthermore, Liao et al. (2020) used PAW as a thawing media, which effectively inactivated the total bacteria, yeast, and molds in beef by 0.83–1.76 logs. Moreover, both PAW and SEAW maintained the quality characteristics such as texture, pH, and color as well as prevented lipid/protein oxidation and protein degradation in beef as compared to traditional thawing methods (air thawing, water thawing, microwave thawing) (Liao et al., 2020).

3. Conclusion

Non-thermal electrochemical activation of water is a relatively new and fast-emerging technology. Non-thermal technologies used in water affect the chemistry of reactive species and their interactions with microorganisms. Non-thermal activation of water allows the use of aqueous solutions at lower dose rates

compared to conventional chemical disinfectants, preventing intoxication and adverse environmental impacts. In the meat industry, electrochemically activated solutions with increased antimicrobial capacity could be applied as an alternative to costly decontamination methods. However, a more detailed study is needed to understand the thermodynamics behind the electro-activation phenomena of water and the interaction of aqueous solutions with food and targeted microorganisms. Further, ozone, electrolyzed water, and cold plasma technologies are concentration-dependent as they can harm health if used beyond their recommended limits, while their electrochemically treated water can be applied at very high concentrations. For instance, ozone is allowed to be used at levels of 5 ppm; however, its activated water could be used at levels as high as 95 ppm. Therefore, non-thermal functional water at low levels has enough active components to ensure meat quality and safety. Furthermore, the synergistic combination of these non-thermal technologies could enhance the functionality of rinsing solutions. However, these treatments could further be enhanced by taking into account the stability of activated water during storage conditions.

Author contributions

UR and RA: conceptualization. UR, GM, and AK: reviewed and refined tables and figures. UR and MR: writing—original draft preparation. UR and MS: writing—review and editing. MA, SS, X-AZ, and RA: supervision. All authors have read and agreed to the published version of the manuscript.

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