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Surface Decontamination of Some Foodborne Pathogens at Bovine Carcasses

Ghada Sh. El-Meleeh[1*], Abobakr M. Idris[2], Reyad R. Shawish[3], Zakaria H. Elbayoumi[3]

(1) Veterinarian at Directorate of Veterinary Medicine, Shebin-Elkom, Menoufia, Egypt.

(2) Department of Food Hygiene and Control, Faculty of Veterinary Medicine, Benha University, Benha City, Al Qalyubiyah Governorate, Egypt.

(3) Department of Food Hygiene and Control, Faculty of Veterinary Medicine, University of Sadat City, Sadat City, Menoufia, Egypt.

[*] *Corresponding author:* ghadasherif86@gmail.com *Received: 15/8/2024 Accepted: 15/9/2024*

Abstract

One green chemistry idea that has gained favor as a disinfection method is electrolyzed water technology. Electrolyzed water (EW) is a new technology that emerged in recent years with potential application in foods, mainly in the microbiological aspects, with variations in application techniques and time of exposure. In the current study, the antimicrobial effect of acidic, alkaline, and a mixture of electrolyzed water was assessed as fresh carcasses' surface decontaminant agent after spraying and swab collection. For this purpose, 30 beef carcasses were used, ten carcasses for each group, where aerobic bacterial count (ABC) was investigated pre- and post- spraying; in the local abattoir of El-Shohadaa, Menoufia governorate, Egypt. Results revealed a significant reduction in the bacterial count in the treated groups rather than the pre-treated samples at ($P<0.05$); where the mean reduction (%) were 98.9, 97.3, and 99.8 for the treated samples with acidic EW, alkaline EW, and both alkaline and acidic EW, respectively. Referring to the obtained results, acidic EW showed a higher antibacterial effect than alkaline EW; whereas, using the alkaline EW followed by acidic EW revealed more decontamination effect than using each alone. So, EW proved that it is not only an inexpensive decontamination agent, but also kills microorganisms, and protects the environment from the adverse impacts of hazardous chemical disinfectants. Therefore, it is highly recommended to be used in the slaughterhouse as a surface decontamination technique.

Keywords: Disinfectant, Electrolyzed water, Slaughterhouse.

INTRODUCTION

The need for food is rising in tandem with the ongoing growth of the human population. An essential component of the human diet is animal proteins, but animal products are highly vulnerable to contamination from foodborne pathogens and other contaminants associated with slaughterhouses, processing facilities and transportation (Saraiva *et al.*, 2022).

In developing nations, food contamination during processing, shipping or storage can result in losses of 75% of the total production (Lung *et al.*, 2015). The profusion of biological macromolecules, such as proteins, lipids, carbohydrates and nucleic acids, provides an ideal setting for the growth of various pathogens that can spoil foods and cause a variety of foodborne diseases (Silva *et al.*, 2020). Meat handling, processing, and storage may pose a risk to public health because they can harbor pathogenic bacteria that may be a cause of foodborne illnesses or intoxicate consumers, or they might harbor rotting bacteria that cause undesirable alterations (Barcenilla *et al.*, 2022).

Raw meat is usually contaminated from the outside as soon as bleeding till be consumed. According to Manyi-Loh and Lues (2023), there are numerous potential sources of meat contamination by microorganisms, particularly foodborne bacteria. These include direct contact with the hide skin, or feet; the contents of gastrointestinal tracts; contaminated water sources; the dressing tool (knives, saws, cleavers, or hooks); and/or airborne contaminations.

Since the food industry and customers share a strong concern about safety, various technologies process has been created to maintain quality. Products with minimal processing and minimal modifications to their organoleptic characteristics are in high demand in the food market. Using electrolyzed water, which is regarded as a non-thermal, non-chemical method, is one substitute (Chakka *et al.*, 2021).

The bactericidal effect of electrolyzed water (EW), a sanitizer, is mostly caused by hypochlorous acid (HOCl) (Yan *et al.*, 2021), its chemical and physical characteristics making it more effective and more popular. Sodium chloride (NaCl) and water are needed for EW production, which has several uses. It was first used to disinfect medical supplies (like dialyzers); however, other uses were later documented, such as the disinfection of fruits and vegetables that are ready to eat, where it helps in prevention of food contamination and microbial deterioration, while enhancing safety and shelf life without compromising organoleptic qualities (Khan *et al.*, 2017). Furthermore, several antimicrobial effects have been documented after short exposures (5 to 20 seconds) (Fan *et al.*, 2013; Gómez-Espinosa *et al.*, 2017). The primary benefit of EW is its environmental friendliness; upon reacting with bacteria and organic matter, it returns to water and salt. Additionally, because EW physically kills bacteria, it prevents them from developing resistance (Mosaka *et al.*, 2023).

Therefore, the current study aims to investigate the antibacterial effect of electrolyzed water spray application on the outer surface of beef carcasses.

MATERIAL AND METHODS

1. Experimental design

Thirty random bovine carcasses (10/group) were examined after dehiding, evisceration, and washing at El-Shohadaa abattoir in Menoufia Governorate, Egypt. Swabs were taken from three points in brisket in area about 25 cm^2, before and after spraying of electrolyzed water. Swabs were collected after ten seconds of application; swabs were identified, packed and transferred to the laboratory in an icebox under complete aseptic conditions without undue delay in which aerobic bacterial count (ABC) was measured as a hygienic indicator.

2. Experimentally used electrolyzed water

Two Preparations of alkaline and acidic EW according to Al-Haq *et al.* (2005); Hricova *et al.* (2008); Athayde *et al.* (2018) depending on passing two poles of electrolysis cell connected 24 volts into sufficient amount of salted drinking water (2 g NaCl / L). Upon the onset of the electrolysis process, NaCl dissolved in water and dissociated into Na^+ and Cl^-. Meanwhile, water was reduced at the cathode pole formed hydroxide (OH^-) and Hydrogen (H^+) ions; where, negatively charged ions (OH^- and Cl^-) move towards the anode where electrons are released and hypochlorous acid (HOCl), hypochlorite ions (OCl^-), oxygen gas (O_2) and chlorine gas (Cl_2) are generated. On the other hand, positively charges ions (Na^+ and H^+) move toward the cathode resulting in the generation of sodium hydroxide (NaOH) and hydrogen gas (H_2) forming alkaline EW; whereas a few drops of vinegar 5% was added to the electrolyzed water to adjust the pH to be acidic EW.

3. Experiment groups

The swabs groups divided into 3 groups. Swabs were token from each carcass/group before spraying tested EW, and then,

Group 1: treated with acidic EW [pH 5.6]

Group 2: treated with alkaline EW [pH 8.3]

Group 3: treated with alkaline EW followed by acidic EW

4. Preparation of swab samples (ISO 18593, 2018).

Swabs were taken from the confined area with a template loop of 5cm x 5cm dimensions (25 cm^2); after swabbing, cotton buds ware immediately placed in 1ml of 0.1% solution of peptone broth and held at $4^{O}C$ until plating was accomplished. After appropriate dilutions, ABC was performed according to ISO 4833-1 (2013) on plate count agar that incubated at $30^{O}C$/72h. Colonies were counted and recorded as CFU/cm^2 of sample.

5. Statistical analysis:

A logarithmic transformation of the obtained results was then analyzed using paired samples T-test on SPSS application version 26-2020 (Statistical Package for the Social Science; IBM Corp, Armonk, NY, USA) for Microsoft Windows 10.

- **Reduction rate (%) =** $\frac{B-A}{A}x100$

  A = mean value of ABC before application of EW.

  B = mean value of ABC after application of EW.

RESULTS

It is obvious that treatment of the fresh carcasses with EW had a significant antibacterial effect appeared as a significant reduction in the bacterial counts after EW spraying. Application of acidic EW revealed reduction ranges (%) from 97.4 to 99.5 with a mean value of 98.9% (Table 1 and Fig. 1); while it was 86.5 to 98.9 with a mean value of 97.3% in alkaline group (Table 2 and Fig. 2), treatment with acidic EW gave more antibacterial effect than the alkaline EW. On the other hand, application of alkaline EW followed by acidic EW had a significant synergistic antibacterial effect from 99.3 to 99.9% with a mean reduction value of 99.8% (Table 3 and Fig. 3).

Table 1. Aerobic bacterial count ($\log_{10}$ cfu/ cm^2) in the examined carcasses pre- and post-treatment with Acidic EW (n=10).

Samples	Control group ($\log_{10}$ cfu/ cm^2)	Treated group ($\log_{10}$ cfu/ cm^2)	Reduction %
1st	6.74 ± 0.05	4.75* ± 0.09	98.9
2nd	6.17 ± 0.11	4.14* ± 0.07	99.1
3rd	6.61 ± 0.13	4.68* ± 0.11	99.5
4th	6.75 ± 0.09	4.80* ± 0.2	98.8
5th	5.81 ± 0.2	3.66* ± 0.12	99.2
6th	5.70 ± 0.11	3.75* ± 0.11	98.8
7th	5.79 ± 0.15	3.89* ± 0.09	98.7
8th	6.70 ± 0.08	4.52* ± 0.11	99.3
9th	6.05 ± 0.11	4.05* ± 0.2	99.0
10th	6.45 ± 0.14	4.87* ± 0.14	97.4
Mean ± SD	6.27 ± 0.42	4.31* ± 0.46	98.9

* Values with asterisk superscript in the same raw are significantly different at (P<0.05) using paired sample t- test.

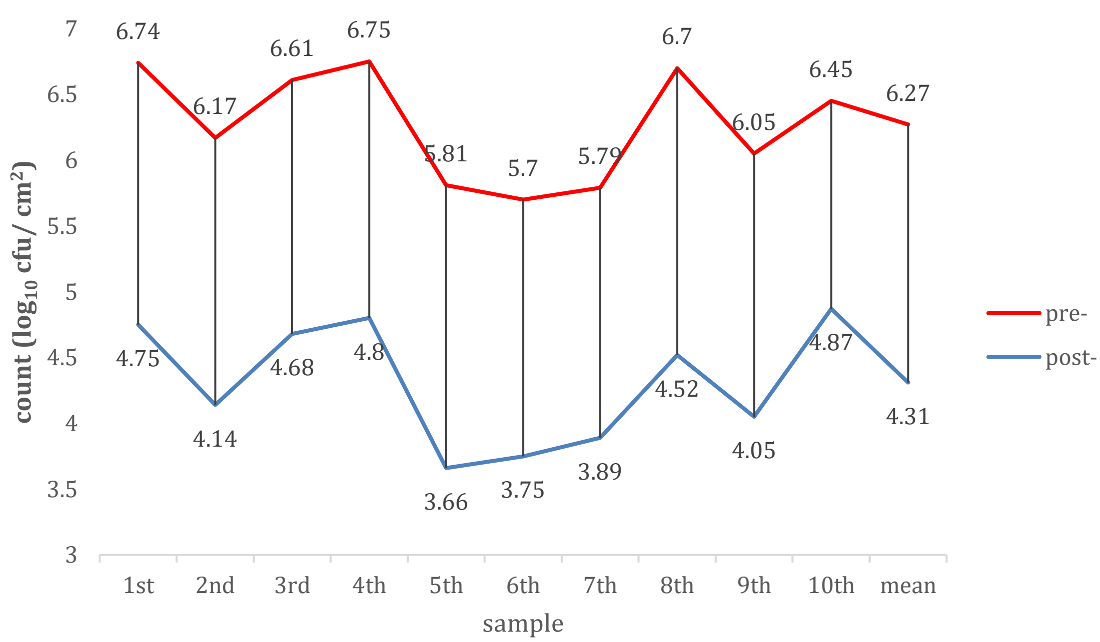

Fig. 1. Aerobic bacterial count ($\log_{10}$ cfu/ cm^2) in the examined carcasses pre- and post-treatment with Acidic EW

Table 2. Aerobic bacterial count (log_{10} cfu/ cm^2) in the examined carcasses pre- and post-treatment with Alkaline EW (n=10).

Samples	Control group (log_{10} cfu/ cm^2)	Treated group (log_{10} cfu/ cm^2)	Reduction %
1st	5.48 ± 0.12	4.61 * ± 0.18	86.5
2nd	6.22 ± 0.14	4.77 * ± 0.11	96.8
3rd	6.18 ± 0.05	4.54 * ± 0.09	97.7
4th	6.67 ± 0.11	4.69 * ± 0.14	98.9
5th	6.70 ± 0.15	4.71 * ± 0.11	96.7
6th	5.94 ± 0.11	4.52 * ± 0.2	96.2
7th	6.74 ± 0.09	4.84 * ± 0.12	98.7
8th	6.53 ± 0.11	4.73 * ± 0.13	98.4
9th	6.40 ± 0.10	4.64 * ± 0.11	98.3
10th	5.48 ± 0.11	4.51* ± 0.15	89.3
Mean ± SD	6.23 ± 0.47	4.65 ± 0.11	97.3

* Values with asterisk superscript in the same raw are significantly different at ($P<0.05$) using paired sample t- test.

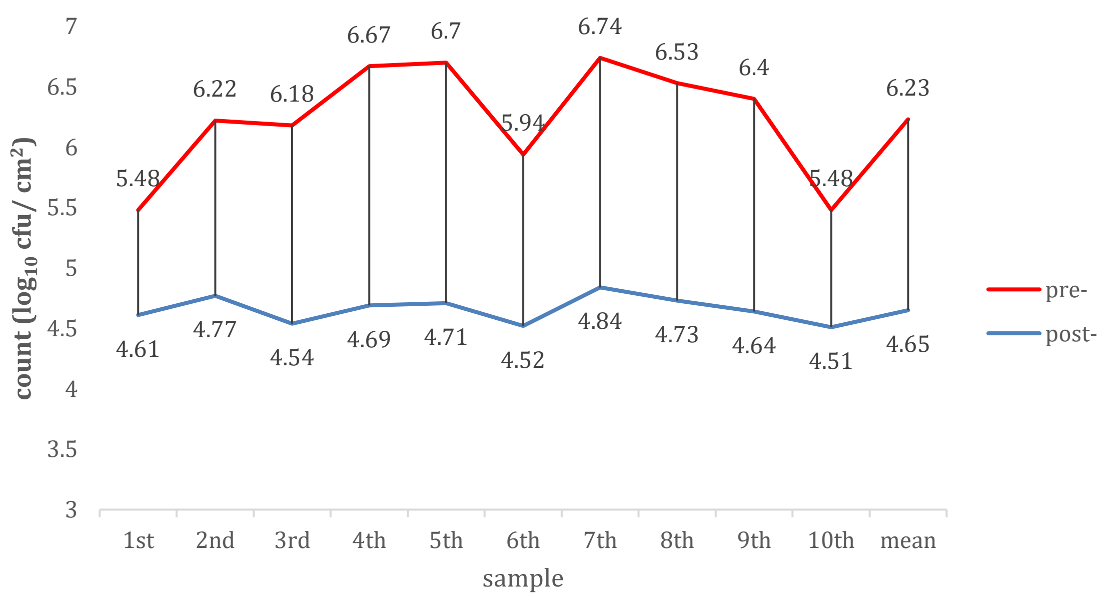

Fig. 2. Aerobic bacterial count (log_{10} cfu/ cm^2) in the examined carcasses pre- and post-treatment with Alkaline EW

Table 3. Aerobic bacterial count (log_{10} cfu/ cm^2) in the examined carcasses pre- and post-treatment with alkaline followed by acidic EW (n=10).

Samples	**Control group (log_{10} cfu/ cm^2)**	**Treated group (log_{10} cfu/ cm^2)**	**Reduction %**
1st	5.94 ± 0.04	3.77* ± 0.08	99.3
2nd	6.74 ± 0.11	3.95* ± 0.2	>99.9
3rd	6.53 ± 0.14	3.28* ± 0.12	99.9
4th	6.40 ± 0.09	3.70* ± 0.11	99.8
5th	5.88 ± 0.06	3.7* ± 0.08	99.3
6th	5.96 ± 0.12	3.71* ± 0.09	99.4
7th	6.49 ± 0.15	3.56* ± 0.20	>99.9
8th	6.24 ± 0.13	3.61* ± 0.16	99.7
9th	6.48 ± 0.11	3.69* ± 0.14	99.8
10th	6.87 ± 0.14	3.75* ± 0.2	99.9
Mean ± SD	6.35 ± 0.34	3.67 ± 0.17	99.8

* Values with asterisk superscript in the same raw are significantly different at (P<0.05) using paired sample t- test.

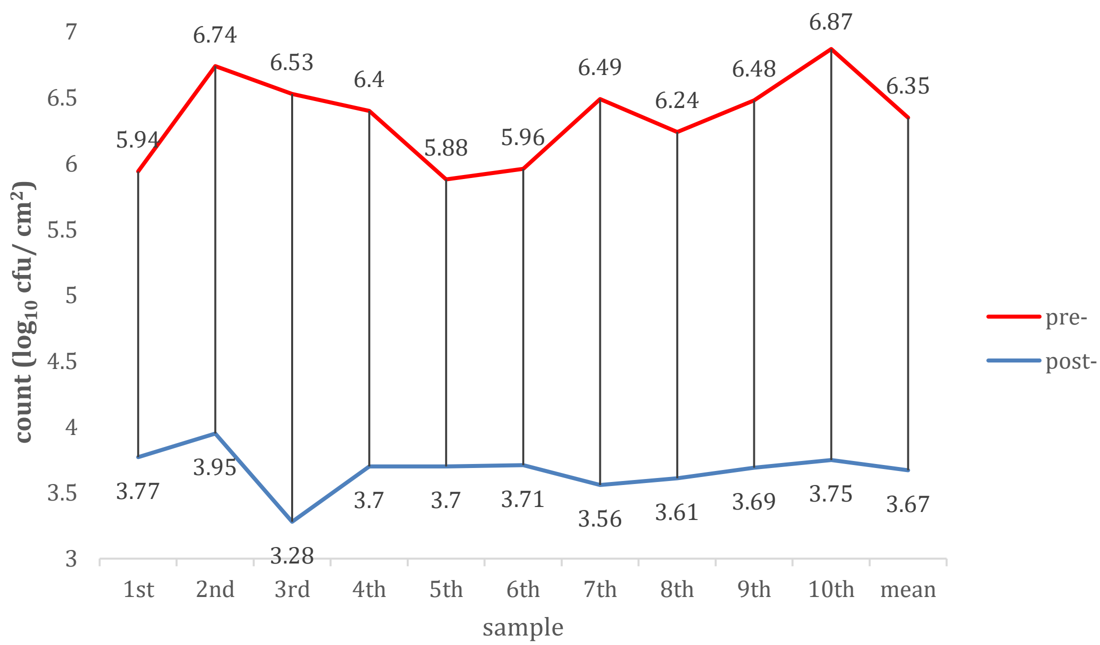

Fig. 3. Aerobic bacterial count (log_{10} cfu/ cm^2) in the examined carcasses pre- and post-treatment with alkaline followed by acidic EW

DISCUSSION

Food poisoning and acute foodborne infections are becoming increasingly common compared to the previous few decades, and a variety of foodborne pathogens are widespread throughout the world, causing great suffering and making safety difficult over time (WHO, 2022).

Many years' worth of literatures has noted that fresh carcasses can become contaminated with a variety of germs from diverse sources. Such contamination could make products harmful for consumers or reduce their usefulness, particularly in developing nations where sanitary regulations are still being implemented. Numerous attempts were made to import beef free of pathogens that pose a risk to public health (Karanth *et al.*, 2023).

Many methods have been developed to date to prevent foodborne illness outbreaks and guarantee a safe food supply. Over the past few years, electrolyzed oxidizing water, also known as electrolyzed strong acid aqueous solution (ESAAS) or strong acidic electrolyzed water (StAEW), has been used all over the world as a novel antimicrobial. Several studies have demonstrated its antimicrobial potential against a wide range of microorganisms (Rebezov *et al.*, 2022).

In the various food industries, electrolyzed water is used as disinfectant for cutting tools, an antimicrobial agent for the carcasses of poultry birds, and for disinfecting eggs and meat decontamination industry (Zang *et al.*, 2019; Hamidi *et al.*, 2020).

Referring to the current results, significant reduction in the bacterial count in the treated groups rather than the pre-treated samples at ($P \leq 0.05$) was recorded; where the mean reduction (%) were 98.9, 97.3 and 99.8 for the treated samples with acidic EW, alkaline EW, and both alkaline and acidic EW, respectively. Referring to the obtained results, acidic EW showed higher antibacterial effect than alkaline EW; whereas, using the alkaline EW followed by acidic EW revealed more decontamination effect than using each alone.

The obtained results came in agreement with those recorded by McCarthy and Burkhardt III (2012) who studied antimicrobial effect of EW in objects related to food preparation in intermittent spray application to reduce or prevent bacterial biofilm formation; Sun *et al.* (2012) who concluded that alkaline EW have capacity to remove *S. aureus* biofilm compared to 2% of NaOH, and acidic EW have bactericidal effect compared to 2% of HCl with a possible synergistic action; Al-Holy and Rasco (2015) who evaluated the action of EW by soaking fish, chicken and beef surfaces that were experimentally contaminated with *E. coli, Salmonella* and *L. monocytogenes*; Jiménez-Pichardo *et al.* (2016) who recorded that the action of EW was better than common sanitizers used in dairy industry hygiene; Athayde *et al.* (2017) who used EW as surface decontaminant on pork carcasses; and Tolba *et al.* (2020) who used EW for its antimicrobial effect on shrimp by soaking. It is worth note that they all agreed in the point of the significant antimicrobial effect of EW with minimal variation on its potency referring to its pH, time of exposure and form of application.

In fact, there are numerous theories regarding the mechanism of action of EW antimicrobials. According to Liao *et al.* (2007), the oxidation reduction potential of EW has the ability to harm both the inner and outer membranes of bacteria, resulting in cell necrosis. Chlorine can also impact bacteria by blocking enzymes involved in the metabolism of carbohydrates that are sensitive to sulfhydryl groups; thus prevented the oxidation of glucose (Eifert and Sanglay, 2002). Microorganisms' EW activity is caused by one or more processes. Key enzyme inactivation, nucleic acid degradation, and damage to the wall and other vitals that may be occurred (White, 2010). Additionally, EW can alter the membrane's permeability, increase the affected microbial cell's conductivity, and decrease dehydrogenase activity (Zeng *et al.*, 2010).

Therefore, electrolyzed water is not only inexpensive but is also far more effective than conventional decontaminants. Electrolyzed water kills pathogenic microorganisms, and protects the environment from the adverse impacts of hazardous chemical disinfectants.

CONCLUSION

Out of the recorded results, it is obvious that application of EW has a potential significant antimicrobial effect on the meat industry, especially on the carcass surface decontamination after spraying technique. Application of alkaline and acidic EW combination revealed to maximize their antimicrobial action with synergistic effect. Electrolyzed water is a green disinfectant with no environmental hazard and harmful residues; so, it is recommended to replace chemical disinfectant with EW in the meat decontamination.

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CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

REFERENCES

Al-Haq, M.I.; Sugiyama, J. and Isobe, S. (2005). Application of electrolyzed water in agriculture and food industries. Food Sci. Technol Res. 11: 135-150.

Al-Holy, M.A. and Rasco, B.A., (2015). The bactericidal activity of acidic electrolyzed water against *Escherichia coli* O157:H7, *Salmonella* Typhimurium, and *Listeria monocytogenes* on raw fish, chicken and beef surfaces. Food Control 54: 317-321.

Athayde, D.; Flores, D.; da Silva, J.; Genro, A.; Silva, M.; Klein, B.; Mello, R.; Campagnol, P.; Wagner, R.; Menezes, C.; Barin, J. and Cichoski, A. (2017). Application of electrolyzed water for improving pork meat quality. Food Res. Int. 100: 757-763.

Athayde, D.R.; Flores, D.R.M.; Silva, J.S.; Silva, M.S.; Genro, A.L.G.; Wagner, R.; Campagnol, P.C.B.; Menezes, C.R. and Cichoski, A.J. (2018). Characteristics and use of electrolyzed water in food industries. Int. Food Res. J. 25: 11-16.

Barcenilla, C.; Álvarez-Ordóñez, A.; López, M.; Alvseike, O. and Prieto, M. (2022). Microbiological safety and shelf-life of low-salt meat products-A Review. Foods 11: 2331.

Chakka, A.K.; Sriraksha, M.S. and Ravishankar, C.N. (2021). Sustainability of emerging green non-thermal technologies in the food industry with food safety perspective: A review. LWT 151: 112140.

Eifert, J.D. and Sanglay, G.C., (2002). Chemistry of chlorine sanitizers in food processing. Dairy, Food and Environmental Sanitation 22: 534-538.

Fan, S.; Zhang, F.; Liu, S.; Yu, C.; Guan, D. and Pan, C. (2013). Removal of aflatoxin B1 in edible plant oils by oscillating treatment with alkaline electrolyzed water. Food Chem. 141: 3118–3123.

Gómez-Espinosa, D.; Cervantes-Aguilar, F.J.; Río-García, J.C.; Villarreal-Barajas, T.; Vázquez-Durán, A. and Méndez-Albores, A. (2017). Ameliorative effects of neutral electrolyzed water on growth performance, biochemical constituents, and histopathological changes in Turkey poults during aflatoxicosis. Toxins. 9: 104.

Hamidi, M.R.; Shekarforoush, S.S.; Hosseinzadeh, S. and Basiri, S. (2020). Evaluation of the effect of neutral electrolyzed water and peroxyacetic acid alone and in combination on microbiological, chemical, and sensory characteristics of poultry meat during refrigeration storage. Food Sci. Technol. Int. 27: 499–507.

Hricova, D.; Stephan, R. and Zweifel, C. (2008). Electrolyzed water and its application in the food industry. J. Food Protec. 71: 1934-1947.

ISO "International Organization for Standardization", (2013). International Organization for Standardization. No. 4833-1. Microbiology of the food chain-Horizontal method for the enumeration of microorganisms-Part 1: Colony count at 30 °C by the pour plate technique".

ISO "International Organization for Standardization", (2018). International Organization for Standardization. No. 18593. Microbiology of the food chain — Horizontal methods for surface sampling.

Jiménez-Pichardo, R.; Regalado, C.; Castaño-Tostado, E.; Meas-Vong, Y.; Santos-Cruz, J. and García- Almendárez, B.E. (2016). Evaluation of electrolyzed water as cleaning and disinfection agent on stainless steel as a model surface in the dairy products. Food Control. 60: 320-328.

Karanth, S.; Feng, S.; Patra, D. and Pradhan, A.K. (2023). Linking microbial contamination to food spoilage and food waste: the role of smart packaging, spoilage risk assessments, and date labeling. Front. Microbiol. 14: 1198124.

Khan, I.; Tango, C.N.; Miskeen, S.; Lee, B.H. and Oh, D.H. (2017). Hurdle technology: A novel approach for enhanced food quality and safety—A review. Food Control 73: 1426–1444.

Liao, L.B.: Chen, W.M. and Xiao, X.M. (2007). The generation and inactivation mechanism of oxidation-reduction potential of electrolyzed oxidizing water. J. Food Engin. 78- 1326-1332.

Lung, H.M.; Cheng, Y.C.; Chang, Y.H.; Huang, H.W.; Yang, B.B. and Wang, C.Y. (2015). Microbial decontamination of food by electron beam irradiation. Trends Food Sci. Technol. 44: 66–78.

Manyi-Loh, C.E. and Lues, R.A. (2023). South African perspective on the microbiological and chemical quality of meat: Plausible public health implications. Microorganisms. 11 : 2484.

McCarthy, S. and Burkhardt III, W. (2012). Efficacy of electrolyzed oxidizing water against *Listeria monocytogenes* and *Morganella morganii* on conveyor belt and raw fish surfaces. Food Control. 24: 214-219.

Mosaka, T.B.M.; Unuofin, J.O.; Daramola, M.O.; Tizaoui, C. and Iwarere, S.A. (2023). Inactivation of antibiotic-resistant bacteria and antibiotic-resistance genes in wastewater streams: Current challenges and future perspectives. Front. Microbiol. 13: 1100102.

Rebezov, M.; Saeed, K.; Khaliq, A.; Rahman, S.J.U.; Sameed, N.; Semenova, A.; Khayrullin, M.; Dydykin, A.; Abramov, Y. and Thiruvengadam, M. (2022). Application of electrolyzed water in the food industry: A Review. Appl. Sci. 12(13): 6639.

Saraiva, S.M.; Lim, K.; do Monte, D.F.M.; Givisiez, P.E.N.; Alves, L.B.R.; de Freitas Neto, O.C.; Kariuki, S.; Júnior, A.B.; de Oliveira, C.J.B. and Gebreyes, W.A. (2022). Antimicrobial resistance in the globalized food chain: a One Health perspective applied to the poultry industry. Brazilian J. Microbiol. 53: 465–486.

Silva, A.; Silva, M. and Ribeiro, B. (2020). Health issues and technological aspects of plant-based alternative milk. Food Res. Int. 131: 108972.

Sun, J.L.; Zhang, S.K.; Chen, J.Y. and Han, B.Z. (2012). Efficacy of acidic and basic electrolyzed water in eradicating *Staphylococcus aureus* biofilm. Canadian J. Microbiol. 58: 448-454.

Tolba, K.; Ali, H. and Omara, N. (2020). Slightly acidic electrolyzed water (SAEW) and its relation with shelf life of chilled shrimps. Anim. Health Res. J. 8: 1-14.

White, G.C. (2010). Chemistry of aqueous chlorine. In: White's handbook of chlorination and alternative disinfectants. 5th ed, p 152-153. New Jersey: John Wiley and Sons.

WHO 'World Health Organization', (2022). Food safety. Available on https://www.who.int/news-room/fact-sheets/detail/food-safety. Accessed in 22/2/2024.

Yan, P.; Daliri, E.B. and Oh, D.H. (2021). New clinical applications of electrolyzed water: A Review. Microorganisms 9, 136.

Zang, Y.T.; Bing, S.; Li, Y.J.; Shu, D.Q.; Huang, A.M.; Wu, H.X.; Lan, L.T. and Wu, H.D. (2019). Efficacy of slightly acidic electrolyzed water on the microbial safety and shelf life of shelled eggs. Poultry Sci. 98: 5932–5939.

Zeng, X.; Tang, W.; Ye, G.; Ouyang, T.; Tian, L.; Ni, Y. and Li, P. (2010). Studies on disinfection mechanism of electrolyzed oxidizing water on *E. coli* and *Staphylococcus aureus*. J. Food Sci. 75: 253-260.