NON-THERMAL TECHNOLOGIES

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NON-THERMAL TECHNOLOGIES

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Antonio Morata holds patents in wine technology specifically related to aging on lees, grape skin separation and brettanomyces analysis. All other Topic Editors declare no competing interests with regard to the Research Topic subject.

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EDITORIAL
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Editorial: Non-thermal Technologies

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Editorial on the Research Topic

Non-thermal Technologies

Non-thermal technologies for food applications comprises technologies such as ultrasound, cold plasma, high-pressure, pulsed electric field, ozone, UV, electrolyzed water, and pulsed UV light. The development of these technologies has been growing in the last decade due to its successful application in sanitization, but also as processes that can improve nutritional and sensory properties of food products. Some technologies, such as high-pressure processing, ozone, UV light, and pulsed electric field have been used in industrial processes for at least two decades. While others, such as ultrasound, cold plasma, pulsed UV light are still under development, and are currently in lab- and pilot-scale phases.

Pasteurization, sterilization, and other thermal technologies used to sanitize food products usually affects negatively the nutritional and sensory properties of many products. As food quality and nutrition becomes a major concern in the modern world, it is not possible anymore to rely on technologies that sanitize our food but that also reduce their sensory and nutritional quality. Studies with non-thermal technologies, at first, aimed at achieving the same level of sanitization, while maintaining the nutritional and sensory properties of the products as similar as possible to natural. The application of these non-thermal processes in sanitization has been well-explored, and the knowledge of the strains of microorganisms that can be inactivated and how this inactivation is achieved was the focus of several research. In this Research Topic, articles on ultra-high pressure homogenization by Morata and Guamis and acidic electrolyzed water by Liu et al. present some aspects involving non-thermal technology and sanitization.

Although non-thermal technologies do not involve heat, several of these technologies induce chemical reactions in the food product. These reactions may alter the food tissue structure, enzymes, metabolic rates, phenolics, antioxidants, vitamins, color, and other sensory and nutritional aspects of our food. Under some circumstances the changes are positive, improving the phenolic and free vitamin content, inactivating color degrading enzymes, improving cloud stability in juices; but there are also reports on negative effects such as color fading, texture loss and production of off-flavor. Reports on these kinds of effects of non-thermal technologies on food is still missing and are the primary focus of this Research Topic.

Articles on the impact of ultra-high pressure homogenization, plasma treated water, ultrasound on different food products can be found in this Research Topic, as well as a review on the theory and application of several non-thermal technologies. The article written by Morata and Guamis shows a good examples of how non-thermal technologies can boost the production of wines and juices with higher antioxidant capacities without resorting to the addition of sulfur dioxide. Schnabel et al. present how plasma treated water can be an effective and low-cost process to extend the shelf-life of vegetables, while Liu et al. report on the application of acidic electrolyzed water to process fish. A review on the theory and application of several non-thermal technologies is presented by Jadhav et al., while Astráin-Redín et al. focus on direct contact ultrasound.

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Non-thermal technologies have also been used in non-invasive quality control, where the product or the process can be monitored. In this Research Topic, Wang et al. demonstrate the efficacy of on low-field nuclear magnetic resonance in monitoring the quality control of aquatic products.

This Research Topic illustrate some of the possibilities that non-thermal emerging technologies open in the new food technology allowing the production of safer, healthier, and more sensory-attractive foods.

AUTHOR CONTRIBUTIONS

FF prepared a draft concept on the present Resource Topic, which was supplemented and corrected by SR and AM. All authors contributed to the article and approved the submitted version.

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MINI REVIEW
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Use of UHPH to Obtain Juices With Better Nutritional Quality and Healthier Wines With Low Levels of SO_2

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Ultra-high pressure homogenization (UHPH) is a high pressure technique in which a fluid is pressurized by pumping at higher than 200 MPa and instantaneously depressurized at atmospheric pressure across a special valve. The full process takes <0.2 s and the in-valve time is <0.02 s. In the valve, extremely intense impacts and shear forces produce the nanofragmentation of biological tissue at a range of 100–300 nm. The antimicrobial effect is highly effective, reaching easily inactivation levels higher than 6-log cycles even at low in-valve temperatures. At in-valve temperatures of 140–150°C (0.02 s) the destruction of thermoresistant spores is possible. Even when the temperature in-valve can be elevated (70–150°C), it can be considered a gentle technology because of the tremendously short processing time. It is easy to get outlet temperatures after valve of 20–25°C by the expansion and assisted by heat exchangers. Thermal markers as hydroxymethylfurfural (HMF) are not formed, nor are deleterious effects observed in sensitive compounds as terpenes or anthocyanins, probably because of the low effect in covalent bonds of small molecules of the high-pressure techniques compared with thermal technologies. Additionally, intense inactivation of oxidative enzymes is observed, therefore protecting the sensory and nutritional quality of fruit juices and avoiding or reducing the use of antioxidants as sulphites. UHPH can be consider a powerful and highly effective continuous and sterilizing technology without thermal repercussions, able to keep fresh juices with most of their initial sensory and nutritional quality and allowing high-quality and natural fermented derivatives as wine.

Keywords: emerging technologies, grape must, winemaking, oxidative enzymes, colloidal stability, additives, sulphites

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INTRODUCTION

Currently there are available at commercial and industrial levels two groups of high pressure technologies: (i) discontinuous high hydrostatic pressure (HHP) and (ii) continuous high pressure homogenization including high pressure homogenization (HPH), microfluidization (MF), and ultrahigh pressure homogenization (UHPH). The first group (i) uses a fluid, usually water,

as pressurizing media, and the antimicrobial effect is produced by the damage produced in membranes and cell walls by the intense pressures ranging from 400 to 600 MPa, during 3–10 min (1, 2). It is possible to process solid and liquid foods. This technology is discontinuous on batch-mode and unable to inactivate spores (3) and with low effect in enzymes (4, 5). However, it is highly protective with molecules with nutritional or sensory impact, e.g., vitamins, aroma compounds, and pigments. The second group (ii) are continuous technologies in which the liquid is pumped at high pressure. In UHPH, which is the most effective, pressure ranges from 200 to 600 MPa (**Figure 1**). The antimicrobial effect is produced by the intense impact and shear forces produced in the valve (2, 6, 7). Liquid suffers strong acceleration when pumped at 300 MPa, reaching Mach 2 speed and extremely intense deceleration to almost zero when crossing the valve during <0.02 s. The effect is a full nanofragmentation to a submicro size range of 100–300 nm of whatever biological structure: cells, tissues, and biomolecules including enzymes (8). Depending on in-valve temperature (140°C), even spores are destroyed. UHPH sterilization is an alternative technique to UHT but with lower sensory and nutritional impact.

The objective of this review is to explain the main features of the processing by UHPH and its advantages compared with the conventional hydrostatic pressurization (HHP). Additionally, it described the impact of UHPH in sensory and nutritional quality and the reduction of conflictive additives as sulphites.

PERFORMANCE OF CONTINUOUS UHPH VS. HHP

UHPH is a powerful technique that can be applied in continuous mode to whatever fluid with a size particle lower than 0.5 mm and a viscosity below 2,000 centipoises (9), and therefore can be used in most food fluids. Currently UHPH technologies can be found at the commercial level; one of the most evolved is patented by UAB and exploded by Ypsicon Advances Technologies (10). Liquids are pumped usually at 300 MPa, which can be considered the limit between HPH and UHPH (11), and the antimicrobial effect and enzyme destruction is produced in a special valve (7, 12) produced with special strong materials as tungsten carbide (weaker) or artificial diamond (highly resistant). The homogenizing valve has a few μm in width favoring mechanical interactions with microbial cells (11, 13). In-valve the intense impact and shear efforts produce several effects at nano-scale: nano-fragmentation, nano-covering, nano-encapsulation, and nano-emulsion that can be modulated to produce sterile or pasteurized foods (2, 7, 8, 14, 15), to increase the accessibility of nutrients and health-promoting compounds (16) and to develop innovative foods with improved colloidal structure or novel properties.

Pressure range in UHPH is 200–600 MPa; however, at 300 MPa the microbial and enzyme inactivation is more intense. Pressure higher than 300 MPa does not produce more intense effects but sometimes increases the level of wearing in-valve and other components. Concerning the temperature, most of the applications can be done at pasteurization or lower temperatures (<70°C). This includes enzyme inactivation, microbial destruction, nano-emulsion, and nano-covering (7, 8, 14, 17).

Concerning microbial inactivation at low pasteurization temperatures (<70°C) or even at room temperature, but considering the shorter processing times of UHPH (0.02 s in-valve and around 0.2 s of total process), it is possible to eliminate vegetative cells of fungi, yeast, and bacteria at populations of 6-log CFU/mL or higher (8, 14). The short in-valve time at this temperature guarantees the absence of thermal markers as hydroymethylfurfural (HMF) (8).

The elimination of bacterial endospores requires higher temperature 140–150°C (7, 18); however, thermal effect is very soft considering the short processing time that is lower than 0.2 s for the full process (14). This allows a higher sensory quality than traditional UHT.

The main features of UHPH in comparison with HHP concern the processing mode, effects on food components, antimicrobial capacity, impact in nutritional and sensory properties, and evaluation of thermal degradation as shown in **Table 1**. UHPH shows some advantages concerning gentle impact in food quality with a more effective antimicrobial and anti-enzymatic performance.

ELIMINATION OF SPOILAGE MICROORGANISMS IN MUST AND WINES AND IMPROVED APPLICATION OF NEW BIOTECHNOLOGIES

Spoilage yeasts that can be present in grapes, such as *Brettanomyces*, are easily destroyed by UHPH processing (14). Lactic or acetic undesired bacteria that can develop in the future wine increasing volatile acidity, or forming biogenic amines or ethyl carbamate, will be eliminated from the grape must using UHPH, ensuring a healthier wine. The use of 300 MPa and temperatures of 60–100°C for 0.2 s eliminate vegetative bacterial cells (8, 14). Sporulated forms need higher temperatures (6, 7, 18). Toxins as patulin were not directly affected by UHPH; however, a significant decrease can be observed in UHPH juices during storage (18).

There is facilitation of new biotechnologies as use of non-*Saccharomyces* yeasts and yeast-bacteria co-inoculations by a better implantation of the starters in absence of indigenous competitive microorganism (14, 23).

DESTRUCTION OF OXIDATIVE ENZYMES (PPO) AND RETENTION OF ANTIOXIDANT CAPACITY IN JUICES

HPH can be used to modulate enzymatic activity (42), but at higher pressure using UHPH the enzymes can be denatured. UHPH processing is able to disrupt quaternary structure of proteins (43). The intense inactivation of oxidative enzymes such as polyphenol oxidase (PPO), usually higher than 90%

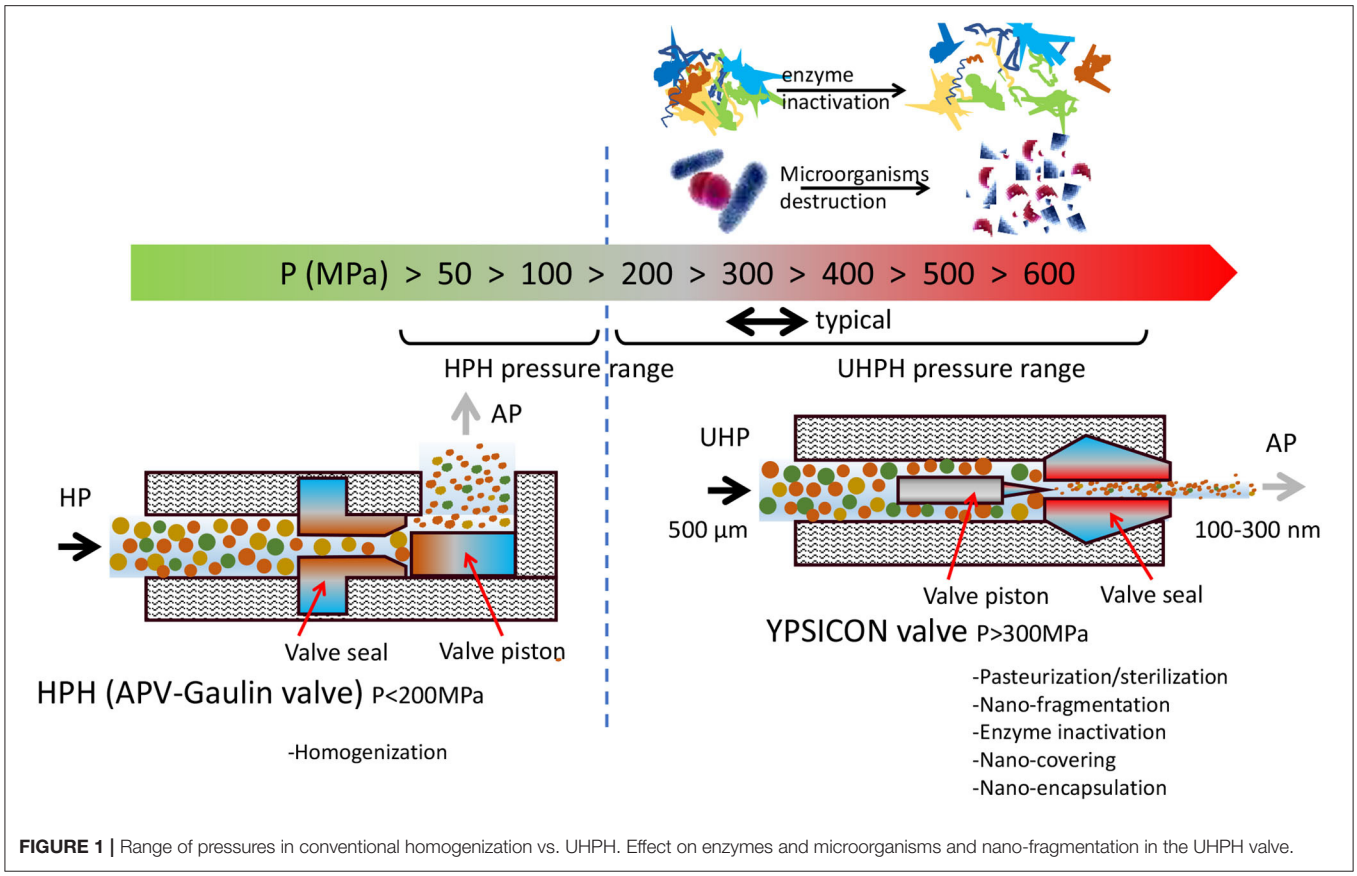

FIGURE 1 | Range of pressures in conventional homogenization vs. UHPH. Effect on enzymes and microorganisms and nano-fragmentation in the UHPH valve.

(14, 18), produces wines with antioxidant capacities increased more than 150% regarding controls (8, 14). Preserved antioxidant capacity has also been observed in apple or strawberry juices (30, 44). Moreover, the stability of vitamin C in HPH and UHPH treatments has been confirmed (30, 45). Additionally, UHPH grape must can be exposed to air with high exchange surfaces (>1 cm^2/mL) without experimenting oxidations or browning processes during several days. The inactivation of pectinmethylesterase has been also observed in apple juices as what contributes to the colloidal stability of turbid juices (18).

The impact of UHPH in the wine industry is the potential reduction of sulfur dioxide (SO_2) in wines, opening the possibility to produce wines with 0 mg/L of sulphites by inactivating oxidative enzymes but also destroying spoilage microorganisms (8, 14). For juices both the antimicrobial and antienzyme effect facilitate the production of low processed juices that can keep their sensory profile in absence of antimicrobials and antioxidative chemical products for long periods from months to years. The production of SO_2 free red wine was studied using discontinuous processing by HHP obtaining a good sensory quality (46). However, recently, it has been reported that it is necessary to have at least 60 mg/L of SO_2 to preserve quality in red wines processed at 350 MPa during 10 min at 8°C (47). Wines with less of 60 mg/L were found both less aromatic and with lower contents of anthocyanins.

SENSORY AND NUTRITIONAL QUALITY

Concerning sensory and nutritional quality, the short thermal effect produced by the in-valve temperature is not affecting the degradation of aroma compound as terpenes (8) or delicate pigments as anthocyanins (25, 48). Vitamin contents remain unaffected. Vitamin C contents remain unaffected after UHPH processing in apple juices (18). Thermal markers as HMF are not detected after the UHPH processing (8, 14).

It has been observed that delicate aroma compounds as several terpenes from a Muscat juice are not affected by UHPH treatments at 300 MPa, 65°C, processing time lower than 0.2 s. Concentrations of linalool, terpinen-4-ol, epoxylinalool, β-citronellol, geraniol, α-terpineol, cis-linalool oxide did not show significant differences with the unprocessed controls (8). Also, differences were not found in the pool of polyoxygenated terpenes. Additionally, the sensory panel also did not detect significant differences in the aromatic varietal profile.

Extraction of phenolic compounds can be improved significantly when juices with pulps or skin fragments are processed by UHPH. The intense nanofragmentation of colloids that are reduced to a size range of 100–300 nm increase the extraction of flavonoids and phenols from solids (18).

TABLE 1 | Comparative features of UHPH and HHP.

Effect	UHPH	HHP	References
Operating mode	Continuous pumping. Treatment time 0.2 s	Discontinuous pressurization with water. Treatment time 3–10 min	(2, 7)
Effect on covalent bonds in small molecules with sensory impact	Unaffected	Unaffected	(19)
Molds and yeasts control	Highly effective at 300 MPa	Highly effective at >400 MPa/5–10 min	(1, 8, 14, 20–23)
Bacteria	Highly effective at 300 MPa	Effective at >600 MPa/5–10 min	(1, 14, 20, 22, 23)
Bacterial endospores	Highly effective at 300 MPa, if in-valve Temperature 140°C 0.2 s	Not applicable. Effective at >1,000 MPa/5–10 min	(7, 18, 24)
Effect on biopolymers	Intense fragmentation. 100–300 nm.	Starch gelatinization. Protein denaturation.	(7, 19)
Oxidative enzymes	Strong inactivation at 300 MPa. PPO inactivation >90%. Absence of browning during more than 5 days in air exposed juices	Weak. Variable, usually needs temperature assistance	(8, 14, 25, 26)
Thermal markers (HMF)	Undetected	Lower than in thermal treatments	(8, 27)
Antioxidant capacity	Increased >150%	Non differences-slight reduction	(14, 28, 29)
Vitamins	Preserved	Preserved	(30, 31)
Terpenes and aroma molecules	Unaffected	Unaffected	(8, 32)
Anthocyanins	Unaffected. Increased extraction from skin colloidal particles	Unaffected. Improved extraction from grapes	(1, 14, 25, 28)
Polyphenols	Improved extraction from apple and grapes	Non-significant differences in total phenolics and flavonoids. Improved extraction from grapes.	(1, 18, 33)
Sensory profile	Unaffected. Better fruitiness	Unaffected	(8, 33–35)
New fermentative biotechnologies as use of non-*Saccharomyces* and yeast-bacteria co-inoculations	Better implantation and lesser competitiveness with indigenous microbiota	Better implantation and lesser competitiveness with indigenous microbiota	(1, 14, 23)
Release of yeast assimilable nitrogen (YAN) and nutritional properties of must	Increased extraction from juice cell fragments. Favors the formation of fermentative esters.	Not described	(14)
Protein digestibility	Improved	Improved	(36, 37)
Allergenicity	Decreased	Decreased	(36, 38, 39)
Colloidal and color stability.	Improved. Higher stability of nanofragmented particles. Color stability. Better protein stability and lower haze formation.	Unaffected. Better protein stability and delayed protein haze.	(8, 14, 15, 34, 36, 40, 41)

The color of white musts is improved by the intense inactivation of oxidative enzymes, making it possible to produce clear and pale juices from white grape varieties (8). Furthermore, the color remains pale even after several days of intense exposure to air.

The intense nano-fragmentation on grape cell walls produces a release of nitrogen compounds that increase the yeast assimilable nitrogen (YAN) available with positive effects in yeast nutrition during fermentation but also affecting the formation of fermentative fruity and floral esters (14).

In vegetal beverages of soya and almond processed by UHPH, there have been observed a better protein digestibility and a lower allergenicity (36). The effect is probably due to denaturation of the protein structure. Contents of lysine were stable after the treatment.

HIGHLIGHT OF FUTURE DIRECTIONS

The inactivation of spores at lower temperatures can be optimized probably by a new design of special valves built with new materials increasing the impact and shear forces. New geometries of valves increase the mechanical effects, promoting fluid jet impact, cavitation, and extreme shear efforts. Emerging materials also can help to improve the possibilities of processing abrasive foods as high fiber or high viscosity juices and smoothies. The conventional tungsten carbide alloy used in the valves must be substituted by ceramic coverings or artificial diamond seats and needles. There must be better knowledge of the nanoscale processes, and how to manage and monitor the nano-covering, nano-encapsulation, and nano-emulsion processes. The co-injection of different fluids at ultrahigh pressure promotes the formation of interactions among proteins, polysaccharides, and lipids creating nano-structures able to content and to increase the bioavailability of nutraceutical compounds. Using UHPH, it is possible to produce functional foods by the nanoencapsulation and stabilization of nutritional or sensory compounds. Frequently, these nano-encapsulates are more stable in solution or colloidal dispersion creating food fluids with same appearance in the long term without coalescence phenomena. Development of high throughput UHPH machines able to process more than

10,000 L/h is another key parameter to reach industrial scale-up. Currently, there are industrial UHPH devices able to process at 10,000 L/h working at 300 MPa, just with a single pump, but it is necessary to have several pumps to increase this processing flow. There must be development of truck portable UHPH devices to rent the processing technology to be used in seasonal food industries. The elimination of chemical preservatives as SO_2, sorbates, benzoates, and others can help to reach a key objective in the food industry: cleaning labels.

AUTHOR CONTRIBUTIONS

AM: conceptualization, writing draft, revision. BG: writing, supervision. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Morata A, Loira I, Vejarano R, Bañuelos MA, Sanz PD, Otero L, et al. Grape processing by high hydrostatic pressure: effect on microbial populations, phenol extraction and wine quality. *Food Bioprocess Tech*. (2015) 8:277–86. doi: 10.1007/s11947-014-1405-8
2. Morata A, Loira I, Vejarano R, González C, Callejo MJ, Suárez-Lepe JA. Emerging preservation technologies in grapes for winemaking. *Trends Food Sci Tech*. (2017) 67:36–43. doi: 10.1016/j.tifs.2017.06.014
3. Sarker MR, Akhtar S, Torres JA, Paredes-Sabja D. High hydrostatic pressure-induced inactivation of bacterial spores. *Crit Rev Microbiol*. (2015) 41:18–26. doi: 10.3109/1040841X.2013.788475
4. Cao X, Zhang Y, Zhang F, Wang Y, Yi J, Liao X. Effects of high hydrostatic pressure on enzymes, phenolic compounds, anthocyanins, polymeric color and color of strawberry pulps. *J Sci Food Agric*. (2011) 91:877–85. doi: 10.1002/jsfa.4260
5. Chakraborty S, Kaushik N, Rao PS, Mishra HN. High-Pressure inactivation of enzymes: a review on its recent applications on fruit purees and juices. *Compr Rev Food Sci Food Saf*. (2014) 13:578–96. doi: 10.1111/1541-4337.12071
6. Dumay E, Chevalier-Lucia D, Picart-Palmade L, Benzaria A, Gràcia-Julià A, Blayo C. Technological aspects and potential applications of (ultra) high-pressure homogenization. *Trends Food Sci Technol*. (2013) 31:13–26. doi: 10.1016/j.tifs.2012.03.005
7. Zamora A, Guamis B. Opportunities for ultra-high-pressure homogenization (UHPH) for the food industry. *Food Eng Rev*. (2015) 7:130–42. doi: 10.1007/s12393-014-9097-4
8. Bañuelos MA, Loira I, Guamis B, Escott C, Del Fresno JM, Codina-Torrella I, et al. White wine processing by UHPH without SO_2. Elimination of microbial populations and effect in oxidative enzymes, colloidal stability and sensory quality. *Food Chem*. (2020) 332:127417. doi: 10.1016/j.foodchem.2020.127417
9. Ypsicon. *Ultra High Pressure Homogenization/Sterilization.* (2018). Retrieved from: www.ypsicon.com (accessed July 2020).
10. Guamis-Lopez B, Trujillo-Mesa AJ, Ferragut-Pérez V, Quevedo-Terré JM, Lopez-Pedemonte T, Buffa-Dunat MN. *Continuous System and Procedure of Sterilization and Physical Stabilization of Pumpable Fluids Bymeans of Ultrahigh Pressure Homogenization*. EP2409583. Barcelona: Universidad Autónoma de Barcelona (2012).
11. Sevenich R, Mathys A. Continuous versus discontinuous ultra-high-pressure systems for food sterilization with focus on ultra-high-pressure homogenization and high-pressure thermal sterilization: a review. *Compr Rev Food Sci Food Saf*. (2018) 17:646–62. doi: 10.1111/1541-4337.12348
12. Patrignani F, Lanciotti R. Applications of high and ultra high pressure homogenization for food safety. *Front Microbiol*. (2016) 7:1132. doi: 10.3389/fmicb.2016.01132
13. Balasubramaniam V, Martínez-Monteagudo SI, Gupta R. Principles and application of high pressure-based technologies in the food industry. *Ann Rev Food Sci Technol*. (2015) 6:435–62. doi: 10.1146/annurev-food-022814-015539
14. Loira I, Morata A, Bañuelos MA, Puig-Pujol A, Guamis B, González C, et al. Use of ultra-high pressure homogenization processing in winemaking: control of microbial populations in grape musts and effects in sensory quality. *Innov Food Sci Emerg Technol*. (2018) 50:50–6. doi: 10.1016/j.ifset.2018.10.005
15. Comuzzo P, Calligaris S. Potential applications of high pressure homogenization in winemaking: a review. *Beverages*. (2019) 5:56. doi: 10.3390/beverages5030056
16. Levy R, Okun Z, Shpigelman A. High-pressure homogenization: principles and applications beyond microbial inactivation. *Food Eng Rev*. (2020). doi: 10.1007/s12393-020-09239-8
17. Velázquez-Estrada RM, Hernández-Herrero MM, Guamis-López B, Roig-Sagués AX. Impact of ultra-high pressure homogenization on pectin methylesterase activity and microbial characteristics of orange juice: a comparative study against conventional heat pasteurization. *Innov Food Sci Emerg Technol*. (2012) 13:100–6. doi: 10.1016/j.ifset.2011.09.001
18. Suárez-Jacobo Á, Saldo J, Rüfer CE, Guamis B, Roig-Sagués AX, Gervilla R. Aseptically packaged UHPH-treated apple juice: Safety and quality parameters during storage. *J Food Eng*. (2012) 109:291–300. doi: 10.1016/j.jfoodeng.2011.09.007
19. Cheftel JC. Effects of high hydrostatic pressure on food constituents: an overview. In: Balny C, Hayashi R, Heremans K, Masson P, editors. *High Pressure and Biotechnology. Colloque INSERM*. Montrouge: John Libbey Eurotext Ltd. (1992). p. 195–209.
20. Delfini C, Conterno L, Carpi G, Rovere P, Tabusso A, Cocito C, et al. Microbiological stabilisation of grape musts and wines by high hydrostatic pressures. *J Wine Res*. (1995) 6:143–51. doi: 10.1080/09571269508718031
21. Maresca P, Donsì F, Ferrari G. Application of a multi-pass high-pressure homogenization treatment for the pasteurization of fruit juices. *J Food Eng*. (2011) 104:364–72. doi: 10.1016/j.jfoodeng.2010.12.030
22. Buzrul S. High hydrostatic pressure treatment of beer and wine: a review. *Innov Food Sci Emerg Technol*. (2012) 13:1–12. doi: 10.1016/j.ifset.2011.10.001
23. Bañuelos MA, Loira I, Escott C, Del Fresno J M, Morata A, Sanz PD, et al. Grape processing by high hydrostatic pressure: effect on use of non-Saccharomyces in must fermentation. *Food Bioprocess Technol*. (2016) 9:1769–78. doi: 10.1007/s11947-016-1760-8
24. Smelt JPPM. Recent advances in the microbiology of high pressure processing. *Trends Food Sci Technol*. (1998) 9:152–8.
25. Marszałek K, Wozniak Ł, Kruszewski B, Skapska S. The effect of high pressure techniques on the stability of anthocyanins in fruit and vegetables. *Int J Mol Sci*. (2017) 18:277. doi: 10.3390/ijms18020277
26. Cheftel JC. High-pressure, microbial inactivation and food preservation. *Food Sci Technol Int*. (1995) 1:75–90. doi: 10.1177/108201329500100203
27. Onür I, Misra NN, Barba FJ, Putnik P, Lorenzo JM, Gokmen V, et al. Effects of ultrasound and high pressure on physicochemical properties and HMF formation in Turkish honey types. *J Food Eng*. (2018) 219:129–36. doi: 10.1016/j.jfoodeng.2017.09.019
28. Corrales M, Butz P, Tauscher B. Anthocyanin condensation reactions under high hydrostatic pressure. *Food Chem*. (2008) 110:627–35. doi: 10.1016/j.foodchem.2008.02.055
29. Varela-Santos E, Ochoa-Martinez A, Tabilo-Munizaga G, Reyes JE, Pérez-Won M, Briones-Labarca V, et al. Effect of high hydrostatic pressure (HHP) processing on physicochemical properties, bioactive compounds and shelf-life of pomegranate juice. *Innov Food Sci Emerg*. (2012) 13:13–22. doi: 10.1016/j.ifset.2011.10.009
30. Suarez-Jacobo A, Rüfer C, Gervilla R, Guamis B, Roig-Sagués A, Saldo J. Influence of ultra-high pressure homogenisation on antioxidant capacity,

polyphenol and vitamin content of clear apple juice. *Food Chem*. (2011) 127:447–54. doi: 10.1016/j.foodchem.2010.12.152
31. Sancho F, Lambert Y, Demazeau G, Largeteau A, Bouvier J-M, Narbonne J-F. Effect of ultra-high hydrostatic pressure on hydrosoluble vitamins. *J Food Eng*. (1999) 39:247–53. doi: 10.1016/S0260-8774(98)00143-5
32. Bermejo-Prada A, Vega E, Pérez-Mateos M, Otero L. Effect of hyperbaric storage at room temperature on the volatile profile of strawberry juice. *LWT - Food Sci. Technol*. (2015) 62:906–14. doi: 10.1016/j.lwt.2014.08.020
33. Briones-Labarca V, Perez-Wom M, Habib G, Giovagnoli-Vicuña C, Cañas-Sarazua R, Tabilo-Munizaga G, et al. oenological and quality characteristic on young white wines (Sauvignon Blanc): effects of high hydrostatic pressure processing. *J Food Qual*. (2017) 2017:8524073. doi: 10.1155/2017/8524073
34. Tao Y, Sun D-W, Górecki A, Błaszczak W, Lamparski G, Amarowicz R, et al. Effects of high hydrostatic pressure processing on the physicochemical and sensorial properties of a red wine. *Innov Food Sci Emerg Technol*. (2012) 16:409–16. doi: 10.1016/j.ifset.2012.09.005
35. Mok C, Sonk K-T, Park Y-S, Lim S, Ruan R, Chen P. High hydrostatic pressure pasteurization of red wine. *J Food Sci*. (2006) 71:265–9. doi: 10.1111/j.1750-3841.2006.00145.x
36. Ferragut V, Hernández-Herrero M, Veciana-Nogués MT, Borras-Suarez M, González-Linares J, Vidal-Carou MC, et al. Ultra-high-pressure homogenization (UHPH) system for producing high-quality vegetable-based beverages: physicochemical, microbiological, nutritional and toxicological characteristics. *J Food Sci Agric*. (2015) 95:953–61. doi: 10.1002/jsfa.6769
37. Vilela RM, Lands LC, Chan HM, Azadi B, Kubow S. High hydrostatic pressure enhances whey protein digestibility to generate whey peptides that improve glutathione status in CFTR-deficient lung epithelial cells. *Mol Nutr Food Res*. (2006) 50:1013–29. doi: 10.1002/mnfr.200600074
38. Li H, Zhu K, Zhou H, Peng W. Effects of high hydrostatic pressure treatment on allergenicity and structural properties of soybean protein isolate for infant formula. *Food Chem*. (2012) 132:808–14. doi: 10.1016/j.foodchem.2011.11.040
39. Briviba K, Gräf V, Walz E, Guamis B, Butz P. Ultra high pressure homogenization of almond milk: physico-chemical and physiological effects. *Food Chem*. (2016) 192:82–9. doi: 10.1016/j.foodchem.2015.06.063
40. Tabilo-Munizaga G, Gordon TA, Villalobos-Carvajal R, Moreno-Osorio L, Salazar FN, Pérez-Won M, et al. Effects of high hydrostatic pressure (HHP) on the protein structure and thermal stability of Sauvignon blanc wine. *Food Chem*. (2014) 155:214–20. doi: 10.1016/j.foodchem.2014.01.051
41. Patrignani F, Mannozzi C, Tappi S, Tylewicz U, Pasini F, Castellone V, et al. (Ultra) High pressure homogenization potential on the shelf-life and functionality of kiwifruit juice. *Front Microbiol*. (2019) 10:246. doi: 10.3389/fmicb.2019.00246
42. Gonçalves dos Santos Aguilar J, Cristianini M, Harumi Sato H Modification of enzymes by use of high-pressure homogenization. *Food Res Int*. (2018) 109:120–5. doi: 10.1016/j.foodres.2018.04.011
43. Harte F. Food processing by high-pressure homogenization. In: Balasubramaniam VM, Barbosa-Cánovas GV, Lelieveld HLM, editors. *High Pressure Processing of Food Principles, Technology and Applications*. 1st ed. New York, NY: Springer (2016). p. 123–41.
44. Karacam CH, Sahin S, Oztop MH. Effect of High Pressure Homogenization (microfluidization) on the quality of Ottoman Strawberry (*F. ananassa*) juice. *LWT-Food Sci Technol*. (2015) 64:932–7. doi: 10.1016/j.lwt.2015.06.064
45. Sharabi S, Okun Z, Shpigelman A. Changes in the shelf life stability of riboflavin, vitamin C and antioxidant properties of milk after (Ultra) High Pressure Homogenization: direct and indirect effects. *Innov Food Sci Emerg Technol*. (2018) 47:161–9. doi: 10.1016/j.ifset.2018.02.014
46. Santos MC, Nunes C, Cappelle J, Gonçalves FJ, Rodrigues A, Saraiva JA, et al. Effect of high pressure treatments on the physicochemical properties of a sulphur dioxide-free red wine. *Food Chem*. (2013) 141:2558–66. doi: 10.1016/j.foodchem.2013.05.022
47. Christofi S, Malliaris D, Katsaros G, Panagou E, Kallithraka S. Limit SO_2 content of wines by applying High Hydrostatic Pressure. *Innov Food Sci Emerg Technol*. (2020) 62:102342. doi: 10.1016/j.ifset.2020.102342
48. Frank K, Köhler K, Schuchmann HP. Stability of anthocyanins in high pressure homogenisation. *Food Chem*. (2012) 130:716–9. doi: 10.1016/j.foodchem.2011.07.086

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The Effect of Plasma Treated Water Unit Processes on the Food Quality Characteristics of Fresh-Cut Endive

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This study evaluated the impact of a defined plasma treated water (PTW) when applied to various stages within fresh-cut endive processing. The quality characteristic responses were investigated to establish the impact of the PTW unit processes and where PTW may be optimally applied in a model process line to retain or improve produce quality. Different stages of application of PTW within the washing process were investigated and compared to tap water and chlorine dioxide. Fresh-cut endive (*Cichorium endivia* L.) samples were analyzed for retention of food quality characteristics. Measurements included color, texture, and nitrate quantification. Effects on tissue surface and cell organelles were observed through scanning electron and atomic force microscopy. Overall, the endive quality characteristics were retained by incorporating PTW in the washing process. Furthermore, promising results for color and texture characteristics were observed, which were supported by the microscopic assays of the vegetal tissue. While ion chromatography detected high concentrations of nitrite and nitrate in PTW, these did not affect the nitrate concentration of the lettuce tissue post-processing and were below the concentrations within EU regulations. These results provide a pathway to scale up the industrial application of PTW to improve and retain quality characteristic retention of fresh leafy products, whilst also harnessing the plasma functionalized water as a process intervention for reducing microbial load at multiple points, whether on the food surface, within the process water or on food-processing surfaces.

Keywords: atmospheric pressure plasma, food quality, leafy greens, microwave-driven discharge, non-thermal processing, ready-to-eat produce

INTRODUCTION

As with many other vegetables, fresh-cut lettuce (e.g., endive) is a minimally processed produce that is harvested, cut, washed, centrifuged, and packaged (1, 2). These activities may be associated with mechanical damage to plant tissue, which causes biochemical and physiological reactions such as enzymatic browning, increased respiration, sensory, and structural decay (3). These changes may

lead to significant losses in quality and thus reduce the shelf life and marketability of the produce (4, 5). Washing the fresh-cut lettuce is used to remove field heat, dirt, microorganisms, possible pesticide residues, and cell exudates, which could otherwise lead to a loss of quality (6, 7). Washing is therefore particularly important for the microbial safety and storage quality of fresh cut lettuce.

However, the process water can also be a source of microorganisms and lead to cross contamination. Therefore, where the use is legally permitted, water additives, mostly chemical, are used to reduce the microbial load in the washing water. Chlorine-based compounds are the most common and widely used disinfectant (8, 9). However, the use of chemicals is not permitted in the production of organic food and in conventional food processing the use of chemical disinfectants is not without concern, as they can lead to the formation of potentially harmful haloform by-products namely chloramines and trihalomethanes (10). Other wash water additives or treatments already in industrial use include chemical sanitizers like ozone, hydrogen peroxide, electrolyzed water, and peracetic acid. Physical treatments such as high hydrostatic pressure, pulsed electric field, oscillating magnetic field, ultra violet (UV)- or gamma irradiation and high-power ultrasound are also possible (4, 11–15). Some innovative process water additives under research are Quillaja saponaria extract (QSE) and Nα-lauroyl-L-arginine ethyl ester (LAE) (16–18).

The development of sustainable disinfection methods is important and challenging, but product quality compatibility, cost, environmental impact, and regulatory must also be met (19). An innovative strategy under research to reduce the bacterial load of process water and subsequently, to keep the food quality and shelf-life of fresh-cut lettuce at high levels is the use of plasma treated water (PTW) as an antimicrobial process stage.

The application of non-thermal plasma (NTP) generated at atmospheric pressure is a promising physical approach (20, 21). Plasmas are ionized gases containing neutral- and free charged particles such as ions and electrons (22, 23). Novel intervention technologies for fresh foods demand minimal processing at low or mild heat temperatures to maintain fresh characteristics, as well as compatibility with high throughput continuous processing, which can be achieved through non-thermal plasma at atmospheric pressure in gas or functionalized liquid mode of delivery (22). PTW can be used as the transport medium of reactive species and antimicrobial components for food, water, and surface sanitation (24). PTW is comparable to ozonized or chlorinated water with regard to mode of application and antimicrobial effects. The chemical composition of PTW concerning the acidic pH, and the reactive oxygen and nitrogen species (RONS) was previously characterized (25). Both, low pH and RONS are known to support and to cause the antimicrobial mechanisms of action, therefore the chemical composition of PTW should be known if PTW is investigated as a process wash water. This study investigates the unexplored aspects of how the PTW effects the food quality of fresh-cut endive and determines where PTW may be optimally applied within a fresh produce washing process for produce quality retention.

MATERIALS AND METHODS

Generation of PTW and Its Chemical Characterization

The PTW generation was previously described in (25). In brief, plasma processed air (PPA) was used to treat distilled water. This leads to the formation of PTW. The used plasma source was a two-stage microwave-driven device (2.45 GHz) based on a single-stage plasma torch (26, 27) and operated at atmospheric pressure. The used technical parameters for the presented experiments to generate PPA and subsequently PTW were 1.3 kW (power) and 12 slm (volume flowrate) for the first stage. For the second stage, 3.0 kW and 60 slm were applied. The chemical composition of PPA and PTW was previously reported (25, 28). Briefly, using emission spectroscopy (ES) analysis, the spectral lines of nitrogen monoxide radical (·NO), nitronium cation (NO_2^+), and hydroxide anion (OH^-) were dominant in the detected spectrum (28). In the FTIR analysis, the main components of PPA were nitrogen monoxide radicals (·NO), nitrogen dioxide radicals ($\cdot NO_2$) and water (H_2O), oxygen (O_2) and nitrogen (N_2). ·NO was determined with a concentration of 2,900 ppm (7.79×10^{22} m^{-3}), $\cdot NO_2$ with 76 ppm (2.04×10^{21} m^{-3}) and H_2O with 9,200 ppm (2.47×10^{23} m^{-3}) (25). The chronoamperometry identified a H_2O_2 concentration of 5.61 mg L^{-1} (29.39 mM) in the PTW (25). Finally, the ion chromatography (IC) measurements identified high values for NO_2^- and NO_3^-, 687 mg L^{-1} NO_2^-, and 1,227 mg L^{-1} NO_3^-, respectively (25).

Investigated Specimen—Fresh-Cut Lettuce With Native Load

The specimen (endive, *Cichorium endivia* L.) was bought at a local organic market in Greifswald, Germany. The lettuce was grown on different fields (loamy sand and sandy loam) in the state of Mecklenburg-Western Pomerania, Germany. The harvest months were October to December 2019. Subsequently, the whole endive heads were stored in the dark for a maximum of 24 h at 7.4 ± 0.1°C before use. The relative humidity inside the fridge was 78% to 99% with a dew point of 3.9 td to 7.3°C td. Produce samples were prepared in accordance with previous studies aligned to industry practice and are briefly presented here (25). For food color and texture analyses, the lettuce was cut before washing. In the case of the microscopy analysis, the lettuce was washed first and subsequently cut. Before experimental use, the outermost leaves were removed, but the stalk was retained. Both, the softer leaf parts and the harder stalk parts of the lettuce were mixed to provide representative. Prepared experimental samples were stored in closed homogenizer bags (polyethylene; VWR International GmbH, Darmstadt, Germany) in air and removed for analyses ay days 1 and 7.

Processing of Fresh-Cut Endive

The processing of the fresh-cut endive was performed on a washing line mimicking a common industrial production process. The washing line ultimately consisted of up to five main sections—pre-bathing, pre-rinsing, pre-washing, main washing, and post-rinsing. The investigated process variants are given in

TABLE 1 | Investigated process variants included the application of PTW at various process stages and unwashed lettuce, tap water and chlorine dioxide (ClO_2, 15 ppm) were the reference treatments.

Process variant	Unwashed (0)	Pre-bathing (1)	Pre-rinsing (2)	Pre-washing (3)	Main washing (4)	Post-rinsing (5)
Time of washing in [s]	0	180	30	180	180	30
Ia—tap water	NA	Tap water	Tap water	Tap water	Tap water	Tap water
Ib—tap water	NA	NA	Tap water	Tap water	Tap water	Tap water
II—PTW	NA	PTW	Tap water	Tap water	Tap water	Tap water
III—PTW	NA	NA	PTW	Tap water	Tap water	Tap water
IV—PTW	NA	NA	Tap water	Tap water	PTW	Tap water
V—PTW	NA	PTW	PTW	Tap water	PTW	Tap water
VI—ClO_2	NA	NA	Tap water	Tap water	ClO_2	Tap water

The washing time for each of pre-bathing, pre-washing and main washing was 180 s and the washing times for pre- and post-rinsing were 30 s. NA, not analyzed. For better visualization, tap water treatment is highlighted in blue, PTW treatment in pink, and ClO_2 treatment in red.

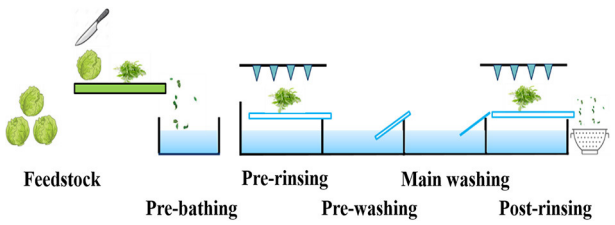

FIGURE 1 | An example process line, the illustrated washing process of a tap water reference (process variant Ia): five washing sections—pre-bathing, pre-rinsing, pre-washing, main washing, and post-rinsing.

Table 1 and as an example, the reference process using tap water is illustrated in **Figure 1**. After the last washing step, the samples were placed over a sieve for draining, but were not spun.

Characterizing the Impact of Process Variants on Food Quality Parameters

Color Analyses

Color analyses on lettuce leaves were performed using a portable colorimeter NH310 of 3nh (PCE Deutschland GmbH, Meschede, Germany) with the CIELab system. Five points of measurement were used for each lettuce leaf, and chroma was expressed as *C*-value. For the process variants listed in **Table 1**, the Chroma was examined after each washing section. These investigations were carried out immediately on the day of treatment (day 0), 24 h (day 1), and 168 h (day 7) later.

Texture Analyses

The texture of the fresh-cut endive samples was examined with the Texture Analyser TAXT+ (WINOPAL Forschungsbedarf GmbH, Elze, Germany) before and after treatment with tap water, PTW, or ClO_2. Samples had a mass of 10 ± 2 g. Five samples were examined for each process variant, washing section, and storage time after treatment (0, 1, and 7 days). The sample to be measured was transferred into a 600 mL beaker and positioned under the probe head. A probe head with three concentric rings (WINOPAL Forschungsbedarf GmbH, Elze, Germany) was used, as the large cross section is appropriate to measure the impact of the force over the whole leaf samples structure. The used protocol was previously described in Schnabel et al. (25).

Determination of Nitrate (NO_3^-) Content

Before and after washing the fresh-cut lettuce with tap water, PTW and ClO_2, the plant tissue was homogenized with a common hand blender. After blending, 5 g of the homogenized plant tissue was mixed with 50 mL sterile tap water (70°C), briefly shaken and incubated for 15 min. Two filtration steps were completed after incubation. For the first step, the whole tissue sample was rinsed over a paper filter (VWR, Darmstadt, Germany; particle retention of 2–3 µm). In the second step, the filtrated solution was filtrated again by a tip filter [Sarstedt, Nümbrecht, Germany; particle retention 0.2 µm, PES (polyester) membrane]. The collected double-filtrated solution was analyzed by IC for NO_3^- concentrations according to the following procedure. Immediately after sample preparation, nitrate was determined by ion chromatography (IC). For this purpose, the IC was performed with the 850 Professional IC (Deutsche METROHM GmbH & Co. KG, Filderstadt, Germany) as previously described in detail in our publication Schnabel et al. (25).

SEM

Fresh samples of fresh-cut endive with an area of 25 mm^2 were retrieved before and after treatment with tap water, PTW and ClO_2. They were then prepared on brass holders with an electrically conductive glue containing silver particles (Ferro GmbH, Germany). The samples were dried under vacuum (1.0 mPa, 24 h) and subsequently coated with thin gold film by the sputter coater SCD 050 (Bal-Tec, Switzerland) in order to adjust the optimal material conditions for observation by means of an electron microscope. Overview and high-resolution images of the samples were taken with the electron microscope JSM 7500F (Jeol, Germany) at magnifications 100; 1,000; and 4,000. A secondary electron detector with a resolution of 1.0 nm was used for this morphological analysis. The microscope setting in this study was applied as follows: accelerating voltage 1 kV, working distance 8 mm, samples surface in the perpendicular position to the beam.

AFM

A fresh-cut lettuce tissue sample was fixed onto a PE-holder ($32 \times 8 \times 2$ mm^3), and placed in a 60 mm diameter petri dish. Following a standard procedure (25), the sample have been submerged in 5 ml of filtered tap water (tip filter, which excludes all particles $>2\,\mu$m) and fixed beneath the measuring head of the AFM. We used a NanoWizard III (JPK BioAFM, Bruker, Berlin, Germany) with a linearized piezo scanner for the scanning-probe topographies. Therefore, the scanning covered a travel path length of the piezos of 100 μm in every direction (*xyz*). Beam-shaped silicon probes without any top coating, a nominal spring constant of 0.29 N m^{-1}, and a pyramidal-shaped tip (nominal aspect ratio: 1.5–3.0) were used. All samples were measured in contact mode with a set point of 15 nN and a line rate of 0.08 Hz by a 90×90 μm^2 scan width. Two recording modes were used; the height micrograph reflects the vertical (*z*-information) extension of the piezo and the horizontal movement of the lateral scanner whilst it scans over the surface. It reflects the exact topographic height differences on the entire scan area. The deflection image, which is also referred as the error signal, reflects the deflection, i.e., the bending, of the probe that has been plotted against its *xyz*-position.

TEM

Transmission-electron microscopic (TEM) micrographs were taken before and after each washing step with tap water, PTW, or ClO_2. Using a sterile razor blade, samples of a 5×20 mm^2-area (5 mm length and a width of maximum 1 mm) were cut from green and healthy lettuce leaves. These pieces were fixed at room temperature for 2 h with a fixative containing 3% glutaraldehyde in a 50 mM cocadylate buffer at pH 7, and stored at 4°C until further processing. Before placing the samples in the high vacuum of the TEM, they were washed for 5, 10, 15, 20, and 30 min, and then embedded in low gelling agarose, further washed with cacodylate buffer for 10 min, prior to fixing in 2% osmium tetroxide (which was dissolved in cocadylate buffer) for 2 h at room temperature. Final samples were washed repeatedly as described above. For dehydration, the specimens were submerged for 15 min in distinct ethanolic solutions with increasing concentrations of 30, 50, 70, and 90%. Finally, we placed the samples in 96% ethanol for duplicate 10 min periods, and then in 100% ethanol thrice. In applying the flat-embedding technique, we transferred the samples stepwise into propylene oxide before they have been ingrained in low-viscous agar-resin (AGAR-LV, plano, Wetzlar, Germany). Subsequently, sections were cut on an ultramicrotome (Reichert Ultracut, Leica UK Ltd., Milton Keynes, UK), transferred onto pioloform-coated slot grids (2×1 mm, plano, Wetzlar, Germany), and stained with 4% aqueous uranyl acetate for 5 min. This was followed by lead citrate incubation for 1 min. The samples were analyzed with a TEM LEO 906 (Zeiss, Oberkochen, Germany) at an acceleration voltage of 80 kV, and micrographs were edited with Adobe Photoshop CS6 (Adobe Systems Inc., San Jose, Cal., US).

Statistics

Our experiments aim to mimic an industrial process as close as it is possible and therefore the leaves are not treated the same way at every extraction point (EP) along the washing process (process variant I–VI). Some process variants (Ib, III, IV, and VI) miss extraction point 1 (pre-bathing). Consequently, no data were obtained for these aspects at EP1, which make a more direct testing based on a distinct hypothesis complicated. Additionally, tests need to be designed to test the data against two references. The colorfulness (C) of the fresh leaves and their texture (T) are basic values, which are used in our hypotheses. We assumed a gaussian distribution among the values of C and T. Consequently, the arithmetic means and a standard deviation were computed and used in a statistical test series. We tested against a statistical significance of $\alpha = 0.05$.

Certainly, it is straightforward to assume that possible changes in C and T appear when the leaves are treated with different sanitizers, which is best observed at EP5. Therefore, we formulate the basic α-hypothesis:

H_0^α = The C or the T of the leaves did not change at EP5,

and the alternative:

H_1^α = The C or the T of the leaves changed at EP5.

For that reason, we tested C and T at EP5 with an ANOVA against the outcome of process variant Ia/b (tap water). By this way, a statistical evaluation of possible differences in C and T of treated leaves based on a *p*-value that mirrors the probability of error for an incorrect rejection of the null hypothesis at that point was possible. A second test series evaluated every single step of each process variant against the input-values (EP0) of C and T based on ANOVA. By this way, a statistical evaluation of possible C and T changes along every process variant was possible. Therefore, the β-hypothesis is formulated:

H_0^β = The C or the T of the leaves do not change due to the treatment and the washing stage along a given process variant,

and the alternative:

H_1^β = The C or the T of the leaves changed due to the treatment and washing stage along a given process variant.

In summary, we have a set of horizontal ANOVA-tests for every process variant (I–VI, β-test) and a vertical ANOVA-test at EP5 (α-test). The β-test evaluated whether any C or T changes occurred along a process variant. The α-test evaluated possible C and T changes at EP5. Combined, α and β revealed information about the experiment of a completely examined day (0, 1, and 7). Set the case, the input of leaves was, regarding their C and T, homogenous and the outcome at EP5 differs, changes were a consequence to a sanitizer treatment along a process variant. A behavior, which could be observed when the α-H_1-hypothesis went through and if at least one β-H_0-hypothesis was ineligible. Generally, we interpreted the *p*-values of the α- and β-tests for every examination day as strong indicators for the behavior of the daily experimental set up. Nevertheless, they alone give not a basis to test a hypothesis addressing the overall interpretation of a completely examined day.

The hypothesis addressing the changes in C and T over a time of 0, 1, and 7 days compared the mean values of the outcomes at EP5 against each other based on an ANOVA evaluation and the hypothesis:

H_0 = The C or the T of the leaves did not change over the time of 0, 1, or 7 days,

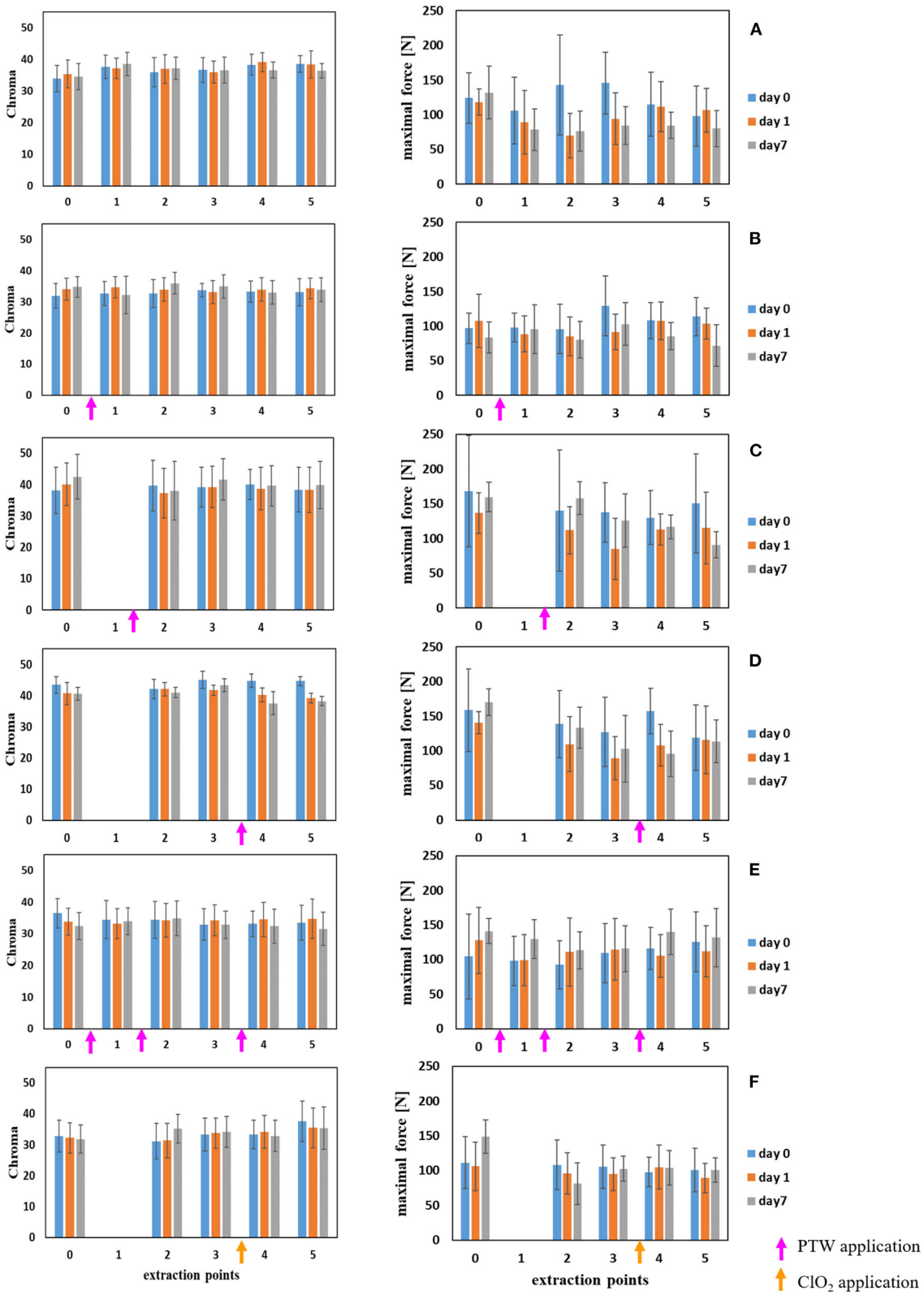

FIGURE 2 | The effect of unit process treatments on color and texture stability of fresh cut endive. Color (left column) and texture measurements (right column) of unwashed (extraction point 0) and tap water, PTW or ClO_2 washed fresh-cut endive (extraction points 1–5). **(A)** Variant I: tap water at all extraction points for storage

(Continued)

FIGURE 2 | days 0, 1 and 7. **(B)** Variant II: PTW at extraction point 1 (180 s pre-bathing), extraction points 2–5 with tap water for storage days 0, 1, and 7. **(C)** Variant III: PTW at extraction point 2 (30 s pre-rinsing), extraction points 3–5 with tap water for storage days 0, 1, and 7. **(D)** Variant IV: PTW at extraction point 4 (180 s main washing), extraction points 2, 3, and 5 with tap water for storage days 0, 1, and 7. **(E)** Variant V: PTW at extraction point 1, 2, and 4 (180 s pre-bathing, 30 s pre-rinsing, and 180 s main washing), extraction points 3 and 5 with tap water for storage days 0, 1, and 7. **(F)** Variant VI: ClO_2 at concentration of 15 ppm at extraction point 4 (180 s main washing), extraction points 2, 3, and 5 with tap water for storage days 0, 1, and 7. All experiments were repeated threefold with $n = 5$ resulting in $n = 15$.

TABLE 2 | Statistical evaluation of storage day 0.

Process variant	*p*-value	H_0 hypothesis
Horizontal β evaluation		
I—tap water	0.59	Not rejected
II—PTW	0.12	Not rejected
III—PTW	0.75	Not rejected
IV—PTW	0.95	Not rejected
V—PTW	0.84	Not rejected
VI—ClO_2	0.32	Not rejected
Vertical α evaluation		
Extraction point 5 (EP5)	0.09	Not rejected

TABLE 3 | Statistical evaluation of storage day 1.

Process variant	*p*-value	H_0 hypothesis
Horizontal β evaluation		
I—tap water	0.22	Not rejected
II—PTW	0.22	Not rejected
III—PTW	0.85	Not rejected
IV—PTW	0.90	Not rejected
V—PTW	0.90	Not rejected
VI—ClO_2	0.39	Not rejected
Vertical α evaluation		
Extraction point 5 (EP5)	0.32	Not rejected

TABLE 4 | Statistical evaluation of storage day 7.

Process variant	*p*-value	H_0 hypothesis
Horizontal β evaluation		
I—tap water	0.17	Not rejected
II—PTW	0.44	Not rejected
III—PTW	0.86	Not rejected
IV—PTW	0.96	Not rejected
V—PTW	0.42	Not rejected
VI—ClO_2	0.69	Not rejected
Vertical α evaluation		
Extraction point 5 (EP5)	0.12	Not rejected

TABLE 5 | Statistical evaluation over all storage days (day 0, 1, and 7) at EP5.

Process variant	*p*-value	H_0 hypothesis
ANOVA evaluation over day 0, 1, and 7 at EP5		
I—tap water	0.53	Not rejected
II—PTW	0.94	Not rejected
III—PTW	0.88	Not rejected
IV—PTW	0.97	Not rejected
V—PTW	0.94	Not rejected
VI—ClO_2	0.81	Not rejected

and the alternative:

H_1 = The C and the T of the leaves changed over the time of 0, 1, or 7 days.

That hypothesis allowed statistical statements about changes in C and T when the samples were stored up to 7 days.

RESULTS

Color

The results for the color analyses are shown in **Figure 2** (left column). The change of green lettuce color after washing with tap water, PTW and ClO_2 was determined directly (day 0), after 1 day and 7 days of storage at 7°C in a closed plastic bag. The Chroma values of the color measurements were not affected by any of the treatments studied within the 7-day storage trial. Statistical analysis revealed no significant color changes for every process variant and analysis day. The results of the statistical evaluation are given in **Tables 2**–**5**.

Texture

The change in texture of fresh-cut endive leaves after each process variant (**Figure 2**—right column) was determined directly (day 0), 1 day, and 7 days after treatment for each process extraction point (0–5). For the texture determination, the complete head of lettuce, except the outer leaves, was prepared for analysis and the stem components were not removed. In texture analysis, the required maximum force for the first breakthrough varied between 100 and 150 N, in general. The relatively large deviations at the individual measuring points may be due to the small sample quantity of 10 g or the mixture of hard and soft leaf parts (Stem components were not sorted out). In comparison to unwashed fresh-cut endive (extraction point 0) the washed lettuce became softer, less force was needed. However, compared to the tap water variant (I), no negative (softening) effect of PTW (variant II to V) or ClO_2 (variant VI) was observed for all process variants at day 0 and 1 ($p > \alpha$). There were moderate variations of texture for the process variant II. A 180 s pre-bathing effected the T [$p = 0.02$ (*)] directly after the treatment on day 0. For the process variant III (30 s pre-rinsing, $p = 0.03$) the texture was effected on examination day 1. Undoubtedly, variations of texture due to a sanitizer treatment (see chapter 2.5) have been observed

TABLE 6 | Statistical evaluation of storage day 0.

Process variant	*p*-value	H_0 hypothesis
Horizontal β evaluation		
I—tap water	0.19	Not rejected
II—PTW	0.02	Rejected
III—PTW	0.74	Not rejected
IV—PTW	0.27	Not rejected
V—PTW	0.98	Not rejected
VI—ClO_2	0.4	Not rejected
Vertical α evaluation		
Extraction point 5 (EP5)	0.13	Not rejected

TABLE 7 | Statistical evaluation of storage day 1.

Process variant	*p*-value	H_0 hypothesis
Horizontal β evaluation		
I—tap water	0.48	Not rejected
II—PTW	0.41	Not rejected
III—PTW	0.03	Rejected
IV—PTW	0.31	Not rejected
V—PTW	0.43	Not rejected
VI—ClO_2	0.64	Not rejected
Vertical α evaluation		
Extraction point 5 (EP5)	0.39	Not rejected

TABLE 8 | Statistical evaluation of storage day 7.

Process variant	*p*-value	H_0 hypothesis
Horizontal β evaluation		
I—tap water	0.001	Rejected
II—PTW	0.99	Not rejected
III—PTW	0.001	Rejected
IV—PTW	<0.001	Rejected
V—PTW	0.06	Not rejected
VI—ClO_2	<0.001	Rejected
Vertical α evaluation		
Extraction point 5 (EP5)	<0.001	Rejected

on day 7. The test scenario clearly shows changes in texture, which appear to be highly significant (*p*-value: <0.001 for process variants IV and VI). The results of the statistical evaluation are given in **Tables 6**–**9**.

Nitrate

Dietary nitrate is mainly derived from the consumption of vegetables, in particular green leafy vegetables (29). Due to their dietary importance, nitrate levels in fresh-cut endive were monitored during storage (**Table 10**). However, the EU has regulated the maximum levels of nitrate for fresh lettuce with 2,500-4,500 mg NO_3 kg^{-1} depending on the harvest time and conditions (30). Since PTW, which was used to wash fresh-cut endive, contained both significant concentrations of nitrite and nitrate, it was necessary to determine whether the nitrate content in lettuce increases after washing and exceeds acceptable limits and guidance values. This could be an exclusion criterion for the approval of this innovative washing method. The absorption of nitrate may be increased by the fresh cut edges. In order to determine the nitrate values, IC analysis was used. The IC-samples were prepared immediately after processing (day 0) and the nitrate content in unwashed, tap water, PTW and ClO_2 washed endive were measured.

TABLE 9 | Statistical evaluation over all storage days (day 0, 1, and 7) at EP5.

Process variant	*p*-value	H_0 hypothesis
ANOVA evaluation over day 0, 1, and 7 at EP5		
I—tap water	0.03	Rejected
II—PTW	0.002	Rejected
III—PTW	0.34	Not rejected
IV—PTW	0.53	Not rejected
V—PTW	0.06	Not rejected
VI—ClO_2	0.25	Not rejected

The nitrate content of unwashed endive (process variant I) was the highest with 1,793.6 ± 373.7 mg kg^{-1}. Within all process variants (I–VI) the nitrate concentration was lower compared to unwashed lettuce. Despite the high water solubility of NO_3, the detected concentrations of nitrate in the variants I to VI were between 377.9 ± 221.6 mg NO_3^- kg^{-1} and 1,206.2 ± 483.8 mg NO_3^- kg^{-1}. The concentrations are summarized in detail for each process variant and extraction point in **Table 10**. For all PTW-application points, an increased NO_3^- concentration compared to the other extraction points was noticeable. As expected, ClO_2 did not lead to an increase in NO_3^-. In PTW variants (II–V), the NO_3^- concentration did not increase compared to unwashed (extraction point 0) and tap water (variant I) washed lettuce at the final extraction point 5. Importantly, all NO_3^- concentrations were significantly below the maximum permitted value of 2,500 mg NO_3^- kg^{-1} for iceberg-type lettuce (2,000–2,500 mg NO_3^- kg^{-1}) and for fresh lettuce (except iceberg-type) of 2,500–4,500 mg NO_3^- kg^{-1} (30).

SEM

Microscopic methods for a deeper characterization of the influence of a PTW on lettuce were chosen. The focus of these investigations was on the food quality of fresh-cut lettuce mirrored by color- and texture analysis. Additionally, SEM and AFM were used to visualize variations on the leaves surface due to a sanitizer treatment. Especially TEM revealed insight into the cell interior and possible changes of cell organelles can be observed. In the texture analysis (see section Texture), only a moderate influence (process variant II, day 0, $p = 0.02$) on the leaf structure directly after washing with PTW was found. This was confirmed by the SEM (**Figure 3**) and AFM (**Figure 4**) analysis, were no sever, structural alterations have been observed. Overall, the SEM analysis showed no clearly visible changes between the samples and the unwashed reference. When washing the lettuce

TABLE 10 | Nitrate concentrations of fresh-cut unwashed and washed endive.

Sample/process variant	Extraction points						Nitrate concentration in
	0	1	2	3	4	5	
Distilled water	0.1 ± 0.09						[mg l^{-1}]
Tap water	1.6 ± 0.05						[mg l^{-1}]
PTW	2.48 ± 0.56						[mg l^{-1}]
ClO_2	2.0 ± 0.1						[mg l^{-1}]
Unwashed	1,793.6 ± 373.7						[mg kg^{-1}]
I—tap water		937.0 ± 750.9	758.0 ± 561.6	671.8 ± 478.2	907.2 ± 724.6	860.9 ± 647.4	[mg kg^{-1}]
II—PTW		1,013.7 ± 387.3[a]	811.6 ± 181.7	729.1 ± 710.2	635.1 ± 355.7	632.6 ± 350.2	[mg kg^{-1}]
III—PTW			709.9 ± 354.2[a]	436.7 ± 319.9	377.9 ± 221.6	403.3 ± 257.6	[mg kg^{-1}]
IV—PTW			509.9 ± 185.9	533.9 ± 174.9	1,019.0 ± 489.5[a]	631.2 ± 298.3	[mg kg^{-1}]
V—PTW		1,134.2 ± 561.4[a]	891.0 ± 289.4[a]	783.7 ± 556.7	1,206.2 ± 483.8[a]	739.4 ± 291.3	[mg kg^{-1}]
VI—ClO_2			380.6 ± 199.2	539.4 ± 274.3	552.7 ± 189.3[b]	422.6 ± 306.4	[mg kg^{-1}]

[a] *PTW application.*
[b] *ClO_2 application.*
All experiments were repeated three-fold with n = 5 resulting in n = 15.

with tap water, or PTW (irrespective of the process variant) there was no noticeable influence and intact stomata—both open and closed—were observed in all samples (**Figure 3** middle and right column). The temperature of the water in all scenarios was 21.5°C ± 0.1°C. Only after the treatment of lettuce with ClO_2, does the lettuce surface appear flatter and smoother, i.e., less rough and structured (**Figure 3G**, magnification ×100). The stomata seemed to be less raised and, more collapsed. In the ×4,000 magnification (**Figure 3G**), the outer edge of the stomata appeared somewhat attacked.

AFM

Our investigations showed results of 8–14 µm for the endive leaves in the height micrograph. The wave-like structure, previously observed through SEM analysis, was clearly seen in the error signal micrograph (**Figure 4**, left column). The slight variations in the height profiles were probably due to the presence of stomata in the scanned section. The slight deviations for ClO_2 observed in the SEM investigations were not visible in the AFM images. This may be due to the small scanned area in the AFM. On the other hand, the reason could also be that the samples are pre-treated for SEM and not for AFM. The proposed advantage of AFM analysis is in sample preparation, where they were not subject to dehydrating steps and were relatively natively analyzed. AFM was proposed as a good alternative for SEM.

TEM

The TEM-analysis (**Figure 5**) of endive leaf tissue from unwashed lettuce showed the typical cell organelles (31–33), with the exception of vacuoles (**Figure 5A**). The absence of vacuoles was probably an artifact of preparation. Differences in the composition of cell organelles were not found for the applied treatments by comparison with the unwashed reference. However, chloroplasts in leaf tissue showed morphological changes after washing with ClO_2 (**Figure 5G**), particularly in the grana the grana (stacks of thylakoid discs). An altered structure of the thylakoid membranes was observed. TEM micrographs of thylakoid membranes clearly revealed the drastic difference in the thylakoid structure in ClO_2 washed fresh-cut lettuce, where the stacks in grana were attached to one another to form large clearances. Further, the stroma thylakoids were disrupted (**Figure 5G**). In contrast to the addition of ClO_2 to the washing water, the use of PTW for lettuce washing had no effect on the grana of the chloroplasts or other organelles, regardless of the application point of PTW (**Figures 5C–F**).

DISCUSSION

Food safety should always be considered in combination with retention of food quality characteristics for approval and adoption of emerging technologies, in order to increase acceptance by stakeholders (industry) and consumers and to identify the optimal process variant taking into account individual needs.

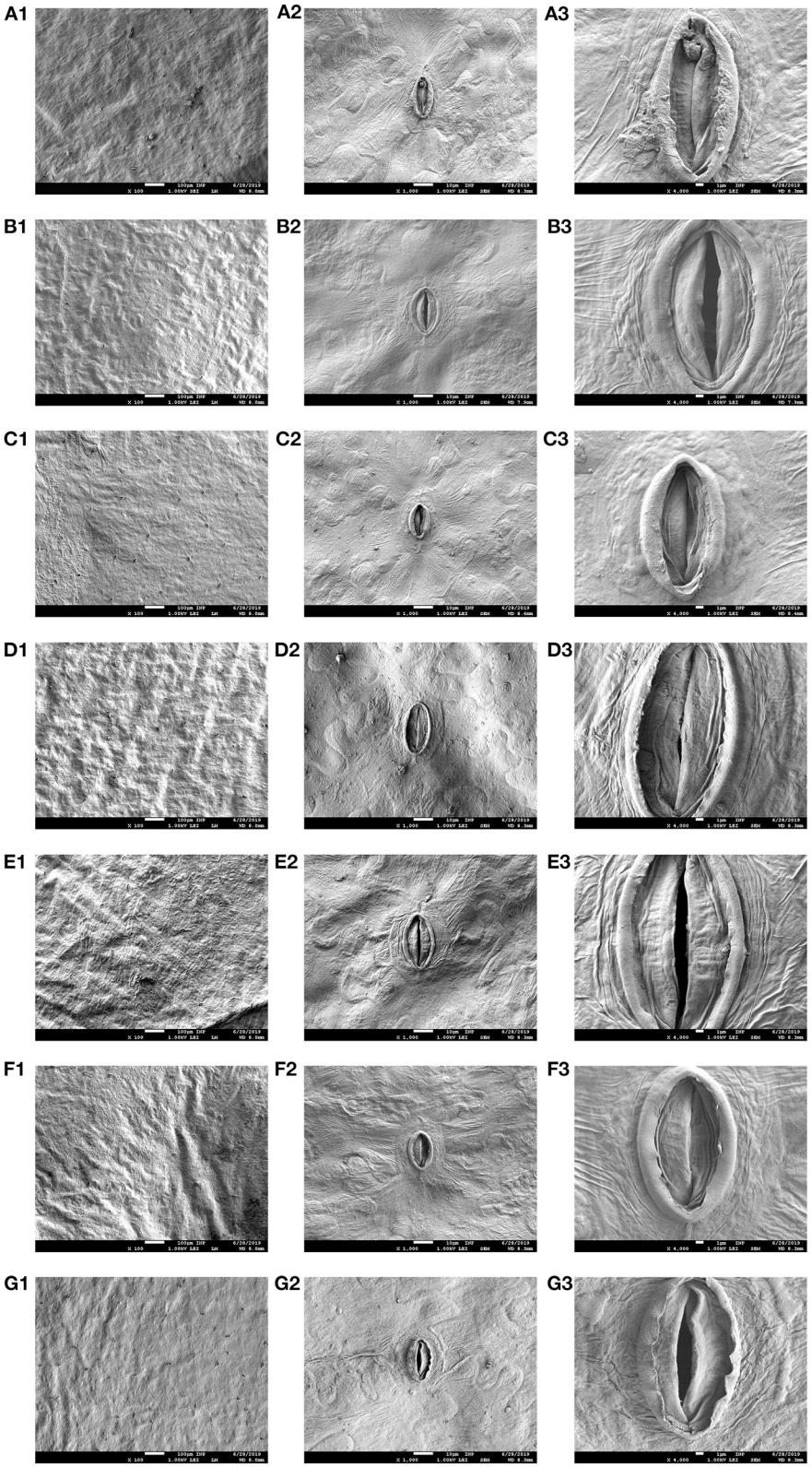

FIGURE 3 | Scanning electron microscopy (SEM) of the fresh-cut lettuce. For each process variant (I–VI), three magnifications are shown: left—×100, middle—×1,000, right—×4,000. The scenarios shown are: **(A)** unwashed (extraction point 0), **(B)** tap water washed (variant I), **(C)** PTW washed—variant II, **(D)** PTW washed—variant III, **(E)** PTW washed—variant IV, **(F)** PTW washed—variant V, and **(G)** ClO_2 washed (variant VI).

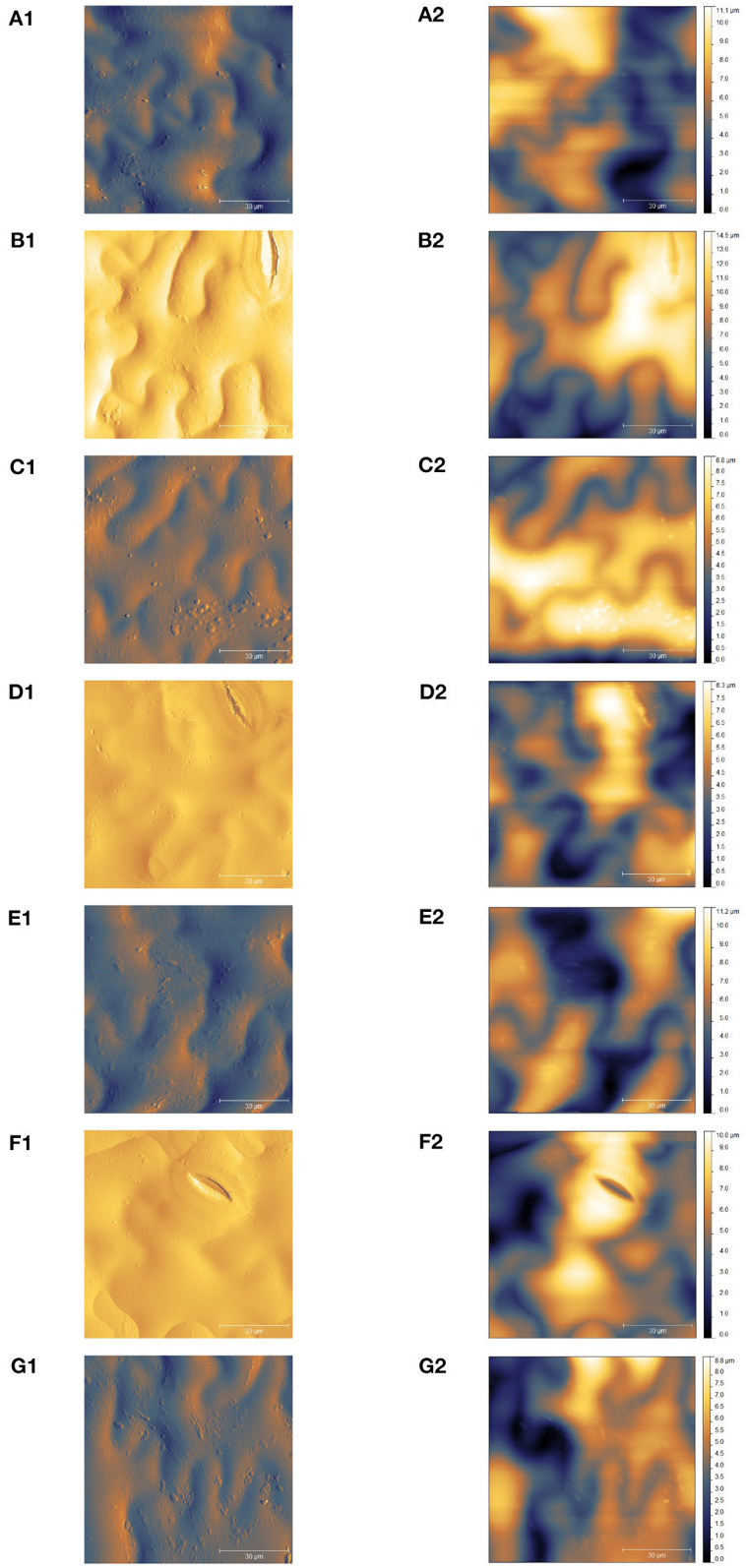

FIGURE 4 | Atomic force microscopy (AFM) of the fresh-cut lettuce. For each process variant (I–VI), two micrographs are shown: left—error signal/deflection micrograph, right—height micrograph. The variants shown are: **(A)** unwashed (extraction point 0), **(B)** tap water washed (variant I), **(C)** PTW washed—variant II, **(D)** PTW washed—variant III, **(E)** PTW washed—variant IV, **(F)** PTW washed—variant V, and **(G)** ClO_2 washed—variant VI.

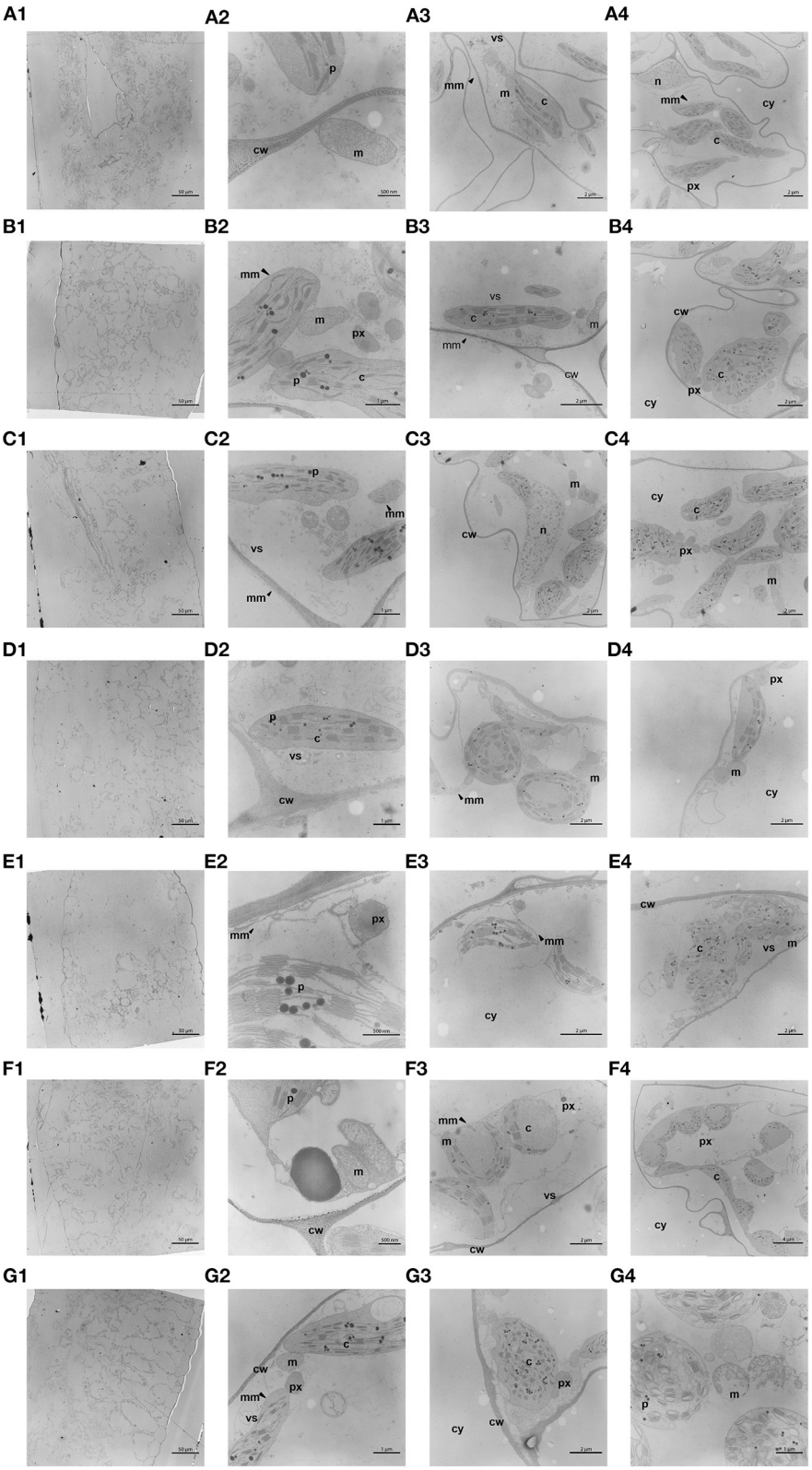

FIGURE 5 | Transmission electron microscopy (TEM) of the ultrastructure of the fresh-cut lettuce leaf tissue. For each process variant, four micrographs are shown: from left to right—overview of the ultrathin section, three times a detailed micrograph of the same section depicting all detected cell organelles. The process variants shown are: **(A)** unwashed (extraction point 0), **(B)** tap water washed (variant I), **(C)** PTW washed—variant II, **(D)** PTW washed—variant III, **(E)** PTW washed—variant IV, **(F)** PTW washed—variant V, **(G)** ClO_2 washed—variant VI. The organelles are: nucleus (n); cytoplasm (cy); cell wall (cw); chloroplast (c); mitochondrion (m); vesicle (vs); plastogloboli (p); peroxisome (px); membrane (mm; plasmalemma or tonoplast).

Previous publications investigated the antimicrobial effects of plasma processed air (PPA) and this PTW for retention of food safety profiles using different target surfaces and microorganisms (25, 26, 34–38). These studies addressed the decontamination potential of PPA or PTW on microorganisms relevant for safe shelf life extension, on the artificial and natural microbial loads of fresh-cut lettuce, and the scalability of the plasma technology and pilot-scale application. However, the impacts on fresh product quality characteristics represented a research lacuna, which was therefore the focus of the current study.

The investigations on color and texture showed only sparsely negative influences of a PTW or ClO_2 treatment, which cannot be detected by the used ANOVA. The systematic color screening of treated leaves revealed no statistically significant support of our assumptions (H_1-hypothesis) and the null-hypotheses cannot be rejected for every single process variant (all $p > \alpha$). On the other hand, statistically meaningful structural changes of product matrix of the fresh-cut endive within the storage trial of 7 days were observed (process variant I, III, IV, and VI; p-values < 0.01). The logical construct when the β-null hypothesis (horizontal) and the α-null hypothesis (vertical) must be rejected states there were structural alterations due to a sanitizer treatment, which become obvious on day 7 for process variant I, III, IV, and VI (p-value < 0.001). Additional, for a storage period of 7 days, structural changes become statistically meaningful for the process variant I (tap water) and the process variant II ($p < 0.03$). However, since the tap water-reference showed a pronounced alteration especially after a storage time of 7 days, changes might attribute to ordinary aging of the biological product matrix. Contrary, ClO_2 and PTW show statistically meaningful texture alterations after their treatment and a storage of 7 days ($P > 0.001$), but distinct changes from day to day could not be supported by statistics ($p > \alpha$). A picture, which states changes due to a sanitizer treatment, but they do not persist over the whole storage time (p-values $> \alpha$). This results in the question if the sanitizer somehow conserves the leaves after they alter their structural appearance in a certain way. Alternatively, the other way around, do ClO_2 and PTW trigger aging processes in the leaves, which lead to massive structural changes after a 7-day storage period? Nevertheless, future investigations with separated lettuce leaf components might provide insight in such processes and support the testing of distinct hypotheses. It is important to have statistically meaningful basic values (e.g., alterations in color and texture) as a basis for logical links answering more holistic questions. Consequently, these quality characteristics are retained using this emerging technology.

Lettuce accumulates nitrate in its leaves during its growth (39, 40). Among the foods consumed by humans, plants represent between 72 and 94% of daily intake of nitrate (41). Dietary nitrate is mainly derived from the consumption of vegetables, in particular green leafy vegetables such as rocket (4,800 mg NO_3^- kg^{-1}) and lamb's lettuce (2,130 mg NO_3^- kg^{-1}) (29). However, nitrate contents in fresh-cut endive can decrease during storage (17), due to a concomitant growth of nitrate-metabolizing microorganisms (29, 42). If nitrate is converted to nitrite, this can have negative health effects, as nitrite can be a source of carcinogenic nitrosamines (43–45). Therefore, the European Union established the maximum permissible levels from 4,000 to 5,000 mg NO_3^- kg^{-1} for the winter season and 3,000 to 4,000 mg NO_3^{-1} kg^{-1} for the summer season (30). The endive samples examined in all our process variants remained within the legally prescribed nitrate values. Therefore, the use of PTW as a washing additive maintained endive food quality concerning the nitrate content. Further investigations about other valuable content compounds such as vitamin C may be also useful.

The promising results for food quality retention of color, texture, and nitrate obtained using PTW as process water additive were supported by the microscopic measurements based on SEM, AFM, and TEM. For trials using tap water or PTW, there was no significant difference noted in the influence compared to each other and to unwashed endive samples. However, for the application of ClO_2 the thylakoid stacks seem to be affected in TEM images, therefore the light-dependent reactions of photosynthesis may be negatively influenced as the interval between photosystem II complexes was expected to be larger. Although harvested and processed lettuce no longer has to undergo photosynthesis, it is still a living tissue, which may be subject to an accelerated aging process due to the change. This may not only affect the shelf life but also the quality of the tissue. Fujii et al. (46) mentioned in their work on photo inhibition of chloroplast genome-modified and common lettuce, that not only were the grana altered, but also that the plastoglobuli, which contain accumulated lipids and lipoproteins, were enlarged (46).

The use of PTW as a new washing and sanitization treatment provides a means to extend safe shelf life while maintaining produce food quality characteristics within regulatory guidelines. To fulfill all requirements of a regulatory environment, further investigations on sensory properties as well as toxicology are necessary, notwithstanding the need to examine marketability and cost benefit with the a product life cycle analysis.

In conclusion, the present study demonstrated that PTW could be used as a process water agent to improve conventional washing methods in fresh-cut processing at diverse stages of a process line, without any impairment of the quality of fresh-cut endive directly after treatment and during subsequent storage for 7 days. The promising results and the advantages of PTW including low-temperature, simple and cheap generation and demonstrated comparability to current procedures such as tap water rinsing and chlorinated water offer a wide range of innovative applications.

DATA AVAILABILITY STATEMENT

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

AUTHOR CONTRIBUTIONS

US and PB: conceptualization. US, TW, CW, JSc, and HB: methodology. US, OH, HW, TW, CW, JSc, and HB: investigation. US: writing–review and editing. OH, DB, PB, and JE: writing–review and editing. JSt: visualization. DB, PB, and JE: supervision. JE: project administration and funding.

All authors have read and agreed to the published version of the manuscript.

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REFERENCES

1. Baur S, Klaiber R, Wei H, Hammes WP, Carle R. Effect of temperature and chlorination of pre-washing water on shelf-life and physiological properties of ready-to-use iceberg lettuce. *Innov Food Sci Emerg Technol.* (2005) 6:171–82. doi: 10.1016/j.ifset.2005.02.002
2. Odumeru JA, Mitchell SJ, Alves DM, Lynch JA, Yee AJ, Wang SL, et al. Assessment of the microbiological quality of ready-to-use vegetables for health-care food services. *J Food Prot.* (1997) 60:954–60. doi: 10.4315/0362-028X-60.8.954
3. Saltveit ME. Physical and physiological changes in minimally processed fruits and vegetables. In: *Phytochemistry of Fruits and Vegetables*, Tomas-Barberan FA, Robins RJ, editors. Oxford: Oxford University Press (1997), p. 205–20.
4. Wulfkühler S, Kurfiss L, Kammerer DR, Weiss A, Schmidt H, Carle R. Impact of different washing procedures on quality of fresh-cut iceberg lettuce (*Lactuca sativa* var. capitata L.) and endive (*Cichorium endivia* L). *Eur Food Res Technol.* (2013) 236:229–41. doi: 10.1007/s00217-012-1878-5
5. Saltveit ME. Wound induced changes in phenolic metabolism and tissue browning are altered by heat shock. *Postharv Biol Technol.* (2000) 21:61–9. doi: 10.1016/S0925-5214(00)00165-4
6. Gil MI, Selma MV, Lopez-Galvez F, Allende A. Fresh-cut product sanitation and wash water disinfection: problems and solutions. *Int J Food Microbiol.* (2009) 134:37–45. doi: 10.1016/j.ijfoodmicro.2009.05.021
7. Zagory D. Effects of post-processing handling and packaging on microbial populations. *Postharv Biol Technol.* (1999) 15:313–21. doi: 10.1016/S0925-5214(98)00093-3
8. Tomas-Callejas A, Lopez-Galvez F, Sbodio A, Artes F, Artes-Hernandez F, Suslow TV. Chlorine dioxide and chlorine effectiveness to prevent *Escherichia coli* O157: H7 and *Salmonella* cross-contamination on fresh-cut red chard. *Food Control.* (2012) 23:325–32. doi: 10.1016/j.foodcont.2011.07.022
9. Ölmez H, Kretzschmar U. Potential alternative disinfection methods for organic fresh-cut industry for minimizing water consumption and environmental impact. *LWT Food Sci. Technol.* (2009) 42:686–93. doi: 10.1016/j.lwt.2008.08.001
10. Gopal A, Coventry J, Wan J, Roginski H, Ajlouni S. Alternative disinfection techniques to extend the shelf life of minimally processed iceberg lettuce. *Food Microbiol.* (2010) 27:210–9. doi: 10.1016/j.fm.2009.10.006
11. Baier M, Görgen M, Ehlbeck J, Knorr D, Herppich WB, Schlüter O. Non-thermal atmospheric pressure plasma: screening for gentle process conditions and antibacterial efficiency on perishable fresh produce. *Innov Food Sci Emerg Technol.* (2014) 22:147–157. doi: 10.1016/j.ifset.2014.01.011
12. Biazotto Bachelli ML, Álvares Amaral RD, Benedetti BC. Alternative sanitization methods for minimally processed lettuce in comparison to sodium hypochlorite. *Braz J Microbiol.* (2013) 44:673–8. doi: 10.1590/S1517-83822013005000065
13. Ramos B, Miller FA, Brandao TRS, Teixeira P, Silva CLM. Fresh fruits and vegetables - an overview on applied methodologies to improve its quality and safety. *Innov Food Sci Emerg Technol.* (2013) 20:1–15. doi: 10.1016/j.ifset.2013.07.002
14. Issa-Zacharia A, Kamitani Y, Muhimbula HS, Ndabikunze BK. A review of microbiological safety of fruits and vegetables and the introduction of electrolyzed water as an alternative to sodium hypochlorite solution. *Afr J Food Sci.* (2010) 4:778–89. doi: 10.5897/AJFS.9000006
15. Hägele F, Nübling S, Schweiggert RM, Baur S, Weiss A, Schmidt H, et al. Quality improvement of fresh-cut endive (*Cichorium endivia* L.) and recycling of washing water by low-dose UV-C irradiation. *Food Bioprocess Technol.* (2016) 9:1979–1990. doi: 10.1007/s11947-016-1782-2
16. Nübling S, Hägele F, Schweiggert RM, Carle R, Schmidt H, Weiss A. Effect of different wash water additives and deep-frozen storage on the quality of curly parsley (*Petroselium crispum* var. crispum). *Food Bioprocess Technol.* (2019) 12:158–65. doi: 10.1007/s11947-018-2201-7
17. Nübling S, Hägele F, Wohlt D, Graf B, Schweiggert RM, Carle R, et al. Effects of *Quillaja saponaria* extract and Nα-lauroyl-L-arginine ethyl ester on reducing selected foodborne pathogens *in vitro* and maintaining quality of fresh-cut endive (*Cichorium endivia* L.) at pilot plant scale. *Food Control.* (2017) 73, 393–400. doi: 10.1016/j.foodcont.2016.08.029
18. Nübling S, Wohlt D, Saile N, Weiss A, Schmidt H. Antimicrobial effect of lauroyl arginate ethyl on Escherichia coli O157:H7 and *Listeria monocytogenes* on red oak leaf lettuce. *Eur Food Res Technol.* (2017) 243:879–87. doi: 10.1007/s00217-016-2802-1
19. Matthews KR. (2006). *Microbiology of Fresh Produce: Emerging Issues in Food Safety*. Washington, DC: American Society for Microbiology Press (ASM Press).
20. Bourke P, Ziuzina D, Boehm D, Cullen PJ, Keener K. The potential of cold plasma for safe and sustainable food production. *Trends Biotechnol.* (2018) 36:615–26. doi: 10.1016/j.tibtech.2017.11.001
21. Schlüter O, Ehlbeck J, Hertel C, Habermeyer M, Roth A, Engel KH, et al. Opinion on the use of plasma processes for treatment of foods. *Mol Nutr Food Res.* (2013) 57:920–7. doi: 10.1002/mnfr.201300039
22. Varilla C, Marcone M, Annor GA. Potential of cold plasma technology in ensuring the safety of foods and agricultural produce: a review. *Foods.* (2020) 9:1435. doi: 10.3390/foods9101435
23. Goldston RJ, Rutherford PH. *Introduction to Plasma Physics*. New York, NY: Taylor and Francis Group. ISBN 978-0-7503-0183-1 (2000).
24. Zhang Q, Liang Y, Feng H, Ma R, Tian Y, Zhang J, et al. A study of oxidative stress induced by non-thermal plasma-activated water for bacterial damage. *Appl Phys Lett.* (2013) 102:203701. doi: 10.1109/PLASMA.2013.6633407
25. Schnabel U, Handorf O, Stachowiak J, Boehm D, Weit C, Weihe T, et al. Plasma-functionalized water: from bench to prototype for fresh-cut lettuce. *Food Eng Rev.* (2020). doi: 10.1007/s12393-020-09238-9. [Epub ahead of print].
26. Ehlbeck J, Schnabel U, Polak M, Winter J, von Woedtke T, Brandenburg R, et al. Low temperature atmospheric pressure plasma sources for microbial decontamination. *J Phys D Appl Phys.* (2011) 44:13002. doi: 10.1088/0022-3727/44/1/013002
27. Krohmann U, Neumann T, Ehlbeck J. *Method and Device for Igniting and Generating an Expanding Diffuse Microwave Plasma and Method and Device for Plasma Treating Surfaces and Substances by Using This Plasma*. US Patent No. 1 US20100001647A1. Washington, DC: U.S. Patent and Trademark Office (2010).

28. Pipa AV, Andrasch M, Rackow K, Ehlbeck J, Weltmann KD. Observation of microwave volume plasma ignition in ambient air. *Plasma Sources Sci Technol.* (2012) 21:035009. doi: 10.1088/0963-0252/21/3/035009
29. European Food Safety Authority (EFSA). Nitrate in vegetables. Scientific opinion of the panel on contaminants in the food chain. *EFSA J.* (2008) 689, 1–79. doi: 10.2903/j.efsa.2008.689
30. EC No 1881/2006. COMMISSION REGULATION (EC) No 1881/2006 of 19 December 2006 setting maximum levels for certain contaminants in foodstuffs. *Off J Eur Union.* (2006) L 364/5–L 364/24. Available online at: https://eur-lex.europa.eu/eli/reg/2006/1881/oj
31. Kongwong P, Boonyakiat D, Poonlarp P. Extending the shelf life and qualities of baby cos lettuce using commercial precooling systems. *Postharv Biol Technol.* (2019) 150:60–70. doi: 10.1016/j.postharvbio.2018.12.012
32. Malinova I, Mahlow S, Alseekh S, Orawetz T, Fernie AR, Baumann O, et al. Double knockout mutants of arabidopsis grown under normal conditions reveal that the plastidial phosphorylase isozyme participates in transitory starch metabolism. *Plant Physiol.* (2014) 164:907–21. doi: 10.1104/pp.113.227843
33. He SY, Feng GP, Yang HS, Wu Y, Li YF. Effects of pressure reduction rate on quality and ultrastructure of iceberg lettuce after vacuum cooling and storage. *Postharv Biol Technol.* (2004) 33:263–73. doi: 10.1016/j.postharvbio.2004.03.006
34. Schnabel U, Andrasch M, Stachowiak J, Weit C, Weihe T, Schmidt C, et al. Sanitation of fresh-cut endive lettuce by plasma processed tap water (PPtW) - Up-scaling to industrial level. *Innov Food Sci Emerg Technol.* (2019) 53:45–55. doi: 10.1016/j.ifset.2017.11.014
35. Niquet R, Boehm D, Schnabel U, Cullen PJ, Bourke P, Ehlbeck J. Characterising the impact of post-treatment storage on chemistry and antimicrobial properties of plasma treated water derived from microwave and DBD sources. *Plasma Process Polym.* (2018) 15:e1700127. doi: 10.1002/ppap.201700127
36. Schnabel U, Niquet R, Schlüter O, Gniffke H, Ehlbeck J. Decontamination and sensory properties of microbiologically contaminated fresh fruits and vegetables by microwave plasma processed air (PPA). *J Food Process Preserv.* (2015) 39:653–62. doi: 10.1111/jfpp.12273
37. Schnabel U, Andrasch M, Weltmann K-D, Ehlbeck J. Inactivation of vegetative microorganisms and *Bascillus atrophaeus* endospores by reactive nitrogen species (RNS). *Plasma Process Polym.* (2014) 11:110–6. doi: 10.1002/ppap.201300072
38. Schnabel U, Niquet R, Krohmann U, Winter J, Schlüter O, Weltmann KD, et al. Decontamination of microbiologically contaminated specimen by direct and indirect plasma treatment. *Plasma Process Polym.* (2012) 9:569–75. doi: 10.1002/ppap.201100088
39. Roorda van Eysinga JPNL. Nitrate and glasshouse vegetables. *Fertilizer Res.* (1984) 5:149–56. doi: 10.1007/BF01052712
40. Andriolo JL. *Fisiologia das Culturas Protegidas*. Santa Maria: UFSM, 142 p.
41. Santamaria P. Contributo degli ortaggi all' assunzione giornaliera di nitrato - nitrito e nitrosamina. *Ind Aliment.* (1997) 36:1.329–1.334.
42. Chung J-C, Chou S-S, Hwang D-F. Changes in nitrate and nitrite content of four vegetables during storage at refrigerated and ambient temperatures. *Food Addit Contam.* (2004) 21:317–22. doi: 10.1080/02652030410001668763
43. Cometti, N. N., and Furlani, P. R. (2009). Hidroponia e segurança alimentar. In: *Tópicos Especiais em Produção Vegetal I*, Ferreira A, Lima ABP, Matta FP, Amaral JAT, Lopes JC, Pezzopane JEM, et al., editors. Alegre, ES: Centro de Ciências Agrárias, p. 517–528.
44. Addiscott T. Is it nitrate that threatens life or the scare about nitrate? *J Sci Food Agricul.* (2006) 86:2005–9. doi: 10.1002/jsfa.2573
45. Wright MJ, Davison KL. Nitrate accumulation in crops and nitrate poisoning in animals. *Adv Agron.* (1964) 16:197–274. doi: 10.1016/S0065-2113(08)60025-5
46. Fujii R, Yamano N, Hashimoto H, Misawa N, Ifuku K. Photoprotection vs. photoinhibition of photosystem ii in transplastomic lettuce (*Lactuca sativa*) dominantly accumulating astaxanthin. *Plant Cell Physiol.* (2016) 57:1518–29. doi: 10.1093/pcp/pcv187

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Direct Contact Ultrasound in Food Processing: Impact on Food Quality

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Consumers' demand for "minimally processed" products that maintain the "fresh-like" characteristics has increased in recent years. Ultrasound (US) is a non-thermal technology that enhances mass and energy transfer processes resulting in improved food quality. A new method of applying US to food without using a liquid or gaseous medium for the propagation of acoustic waves has recently been under research. It is known as direct contact US, since the food is directly placed on a plate where the transducers are located. In this type of systems, the main effect is not cavitation but acoustic vibration, which encourages mass and energy transfer processes due to the "sponge effect." Furthermore, as the product is not immersed in a liquid medium, the loss of hydrophilic nutritional compounds is reduced; systems such as these can thus be more easily implemented on an industrial level. Nevertheless, the very few studies that have been published about these systems mainly focus on dehydration and freezing. This article summarizes published research on the impact of direct contact US in nutritional and organoleptic quality of food in order to assess their potential to meet new market trends.

Keywords: ultrasound, direct contact, quality, food compounds, nutritional compounds

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INTRODUCTION

In recent years, consumers are demanding safe, healthy and long shelf-life products that maintain their "fresh-like" characteristics but without any chemical preservatives. However, this cannot be achieved through the application of thermal technologies, which, although longer shelf-life is possible, nutritional and quality losses are caused due to the high temperatures and long processing time. Therefore, since the twentieth century, non-thermal food processing technologies such as pulsed electric fields (PEF), high hydrostatic pressure (HPP), ultrasound (US), UV light, cold plasma and irradiation (IR) have been widely investigated (1). These technologies allow extending the shelf-life of the food but with small increase in the temperature, affecting minimally the nutritional properties, texture, color, taste and aroma of the food, which means, that products with similar characteristics to those of fresh food are obtained (2). However, despite consumers' demand for "minimally processed" products, awareness of novel technologies is still very low and there is a lack of trust in them (3).

One of these non-thermal food processing technologies is US, and its potential to improve mass and energy transfer processes has attracted great attention. Moreover, US is included within the "Green Food Processing" concept proposed by Chemat et al. (4) to refer to those technologies that allow to process food with a lower consumption of energy and water, thereby obtaining processing methods that are more sustainable and environmentally friendly.

In the food industry, most research on the application of high power US (20–100 kHz, > 1 W/cm^2) is focused on systems in which a liquid or a gaseous medium (such as air, then called airborne US) is used for the propagation of US waves (5). Most of the applications of this technology (such as cleaning, atomization, homogenization and emulsification, defoaming, drying, and freezing) are based on that manner of applying US due to its capability to produce permanent changes in the propagation medium (6). The mechanisms of action behind these effects are the cavitation phenomenon, microcurrents, microjets, the sponge effect, and the primary radicals H· and ·OH, which occur in the food matrix (**Figure 1**) (7, 8).

Several studies have been conducted over the last few years on the potential of ultrasound to obtain food with greater nutritional value and better organoleptic properties (9). This technology favors mass and energy transfer processes, assisting i.e., the elaboration of infusions at lower temperatures (30°C) with a higher content of total polyphenols (6–10 folds higher) and anthocyanins (8–10 folds higher) (10), and red wines with a greater content of polyphenols (11). Moreover, US also promote the elimination of compounds naturally present in food that are potentially harmful to human health, such as oligosaccharides from pulses (12) or heavy metals such cadmium from edible crabs (13), and even carcinogenic compounds such as acrylamide from fried potatoes (14). One of the most commonly used food preservation process is dehydration in which US application reduces the loss of bioactive compounds and improves the color of dehydrated products (15). In the case of freezing, in addition to reducing processing times, the US favors the formation of small ice crystals that, when thawed, reduce the loss of water, resulting in a product with better texture (16, 17).

Nonetheless, consumers not only demand "minimally processed" food, but also have great interest in functional food or nutraceutical ingredients that have additional healthy benefits beyond basic nutrition (18). However, the conventional extraction of natural food additives is quite limited due to the high-energy cost, the use of toxic solvents or the high consumption of water (19). US allows the extraction of bioactive compounds in an environmentally friendly way (4) reducing the use of solvents or with lower energetic costs. In fact, the potential of US to improve the extraction of bioactive compounds (such as polyphenols, carotenoids and anthocyanins) has been demonstrated in many studies (18–25). In addition, US also favor the extraction of functional compounds from foods that give them specific characteristics (18, 26). Within this group are the polysaccharides such as pectins (27, 28), gums (29), alginate and carrageenans (30, 31) and cellulose (32) that provide structure, stability and viscosity to the products. Finally, US also improved the extraction of proteins used to enrich food with low protein content or those used as functional additives to stabilize emulsions or foams (33–35).

Therefore, US is a non-thermal technology with great potential for the food industry and, in fact, there is already some equipment operating in industries e.g., for extraction, cutting soft products and filtration (36). However, there are still many limitations that make this not always possible, and that is why new US application systems are sought such as direct contact or contacting US system, in which the food sample is in close contact with the transducer. Differently to traditional US in which the product is immersed in a liquid, usually water, or applied to air (airborne US), US is applied in dried conditions. In this case, the acoustic vibrations that reach the solid matrix cause successive compressions and expansions of the material, which behaves as a sponge (**Figure 1**) (37). This mechanical stress ("sponge effect") may result in microcracks and microchannels in the internal structure. Acoustic vibration can also improve energy transfer,

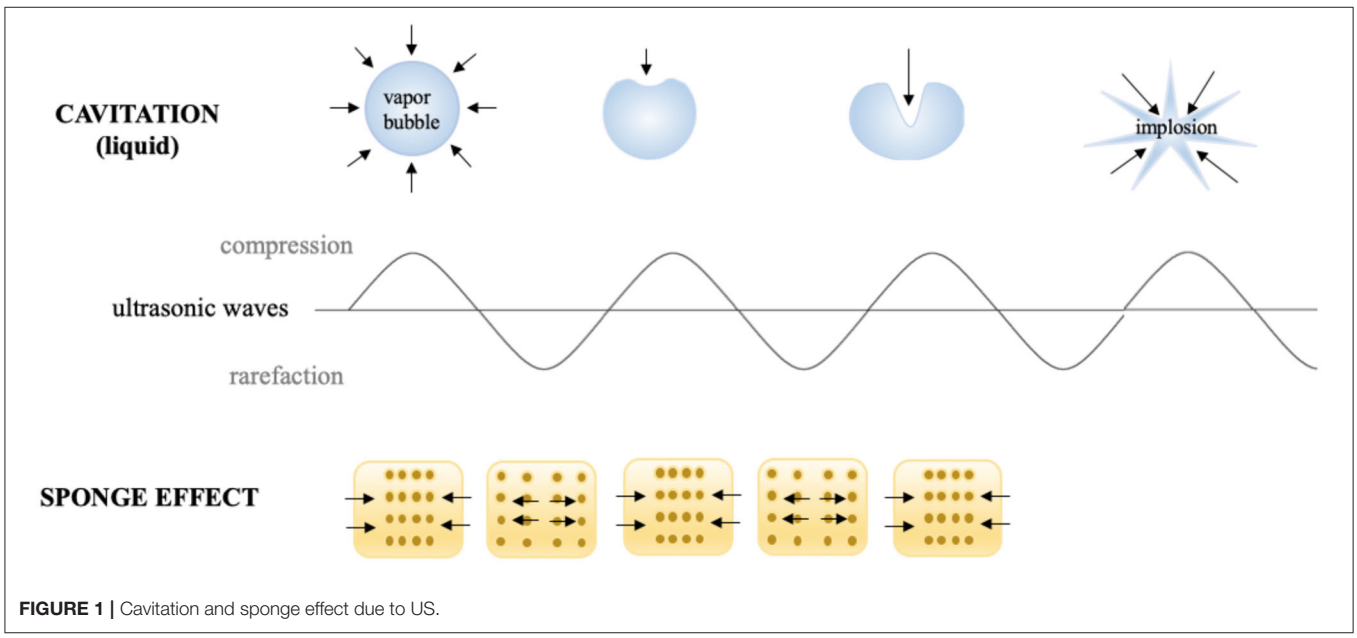


FIGURE 1 | Cavitation and sponge effect due to US.

as reported in different processes such as freezing, drying, etc. (38). As indicated, the main advantage of this system is that the loss of hydrophilic macro or micronutrients would be reduced (39), although this point has not been specifically investigated. In addition, it can be applied to any product without the need to be immersed in water. However, very few studies have been published in this field (40) and thus, this review focuses on describing direct contact US systems and analyzing their impact on food nutritional, quality and sensory properties.

DIRECT CONTACT US SYSTEMS

Similar to water-immersed or airborne US systems, frequency, vibrational amplitude and power intensity are the key parameters (5) for direct contact US. In general, low frequencies are used (close to 20 kHz) where physical and mechanical effects are mostly observed. However, there is not specific studies of the effect of this parameter. Similarly, it occurs with the other parameters. Any case, associated to them, thermal effect can occur if high intensities are applied. This is a crucial parameter in direct contact US which has to be considered for its scaling-up, since it can affect product quality by losing thermosensitive nutritional compounds (i.e., ascorbic acid) or degrading certain pigments (i.e., anthocyanins, carotenoids) affecting negatively the color of the food. Although US is a non-thermal technology by definition, the US-treated product may become heated due to friction among particles, dispersion, and the viscous absorption that takes place when sound waves are transmitted through food products (41), and also due to expansions and contractions generated by the piezoelectric ceramic of the transducers (42). Due to this, to minimize the heat emitted by the transducers, a series of cooling systems or US ON/OFF activation protocols are applied (43, 44).

Several systems have been developed to apply US by direct contact in a series of different food producing processes such as drying, freezing, etc. (**Table 1**). All of them have the same basic elements; moreover, in all cases, the transducers or horns are in direct contact with the plate (emitter) on which the samples are placed.

Drying

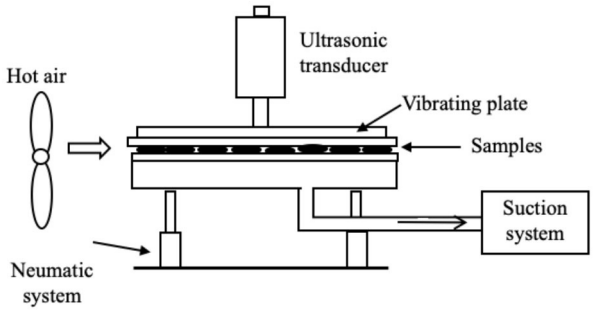
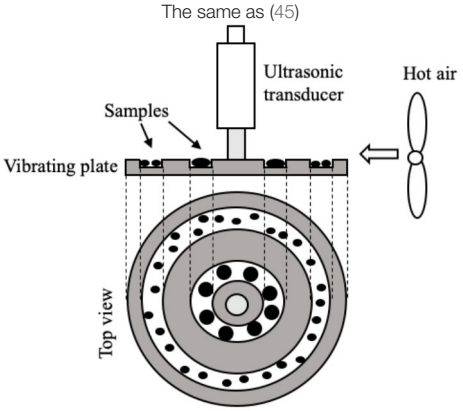
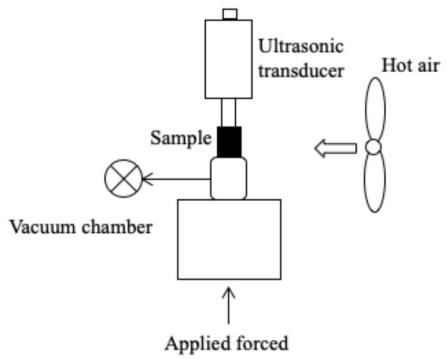
Drying is a preservation process that has a great effect on organoleptic properties and heat-sensitive nutritional compounds such as antioxidants and vitamins (39, 59). Numerous studies have focused on the study of US-assisted drying of fruits and vegetables and its effect on the physical (water activity, shrinkage, rehydration, color, porosity, among others) and chemical (nutrients, antioxidants, vitamins) quality of the dried product (60–62).

All the studies included in **Table 1** reported that US improved the drying rate (up to 70% in some cases), reduced drying time, and enhanced the quality of the dehydrated food. For example, Liu et al. (50) studied the impact of contact-US-assisted drying (28 kHz) on the color of purple-fleshed sweet potato slices by applying 30 W and 60 W US treatments and four air temperatures (40, 50, 60 and 70°C). The effect of the US was more noticeable at high temperatures, as drying times were greatly reduced: at 70°C, time reduction was 18.7 % (30 W) and 37.5 % (60 W), and the dried potato samples were brighter, redder, and less yellowish than control. Tao et al. (54) also observed improvements in the color (whiter values) of dried garlic assisted by US reducing drying time by 48.5% at 60°C. Similar conclusions have been reported using airborne systems for carrots (63), strawberries (64), and green peppers (65).

An important aspect to be considered in the traditional heat-dried process is the potential loss of thermosensitive bioactive compounds (66). Thus, the reduction of drying times by accelerating mass and energy transfer processes minimizes the loss of nutritional compounds. Liu et al. (51, 52) studied the effect of direct-contact-US-assisted convective drying on total phenolic content (TPC), flavonoids, and ascorbic acid of pear slices by applying hot air flow (35, 45 and 55°C) or far infrared radiation (FIR) (100, 220, 340 W). It was observed that the higher the ultrasonic power the lower the loss in TPC: e.g., at 45°C and ultrasonic powers of 24 and 48 W, the retention of TPC was 14.7 and 39.7%, respectively, whereas at 220 W FIR and ultrasonic powers of 30 and 60 W, the improvement compared to control was 6.7 and 16.7%, respectively. However, no beneficial effect of US was observed at 55°C and 340 W FIR; it even had a negative effect as compared to control. According to the authors, this was related to oxidation reactions, since at elevated temperatures the tissue was more sensitive to damage; when US was applied, associated mechanical effects could intensify heat damage, while oxidative reactions occurred more easily due to increased contact between phenolic compounds and oxygen (67). Similar results were found for flavonoids when US was applied at low temperatures (35°C; 48 W US) or at low FIR powers (100 W, 220 W; 60 W US), thereby leading to increases in flavonoid content of 21.1, 45.5 and 26.6%, respectively. However, at higher temperatures or FIR powers, the effect of US was harmful. The effect of the US treatment on ascorbid acid content was always positive, and increased along with power. The highest ascorbic acid contents were observed at 35°C and 48 W US (US samples, 42.5 mg vitamin C/100 g vs. non-US samples, 30.0 mg vitamin C/100 g), and at 100 W FIR and 60 W US (US samples, 265.5 mg ascorbic acid/100g vs. non-US samples, 226.1 mg ascorbic acid/100 g). Another example is that of Tao et al. (54) applying 20 kHz-US during the drying at 60°C of garlic slices (*Allium sativum* L.). Garlic has healthy benefits associated with thiosulfinates that have anti-inflammatory, antioxidant and antimicrobial properties (68). In this study, the total thiosulfine content was 16% higher at an ultrasonic intensity of 902.7 W/m^2 compared to non-sonicated samples, The TPC was also improved at 902.7 W/m^2 (12 %), while at a higher ultrasonic intensity (1,513 W/m^2) the content was even lower than control. Nevertheless, the antioxidant capacity was very similar between non-sonicated and sonicated samples, showing a small improvement when applying 902.7 W/m^2. The application of direct contact US to food drying systems can therefore increase the retention of thermosensitive bioactive compounds but the treatment conditions need to be optimized, mainly the ultrasonic power.

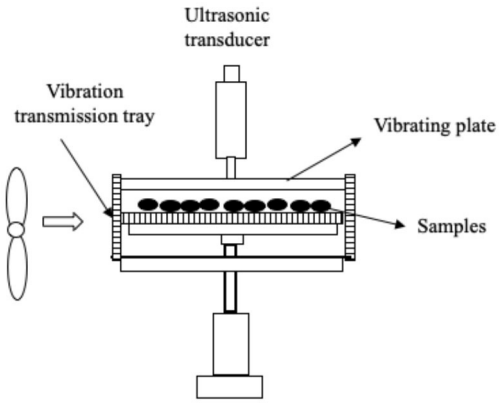
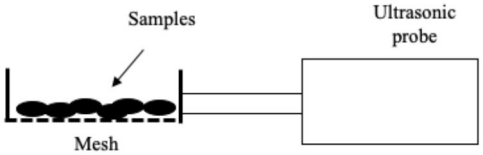
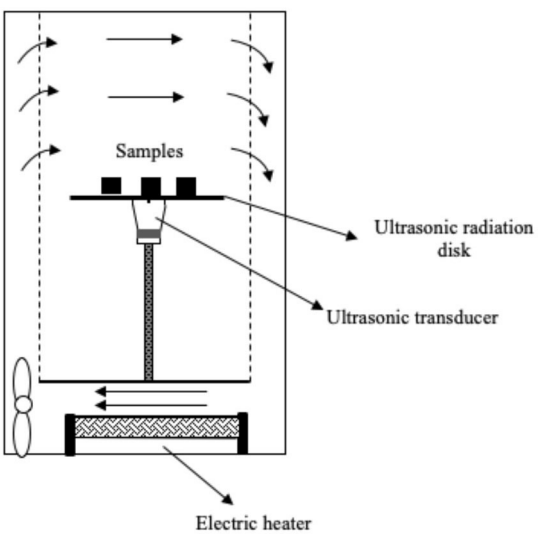
Finally, in the studies by Liu et al. (50, 52) it was observed that rehydration capacity, one of the most important parameters that

TABLE 1 | Different systems of application of US by direct contact with food.

Process	US system	Study	Results	References
Drying	 Adapted from (45)	Carrot slices US parameters: - Frequency: 20 kHz - Power: 100 W - Static pressure Conditions evaluated: - Airflow: 1 and 3 m/s - Air temperature: 22°C	Improvement in the drying rate (70.0%) Lower final moisture	(45)
Drying	The same as (45)  Adapted from (46)	Carrot, apple, and mushroom slices US parameters: - Frequency: 20 kHz - Power: 100 W - Pressure static (1): 0.05 kg/cm^2 Conditions evaluated: - Airflow: 1.7–2 m/s - Air temperature: 20 and 55°C	Reduction of drying time (carrots: up to three times, apples: 50.0–76.7% and mushrooms: 68.3–83.3%) Reduction of drying time (carrots: 50.0–58.3%, apples: 66.7–233.7% and mushrooms: 50.0–75.0%)	(46)
Drying	 Adapted from (47)	Apples and potatoes slices US parameters: - Frequency: 20 kHz - Power: 25 and 50, W - Static pressure: 0.0155–0.050 kg/cm^2 - Suction pressure: 10 and 20 mbar Conditions evaluated: - Airflow: 1 m/s - Air temperature: 31°C	Increase in the effective diffusivity coefficient	(6, 37)

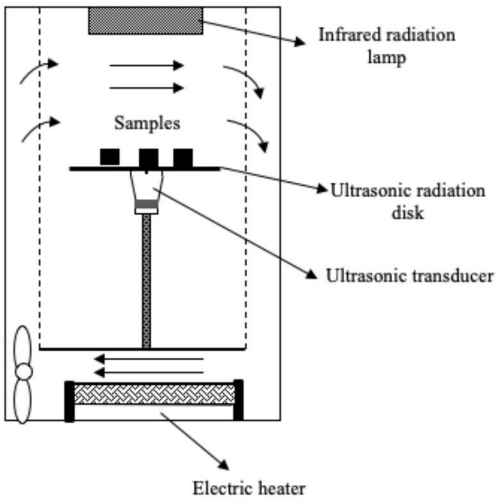
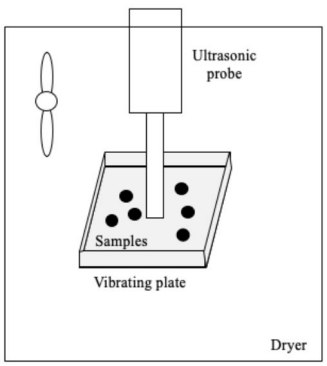
(Continued)

TABLE 1 | Continued

Process	US system	Study	Results	References
Drying	 Adapted from (48)	Apple slices US parameters: - Frequency: 20 kHz - Power: 75 and 90 W Conditions evaluated: - Air temperature: 40 and 60°C - RH% air: 25% - Airflow: 1 m	Reduction of drying time (46.0–57.0 %) No differences in texture	(48)
Drying	 Adapted from (43)	Red bell peppers and apples US parameters: - Frequency: 24 kHz - Power: 42 W - Effective amplitude: 6–13 μm Conditions evaluated: - Air temperature: 70°C - Continuous US treatment - Intermittent US treatment: • 50% net sonication time • 10% net sonication time	No impact on final relative water content Intermittent US treatment at net sonication of 10 % did not improve the process, but at net sonication of 50% there was a reduction in drying time (18–20%) Continuous US treatment allowed to reduce drying time (18–27%)	(43)
Drying	The same as (43)	Potato cylinders US parameters: - Frequency: 24 kHz Conditions evaluated: - Air temperature: 70°C	US effect was strongest in the outermost layer (0.0–0.6 mm) and at the sonicated surface US treatment allowed to reduce drying time (by 10.3%)	(49)
Drying	 Adapted from (50)	Purple-fleshed sweet potato slices US parameters: - Frequency: 28 kHz - Power: 30 and 60 W Conditions evaluated: - Air temperature: 40, 50, 60, and 70°C - Airflow: 1 m/s	Drying time was reduced by increasing the US power (31.5–47.7 %) but the US effect was less pronounced at higher air temperature The drying rate was improved (50.8–100.0 %) at high US power and low temperature Increase in the effective moisture diffusivity (D_{eff}) (17.6–48.1%) Distortions of the cellular tissue and the appearance of large cavities Improvement of the rehydration capacity	(50)

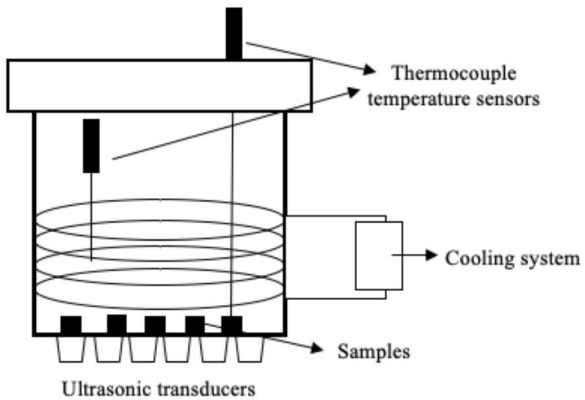
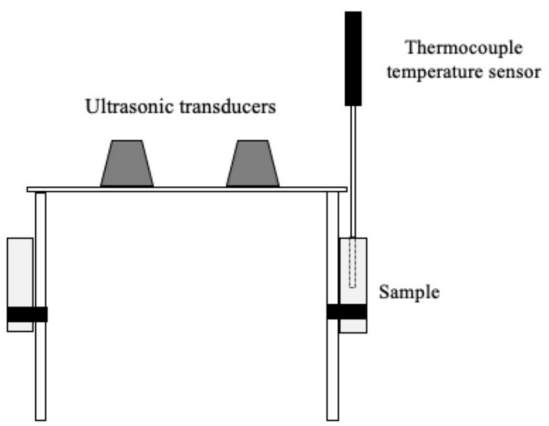
(Continued)

TABLE 1 | Continued

Process	US system	Study	Results	References
Drying	 Adapted from (51)	Pear slices US parameters: - Frequency: 28 kHz - Power: 30 and 60 W Conditions evaluated: - FIR power: 100, 220, and 340 W - Air flow: 1.5 m/s	Increase in the drying rate (at 45°C, the increase was 33.3% at 24 W and 140.1% at 48 W) Positive impact on total phenolic content, flavonoids, and ascorbic acid Appearance of more numerous and larger microchannels in the cell tissue	(51)
Drying	The same as (51)	Kiwi slices US parameters: - Frequency: 28 kHz - Power: 18, 36, and 54 W Conditions evaluated: - FIR temperature: 120, 200, and 280°C - Airflow: 1.5 m/s	Reduction of drying time (the increase at 120, 200, and 280°C was 32.2–48.4%, 22.2–38.9%, 14.3–33.3%, respectively) The drying rate was improved (66.7%) by increasing US power US decreased the resistance to internal diffusion, facilitated the migration and removal of the immobilized and bound water	(52)
Drying	The same as (50)	Pear slices US parameters: - Frequency: 28 kHz - Power: 24 and 48 W Conditions evaluated: - Air temperature: 35, 45, and 55°C - Airflow: 1 m/s	Best increase in drying rate (33.3–140.1 %) at low air temperature The microstructure of the pear samples showed more numerous and larger cavities Positive impact on total phenolic content, flavonoids, and vitamin C Improvement in rehydration capacity	(53)
Drying	 Adapted from (54)	Garlic (*Allium sativum* L.) US parameters: - Frequency: 20 kHz - US treatment: 3 s on/ 1 s off Conditions evaluated: - Power: 216.8, 902.7, and 1513.5 W/m^2. - Air temperatura 50, 60, and 70°C - Airflow: 2.5 m/s	Reduction of drying time (the increase at 216.8, 902.7, and 1513.5 W/m^2 was 5.0%, 12.5%, 35.0% respectively, at 50°C) The drying time was reduced by increasing air temperature Positive impact on thiosulfinate and TPC at 216.8 and 902.7 W/m^2 Greater retention of organosulfur compounds Color improvement	(54)

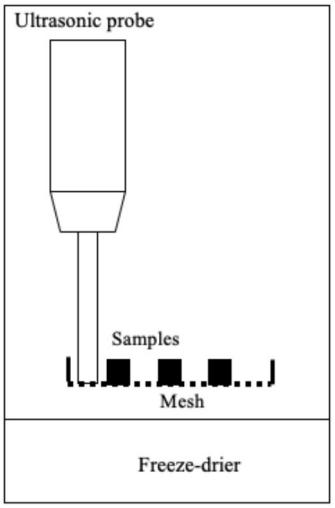
(Continued)

TABLE 1 | Continued

Process	US system	Study	Results	References
Drying	The same as (54)	White cabbage (*Brassica oleracea* L. variety Capitana L.) US parameters: - Frequency: 20 kHz - US treatment: 4 s on/2 s off Conditions evaluated: - Power: 492.3 and 1131.1 W/m^2 - Air temperature: 60 °C - Airflow: 2.5 m/s - Pre-blanching treatment (100°C/30 s)	Synergistic effect of blanching and subsequent US drying to intensify drying process No color differences Higher TPC (12.6 %) in un-blanched sonicated samples at 492.3 W/m^2 No positive effect on Vitamin C content No clear effect on glucosinolate	(55)
Freezing	 Adapted from (56)	Mushroom (*Agaricus bisporus*) US parameters: - Frequency: 20 kHz - Power: 300 W - 12 transducers Conditions evaluated: - US treatment: 10 s on/20 s off when the sample temperature reached −1°C - US treatment: 10 s on/ 10 min off during 3 weeks of frozen storage	Earlier nucleation Smaller crystal size and more uniform shape The microstructure was more uniform, featuring more numerous and more dense pores	(56)
Freezing	 Adapted from (57)	Chicken breasts US parameters: - Frequency: 40 kHz - Power: 50 W Conditions evaluated: - US treatment: 3s on/ 5s off throughout the entire freezing process - Air temperature −13 to −25° C - Air flow: < 0.4 m/s	Reduction of freezing time (19.9%) No difference in quality attributes such as WHC, CL and protein digestibility	(57)

(Continued)

TABLE 1 | Continued

Process	US system	Study	Results	References
Freeze-drying	 Adapted from (58)	Red bell peppers Samples were frozen in a cooling chamber to reach a temperature of −20°C Then they were dried by applying US US parameters: - Frequency: 20 kHz Freeze-drying pressure was 46 Pa Conditions evaluated: - Power: 76, 90, and 110 W - Net sonicated time: continuo (100%), 25%, 14% and 10%	Minimum US thermal effect at 76 W and net sonication time of 10 % Reduction of drying time No difference in quality attributes such as bulk density, color, ascorbic acid, and rehydration capacity	(58)

defines the quality of dehydrated food (69, 70), was improved by 10.6 % in samples of purple-fleshed sweet potato dried at 40°C and 60W (US), and by 36.4, 15.7 and 13.2% in samples of pear slices dried at 35, 45 and 55°C, respectively, and applying a US power of 48 W. These results can be explained by the fact that the application of US by direct contact in solid food, as reported in liquid immersion systems and air systems (19, 71), leads to the formation of cavities and microchannels in plant tissues via mechanical effects (49–52) that reduce internal resistance to the flow of water and enhance its incorporation during rehydration.

Freeze-Drying

Freeze-drying is a process widely used to obtain high-quality dehydrated food by preserving shape and color while minimizing the loss of nutrients (72). However, extended processing times and high energy costs are involved. The application of US could thus serve as a useful alternative in order to accelerate mass and energy transfer process. To the best of our knowledge, only one published study deals with the application of US by direct contact to vacuum freeze-drying. This would probably be due to the technical difficulties involved in applying US in vacuum freeze-drying systems (58). US equipment in that study consisted in two sonotrodes, in the tip of which a mesh was fixed to hold the samples (**Table 1**). An intermittent (10s on/90s off) application of US (from 76 to 110 W) led to a reduction in the freeze-drying time of red bell peppers of 11.5%, but no differences were observed in terms of rehydration capacity, bulk density, color, or ascorbic acid content of the treated samples compared to the conventionally freeze-dried samples. Since the application of US in the freeze-drying process allows reducing the processing time, it is necessary to conduct more studies to evaluate its impact on the nutritional and organoleptic quality of the food.

On the other hand, it is worth mentioning that US airborne systems have been tested in atmospheric drying at low temperature processes (an alternative to freeze-drying) with the aim of improving the quality of air-dried food. Bantle and Eikevik (44) did not observe any differences in color or shrinkage of green peas when US was applied. Similarly, Colucci et al. (73) investigated the impact of US-assisted atmospheric drying at freezing temperatures on the antioxidant properties of eggplant samples, and likewise did not find significant differences when applying US (25 and 50 W). Moreover, although differences were not significant, the application of US promoted the degradation of ascorbic acid (1.5–7%), TPC (4.2–15%) and antioxidant capacity (3–13.8%) in samples dried at −10°C and 2 m/s.

Freezing

The quality of frozen food is determined by the shape, location, and distribution of ice crystals inside the product (74, 75). Therefore, rapid freezing is sought in order to allow the formation of small and numerous intra- and extra-cellularly located ice crystals that minimize quality losses after thawing (76, 77). Many studies have shown that immersion freezing in ultrasonic baths improves the quality (microstructure, weight loss, texture, color, and nutritional components) of frozen food by promoting the initiation of nucleation, thereby controlling the growth of ice crystals and accelerating the transfer of mass and heat (19, 40, 78).

Islam et al. (56) studied the effect of direct contact US on the freezing process of mushroom (*Agaricus bisporus*) cubes by applying a frequency of 20 kHz and a power of 300 W with intermittent treatment of 1 s on/20 s off once the sample temperature reached −1°C, and treatments of 10 s on/10 min off

in the course of storage during 3 weeks. They observed that the sonicated samples displayed earlier nucleation at temperatures of $-2.0 \pm 0.05^{\circ}C$ compared to control samples, in which it occurred at $-2.6 \pm 0.01^{\circ}C$. Differences in morphology and size of the ice crystals were also detected by cryo-electron microscopy. The crystals of the sonicated samples were smaller, thinner, and columnar shaped, while those of control were larger, more irregular, and featured dendrites. Although no quality parameters were analyzed in this study, the characteristics of the ice crystals strongly suggest that the US-assisted process would have a lower impact on the quality of the frozen/thawed products. Recently, Astráin-Redín et al. (57) studied the influence on Water Holding Capacity (WHC), Cook Loss (CL) and protein digestibility of meat when applying direct contact US (40 kHz, 50 W) in freezing chicken breasts while applying an intermittent US treatment. No differences in terms of those quality parameters were observed between sonicated and control samples. These results may be due to the fact that the sample size was small (5–6 g), and, although US-assisted freezing was more rapid (9.9–11.3%), the process was already rapid enough in the control samples to have a negative impact on quality. Indeed, in larger pork loin samples (120 g), Zhang et al. (79) observed smaller and more uniformly distributed crystals resulting from immersion US freezing (180 W and 30 kHz), and they obtained 61% and 12.3% lower weight losses after thawing and cooking, respectively, when applying US compared to a forced air system. Moreover, Li et al. (17) evaluated the influence of immersion-US-assisted freezing (20 kHz) on chicken breast meat, and observed an increase in the proportion of water retained within the myofibrillar protein, thereby resulting in a higher WHC.

CONCLUSIONS

This review summarized the current state of knowledge regarding a new method of applying US to food samples, known as direct contact US systems. Although very few articles have been published on this subject, the application of US has already achieved considerable improvements in mass and energy transfer processes in the food industry, such as dehydration and freezing. In the case of dehydration, the application of US leads to a reduction in drying times, resulting in dehydrated food with a higher content of TPC, flavonoids and ascorbic acid, as well as improved sensory attributes such as color, along with improved functional properties (i.e., rehydration). However, most of the studies did not analyse the thermal effect that these systems could have on the samples; thus, the effect of the US treatment cannot be evaluated correctly as it can be hidden or misleading. As far as freezing processes are concerned, it has been reported that direct US contact freezing promotes the formation of small ice crystals, although no improvements in certain highly relevant quality parameters of defrosted foods such as WHC and CL have been observed. For all these reasons, the application of US by direct contact can be regarded as a thoroughly useful technique to improve mass and energy transfer processes of food. However, due to the scarce number of articles on this subject, further research is required in order to gain a better understanding of this system's effect on food nutritional and organoleptic quality.

AUTHOR CONTRIBUTIONS

LA-R: conceptualization and writing—the original draft. MA and JR: writing—review and editing. GC: writing—review and editing and supervision. IÁ: conceptualization, writing—review and editing, and supervision. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Smith PG. *Introduction to Food Process Engineering*. New York, NY: Springer. (2011) 467–80. doi: 10.1007/978-1-4419-7662-8_16
2. Zhang ZH, Wang LH, Zeng XA, Han Z, Brennan C. Non-thermal technologies and its current and future application in the food industry: a review. *IJFST*. (2018) 54:1–13. doi: 10.1111/ijfs.13903
3. Song X, Pendenza P, Díaz Navarro M, Valderrama García E, Di Monaco R, Giacalone D. European Consumers' perceptions and attitudes towards non-thermally processed fruit and vegetable products. *Foods*. (2020) 9:E1732. doi: 10.3390/foods91 21732
4. Chemat F, Rombaut N, Meullemiestre A, Turk M, Perino S, Fabiano-Tixier AS, et al. Review of green food processing techniques. Preservation, transformation, and extraction. *Innov Food Sci Emerg Technol*. (2017) 41:357–77. doi: 10.1016/j.ifset.2017.04.016
5. Bermúdez-Aguirre D, Mobbs T, Barbosa-Cánovas GV. Ultrasound applications in food processing. In: Feng H, Barbosa-Cánovas GV, Weiss J, ediotrs. *Ultrasound Technologies for Food and Bioprocessing*. New York, NY: Springer (2011) 65–105. doi: 10.1007/978-1-4419-7472-3_3
6. Gallego-Juárez JA. Basic principles of ultrasound, In: Villamiel M, Montilla A, García-Pérez JV, Cárcel JA, and Benedito J, editors. *Ultrasound in Food Processing*. Chichester: Wiley Blackwell (2017). 1–26.
7. Kentish S, Ashokkumar M. The physical and chemical effects of ultrasound. In: H Feng, GV Barbosa-Cánovas, J Weiss *Ultrasound Technologies for Food and Bioprocessing* New York, NY: Springer (2011). 1-12. doi: 10.1007/978-1-4419-7472-3_1
8. Alarcon-Rojo AD, Carrillo-Lopez LM, Reyes-Villagrana R, Huerta-Jiménez M, Garcia-Galicia IA. Ultrasound and meat quality: a review. *Ultrason Sonochem*. (2019) 55:369–82. doi: 10.1016/j.ultsonch.2018.09.016
9. Charoux C, M. G, O'Donnell CP, Tiwari BK. Ultrasound Processing Food Quality. In: Bermudez-Aguirre D, Ediotors. *Ultrasound Advances in Food Processing Preservation*. London: Elservier Academic Press. (2017). p. 215–236. doi: 10.1016/B978-0-12-804581-7.00009-9
10. Ciudad-Hidalgo S, Astráin-Redín L, Raso J, Cebrián G, Álvarez I. Aplicación de ultrasonidos para la preparación de infusiones de té verde a baja temperatura. Nutrición Clínica en Medicina. (2020). Available online

at: https://congresofesnad2020.com/docs/revista.pdf (accessed December 18, 2020).
11. El Darra N, Grimi N, Maroun R, Louka N, Vorobiev E. Pulsed electric field, ultrasound, and thermal pretreatments for better phenolic extraction during red fermentation. *Eur Food Res Technol.* (2012) 236:47–56. doi: 10.1007/s00217-012-1858-9
12. Han IH, Baik B. Oligosaccharide content and composition of legume and their reduction by soaking, cooking, ultrasound and high hydrostatic pressure. *Cereal Chem.* (2006) 83:428–33. doi: 10.1094/CC-83-0428
13. Condon-Abanto S, Raso J, Arroyo C, Lyng JG, Condón S, Álvarez I. Evaluation of the potential of ultrasound technology combined with mild temperatures to reduce cadmium content of edible crab (*Cancer pagurus*). *Ultrason Sonochem.* (2018) 48:550–4. doi: 10.1016/j.ultsonch.2018.07.019
14. Antunes-Rohling A, Ciudad-Hidalgo S, Mir-Bel J, Raso J, Cebrián G, Álvarez I. Ultrasound as a pretreatment to reduce acrylamide formation in fried potatoes. *IFSET.* (2018) 49:58–169. doi: 10.1016/j.ifset.2018.08.010
15. Charoux C, Ojha S, O'Donnell C, Cardoni A, Brijesh T. Applications of airborne ultrasonic technology in the food industry. *J Food Eng.* (2017) 208:28–36. doi: 10.1016/j.jfoodeng.2017.03.030
16. Zhang M, Xia X, Liu Q, Chen Q, Kong B. Changes in microstructure, quality and water distribution of porcine longissimus muscles subjected to ultrasound- assisted immersion freezing during frozen storage. *Meat Sci.* (2019) 151:24–32. doi: 10.1016/j.meatsci.2019.01.002
17. Li K, Kang ZL, Zou YF, Xu XL, Zhou GH. Effect of ultrasound treatment on functional properties of reduced-salt chicken breast meat batter. *J Food Sci Tech.* (2015) 52:2622–33. doi: 10.1007/s13197-014-1356-0
18. Laurora A, Lund MN, Tiwari BK, Poojary MM. Application of ultrasounds to obtain food additives and nutraceuticals. In: Barba FJ, Cravotto G, Chemat F, Lorenzo Rodriguez JM, Sichetti Munekata PE, editors. *Design and Optimization of Innovative Food Processing Techniques Assisted by Ultrasound*. London: Academic Press (2021). p. 111–42. doi: 10.1016/B978-0-12-818275-8.00012-X
19. Chemat F, Rombaut N, Sicaire AG, Meullemiestre A, Fabiano-Tixier AS, Abert-Vian M. Ultrasound assisted extraction of food and natural products. Mechanisms, techniques, combinations, protocols and applications. A review. *Ultrason Sonochem.* (2017) 34:540–60. doi: 10.1016/j.ultsonch.2016.06.035
20. Goula AM, Ververi M, Adamopoulou A, Kaderides K. Green ultrasound-assisted extraction of carotenoids from pomegranate wastes using vegetable oils. *Ultrason Sonochem.* (2017) 34:821–30. doi: 10.1016/j.ultsonch.2016.07.022
21. Wen C, Zhang J, Zhang H, Dzah CS, Zandile M, Duan Y, et al. Advances in ultrasound assisted extraction of bioactive compounds from cash crops–A review. *Ultrason Sonochem.* (2018) 48:538–49. doi: 10.1016/j.ultsonch.2018.07.018
22. Alexandre EM, Moreira SA, Castro LM, Pintado M, Saraiva JA. Emerging technologies to extract high added value compounds from fruit residues: sub/supercritical, ultrasound-, and enzyme-assisted extractions. *Food Rev Int.* (2018) 34:581–612. doi: 10.1080/87559129.2017.1359842
23. Soquetta MB, Terra LDM, Bastos CP. Green technologies for the extraction of bioactive compounds in fruits and vegetables. *CyTA J Food.* (2018) 16:400–12. doi: 10.1080/19476337.2017.1411978
24. Dzah CS, Duan Y, Zhang H, Wen C, Zhang J, Chen G, et al. The effects of ultrasound assisted extraction on yield, antioxidant, anticancer and antimicrobial activity of polyphenol extracts: a review. *Food Biosci.* (2020) 35:100547. doi: 10.1016/j.fbio.2020.100547
25. Kumar K, Srivastav S, Sharanagat VS. Ultrasound assisted extraction (UAE) of bioactive compounds from fruit and vegetable processing by-products: a review. *Ultrason Sonochem.* (2021) 70:105325. doi: 10.1016/j.ultsonch.2020.105325
26. Bleakley S, Hayes M. Algal proteins: extraction, application, and challenges concerning production. *Foods.* (2017) 6:33. doi: 10.3390/foods6050033
27. Grassino AN, Brnčić M, Vikić-Topić D, Roca S, Dent M, Brnčić SR. Ultrasound assisted extraction and characterization of pectin from tomato waste. *Food Chem.* (2016) 198:93–100. doi: 10.1016/j.foodchem.2015.11.095
28. Encalada AMI, Pérez CD, Calderón PA, Zukowski E, Gerschenson LN, Rojas AM, et al. High-power ultrasound pretreatment for efficient extraction of fractions enriched in pectins and antioxidants from discarded carrots (*Daucus carota* L.). *J Food Eng.* (2019) 256:28–36. doi: 10.1016/j.jfoodeng.2019.03.007
29. Niknam R, Ghanbarzadeh B, Ayaseh A, Rezagholi F. Barhang (*Plantago major* L.) seed gum: Ultrasound-assisted extraction optimization, characterization, and biological activities. *J Food Process Preserv.* (2020) 44:e14750. doi: 10.1111/jfpp.14750
30. Youssouf L, Lallemand L, Giraud P, Soul,é F, Bhaw-Luximon A, Meilhac O, et al. Ultrasound-assisted extraction and structural characterization by NMR of alginates and carrageenans from seaweeds. *Carbohydr Polym.* (2017) 166:55–63. doi: 10.1016/j.carbpol.2017.01.041
31. Flórez-Fernández N, Domínguez H, Torres MD. A green approach for alginate extraction from *Sargassum muticum* brown seaweed using ultrasound-assisted technique. *Int J Biol Macromol.* (2019) 124:451–9. doi: 10.1016/j.ijbiomac.2018.11.232
32. Singh SS, Lim LT, Manickavasagan A. Ultrasound-assisted alkali-urea pre-treatment of Miscanthus× giganteus for enhanced extraction of cellulose fiber. *Carbohydr Polym.* (2020) 247:116758. doi: 10.1016/j.carbpol.2020.116758
33. Damodaran S. Protein stabilization of emulsions and foams. *J Food Sci.* (2005) 70:R54–R66. doi: 10.1111/j.1365-2621.2005.tb07150.x
34. Jain S, Anal AK. Optimization of extraction of functional protein hydrolysates from chicken egg shell membrane (ESM) by ultrasonic assisted extraction (UAE) and enzymatic hydrolysis. *LWT.* (2016) 69:295–302. doi: 10.1016/j.lwt.2016.01.057
35. Lafarga T, Álvarez C, Bobo G, Aguiló-Aguayo I. Characterization of functional properties of proteins from G*anxet* beans (*Phaseolus vulgaris* L. var. Ganxet) isolated using an ultrasound-assisted methodology. *LWT.* (2018) 98:106–12. doi: 10.1016/j.lwt.2018.08.033
36. Chemat F, Huma Z, Khan MK. Applications of ultrasound in food technology: processing, preservation and extraction. *Ultrason Sonochem.* (2011) 18:813–35. doi: 10.1016/j.ultsonch.2010.11.023
37. Gallego-Juárez JA, Riera E, de la Fuente S, Rodríguez G, Acosta V, Blanco A. Application of high-power ultrasound for dehydration of vegetables: processes and devices. *Dry Technol.* (2007) 25:1893–901. doi: 10.1080/07373930701677371
38. Astráin-Redín L, Raso J, Condón S, Cebrián G, Álvarez I. Application of high-power ultrasound in the food industry. In: Karakuş S, ediotrs. *Sonochemical Reactions*. London: IntechOpen. (2019) 103–126. doi: 10.5772/intechopen.90444
39. Dadan M, Nowacka M, Wiktor A, Sobczynska A, Witrowa-Rajchert D. Ultrasound to improve drying processes and prevent thermolabile nutrients degradation. In: Barba FJ, Cravotto G, Chemat F, Lorenzo Rodriguez JM, Sichetti Munekata PE. *Design and Optimization of Innovative Food Processing Techniques Assisted by Ultrasound.* London: Academic Press (2021) 55–110. doi: 10.1016/B978-0-12-818275-8.00010-6
40. Fu X, Belwal T, Cravotto G, Luo Z. Sono-physical and sono-chemical effects of ultrasound: Primary applications in extraction and freezing operations and influence on food components. *Ultrason Sonochem.* (2020) 60:104726. doi: 10.1016/j.ultsonch.2019.104726
41. Ensminger D. Acoustic and electroacoustic methods of dewatering and drying. *Drying Technol Int J.* (1988) 6:473–99. doi: 10.1080/07373938808916394
42. Graff KF. Power ultrasonic transducers: principles and design. In: Gallego-Juárez JA, Graff KF, editors. *Power Ultrasonics. Aplication of High-intensity Ultrasound* Oxford: Woodhead publising (2015). p. 127–58. doi: 10.1016/B978-1-78242-028-6.00006-5
43. Schössler K, Jäger H, Knorr D. Effect of continuous and intermittent ultrasound on drying time and effective diffusivity during convective drying of apple and red bell pepper. *J Food Eng.* (2012) 108:103–10. doi: 10.1016/j.jfoodeng.2011.07.018
44. Bantle M, Eikevik TM. Parametric study of high-intensity ultrasound in the atmospheric freeze drying of peas. *Dry Technol.* (2011) 29:1230–9. doi: 10.1080/07373937.2011.584256
45. Gallego-Juárez JA, Rodríguez-Corral G, Gálvez Moraleda JC, Yang TS. A new high-intensity ultrasonic technology for food dehydration. *Dry Technol.* (1999) 17:597–608. doi: 10.1080/07373939908917555

46. Riera E, Gallego-Juárez JA, Rodríguez Corral G, Acosta Aparicio VM, Andrés Gallego E. Application of high-power ultrasound for drying vegetables. In: *Forum Acusticum*. Sevilla (ULT-05-004-IP) (2002).
47. De la Fuente S, Riera E, Gallego JA, Gómez TE, Acosta VM, Vázquez F. Parametric study of ultrasonic dehydration processes. In: *Proceedings of the 5th World Congress on Ultrasonics*. Paris: WCU (2003). 61–64.
48. Sabarez HT, Gallego-Juárez JA, Riera E. Ultrasonic-assisted convective drying of apple slices. *Dry Technol.* (2012) 30:989–97. doi: 10.1080/07373937.2012.677083
49. Schössler K, Thomas T, Knorr D. Modification of cell structure and mass transfer in potato tissue by contact ultrasound. *Food Res Int*. (2012) 49:425–31. doi: 10.1016/j.foodres.2012.07.027
50. Liu Y, Sun Y, Yu H, Yin Y, Li X, Duan X. Hot air drying of purple-fleshed sweet potato with contact ultrasound assistance. *Dry Technol.* (2017) 35:564–76. doi: 10.1080/07373937.2016.1193867
51. Liu Y, Sun C, Lei Y, Yu H, Xi H, Duan X. Contact ultrasound strengthened far-infrared radiation drying on pear slices: effects on drying characteristics, microstructure, and quality attributes. *Dry Technol.* (2018) 37:745–58. doi: 10.1080/07373937.2018.1458317
52. Liu Y, Zeng Y, Wang Q, Sun C, Xi H. Drying characteristics, microstructure, glass transition temperature, and quality of ultrasound-strengthened hot air drying on pear slices. *J Food Process Pres*. (2019) 43:e13899. doi: 10.1111/jfpp.13899
53. Liu Y, Zeng Y, Hu R, Sun X. Effect of contact ultrasonic power on moisture migration during far-infrared radiation drying of kiwifruit. *J Food Process Eng*. (2019) 42:e13235. doi: 10.1111/jfpe.13235
54. Tao Y, Zhang J, Jiang S, Xu Y, Show PL, Han Y, et al. Contacting ultrasound enhanced hot-air convective drying of garlic slices: mass transfer modeling and quality evaluation. *Int Food Res J.* (2018) 235:79–88. doi: 10.1016/j.jfoodeng.2018.04.028
55. Tao Y, Han M, Gao X, Han Y, Show PL, Liu C, et al. Applications of water blanching, surface contacting ultrasound-assisted air drying, and their combination for dehydration of white cabbage: drying mechanism, bioactive profile, color and rehydration property. *Ultrason Sonochem.* (2019) 53:192–201. doi: 10.1016/j.ultsonch.2019.01.003
56. Islam MN, Zhang M, Fang Z, Sun J. Direct contact ultrasound assisted freezing of mushroom (*Agaricus bisporus*): growth and size distribution of ice crystals. *Int J Refrigeration.* (2015) 57:46–53. doi: 10.1016/j.ijrefrig.2015.04.021
57. Astráin-Redín L, Abad J, Rieder A, Kirkhus B, Raso J, Cebrián G, et al. Direct contact ultrasound assisted freezing of chicken breast samples. *Ultrason Sonochem.* (2021) 70:105319. doi: 10.1016/j.ultsonch.2020.105319
58. Schössler K, Jäger H, Knorr D. Novel contact ultrasound system for the accelerated freeze-drying of vegetables. *Innov Food Sci Emerg Technol*. (2012) 16:113–20. doi: 10.1016/j.ifset.2012.05.010
59. Musielak G, Mierzwa D, Kroehnke J. Food drying enhancement by ultrasound–A review. *Trends Food Sci Tech.* (2016) 56:126–41. doi: 10.1016/j.tifs.2016.08.003
60. Fan K, Zhang M, Mujumdar AS. Application of airborne ultrasound in the convective drying of fruits and vegetables: a review. *Ultrason Sonochem.* (2017) 39:47–57. doi: 10.1016/j.ultsonch.2017.04.001
61. Zhang Y, Abatzoglou N. Fundamentals, applications and potentials of ultrasound-assisted drying. *Chem Eng Res Des.* (2020) 154:21–46. doi: 10.1016/j.cherd.2019.11.025
62. Huang D, Men K, Li D, Wen T, Gong Z, Sunden B, et al. Application of ultrasound technology in the drying of food products. *Ultrason Sonochem.* (2020) 63:104950. doi: 10.1016/j.ultsonch.2019.104950
63. Kroehnke J, Szadzińska J, Stasiak M, Radziejewska-Kubzdela E, Biegańska-Marecik R, Musielak G. Ultrasound-and microwave-assisted convective drying of carrots–Process kinetics and product's quality analysis. *Ultrason Sonochem.* (2018) 48:249–58. doi: 10.1016/j.ultsonch.2018.05.040
64. Kowalski SJ, Pawłowski A, Szadzińska J, Łechtańska J. Stasiak M. High power airborne ultrasound assist in combined drying of raspberries. *Innov Food Sci Emerg Technol*. (2016) 34:225–33. doi: 10.1016/j.ifset.2016.02.006
65. Szadzińska J, Łechtańska J, Kowalski SJ, Stasiak M. The effect of high power airborne ultrasound and microwaves on convective drying effectiveness and quality of green pepper. *Ultrason Sonochem*. (2017) 34:531–9. doi: 10.1016/j.ultsonch.2016.06.030
66. Mbondo NN, Owino WO, Ambuko J, Sila DN. Effect of drying methods on the retention of bioactive compounds in African eggplant. *Food Sci Nutr.* (2018) 6:814–23. doi: 10.1002/fsn3.623
67. do Nascimento EMGC, Mulet A, Ascheri JLR, de Carvalho CWP, Carcel JA. Effect of high-intensity ultrasound on drying kinetics antioxidant properties of passion fruit peel. *J. Food Eng.* (2016) 70:108–18. doi: 10.1016/j.jfoodeng.2015.09.015
68. Santhosha SG, Jamuna P, Prabhavathi SN. Bioactive components of garlic and their physiological role in health maintenance: a review. *Food Biosci.* (2013) 3:59–74. doi: 10.1016/j.fbio.2013.07.001
69. Siucinska K, Mieszczakowska-Frac M, Połubok A, Konopacka D. Effects of ultrasound assistance on dehydration processes and bioactive component retention of osmo-dried sour cherries. *J Food Sci.* (2016) 81:1654–61. doi: 10.1111/1750-3841.13368
70. Chen ZG, Guo XY, Wu T. A novel dehydration technique for carrot slices implementing ultrasound and vacuum drying methods. *Ultrason Sonochem.* (2016) 30:28–34. doi: 10.1016/j.ultsonch.2015.11.026
71. Ortuño C, Pérez-Munuera I, Puig A, Riera E, Garcia-Perez JV. Influence of power ultrasound application on mass transport and microstructure of orange peel during hot air drying. *Phys Procedia.* (2010) 3:153–9. doi: 10.1016/j.phpro.2010.01.022
72. Ratti C. Hot air and freeze-drying of high-value foods: a review. *J Food Eng.* (2001) 49:311–9. doi: 10.1016/S0260-8774(00)00228-4
73. Colucci D, Fissore D, Rossello C, Carcel JA. On the effect of ultrasound-assisted atmospheric freeze-drying on the antioxidant properties of eggplant. *Food Res Int.* (2018) 106:580–8. doi: 10.1016/j.foodres.2018.01.022
74. Arvanitoyannis IS, Kotsanopoulos KV, Savva AG. Use of ultrasounds in the food industry-Methods and effects on quality, safety, and organoleptic characteristics of foods: a review, crit. *Rev Food Sci Nutr.* (2017) 57:109–28. doi: 10.1080/10408398.2013.860514
75. Tao Y, Sun DW. Enhancement of food processes by ultrasound: a review. *Crit Rev Food Sci Nutr.* (2015) 55:570–94. doi: 10.1080/10408398.2012.667849
76. Gaukel V. Cooling freezing of foods. In: Smithers G, editor. *Reference Module in Food Science*. Amsterdam: Elsevier. (2016). doi: 10.1016/B978-0-08-100596-5.03415-6
77. Cheng L, Sun DW, Zhu Z, Zhang Z. Emerging techniques for assisting and accelerating food freezing processes: a review of recent research progresses. *Crit Rev Food Sci Nutr.* (2017) 57:769–78. doi: 10.1080/10408398.2015.1004569
78. Qiu L, Zhang M, Chitrakar B, Bhandari B. Application of power ultrasound in freezing and thawing processes: effect on process efficiency and product quality. *Ultrason Sonochem*. (2020) 68:105230. doi: 10.1016/j.ultsonch.2020.105230
79. Zhang M, Niu H, Chen Q, Xia X, Kone B. Influence of ultrasound-assisted immersion freezing on the freezing rate and quality of porcine longissimus muscles. *Meat Sci*. (2018) 136:1–8. doi: 10.1016/j.meatsci.2017.10.005

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Applications of Non-invasive and Novel Methods of Low-Field Nuclear Magnetic Resonance and Magnetic Resonance Imaging in Aquatic Products

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Aquatic products, such as fish, are popular throughout the world due to their satisfying flavor characteristics as well as rich animal nutrition, and they provide high-value food therapy, but they are easily oxidized and spoiled. It is necessary to detect aquatic products through rapid and accurate technology. Low-field nuclear magnetic resonance (LF-NMR) and magnetic resonance imaging (MRI) have been widely used in the aquatic product industry due to their sensitivity, fast analysis, non-destructive nature and low cost. The applications of LF-NMR in the measurement of aquatic product quality and nutrients (water, fat, and protein) are summarized in this paper. Applications in aquatic products have been shown to depend on deep processing, storage and authentication. This review discusses the application of MRI technology in the quality control of aquatic products. Therefore, this review will guide the application of the aquatic products industry and aims to supply the reader with both the theory of the method and practical applications of the method for use as a rapid and non-destructive technology in scientific research and the industrial industry.

Keywords: low field nuclear magnetic resonance, aquatic products, magnetic resonance imaging, quality, non-destructive testing, real-time monitor

INTRODUCTION

Aquatic products, a relatively high-nutrition and economically valuable food, have become a popular food among consumers and producers around the world (1, 2). Due to their high water content, the flesh of aquatic products spoils easily. Therefore, to ensure the quality and safety of aquatic foods, non-destructive and fast technologies are applied with quality monitoring since they maintain the quality of the traditional character, including texture, taste, and flavor. At present, it is reported that 30% of aquatic products cannot be consumed each year due to spoilage, which accounts for 25% of total agricultural losses (3). Therefore, it is essential to attend to the quality monitoring of aquatic products during production, storage, processing, and transportation.

With the introduction of advanced technologies, low-field nuclear magnetic resonance (LF-NMR) has become a powerful tool that is gradually being applied for the detection of water content, distribution, and migration in aquatic products (4, 5). It is considered to be an accurate and non-destructive method for visualizing the internal food structure (6, 7). Meanwhile, magnetic resonance imaging (MRI) has been widely used in life sciences and food science research (6, 8). Moreover, the transverse relaxation time (T_2)-weighted nuclear magnetic resonance (NMR) signal-obtained relaxation time has been proven to be very informative for water dynamics and its correlation with quality changes, as has been demonstrated using the chemometrics model to predict the shelf life of aquatic products (9–12).

In this paper, our review covers the progress and applications of LF-NMR in aquatic products for the determination of water and fat as well as protein. This review has summarized the latest application progress of LF-NMR in deep processing, storage and authentication. The applications of MRI technology in the non-destructive visualization of aquatic products are introduced. Finally, the potential of using LF-NMR/MRI as a technique for non-destructive testing in aquatic products was shown, which will strongly contribute to applications and developments in the aquatic product industry.

OVERVIEW OF LOW-FIELD NUCLEAR MAGNETIC RESONANCE TECHNOLOGY

NMR spectroscopy probes into the interaction of a nucleus with an applied external magnetic field (13) and was initially used to illustrate the structure of molecules and their chemical properties in the 1970s (14). It can qualitatively and quantitatively analyze the composition and structure of organic and inorganic materials (15). NMR can be used to analyze the behavior of NMR-active nuclei (i.e., ^{1}H and ^{13}C, which are most commonly used for food and processed product applications) (16) in a magnetic field or exposed to pulsed radiofrequency (RF) irradiation (17). Relaxation is the complicated process whereby nuclei transition from an excited state [owing to the splitting of the nuclear spin levels (Zeeman effect) of an applied magnetic field] to equilibrium (18).

According to the strength of the magnetic field, NMR technology is divided into high-field NMR ($\geq$1.0 Tesla, T: unit indicating the magnitude of a magnet), middle-field NMR (0.5–1 T) and low-field NMR ($\leq$0.5 T) (**Figure 1**). High-field NMR technology has the advantages of high sensitivity, high resolution and a high signal-to-noise ratio; however, a significant limitation of the related instruments is that they are expensive and require periodic replenishment of liquid nitrogen (19, 20) (**Figure 2**). The price of high-field NMR is 10 times that of LF-NMR. Compared with high-field NMR technology, LF-NMR not only requires low instrument costs and no special sites for installation but also contains shields inside the instrument and does not need refrigeration. LF-NMR belongs to the submicroscopic field (between molecules) *via* spin-lattice relaxation (i.e., longitudinal relaxation time, T_1) and spin–spin relaxation (i.e., transverse relaxation time, T_2). NMR relaxation measurements of water protons provide a great deal of information about the dynamics of water (21). The RF pulse is assumed to resonate with the hydrogen proton. Some of the low-energy hydrogen proton absorption energy transitions to a high-energy state. After the RF pulse disappears, the hydrogen proton returns in a non-radiative manner to the ground state and reaches a Boltzmann equilibrium; the time required for this process is the relaxation time, and it is used to obtain kinetic information between molecules (22–24).

In 1973, Dr. Paul C. Lauterbur first acquired magnetic resonance images by spatial coding. This established a branch of magnetic resonance imaging (MRI) (25). MRI is a non-destructive and non-invasive detection technology that visually reflects the internal structure of a sample. MRI has significant importance in the real-time monitoring of quality changes in aquatic products during processing, transportation, and storage.

The number of references analyzing foods with LF-NMR and MRI over the last 5 years shows that there has been an increasing number of publications over time, indicating that LF-NMR and MRI have been used more frequently in food research. Furthermore, many publications have proven that LF-NMR and MRI techniques have played a key role in investigating the most abundant chemical components (water, protein, and fat), deep processing, storage, and authentication of aquatic products. The number of scientific works regarding the use of LF-NMR for chemical components increased from 163 papers to 291 during the period 2017–2020. The current paper covered most studies that shed light on the deep processing and storage of aquatic products. The number of published works on deep processing and storage of aquatic products increased from 33 papers to 82 during the period 2017–2020.

APPLICATIONS OF LF-NMR IN AQUATIC PRODUCT NON-DESTRUCTIVE TESTING

Aquatic products are easily oxidized and spoiled. Traditional measurement methods used to analyze aquatic product quality are not only time-consuming ones but also destroy the sample. With the increasing demand for international aquatic products, it is necessary to develop a fast, non-destructive detection technology. According to these technical requirements, LF-NMR was used for the detection of aquatic product quality and opportunities.

Measurement of Water in Aquatic Products

Water is the most abundant chemical component in aquatic products, and water content change is one of the important indicators used to evaluate the quality of aquatic products (26). Based on the state of the water, it can be divided into bound water (tightly bound to macromolecules, such as proteins), trapped water (within the myofibrillar structure), and free water (the water outside the myofibrils) (24) (**Figure 3**). There are a variety of traditional moisture measurement methods, such as the direct drying method, vacuum drying method, distillation method, and Karl Fischer method. Traditional measurement methods are time

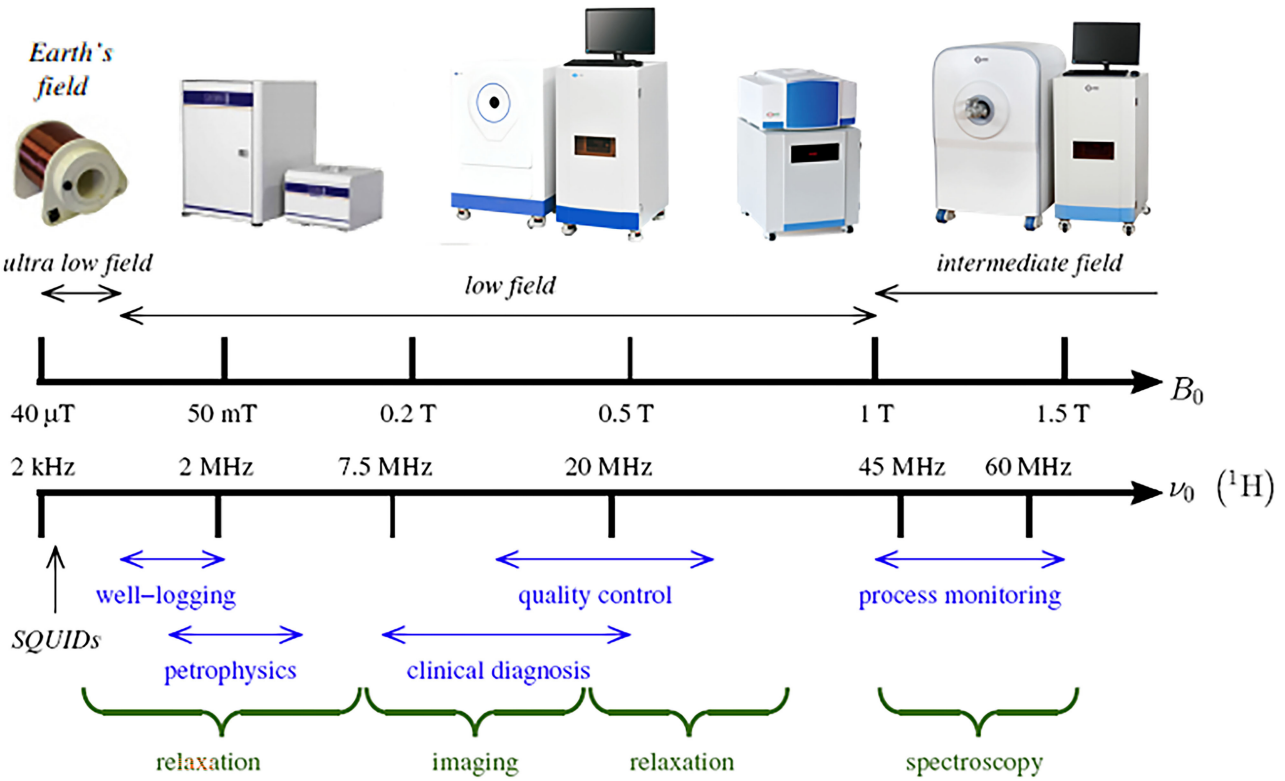

FIGURE 1 | Schematic illustration of the strength of the magnetic field.

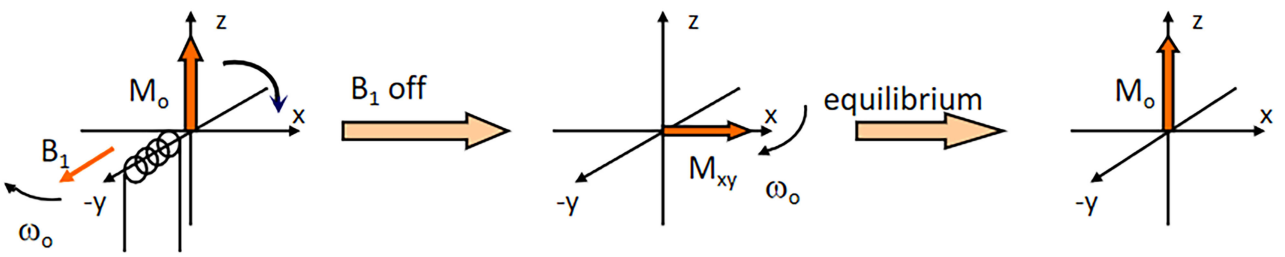

FIGURE 2 | Schematic illustration of the relaxation time principle.

consuming and can only measure the water content, while the state and distribution of water cannot be determined. LF-NMR can be applied to the detection of moisture content, distribution, mobility, and water-binding state in aquatic products. The T_2 relaxation time can be detected by LF-NMR, and water migration and the water content are analyzed according to the changes in T_2 in aquatic products. At present, LF-NMR has been reported in the literature for analyzing water dynamics and its correlations with the water holding capacity, texture, and flavor of fish (10, 27).

The changes in the water state of dried sea cucumber during rehydration, as well as the interaction of water with the surrounding macromolecules, can significantly affect T_1 during LF-NMR analysis (28). The moisture content was determined by T_2 transverse time. The proper presoaking and rehydration times were estimated to be 24 and 96 h, respectively. The rehydration ratio of dried sea cucumber was analyzed by principal component analysis (PCA). There was a good correlation between water content and chewiness. The dry and salt-containing sea cucumbers were clearly distinguished on the PCA score map. LF-NMR can indirectly reflect the texture characteristics of aquatic products, and the results were consistent with the texture parameters of aquatic products. The change in the quality of the wet surface of kelp was recorded as the T_2 relaxation time during storage at room temperature, 4 and −18°C. The amount of trapped water on the wet surface of kelp increased with the extension of storage time. The critical quality

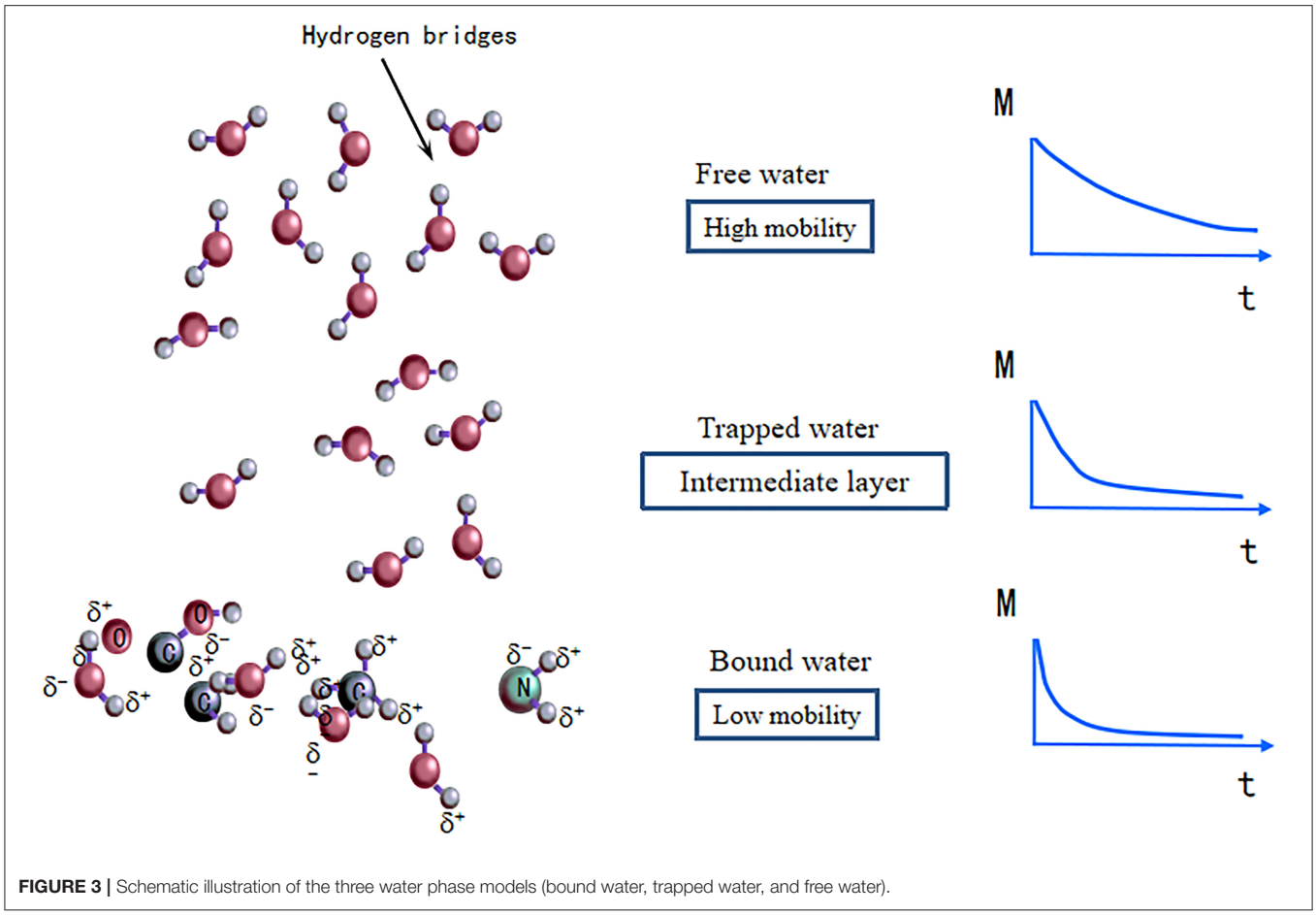

FIGURE 3 | Schematic illustration of the three water phase models (bound water, trapped water, and free water).

control period was between 28 and 35 d at room temperature, and the critical periods of quality control at 4 and −18°C were between 35 and 42 d and between 49 and 56 d (29). The above results indicated that LF-NMR could be used to measure water changes. Water content can be used as a reference standard for predicting the shelf life of aquatic products. Liu et al. (30) used LF-NMR combined with water-holding capacity, Ca^{2+}-ATPase activity and texture analyses, and this combined method provided a great correlation with trapped water (T_{21}), water-holding capacity, free water (T_{22}), and elasticity. The results were consistent with those of Wang et al. (31), who studied the mechanism of water changes by LF-NMR. This result suggested that T_{21} was directly related to the water holding capacity. The elasticity of aquatic products was consistent with the variation trend of free water content, and water changes would affect the flavor of aquatic products.

Measurement of Fat Content in Aquatic Products

Fat is an essential nutrient, and fat content can thus be used to evaluate the quality of aquatic products. There are a variety of traditional methods used for fat content determination, such as the oil weight method, residual method, Soxhlet extraction method, Babcock method, and Gabb's method (32). Traditional measurement methods can be complicated, inaccurate, time-consuming, and labor-intensive. However, samples are qualitatively analyzed by these traditional measurement methods, so corresponding chromatographic peaks must be compared using known data or combined with mass spectrometry/infrared spectroscopy data. A significant limitation of mass spectrometry/infrared spectroscopy methods is their lowered sensitivity for less polar compounds (e.g., hydrocarbons and organometallic compounds). In the quantitative analysis of a sample, standard substances are required to calibrate the output signal before detection, the cost is high, a professional technician is required, and mass spectrometry/infrared spectroscopy is also time-consuming and laborious. The national standard GB/T 31743-2015 allowed for the direct detection of solid fat content by NMR, and the solid fat signal and liquid fat signal observed for the sample were directly determined by NMR and calculated to obtain the solid fat content (32).

At present, LF-NMR has been applied as a technique to rapidly and non-destructively detect fat content. LF-NMR combined with stoichiometry could be used to analyze the fat content of aquatic products, which could be characterized by T_1-weighted imaging and the T_2 relaxation time (33, 34). The fat content of salmon was quantitatively analyzed by pulsed nuclear magnetic resonance relaxation signals (range of 90–182 g kg^{-1}), and this

technique was combined with the novel software "Norwegian mass cutting (NQC)" to obtain a "fat image." Therefore, the NMR spectrometer monitored the fat changes online and can thus be expected to be an increasingly popular research tool (35). Moreover, LF-NMR is more convenient and accurate than traditional Soxhlet extraction (28, 36). The relationships between water migration and the fat content of aquatic products were investigated with were investigated. Fat content was visually reflected by changes in T_1-weighted imaging, and CPMG echo peaks and fat prediction models, including principal component regression (PCR) and partial least squares regression (PLSR), have been rapidly established. LF-NMR, which is combined with chemometric methods, has been used for the quantitative analysis and quality control of fat content in aquatic products.

Measurement of Protein in Aquatic Products

Protein is the main component of aquatic products. Three kinds of proteins from aquatic products are myofibrillar protein, myogen, and matrix protein. The quality changes of aquatic products are mainly caused by the degradation of the myosin heavy chain, α-actin, actin, and tropomyosin. The combination of physical, chemical, and microbiological reactions resulted in a decrease in the freshness of aquatic products (37–40). There are a variety of traditional methods used for analyzing proteins, such as SDS-polyacrylamide gel electrophoresis, mass spectrometry, and near-infrared spectroscopy (27, 41–43). These techniques are complicated, time-consuming and laborious. Many factors that influence protein changes and are of significant importance are protein oxidization and degradation. At present, the T_2 transverse relaxation time determined by LF-NMR can reflect the moisture distribution and migration in the intramyofibrillar space and extramyofibrillar water population. Water molecules are capable of interacting with surrounding surface proteins, leading to relaxation decay rate changes and relaxation declines (44). Meanwhile, the changes in myofibrillar proteins were analyzed from the perspective of water migration and combined with traditional methods used to detect proteins, enzyme activities, texture, and other indicators to evaluate the quality of the aquatic product (27).

Based on the indicators of emulsifying activity, WHC and LF-NMR, the differences in the fatty acid compositions and the acylglycerol structures of the lipid phase significantly affected the emulsifying capacity of the myofibrillar proteins (45). The results above indicated that LF-NMR could be used to explore the mechanism of protein changes. Ozel et al. (45) applied the T_2 transverse relaxation time to investigate the effects of different polysaccharides on the swelling ratios of whey protein hydrogel composites. LF-NMR is capable of monitoring the hydrogel swelling structure. In addition, LF-NMR provided more information on swelling mechanisms than conventional methods, establishing its potential for further investigation. Greiff et al. (46) used LF-NMR to study the effects of different concentrations of salt additives on the protein structure of carp surimi. As the concentration increased, T_{21} and T_{22} gradually increased, and the cooking loss rate and WHC decreased because the interaction between water molecules and proteins in the muscle fibers was suppressed. The LF-NMR results showed that the difference in protein structures was mainly related to the salt additive concentration.

APPLICATIONS OF LF-NMR IN DEEP PROCESSING, STORAGE AND AUTHENTICATION OF AQUATIC PRODUCTS

Note that the signal obtained through LF-NMR studies on the water dynamics of aquatic products comes directly from the water signal of the aquatic product samples. Moreover, the T_2 transverse relaxation time has been used by many researchers to investigate the water content and distribution in traditional and modern processing, including deep processing, storage, and authentication. The water content and distribution were monitored in real time for aquatic products. LF-NMR can accurately measure water content changes and migration and determine the types of aquatic products and their byproducts. Therefore, it has been gradually applied to the aquatic products industry.

Applications of LF-NMR in the Preservation of Aquatic Products

Gudjónsdóttir et al. (47) used LF-NMR to study the effects of different fresh-keeping methods (without polyphosphate and brine pickling) on the moisture and salinity of Atlantic salmon filets. The study showed that T_{21} increased as the water content of fish filets increased. T_{21} has a significant correlation with the water content, salt content and water holding capacity. Moreover, similar WHC results were related to water located outside the myofibrillar network (extramyofibrillar) (48, 49). Sánchezalonso et al. (50) used LF-NMR to study the effects of different freezing methods (air blast, liquid nitrogen, and walk-in freezer methods) on the quality of squid slices. The T_2 transverse relaxation time effectively reflects the water distribution and migration of squid slices in freezer storage due to the wide band of T_{21} within the range 120–360 ms. Moreover, the results showing that these T_2 changes exist may indicate quality changes because it has been recorded that the freezing rate was optimized and that the temperature used for quality parameters was controlled. Idag et al. (51) studied salt uptake in Atlantic salmon filets and showered that salt uptake was affected by antemortem stress and rigor mortis. The T_2 transverse relaxation time was used to show that salt diffusion and distribution strongly depended on the fat distribution during the curing process. Da Silva et al. (7, 48) and Ghidini et al. (49) studied the quality changes of aquatic products preserved by different methods (pickling and adding sodium polyphosphate, polyphosphates, or sulphites). They found that the internal aquatic product tissue of proteins degraded, denatured, and aggregated during storage, and the value of trapped water and free water decreased, which was attributed to chemical conversion between water and protein protons. It is well-documented that the T_{21} and T_{22} of frozen shrimp significantly increase with increasing

additive concentrations compared with those of the CK group. This further demonstrates the importance of LF-NMR, which can significantly influence the quality of frozen shrimp during preservation. Reseachers (47, 52) studied the effects of injecting additives (salt and protein) and different preservation methods (salt content and modified atmosphere packaging) to evaluate the quality of fish filets during the freezing process and revealed that the infusion of various salts into the fish and the protein distribution were more uniform than those in the untreated group. The addition of protein powder enhanced muscle protein electrostatic force, resulting in increased water amplitude (A_{2b}) in myofibrils. Comparing the other processing conditions, the modified atmosphere in the packaging caused the T_{22} migration rate of fish to slow as well, indicating that the modified atmosphere in the packaging had an increasingly protective effect on fish meat losses.

Applications of LF-NMR in the Heating Process of Aquatic Products

Wang et al. (8) used LF-NMR to reveal that clams treated at 80°C in the water state significantly changed. T_1 and T_2 relaxation times are highly correlated with water dynamics in clams during the heating process. Bi et al. (53) used LF-NMR and observed that only one water population was present in sea cucumber (*Stichopus japonicus*) preheated at 40°C for 120 min, and the water did not change dramatically during the heating process. For sea cucumber postheated at 80°C, three distinct populations were shown, and the T_2 relaxation time of the bulk water decreased dramatically, indicating some changes in internal structures and loss in WHC. A good correlation between the T_2 relaxation time and TPA analysis parameters was shown for sea cucumber after both preheating and postheating. Therefore, LF-NMR is presented as a new means of assessing quality and understanding structural changes of seafood during the heating process.

Applications of LF-NMR in the Low-Temperature Storage of Aquatic Products

Wang et al. (10) used LF-NMR to monitor the water mobility of bigeye tuna during low-temperature storage and combined it with texture profile, quality indicator, microorganism and T_2 relaxation time analyses to establish multiple linear regression equations and predict shelf life. The results showed that LF-NMR could be used to dynamically monitor the water migration of fish during cold storage. Shu-Min et al. (54) studied the water change mechanism of vacuum-packed cucumber juice fish balls during cold storage. The results showed that T_{23} showed a decreasing trend, and free water dynamically changed with extended storage time. Sánchez-Alonso et al. (50) analyzed the quality changes in hake stored at −10°C for 6 months. As the storage time increased, T_{22} and T_{21} decreased, the WHC and viscosity apparently decreased, and the shear force increased, reflecting the juice losses of hake. The relaxation time (T_{21}, T_{22}), amplitude (A_{21}, A_{22}), and related quality indicators were used to establish a PLS mathematical model to predict the shelf life. Therefore, LF-NMR can be used as an important detection method for evaluating the quality change of aquatic products during low-temperature storage.

Applications of LF-NMR in the Dry Storage of Aquatic Products

Water dynamics of abalone (*Haliotis discus hannai Ino*) were assessed using LF-NMR and MRI in which dried abalones were rehydrated for 120 h. There was a good correlation between the hardness, chewiness, rehydration ratio, and T_{22} relaxation time of dried abalone (4). Cheng et al. (55, 56) studied shrimp meat (*Penaeus vannamei*) and *Pacific oyster* (*Crassostrea gigas*) as the research objects and analyzed the drying process by performing LF-NMR. T_{21} decreased, indicating that the water mobility and water freedom of the shrimp and oyster decreased gradually during the drying process. This migration of water from the extramyofibrillar space into the intramyofibrillar space indicates that this shrinkage of myofibrils significantly influences water mobility by decreasing the space available to keep the water due to the drying process, which is implied by the relaxation times of the different water populations. The difference in texture and color and its correlation with water dynamics were evaluated by LF-NMR. In conclusion, the effect of the drying method on the quality of aquatic products was analyzed from multiple angles.

Applications of LF-NMR in the High-Pressure Treatment of Aquatic Products

Shang et al. (57) used LF-NMR to study pressures of 300, 500, and 600 MPa, causing the relaxation time to increase in sea bass skeletal muscle and making the dynamics of bound water remarkable. However, the pressure of 100 MPa had little effect on the dynamics of bound water, and 200 MPa could cause the relaxation time of bound water to reach a minimum and the bound water to become more stable. High pressure could retain water in different states; thus, the gel-forming capacity and water holding capacity of aquatic products would change. Similar results were obtained in both fresh and smoked salmon samples treated at 100 and 150 MPa in which the T_2 values of the 150 MPa-treated samples were different to those of the samples treated at the other pressure levels used. High pressure might lead to a slight decrease in dryness, hardness, color, and appearance in fish and affect T_2 relaxation time, in terms of both fish and relaxation time changes, which could be caused by changes in the structure of the fish proteins. We concluded with LF-NMR that high pressure contributes to some changes related to the texture, WHC and T_2 relaxation time of fish due to protein denaturation (51). Combined with physicochemical indicator changes and organizational structures, it was observed that high pressure could extend the shelf life of aquatic products.

Applications of LF-NMR in the Ultrasound Treatment of Aquatic Products

Zhang et al. (58) studied the effect of ultrasonic treatment on the rehydration capacity. As the ultrasonic power increased, T_{22} and T_{23} (A_{22} and A_{23}) increased, revealing that the ultrasonic treatment of sea cucumber could lead it to absorb more free

water during the rehydration process. The LF-NMR results demonstrated that ultrasonic technology was beneficial to water absorption in aquatic products. Additionally, a study using LF-NMR revealed that ultrasound-assisted immersion freezing reduced the mobility and loss of immobilized and free water in common carp (*Cyprinus carpio*) (59). LF-NMR is an effective way to evaluate the deterioration of fish during ultrasound storage.

Applications of LF-NMR in the Authentication of Aquatic Products

Liu et al. (60) used LF-NMR to identify fish gills, miscellaneous fish gills, red snapper, and copper pot fish gills. T_2 was combined with the traditional drying method to determine the moisture content, state, and distribution of surimi, providing a new method for the rapid and non-destructive identification of aquatic products. In addition, T_2 relaxation time measurements are capable of measuring and mapping prawns injected with different hydrocolloids, such as gelatine, carrageenan, agar, Amophophallus konjac, and xanthan gum. In addition, LF-NMR could be used to study the different states of water in the muscle (head, tail, paw, and back), which could be used to authenticate adulterated prawns (61). Geng et al. (28) and Hassoun et al. (62) used LF-NMR combined with PCA/PLSR to identify dried sea cucumber/salt-dried sea cucumber and adulterated shrimp, and the PCA/PLSR score map can clearly distinguish between the different kinds of sea cucumber and adulterated shrimp. The correlations between NMR parameters, rehydration rate and texture characteristics of dried sea cucumber/adulterated shrimp were analyzed by the linear regression mathematical method. Marciani et al. (63) applied ^{1}H NMR to perform data processing and distinguish between wild and farmed salmon, as well as determine their origins, with a high accuracy based on their tissue lipid species and profiles. These studies suggested that they contained complementary functions that would improve the authentication of aquatic products according to their geographical areas and kinds, as well as treatment methods.

MRI Used for Visual Observation in Aquatic Product Storage and Processing

MRI non-destructively provides a great deal of information on internal molecule distribution for use in development research and estimating the quality of aquatic food (52, 64). X-ray diffraction, scanning electron microscopy, transmission electron microscopy, optical microscopy, and atomic force microscopy have been frequently used in the visual analysis of aquatic products (53). However, these methods cause significant damage to the molecules detected. MRI technology can be used to non-destructively see the sample through sliced images of aquatic products, improving the processing conditions and quality by real-time dynamic information (65).

Geng et al. (28) used MRI and LF-NMR to study the water dynamic changes in the presoaking and rehydration process of dried sea cucumber. MRI imaging results showed that the free water content was more than the trapped water content of the dried sea cucumber in the rehydration process. The rehydrated sea cucumber mainly contained free water. The stoichiometric method combined with MRI image information was used to analyze the moisture content of the aquatic product and predict the shelf life. Zhang et al. (58) found that different powerful ultrasonic methods affected the internal water distribution during the rehydration of sea cucumber. The study showed that the higher the ultrasonic power was, the stronger the signal of hydrogen protons in the rehydration of sea cucumber, the greater the moisture content inside the sea cucumber and the stronger the ability of sea cucumber to rehydrate. This was combined with SEM and pseudocolour images for comparative analysis and the more comprehensive monitoring of aquatic product quality changes. MRI was used to analyze the water distribution and content changes of fish during low-temperature storage (8, 10). The water migration of tuna could be observed visually. Physical and chemical indicators were used to analyze the quality changes and monitor tuna dynamically. Bian et al. (66) used MRI greyscale images and pseudocolour maps to find changes in the water-oil balance of dried saury samples, which was consistent with the sensory evaluation results. At the same time, MRI was combined with sensory scores, microbial indicators and physical-chemical indicators, and the saury's shelf life was predicted. Relying on MRI results is one of the best tools for predicting the shelf life of aquatic products. Wu et al. (67) used MRI to analyze the distribution and content of fish fat tissue under different feeding habits and aquaculture conditions. Therefore, MRI can realize the real-time online, non-destructive and non-invasive detection of moisture mobility in aquatic products to better control the quality of aquatic products.

CONCLUSION AND FUTURE RESEARCH

LF-NMR and MRI are useful for a wide range of applications related to food property and food authentication. While there are many methods that can be used to measure food components, LF-NMR/MRI technology can not only measure the moisture, protein and fat content in aquatic products but also detect quality changes in real time and in a rapid, non-destructive and accurate manner. Examples of applications include using LF-NMR/MRI as a substitute for the conventional method to analyze aquatic products faster; it can distinguish between complex aquatic product types; it can detect water content and monitor water migration; and it can evaluate aquatic product quality to reveal the mechanism of aquatic product spoilage. In addition, nuclear magnetic data can be combined with mathematics and stoichiometry to establish relevant quality models, providing new ideas for evaluating the quality of aquatic products. However, applications of LF-NMR are still relatively scarce, and the applications of combining LF-NMR with related testing instruments are even scarcer due to technical and testing limitations. Nevertheless, LF-NMR/MRI techniques are still applied in aquatic product research and are good candidates for assessing quality control during industrial processes. This

future direction has rapidly promoted the applications of LF-NMR, combined with other related instruments, in the real-time monitoring of aquatic products, thereby improving the efficiency of applying non-destructive testing to aquatic products and introducing LF-NMR technology into the corresponding standard system to promote the development of the aquatic product industry.

AUTHOR CONTRIBUTIONS

X-YW analyzed the data, wrote the manuscript, and performed the experiments. JX and X-JC made suggestions for revisions and guided the experiments. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Li J, Lu H, Zhu J, Wang Y, Li X. Aquatic products processing industry in China: challenges and outlook. *Trends Food Sci Technol*. (2009) 20:73–7. doi: 10.1016/j.tifs.2008.09.008
2. Li C, Zeng H, Zhang J, Luo D, Chen M, Lei T, et al. *Cronobacter* spp. isolated from aquatic products in China: incidence, antibiotic resistance, molecular characteristic and CRISPR diversity. *Int J Food Microbiol*. (2020) 335:108857. doi: 10.1016/j.ijfoodmicro.2020.108857
3. Zhang Y. *China's Aquatic Products Supply and Demand Situation over the Past Decade. Proceedings of 2013 World Agricultural Outlook Conference*. Heidelberg: Springer (2014). p. 89–99.
4. Wang H, Wang R, Song Y, Kamal T, Lv Y, Zhu B, et al. A fast and non-destructive LF-NMR and MRI method to discriminate adulterated shrimp. *J Food Meas Charact*. (2018) 12:1340–9. doi: 10.1007/s11694-018-9748-x
5. Yu D, Feng T, Jiang Q, Yang F, Gao P, Xu Y, et al. The change characteristics in moisture distribution, physical properties and protein denaturation of slightly salted silver carp (*Hypophthalmichthys molitrix*) fillets during cold/hot air drying processing. *LWT Food Sci Technol*. (2021) 137:110466. doi: 10.1016/j.lwt.2020.110466
6. Marcone MF, Wang S, Albabish W, Nie S, Somnarain D, Hill A. Diverse food-based applications of nuclear magnetic resonance (NMR) technology. *Food Res Int*. (2013) 51:729–47. doi: 10.1016/j.foodres.2012.12.046
7. da Silva Carneiro C, Mársic ET, Ribeiro RDOR, Conte-Júnior CA, Mano SB, Augusto CJC, et al. Low-Field Nuclear Magnetic Resonance (LF NMR 1H) to assess the mobility of water during storage of salted fish (*Sardinella brasiliensis*). *J Food Eng*. (2016) 169:321–5. doi: 10.1016/j.jfoodeng.2015.09.010
8. Wang S, Lin R, Cheng S, Tan M. Water dynamics changes and protein denaturation in surf clam evaluated by two-dimensional LF-NMR T1-T2 relaxation technique during heating process. *Food Chem*. (2020) 320:126622. doi: 10.1016/j.foodchem.2020.126622
9. Wang S, Xiang W, Fan H, Xie J, Qian YF. Study on the mobility of water and its correlation with the spoilage process of salmon (*Salmo solar*) stored at 0 and 4 degrees C by low-field nuclear magnetic resonance (LF NMR(1)H). *J Food Sci and Tech*. (2018) 55:173–82. doi: 10.1007/s13197-017-2880-5
10. Wang X, Geng L, Xie J, Qian Y-F. Relationship between water migration and quality changes of yellowfin tuna (*Thunnus albacares*) during storage at 0°C and 4°C by LF-NMR. *J Aquat Food Prod Techol*. (2017) 27:35–47. doi: 10.1080/10498850.2017.1400630
11. Pearce KL, Rosenvold K, Andersen HJ, Hopkins DL. Water distribution and mobility in meat during the conversion of muscle to meat and ageing and the impacts on fresh meat quality attributes-a review. *Meat Sci*. (2011) 89:111–24. doi: 10.1016/j.meatsci.2011.04.007
12. Brown RJ, Capozzi F, Cavani C, Cremonini MA, Petracci M, Placucci G. Relationships between (1)H NMR relaxation data and some technological parameters of meat: a chemometric approach. *J Magn Reson*. (2000) 147:89–94. doi: 10.1006/jmre.2000.2163
13. Winkler M, Mccarthy MJ, German JBJJoFS. Noninvasive measurement of lipid and water in food using magnetic resonance imaging. *J Food Sci*. (1991) 56:811–5. doi: 10.1111/j.1365-2621.1991.tb05388.x
14. Massi F, Johnson E, Wang C, Rance M. NMR R1 rho rotating-frame relaxation with weak radio frequency fields. *J Am Chem Soc*. (2004) 126:2247–56. doi: 10.1021/ja038721w
15. Hills BP, Takacs SF, Belton PS. A new interpretation of proton NMR relaxation time measurements of water in food. *Food Chem*. (1990) 37:95–111. doi: 10.1016/0308-8146(90)90084-H
16. Hatzakis E. Nuclear magnetic resonance (NMR) spectroscopy in food science: a comprehensive review. *Compre Rev Food Sci F*. (2019) 18:189–220. doi: 10.1111/1541-4337.12408
17. van Beek TA. Low-field benchtop NMR spectroscopy: status and prospects in natural product analysis. *Phytochem Anal*. (2020) 32:24–37. doi: 10.1002/pca.2921
18. Cornillon P, Salim LC. Characterization of water mobility and distribution in low-and intermediate-moisture food systems. *Magnetic Resonance Imaging*. (2000) 18:335–41. doi: 10.1016/S0730-725X(99)00139-3
19. Granizo DP, Reuhs BL, Stroshine R, Mauer LJ. Evaluating the solubility of powdered food ingredients using dynamic nuclear magnetic resonance (NMR) relaxometry. *LWT Food Sci Technol*. (2007) 40:36–42. doi: 10.1016/j.lwt.2005.08.012
20. Chung MS, Ruan R, Chen P, Kim JH, Ahn TH, Baik CK. Predicting caking behaviors in powdered foods using a low-field nuclear magnetic resonance (NMR) technique. *LWT Food Sci Technol*. (2003) 36:751–61. doi: 10.1016/S0023-6438(03)00096-3
21. Renou JP, Foucat L, Bonny JM. Magnetic resonance imaging studies of water interactions in meat. *Food Chem*. (2003) 82:35–9. doi: 10.1016/S0308-8146(02)00582-4
22. Kirtil E, Oztop MH. 1H nuclear magnetic resonance relaxometry and magnetic resonance imaging and applications in food science and processing. *Food Eng Rev*. (2015) 8:1–22. doi: 10.1007/s12393-015-9118-y
23. Hamilton G, Middleton MS, Hooker JC, Haufe WM, Forbang NI, Allison MA, et al. *In vivo* breath-hold (1) H MRS simultaneous estimation of liver proton density fat fraction, and T1 and T2 of water and fat, with a multi-TR, multi-TE sequence. *J Magn Reson Imaging*. (2015) 42:1538–43. doi: 10.1002/jmri.24946
24. Chinachoti P, Steinberg MP. Correlation of proton T1 with polymer and solute waters in starch-sucrose mixtures. *J Food Sci*. (1989) 54:691–4. doi: 10.1111/j.1365-2621.1989.tb04683.x
25. Koizumi M, Naito S, Ishida N, Haishi T, Kano H. A dedicated MRI for food science and agriculture. *Food Sci Technol Res*. (2008) 14:74–82. doi: 10.3136/fstr.14.74
26. Wang X-Y, Xie J, Qian Y-F. A non-invasive method for quantitative monitoring of quality changes and water migration in bigeye tuna (*Thunnus obesus*) during simulated cold chain logistics using low-field nuclear magnetic resonance coupled with PCA. *Food Sci Technol Int*. (2020) 26:475–84. doi: 10.1177/1082013220903148
27. Wang X-Y, Xie J. Evaluation of water dynamics and protein changes in bigeye tuna (*Thunnus obesus*) during cold storage. *LWT Food Sci Technol*. (2019) 108:289–96. doi: 10.1016/j.lwt.2019.03.076
28. Geng S, Wang H, Wang X, Ma X, Xiao S, Wang J, et al. A non-invasive NMR and MRI method to analyze the rehydration of dried sea cucumber. *Anal Methods*. (2015) 7:2413–9. doi: 10.1039/C4AY03007A

29. Yan L, Lin X, Jia W, Dan L, Ruan R. Monitoring the quality of kelp fresh noodles during the storage using the low-field NMR. *J Chin Inst Food Sci Technol*. (2015) 15:254–60. doi: 10.16429/j.1009-7848.2015.05.035
30. Liu CY, Mei-Hu MA, Wang QL, Jin GF, Jin YG. New advances of application of nuclear magnetic resonance technology in food lipid research. *Sci Technol Food Indus*. (2017) 38:342–6. doi: 10.13386/j.issn1002-0306.2017.12.064
31. Wang JH, Liu DM, Liu YL. Factors influencing lipid degradation of grass carp muscle during cold storage. *Food Sci*. (2013) 34:276–9. doi: 10.7506/spkx1002-6630-201318057
32. Xia L, Jiang, Lu, Li, Wen, Li. Changes of pork water holding capacity processed by different frozen methods during storage. *Food Machinery*. (2013) 29:154–8. doi: 10.3969/j.issn.1003-5788.2013.02.037
33. Veliyulin E, van der Zwaag C, Burk W, Erikson U. *In vivo* determination of fat content in Atlantic salmon (*Salmo salar*) with a mobile NMR spectrometer. *J Sci Food Agr*. (2005) 85:1299–304. doi: 10.1002/jsfa.2117
34. Gao XS, Yan-Ru LI, Guo WH. Prediction models established of moisture and fat content by low field NMR combined with chemometrics in abalone. *Modern Agric Sci Technol*. (2016) 08:260–74.
35. Jing X, Lan W. Research progress of aquatic products preservation technology during circulation. *J Chin Inst Food Sci Technol*. (2017) 17:1–8. doi: 10.16429/j.1009-7848.2017.07.001
36. Gao ZL, Xie J. Research progress on low-temperature preservation of aquatic products. *Guangdong Agric Sci*. (2012) 39:98–101. doi: 10.1007/s11783-011-0280-z
37. Magro SL, Summa S, Iammarino M, D'Antini P, Marchesani G, Chiaravalle AE, et al. A 5-years (2015–2019) control activity of an EU Laboratory: contamination of histamine in fish products and exposure assessment. *Appl Sci*. (2020) 10:8693. doi: 10.3390/app10238693
38. Peivasteh-Roudsari L, Rahmani A, Shariatifar N, Tajdar-Oranj B, Mazaheri M, Sadighara P, et al. Occurrence of histamine in canned fish samples (Tuna, Sardine, Kilka, and Mackerel) from markets in Tehran. *J Food Pro*. (2019) 83:136–41. doi: 10.4315/0362-028X.JFP-19-288
39. Ironside JIM, Love RMJN. Measurement of denaturation of fish protein. *Nature*. (1956) 178:418–9. doi: 10.1038/178418a0
40. Zhang X, Zhu F, Yong H. Fast measurement of crude protein content in fish feed based on visible and near infrared spectroscopy. In: *2010 World Automation Congress*. IEEE (2010). p. 461—4.
41. Kristinsson HG, Rasco BA. Fish protein hydrolysates: production, biochemical, and functional properties. *Crit Rev Food Sci Nutr*. (2000) 40:43–81. doi: 10.1080/10408690091189266
42. Beauvallet C, Renou J-P. Applications of NMR spectroscopy in meat research. *Trends Food Sci Tech*. (1992) 3:241–6. doi: 10.1016/S0924-2244(10)80001-9
43. Zhang K, Guan ZQ, Min LI, Hong PZ, Yang-Yang WU. Assessment of the effect of trehalose on frozen tilapia fillets by low field nuclear magnetic resonance (LF-NMR) and conventional physicochemical analytical methods. *Food Fermentation Indus*. (2015) 41:196–201.
44. Miklos R, Cheong L-Z, Xu X, Lametsch R, Larsen FH. Water and fat mobility in myofibrillar protein gels explored by low-field NMR. *Food Biophys*. (2015) 10:316–23. doi: 10.1007/s11483-015-9392-5
45. Ozel B, Uguz SS, Kilercioglu M, Grunin L, Oztop MH. Effect of different polysaccharides on swelling of composite whey protein hydrogels: a low field (LF) NMR relaxometry study. *J Food Process Eng*. (2017) 40:e12465. doi: 10.1111/jfpe.12465
46. Greiff K, Fuentes A, Aursand IG, Erikson U, Masot R, Alcañiz M, et al. Innovative nondestructive measurements of water activity and the content of salts in low-salt hake minces. *J Agric Food Chem*. (2014) 62:2496–505. doi: 10.1021/jf405527t
47. Gudjónsdóttir M, Arason S, Rustad T. The effects of pre-salting methods on water distribution and protein denaturation of dry salted and rehydrated cod–a lowfield NMR study. *J Food Eng*. (2011) 104:23–9. doi: 10.1016/j.jfoodeng.2010.11.022
48. da Silva Carneiro C, Mársico ET, Ribeiro RDOR, Júnior CAC, Álvares TS, de Jesus EFO. Studies of the effect of sodium tripolyphosphate on frozen shrimp by physicochemical analytical methods and low field nuclear magnetic resonance (LF 1H NMR). *LWT Food Sci Technol*. (2013). 50:401–7. doi: 10.1016/j.lwt.2012.09.009
49. Ghidini S, Varra MO, Zanardi E. Approaching authenticity issues in fish and seafood products by qualitative spectroscopy and chemometrics. *Molecules*. (2019) 24:1812. doi: 10.3390/molecules24091812
50. Sánchezalonso I, Moreno P, Careche M. Low field nuclear magnetic resonance (LF-NMR) relaxometry in hake (*Merluccius merluccius*, L.) muscle after different freezing and storage conditions. *Food Chem*. (2014) 153:250–7. doi: 10.1016/j.foodchem.2013.12.060
51. Idag A, Ulf E, Emil V. Water properties and salt uptake in Atlantic salmon fillets as affected by ante-mortem stress, rigor mortis, and brine salting: a low-field ^{1}H NMR and ^{1}H/^{23}Na MRI study. *Food Chem*. (2010). 120:482–9. doi: 10.1016/j.foodchem.2009.10.041
52. Li N, Shen Y, Liu W, Mei J, Xie J. Low-field NMR and MRI to analyze the effect of edible coating incorporated with map on qualities of half-smooth tongue sole (*Cynoglossus semilaevis Günther*) fillets during refrigerated storage. *Appl Sci*. (2018) 8:1391. doi: 10.3390/app8081391
53. Bi J, Li Y, Cheng S, Dong X, Kamal T, Zhou D, et al. Changes in body wall of sea cucumber (Stichopus japonicus) during a two-step heating process assessed by rheology, LF-NMR, and texture profile analysis. *Food Biophys*. (2016) 11:257–65. doi: 10.1007/s11483-016-9437-4
54. Shu-Min YI, Wang XQ, Jian-Rong LI, Ying-Chang LI, Zhu JL, Xiao-Feng YU, et al. Quality changes of cucumber juice fish balls during refrigerating storage. *Sci Technol.Food Ind*. (2013) 34:280–4. doi: 10.13386/j.issn1002-0306.2013.22.081
55. Cheng S, Tang Y, Zhang T, Song Y, Wang X, Wang H, et al. Approach for monitoring the dynamic states of water in shrimp during drying process with LF-NMR and MRI. *Drying Technol*. (2018) 36:841–8. doi: 10.1080/07373937.2017.1357569
56. Cheng S, Zhang T, Yao L, Wang X, Song Y, Wang H, et al. Use of low-field-NMR and MRI to characterize water mobility and distribution in pacific oyster (*Crassostrea gigas*) during drying process. *Drying Technol*. (2018) 36:630–6. doi: 10.1080/07373937.2017.1359839
57. Shang X, Liu A, Zheng J, Wang P, Yin S. High pressure processing and water holding capacity of sea bass skeletal muscle. *J Aquat Food Prod Technol*. (2015) 24:740–51. doi: 10.1080/10498850.2013.809831
58. Zhang L, Huang X, Miao S, Zeng S, Zhang Y, Zheng B. Influence of ultrasound on the rehydration of dried sea cucumber (*Stichopus japonicus*). *J Food Eng*. (2016) 178:203–11. doi: 10.1016/j.jfoodeng.2016.01.024
59. Sun Q, Sun F, Xia X, Xu H, Kong B. The comparison of ultrasound-assisted immersion freezing, air freezing and immersion freezing on the muscle quality and physicochemical properties of common carp (*Cyprinus carpio*) during freezing storage. *Ultrason Sonochem*. (2019) 51:281–91. doi: 10.1016/j.ultsonch.2018.10.006
60. Liu, Yuan, Wang, Xichang, Xu, Changhua, et al. Accelerated chemotaxonomic discrimination of marine fish surimi based on Tri-step FT-IR spectroscopy and electronic sensory. *Food Control*. (2017) 73:1124–33. doi: 10.1016/j.foodcont.2016.10.030
61. Li M, Li B, Zhang W. Rapid and non-invasive detection and imaging of the hydrocolloid-injected prawns with low-field NMR and MRI. *Food Chem*. (2018) 242:16–21. doi: 10.1016/j.foodchem.2017.08.086
62. Hassoun A, Mge I, Schmidt WF, Temiz HT, Li L, Kim HY, et al. Fraud in animal origin food products: advances in emerging spectroscopic detection methods over the past five years. *Foods*. (2020) 9:1069. doi: 10.3390/foods9081069
63. Marciani L, Wickham M, Singh G, Bush D, Spiller RC. Enhancement of intragastric acid stability of a fat emulsion meal delays gastric emptying and increases cholecystokinin release and gallbladder contraction. *Am J Physiol Gastrointestinal Liver Physiol*. (2007) 292:1607–13. doi: 10.1152/ajpgi.00452.2006
64. Ezeanaka MC, Nsor-Atindana J, Zhang M. Online low-field nuclear magnetic resonance (LF-NMR) and magnetic resonance imaging (MRI) for food quality optimization in food processing. *Food Bioprocess Tech*. (2019) 12:1435–51. doi: 10.1007/s11947-019-02296-w
65. Kaneko G, Ushio H, Ji H. Application of magnetic resonance technologies in aquatic biology and seafood science. *Fisheries Sci*. (2019) 85:1–17. doi: 10.1007/s12562-018-1266-6
66. Bian R, Cao R, Zhao L, Liu Q, Ren D. Changes in moisture status of Cololabis saira during salting and drying by low-field nuclear magnetic resonance. *J Food Safety Qual*. (2017) 8:1698–703. doi: 10.19812/j.cnki.jfsq11-5956/ts.2017.05.029
67. Wu J-L, Zhang J-L, Du X-X, Shen Y-J, Lao X, Zhang M-L, et al. Evaluation of the distribution of adipose tissues in fish

using magnetic resonance imaging (MRI). *Aquaculture*. (2015) 448:112–22. doi: 10.1016/j.aquaculture.2015.06.002

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REVIEW
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Non-thermal Technologies for Food Processing

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Food is subjected to various thermal treatments during processes to enhance its shelf-life. But these thermal treatments may result in deterioration of the nutritional and sensory qualities of food. With the change in the lifestyle of people around the globe, their food needs have changed as well. Today's consumer demand is for clean and safe food without compromising the nutritional and sensory qualities of food. This directed the attention of food professionals toward the development of non-thermal technologies that are green, safe, and environment-friendly. In non-thermal processing, food is processed at near room temperature, so there is no damage to food because heat-sensitive nutritious materials are intact in the food, contrary to thermal processing of food. These non-thermal technologies can be utilized for treating all kinds of food like fruits, vegetables, pulses, spices, meat, fish, etc. Non-thermal technologies have emerged largely in the last few decades in food sector.

Keywords: food preservation, pulse electric field, ultrasound, cold plasma, microwave, high pressure processing, irradiation

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INTRODUCTION

Food quality is a great concern when processing food for preservation. Conventional food preservation processes expose food to a very high temperature, which no doubt reduces the contamination or microbial load from food, but it also results in some undesirable changes in food, such as loss of nutritional components that are temperature-sensitive, change in the texture of food due to heat, and changes in the organoleptic characteristics of food (1). In thermal processing, food is exposed to heat for a long duration of time, which causes observable changes in food and results in the production of low-grade food (2, 3). The thermal techniques used for preservation result in the formation of chemical toxicants in food that are carcinogenic and harm the human body (4, 5). The amount and the type of toxicants formed also depend on the type of thermal method used for cooking food. Microwave cooking and deep fat frying result in the formation of heterocyclic aromatic amines, which can even cause mutagenic changes in the body (5, 6). Thermal treatment can also cause loss of water from food, oxidation of lipids, and changes in the composition of fatty acids. Barbequing of meat causes loss of meat juices that mainly contain saturated lipids stored in the form of adipose tissue, leading to a decrease in saturated fatty acid and an increase in polyunsaturated fatty acid in the final product. The presence of polyunsaturated fatty acid makes the final product more susceptible to lipid oxidation and decreases the quality of product, imparting an off-flavor with a reduced mouthfeel (7). But now, consumers' awareness regarding food safety has increased and they demand food free from microorganism and with high nutritional qualities and excellent mouthfeel. This led food professionals to search for a better alternative, like non-thermal treatments. In non-thermal processing, food is exposed to ambient temperature for a

very limited period of time, i.e., for ∼l min or less, which causes no change in the nutritional composition of food, the texture remains intact, and the mouthfeel is not lost (8–10). The rise in consumer demand for fresh food with longer shelf-life and good sensory qualities led to extensive research in the field of non-thermal treatment of food (11). Thermal technologies that require huge energy consumption and produce low-grade food can be fully or partly replaced by the consumer-, environment-, and pocket-friendly (since they are economical) non-thermal technologies for food processing and preservation (12–14). Various non-thermal food processing treatments came into light since the last few decades, which included pulsed electric field, cold plasma, ultrasonication, microwave, supercritical technology, etc. These non-thermal treatments unmask food to treatment conditions for a fraction of seconds, which results in the reduction of the microbial load in food with an increase in shelf-life, with good sensory and textural characteristics (15, 16). The preservation effect of non-thermal technologies is more than that of thermal technologies because there is no chance for the formation of any undesirable products/by-products in food or on the surface of food since it is not exposed to higher temperatures (17). Pulsed electric field is an extensively used non-thermal processing treatment in the food sector. It is mostly exploited for liquid food including fruit juices, alcoholic beverages, non-alcoholic beverages, etc. It can be directly applied on the entire fruit. It damages the cell wall of microorganisms, leading to the death of microbes and the reduction of the microbial load (18). The intensity of pulse and pulse width play important roles in microbial reduction in food exposed to pulse electric field treatment (19). Non-thermal treatment can also arrest the activity of enzymes, leading to the spoilage of fruits and vegetables. Cold plasma technology is extensively used to enhance the physiological properties of proteins and carbohydrates in food, so that they can be used in numerous applications in food processing. Gaseous cold plasma processing has been used for improving the cooking and textural properties of food grains (20, 21). It also inactivates the microbes present on the surface of the food product. Cold plasma treatment time plays an important role in achieving the desired results (22–24). Ultrasonication is an energy-efficient non-thermal treatment usually used for the intensification of processes like synthesis, extraction, and preservation of food and allied products. Ultrasonication duty cycle and exposure time have positive effects on food. A perfect combination of duty cycle and exposure time can be utilized in developing safe and nutritious food with ultrasonication (25, 26). Other technologies such as ultra-pressure treatment and irradiation are also exploited in the food processing sector to achieve food safety with minimal or no loss of the nutritional, textural, and organoleptic characteristics of food (27, 28). These non-thermal treatments result in a decrease of the microbial load by altering the structure of the membranes in bacterial cells and unfolding of the helical structure of the DNA of the genetic material of microbial cells, leading to the death of microbial cells in a short period of time. Apart from the reduction of the microbial load, these non-thermal treatments are also used for the extraction of bioactives from plant and animal sources having nutraceutical food application for the intensified synthesis of the nutraceutical components, dehydration, for enhancing the physical and chemical properties of food constituents, etc. (29–35). In spite of the many advantages of these non-thermal technologies in the food sector, they are rarely used in food industries and remain at laboratory scale only. There is a great need for the understanding of the construction and workings of these non-thermal technologies and their action on food. There is enough scientific literature available on these technologies. The present review focuses on the recent status of non-thermal techniques in food processing industries to enhance the quality of food products, the effects of these non-thermal techniques on food components, instrumentation used for these non-thermal techniques with a focus on the limitations of these techniques for large-scale production and how they could be overcome, and the future prospects of these techniques in food processing industries. This comprehensive review will definitely help food scientists and technologists working in the field of non-thermal technology since non-thermal treatment is gaining research interest due to its numerous merits over thermal techniques.

NON-THERMAL TECHNOLOGIES

Ultrasonication

Theory

Ultrasonication is an emerging non-thermal technology in the food sector, but it is already a well-established technique in other processing sectors (36). In simple words, ultrasound is a sound wave bearing certain frequency that is more than the normal human hearing frequency, i.e., above 20 kHz (37). When ultrasonic waves oscillate through the medium, they generate many expansion and compression effects in the medium. There is a formations of small cavities due to the presence of air. The cavities formed grow to a desired size and then collapse. When these cavities collapse, they generate both a large amount of energy and local hot spots, and thus there is an increase in heat and mass transfer rates (38). Ultrasonication is used to speed up the chemical synthesis of organic compounds and increase the yield of reaction because the ultrasonication effect results in enhanced heat and mass transfer. Ultrasonication is used with different frequencies, which are classified as low-frequency, medium-frequency, and high-frequency ultrasonication, with frequency ranges of 20 kHz–100 kHz, 100 kHz−1 MHz, and 1 MHz–100 MHz, respectively (39). Low-frequency ultrasonication produces large shear forces in the medium, whereas high-frequency ultrasonication produces less shear forces in the medium. The medium frequency results in the formation of radical species, and this frequency range is considered to be optimum for various sonochemical-assisted process, but the formation of chemical radicals can bring about undesirable changes in food, such as oxidative changes in lipids and proteins (40). Ultrasonication is done using an ultrasonic horn, which is dipped in the liquid solution or juice and is treated with certain treatment frequency. Ultrasonication can be done using an ultrasonic bath, in which the food material or packaged food is kept and the sound waves are generated in a bath that creates ultrasound effect and brings about desired changes in food (41).

Applications

In food processing, frequency in the range of 20 kHz–100 kHz is used for the extraction of bioactives, emulsification, cooking, debittering, intensified synthesis, etc. Jadhav et al. (42) reported on the synthesis of designer lipids using sonication as an excellent alternative for intensified yield. The authors reported a maximum yield of 92% in 6 h of reaction. Due to the increase in energy, there is a generation of high-energy spots that increase the rate of mass transfer, and the reaction is completed in a shorter period of time. Ultrasonication-assisted synthesis is rapid compared to the conventional synthesis process (43). Ultrasound also assists the interfacial transfer of molecules, which enhances the efficiency of the process of extraction of bioactives from plant and animal sources. The extraction process not only increases the yield of the extraction process but also improves the physical and chemical properties of the extracted compound. One such recent study by Sun et al. (44) reported that protein extracted using ultrasonication showed superior properties in terms of the size of the particle, emulsification power, and structure. Ultrasonication-extracted particles with small particle size and larger α helix structure have improved emulsifying power when treated with sonication for 30 min at 20 kHz. Cheila et al. (45) designated ultrasonication as a greener approach for the extraction of bioactives from the leaves of velame. Ultrasound increased the yield of extracted bioactives to 94% in 39.5 min using indirect ultrasonication. Ultrasound has been proven to be an intensified extraction process for the extraction of oil from olive fruit, soybean, and flaxseed (46, 47). It is also employed for the extraction of bioactives from different parts of plants, fruits, and vegetables (48–51). Ultrasonic-assisted filtration process is also very effective and is of importance to dairy and beverage industries. In cheese making, the membrane filtration process is used for the complete separation of milk protein from other milk solids (52). Ultrasonication also aids in the processes of freezing, drying, and thawing of food products (53–55). Mothibe et al. (56) used ultrasonication as a preliminary processing step before the dehydration of apples and reported that the drying time was reduced and the dried apple had a good texture with less water activity. The authors reported that treatment at 25 kHz and time of 15 min showed good results. As the treatment time increased, there was more loss of soluble solids from apple. The ultrasound-assisted process not only decreases the drying time but also enhances and retains the texture after rehydration. Rehydration refers to the absorption of moisture by the dried food (57). Tao et al. (58) showed that ultrasound-rehydrated white cabbage showed a higher rate of rehydration compared to the untreated sample. Similar studies were reported for the rehydration of carrot and green pepper (59, 60). It is also used as a pretreatment for convective drying and freeze drying (61, 62). Ultrasonication is also effective for the preservation of food products by using brine solution. Carcel et al. (63) reported on the use of ultrasound in the treatment of pork loin with a brine solution; ultrasound was applied to this solution. The authors reported that the ultrasound-assisted brine sample has more concentration of brine in it with good color and texture of pieces of pork loin compared to the untreated sample. Ultrasonication is also beneficial for the process of degassing in carbonated beverages and is a good replacement for the processes of pasteurization and sterilization in the reduction of microbial load in food and food products (26). Ultrasound has successfully proven its potential in the food sector in various critical areas like food preservation, extraction, intensified synthesis, and improvement of the physical and chemical properties of food. The very limited technical information about ultrasonication and consumer awareness about ultrasonic-processed food have been the hindrance in the commercialization of this process in food industries. However, the treatment must be studied on bulk food to understand its effect so that it can be implemented at industrial scale.

Cold Plasma Technology

Theory

Plasma is the fourth state of matter after solid, liquid, and gas. The term plasma was used by Langmuir in the year 1925 (64). An increase in the kinetic energy of solids leads to the heating up of molecules, and there is phase transformation from solid to liquid, further increasing the energy of liquid and converting liquid to gas. The increase in energy causes disintegration in the intermolecular structure. When the energy of gases crosses a certain value, it results in the ionization of gas molecules (65). Ionization of gas molecules gives rise to plasma. Hence, it is known as the fourth state of matter. Basically, plasma treatment is divided into two types: thermal plasma and cold plasma (non-thermal). Thermal plasma produces huge energy by utilizing high temperature. Cold plasma is a non-thermal treatment that works in the temperature range 25–65°C (66). When gas is ionized, free radicals (ions, electrons, etc.) are formed. The composition of the plasma reactive species largely depends on the composition of gas which is ionized (67). The gases commonly used for the generation of plasma include argon, helium, oxygen, nitrogen, and air (68). These gases are subjected to any of the types of energy like thermal, electrical, magnetic field, etc., to generate plasma containing positive ions, negative ions, and reactive species like ozone and singlet oxygen (O) (69). Based on the nature of plasma, it has found various applications in the fields of chemistry, chemical engineering, textile, electronics, surface coating, and pharmaceuticals and in food sectors (70). In the food sector, cold plasma can be used for the reduction of the microbial load in food or on the surface of food, enhancing the physical and chemical properties of food constituents like lipids and proteins, and for the sterilization of food processing equipment, inactivation of food spoilage enzymes, treatment of food packaging material, and treatment of wastewater (71). Cold plasma is produced at near ambient temperature and does not depend on high temperature for microbial inactivation. Since the temperature used is ambient, there are no chances of thermal damage to heat-sensitive food material (16).

Applications

Microbial inactivation in cold plasma is due to the effect of reactive species on the microbial cell. Reactive species damage the DNA of cells, induce oxidation in protein, and damage the

cellular components of microbes, causing cell death (72). Lin et al. (73) have reported that cold nitrogen plasma shows inhibitory action on *Salmonella enterica* serovar Typhimurium biofilms formed on the outer surface of an egg shell. The sample was treated at 600 W for 2 min, which reduced the catabolic and anabolic activities of the *S. enterica* serovar Typhimurium by 82.2%. Devi et al. (74) showed 97.9% and 99.3% reductions in the growth of fungal species such as *Aspergillus parasiticus* and *Aspergillus flavus*, respectively, on the ground nut surface when treated at 60 W plasma power. In the food sector, atmospheric pressure cold plasma is used in combination with other gases like helium, argon, etc. Recently, Bang et al. (75) reported on the combination of antimicrobial washing and in-package cold plasma treatment to mandarin oranges for reduction of the microbial load. Treatment at 26 and 27 kV for 1–4 min inactivated *Penicillium digitatum*. The combined effect of washing with an antimicrobial solution and cold plasma treatment reduced the load of *P. digitatum* in the package without affecting the texture, sensory, and nutritional qualities of the oranges. The treated oranges showed a decrease in ripening damage compared to the untreated oranges. Liao et al. (76) reported the use of cold atmospheric pressure-activated water or plasma-activated ice as a cold storage medium for seafood. Shrimps stored in plasma-activated water showed longer shelf-life due to bacterial inactivation, and there was no observable change in the texture of shrimps. The total volatile base nitrogen value for shrimps stored in plasma-treated ice was lower than 20 mg/100 g on the ninth day, which was higher than the 30 mg/100 g for shrimps stored in untreated water or ice. Cold plasma treatment is also effective against the pathogenic microbes present in food and processed food products. One such recent study reported by Gan et al. (77) showed the effectiveness of cold plasma against *Escherichia coli* and *Saccharomyces cerevisiae* in the juice of chokeberries. The authors reported that treatment of 4 min decreased the loads of *E. coli* and *S. cerevisiae* by 2.27 and 1.23 log CFU/ml, respectively. The treatment was seen to be more effective against the inactivation of *E. coli* compared to *S. cerevisiae*. Similar studies on the inactivation of *E. coli* were also reported by Shah et al. (78). Cold plasma is also used for disinfection of the surfaces of food processing equipment to remove the microbial load before the processing of food. Hou et al. (79) investigated the effect of atmospheric pressure cold plasma on bacterial inactivation and the quality of blueberry juice. There was a decrease in the load of *Bacillus* spp. in juice with exposure to cold plasma for 6 min by 7.2 log CFU/ml. The short exposure time resulted in good color and bioactive component retention in juice. Similar results were reported for the preservation of fresh tomato juice (80), cloudy apple juice (81), and apple, tomato, orange, sour cheery nectar (82), and whey grape (83) juice. It is also used for the preservation of meat and related products by reducing their microbial load. Roh et al. (84) studied the effect of cold plasma treatment of 3.5 min against pathogenic microbes in chicken breast. The treatment resulted in decreases in the loads of *E. coli* by 3.9 log CFU/g of chicken, *Listeria monocytogenes* by 3.5 log CFU/g, and Tulane virus by 2.2 CFU/g of chicken. Similar results were reported for the inactivation of *Salmonella* in chicken breast (68, 85, 86) and the microbial load in sea snail (87). The technology is also used for enhancing the physical and chemical properties of food constituents (16, 23). This technology also finds application in enhancing the physical and chemical properties of carbohydrates and proteins in order to increase their functionality and application in food. In the recently published research by Jahromi et al. (88), sodium caseinate in granular form was subjected to 10-kHz treatment for 0, 2.5, 5, and 10 min. With the increase in treatment time, the physical and chemical properties were enhanced. The hydrophilicity of protein increased due to unfolding of the protein structure. Water solubility increased from 20.6 to 30.28%. Tensile strength increased from 5.04 to 7.17 MPa for the 10-min treatment and decreased to 4.73 MPa at 15 min. The effect of cold plasma especially on milk protein is reported by Sharma et al. (89). Cold plasma contains various reactive species, and it is found that these reactive species may trigger the process of lipid oxidation during storage. Gao et al. (90) reported that cold plasma treatment at 70 kV for 180 s triggers the oxidation of lipids during storage. The thiobarbituric acid-reactive substance (TBARS) value increased to 2.48 from 1.43 mg MDA/kg when stored at refrigeration temperature for 5 days, which was 0.37 mg MDA/kg for the control sample of chicken patties. In the TBARS assay, malondialdehyde (MDA) is measured. MDA is a by-product resulting from the process of lipid peroxidation. This MDA reacts with thiobarbituric acid and forms a pink chromogen, known as TBARS. This oxidative degradation of lipids in food can be controlled by altering the treatment conditions, such as the exposure of food to plasma for a short duration of time or the addition of antioxidants in food to overcome these disadvantages of cold plasma on lipids in food. Food containing higher lipid levels can be exposed for a shorter time to cold plasma compared to food with a low lipid content (91, 92).

Supercritical Technology

Theory

Supercritical technology makes use of supercritical fluids, which are considered as a good replacement for organic solvents used in various operations (93). When a fluid is heated beyond its critical temperature and critical pressure, it attains a supercritical state and is referred to as a supercritical fluid. The supercritical fluid shows some properties of gas and some properties of liquid. It shows density like liquids and diffusivity and viscosity like gas (35). Supercritical fluid shows enhanced properties similar to liquid, and hence it can be used as a solvent with an increased rate of mass transfer during the extraction of bioactives from various plant and animal sources. The properties of fluids can be altered with changes in temperature and pressure. Many fluids are used for supercritical operations, but carbon dioxide finds special attention as an excellent supercritical fluid in the food processing sector because it can achieve a supercritical state at a modest temperature and pressure (31.1°C and 7.4 MPa, respectively). Supercritical fluids are extensively used in food industries for extraction, microbial inactivation, enhancement of mass transfer in synthesis, etc. Among all the applications, the supercritical technology is extensively employed for extraction purposes.

Applications

Supercritical carbon dioxide is used for the purpose of extraction since it is non-toxic and can be separated from the final product without much effort (94). Natural bioactives that are extracted are sensitive to temperature and oxygen. In the presence of carbon dioxide, the supercritical extraction temperature is very low and there is no chance of the presence of oxygen; hence, the quality of the extracted material is high and can be used as a functional ingredient in various nutraceutical formulations. Recent studies reported by Lefebvre et al. (95) showed that supercritical carbon dioxide is effectively utilized as an excellent tool for the selective extraction of antioxidants from rosemary. The temperature and pressure of CO_2 were 25°C and 20 MPa, respectively, which were ideal and did not affect the purity of the extracted products. Santos et al. (96) investigated the extraction of bioactives from feijoa leaves using supercritical and pressurized liquid extraction. The authors reported that pressurized extraction gave more yield of antioxidant and antibacterial components, but these extracted components were not effective in their function, while supercritical extraction of antioxidant and antibacterial components at 55°C and 30 MPa showed higher effectiveness against pathogenic bacteria including *E. coli*. The technique is also used for the extraction of functional and nutraceutical ingredients from microalgae (97), oil from fruit seeds (98–101), oil from olives (102), oil from ginger (103), extraction of corn germ oil and green coffee oil (104, 105), essential oil extraction (106, 107), and extraction of bioactives such as carotenoids, lycopene, astaxanthin, anthocyanins, and quercetin (108–110), which can be used as components in nutraceutical formulations. Extraction using supercritical carbon dioxide has been common for many years in the food processing industry. Apart from this, supercritical technology is also used for reducing the microbial load in food. Since the operating temperature in supercritical treatment is low, the original characteristics of food, along with its organoleptic characteristics, is retained (111). Supercritical fluid treatment reduces the pH of bacterial cell, which leads to the rupture or bursting of cells and the inactivation of bacterial enzymes that are responsible for catabolism and anabolism; thus, the bacterial cell dies and reduces the load of microbes in food and related products (112). It is extensively used for the preservation of fresh agricultural products including fruits, vegetables, and their juices (113). Bertolini et al. (114) studied the effect of supercritical carbon dioxide on the decrease of microbial load in pomegranate juice and compared it to traditional pasteurization and high-pressure processing. The authors reported that supercritical-treated juice showed bacterial growth below the detection level after storage for 28 days. The total phenolic content increased by 22%, but it decreased in traditional pasteurization by 15%. The antioxidant activity of the phenolic components was more in supercritical-treated juice compared to that in high-pressure processing and traditional pasteurization. Similar results were reported for the preservation of coconut water (115), sports drink (116), and liquid food (117). Supercritical fluids are also used for the preservation of ground meat. Yu and Iwahashi (118) treated ground beef with high-pressure carbon dioxide at 1 MPa pressure for 26 h and found a reduction in the microbial load. The critical review of the literature showed that the supercritical technology has bright prospects in the food processing sector not only for extraction but also for the preservation and enhancement of the physiological properties of food constituents to be used as functional ingredients in functional and nutraceutical formulations.

Irradiation

Theory

Gamma rays with high energy, X-rays, and high-speed electrons are approved irradiations to be used in food processing industries. Radionuclide ^{60}CO and ^{137}Cs producing gamma rays are used for the production of elevated energy photons. X-rays with energies up to 5 MeV are used in the food processing sector. High-speed electrons with energy of 10 MeV are used in food industries for various applications (119). Irradiation effects are achieved without an increase in the temperature of food. Since the temperature of food is not raised, there is no chance of damage to the components in food that are sensitive to heat (120). The penetration ability of a high-speed 10-MeV electron is up to 39 mm deep in food with high moisture content. X-rays and gamma rays can reach deep into the food material (119, 121). These radiations result in the unfolding of DNA and damage to the nucleic acid, and the ionization of water molecules results in oxidative damage to the microbial cells; thus, there is reduction in the microbial load of food (122).

Applications

Irradiation is mostly employed in the food processing sector for the preservation of food products. It is effective against pathogenic microbes including *E. coli*, *Staphylococcus*, and *Salmonella* (123, 124). Changing the intensity of irradiations shows more intense effects on the inactivation of microbes in food. Irradiation is also used in the preservation of meat for several days. Ready-to-cook chicken stored for 15 days treated with gamma radiations of intensities 0, 1.5, 3, and 4.5 kGy showed excellent result for the inactivation of *L. monocytogenes*, *E. coli*, and *Salmonella typhimurium*, with D_{10} values of 0.680, 0.397, and 0.601, respectively. The ready-to-eat chicken showed good sensory and textural characteristics even after 15 days of storage (125). Irradiation technology also enhances keeping qualities of food and keeping food fresh by the inactivation of microbes causing foodborne diseases (126). It has been found that the use of irradiation scan results in some undesirable changes in food if treated at high irradiation doses, mostly seen in food like meat whose color and lipids are the main defining factors and a slight change in color and lipids may lead to rejection by consumers (127). It is also seen in cereals and food grains (128). Thus, to achieve the desired inactivation in food with no or little change in the food composition and processed food products, irradiation is usually done with a low dose, and the irradiation effect is combined with the use of antimicrobial agents (129). Irradiation is successfully used for achieving microbial inactivation, like the microbial load in fresh pasta (130) and for enhancing the physical and chemical properties of food, such as those of wheat

(131), garlic bulbs (132), grape juice (133), mangosteen fruit (134), apple juice (135), etc. Despite the many advantages of irradiation technology mostly in food preservation, consumer acceptability of irradiation-processed food is low because of the wrong perception of the word "irradiation." For a non-food technologist, irradiation is the generation of some carcinogens in food, as the word is similar to "radiation therapy" (122). Low consumer acceptance is the great hindrance in the development of this technology in the food industry. Changing the views of consumers and encouraging them to buy irradiated food could be solutions for the development of this technique, and designing simpler and reliable instrumentation and overcoming the myths about this technology among consumers will greatly influence the market of irradiated food in the coming years.

Pulsed Electric Field

Theory

Pulsed electric field (PEF) is an emerging non-thermal technology and finds various applications in the food sector. The growing demand for safe food with nutritional qualities has influence on the use of pulsed electric field in the food sector. In pulsed electric field, a pulse of high field intensity is applied to food for a very short duration of time (19). Usually, for the treatment of food, the field intensity is from 25 to 85 kV/cm, and the exposure time is a few milliseconds or nanoseconds. Since food is exposed to pulsed electric field for a very short duration of time, there is no chance of heating; hence, undesirable changes in food due to high temperature are eliminated (18). In the early 1950s, PEF was used for preservation by inactivating microbes. Since then, it has developed a lot in recent years and has been widely used for microbial inactivation in food. A typical PEF device consists of a food treatment chamber, a control system, and a pulse generation unit. The food is kept in the treatment chamber in between two electrodes generally made of stainless steel (136). Pulsed electric field is generally used for liquid food or semi-solid food that can flow easily (137). There is a damage to the cell membrane of microbes due to the high field intensity. Hydrogen peroxide is found in a PEF-treated sample, and it brings about oxidative changes in the cell lipids and protein of the bacterial cell, which also inactivates the metabolic enzymes, thus causing cell death (138). The efficiency of PEF in reducing microbial load largely depends on the intensity of field applied, the total exposure time, temperature, and energy.

Applications

PEF is extensively employed for increasing the shelf-life of food by decreasing the microbial load. A recently published study by Preetha et al. (139) showed that PEF with an intensity of 5.6 W/cm^2 was effective against *E. coli* in flowable food like pineapple and orange juice and coconut water, with decreases in the *E. coli* load of 4.5, 4, and 5.3 log CFU/ml juice respectively. Similar results with moderate PEF intensity are also effective for microbial inactivation in fruit juices (140). Microbial cells that are larger in size are exposed to PEF easily, but smaller microbial cells may resist the treatment and remain unaffected (141). Apart from microbial inactivation, PEF is also effective in the deactivation of food spoilage enzymes. Similar studies are reported for the inactivation of enzymes in apple and carrot juice (142) and pine nut (143). López-Gámez et al. (144) investigated PEF treatment of 580 J/kg on carrot, which showed enhanced anabolism for the production of phenolic compounds over a storage period of 36 h. PEF is also extensively used in the extraction of bioactives from many natural sources. A recent study was reported by Käferböck et al. (145) on the extraction of functional components from microalgae using PEF with a frequency of 300 Hz and pulse width of 4–32 μs. The PEF treatment resulted in enhanced extraction of the functional components with high purity, which can be used for their nutraceutical application in food industries without purification. A similar study for extraction from the microalgae *Haematococcus pluvialis* is reported by Gateau et al. (146). Other studies were on extraction from apple peels (147), cyanobacteria (148), tomato (149), and from cinnamon (150). Apart from the known application of PEF in microbial inactivation, extraction, and physiochemical changes, nowadays, PEF is also employed for unit operations like dehydration and freezing. Liu et al. (151) reported on PEF with an intensity of 0.6 kV/cm and exposure time of 0.1 s that resulted in a decrease in the drying time of carrot by 55% at 25°C and 33% at 90°C. The PEF-treated sample showed good textural properties and color after rehydration compared to the untreated sample. PEF pretreatment before the drying operation results in enhancing the vitamin and mineral contents in food (152). It is also used in the freezing of food and improves the quality of food during the thawing process (153). PEF treatment leads to an improvement in the coefficient of diffusion for water before drying, reduces the time required for freezing and drying food, and maintains the quality of rehydrated and thawed food for a longer period. PEF is also employed for enhancing the physical and chemical properties of major food components such as polysaccharides, proteins, etc. (154–156), and for the modification of potato starch (157) and the properties of oat flour (158). PEF also enhances the rate of reaction; because of the high intensity, there is an enhanced heat transfer, which increases mass transfer in the esterification (159) and chelation (160) reactions. The numerous good effects of PEF on food products make it a good non-thermal treatment method. There is a need for the development of high-strength PEF instrumentation for commercial application.

High Hydrostatic Pressure

Theory

High hydrostatic pressure (HHP) utilizes a very common medium, i.e., water, to apply the pressure on the product to be treated. HHP can bring about a significant decimal decrease in the population of pathogenic Gram-negative bacteria, Gram-positive bacteria, yeast, and mold and helps in food preservation for a longer duration. The reduction in microbial load depends on the pressure and temperature during treatment. It largely depends on the type of food processed. Food, when subjected to HHP treatment, undergoes high pressure for a short duration of time. The pressure applied to food during treatment is in the range of 200–700 MPa (161). The quality in terms of nutritional components, sensory, and texture of HHP-processed food is excellent since the food is exposed to treatment conditions for a very short period of time (162). It is found that the HHP

treatment is more effective against eukaryotes, Gram-negative bacteria, protozoa, and parasites than yeast and mold, which are inactivated at much higher pressure (163). The instrumentation required for HHP is very simple and easy to operate. It consists of a pressure compartment in which food is kept and water is introduced into the chamber; food is then pressurized using this water (164). Thus, HHP-treated food shows fresh-like attributes since there is no intervention of high temperature and chemical additives. Pressure of 350–450 MPa is sufficient for the inactivation of Gram-negative bacteria, yeast, and mold at room temperature, but to inactivate Gram-positive bacteria, pressure more than 1,100 MPa is required (165). The high pressure results in damage to the cell membrane of microbial cells, which changes the permeability of the microbial cell wall and membranes. The coiled protein structure breaks and there is destruction to microbial cell enzymes, which alter the metabolic pathways; finally, the microbial cell dies, leading to a decrease in the microbial population in food (161).

Applications

HHP treatment has been proven to be efficient in the inactivation of microbes in a wide range of food products, including processed fruits, meat and meat products, and dairy products, which serve as an excellent medium for microbial growth. The recently published study by Bulut and Karatzas (166) investigated the effectiveness of HHP against *E. coli* in liquid food. Orange juice was stored at −80°C, and then HHP treatment was applied to the juice with pressure of 250 MPa for 900 s. The HHP treatment reduced the microbial load by 4.88, 4.15, and 4.61 log CFU/ml for orange juice with pH 3.2, 4.5, and 5.8, respectively. Cap et al. (167) investigated a decrease in *Salmonella* spp. load in meat using HHP treatment. The HH pressure of 500 MPa for 60 s was enough to inactivate the *Salmonella* spp. in a chicken breast sample without any effect on the organoleptic and sensory attributes of chicken. Cava et al. (168) showed that dry cured sausage can be preserved for more than 60 days with inactivation of *L. monocytogenes* by 3.2 log CFU/g with HHP treatment of 600 MPa for 480 s. There was no oxidative damage to the lipids and proteins in food up to 60 days. HHP has no effect on the oxidation of lipids; thus, it does not contribute to the development of rancidity in food. de Jesus et al. (169) reported that HHP is not only effective in the reduction of the microbial load but it can also be effectively utilized for the extraction of antioxidant, anthocyanin, and phenolic compounds with various nutraceutical properties in food. Similar studies on extraction were reported from tomato waste (170), pomace of grapes (171), red microalgae (172), egg yolk (173), and from gooseberry juice (110). HHP also enhances the physical and chemical properties of fermented juices and increases bioactives in the fermented juice (174). HHP is also effective in the preservation of human breast milk (175, 176). It is also beneficial for intensifying the technical and functional properties of milk proteins for their increased application in various functional and nutraceutical foods (177). HHP has been the potential treatment not only for bacterial inactivation but also for the extraction and enhancement of antioxidant, phenolic, bioactive, and functional components from various sources, suggesting its application potential in various nutraceutical, pharmaceutical, health, food, and related industries. There are many technical obstacles in building the HHP units that are feasible for the treatment of high-volume food, and hence there are very few or no HHP-treated food available in the market today.

Pulsed Ultraviolet Technology

Theory

Ultraviolet technology is a very economical, non-thermal technology. It is basically used to reduce the microbial load on the surface of food materials that are indirectly exposed to radiation, which are grouped as UV-A in the electromagnetic spectrum in range of 320–400 nm, UV-B in the range of 280–320 nm, and UV-C in the range of 200–280 nm (178). When food is exposed to UV-C, with 200–280 nm, these short wavelengths are absorbed by the microbial cell nucleic acids. These absorbed photons cause the breakage of the bond and interlinking between thymine and pyrimidine of different strands and the formation of dimers of pyrimidine. These dimers prevent DNA transcription and translation, thus leading to the malfunctioning of the genetic material, which causes microbial cell death (179). The photons of UV-A and UV-B result in the destruction of the cellular membranes, proteins of microbial cells, and other cellular organelles, which causes death of the microorganisms present in food (180).

Applications

Pulsed UV technology is among the most popular non-thermal technologies in the food processing sector. Owing to its economical nature, it is also being experimented on a pilot scale for the inactivation of microbes. A recent study by Fenoglio et al. (181) on a pilot-scale UV inactivation of pathogenic microbes showed that the intensity of UV-C with 390 mJ/cm^2 leads to the inactivation of pathogenic bacteria in fruit juices, with log reductions of 6.3 for *Lactobacillus plantarum*, 5.1 log CFU for *E. coli*, and 5.5 for *S. cerevisiae*. Similar studies on the inactivation of microorganisms in fruit juices are reported in apple juice (182), orange juice (183), and cantoloupe melon juice (184). Ultraviolet inactivation is also extensively used for the inactivation of microbes present in milk and milk products (185). Ultraviolet radiation also shows useful effects on the chemical and physical properties of food. Kumar et al. (186) showed that UV-C radiation with 254 nm was able to enhance the physical and chemical properties of protein from wheat. Therefore, it can be used for many applications in food industries. Recent studies have shown that ultraviolet treatment of fresh fruits and vegetables (after harvesting) not only results in microbial inactivation but also increases the antioxidant content and enhances its activity (187). UV treatment is also used for the reduction of toxins in food (188). With many positive effects on food, there are some studies reported in the literature which showed that high-dose UV treatment can lead to a decrease in the color of food and adversely affects the texture of solid food (189). All food products have different textures with uneven and rough surfaces, so the ability of radiation to reach inside the food material may be reduced, decreasing the efficiency of the inactivation process. Thus, to increase the efficiency of the

process and achieve higher inactivation, non-thermal processes are usually coupled or antibacterial agents are used along with UV treatment (190, 191). Due to its simple operation, UV is one of the well-established non-thermal processing technologies adopted by food processing industries to produce food with longer shelf-life. The effect of UV can be more intensified if the process is coupled with other processes to bring about desired changes.

Ozone

Theory

Ozone, chemically written as O_3, contains three molecules of oxygen. It is a colorless gas with a typical odor. It is formed when molecular oxygen (O_2) combines with singlet O. Ozone is denser in gas form than air. Ozone is a very reactive gas, and it is very much unstable and cannot be stored and needs to be produced on the spot when needed. Ozone is extensively employed as an effective antibacterial against many bacteria in food. It can be used in gas form or it can be mixed with water to form ozonated water. There are many ways by which ozone causes microbial cell death. Ozone alters the permeability of cells by damaging the microbial cell membranes. Ozone is also known to damage the structure of proteins, leading to the malfunctioning of microbial enzymes, which affects the metabolic activity and finally results in microbial cell death (192, 193).

Applications

Gimenez et al. (194) reported on the effectiveness of ozone against *L. monocytogenes* present in meat. Treatment of 280 mg O_3/m^3 for 5 h with pulse of ozone passed after 10 min for 30 min duration was effective, but an increase in the treatment time showed a change in color and oxidative damage to the lipids present in meat. Thus, to reduce the exposure time of ozone, it is combined with other treatments of food additives in order to enhance its effectiveness without any damage to food. Such studies were reported for the inactivation of *Salmonella* (195) and spoilage microorganisms (196). Ozone treatment of fruits after harvest enhances their physical, chemical, and textural properties with a reduction in the microbial load when stored in modified atmosphere packaging for 15 days (197). It is extensively used in the reduction of microbes in fruit juices (198–200). Ozone treatment is also effective in the inactivation of toxins present in food (193). There are many research reports in the literature proving the potential of ozone in the food sector, but these studies are on a laboratory scale and are not commercialized. Ozone is used in the industry for the disinfection of processing equipment. It is a very reactive molecule that reacts with many components in food, leading to undesirable changes. It also induces oxidation in food lipids; thus, it can be used in combination with other techniques. There is a need for thorough studies regarding the doses of ozone in order to reduce undesirable changes in food and to improve its acceptability. Efforts are required to increase consumer acceptability of ozone-treated food, which will force the food industry to adopt this technology to process food and market ozone-treated food.

CONCLUSION

Non-thermal treatments are among the most focused research areas in the food sector due to consumer demands for safe and nutritious food free from microbes. The food product is exposed to non-thermal treatment for a very short period of time and food is treated at ambient temperature. Since the exposure time is short and the temperature is low, there are no chances of damage to heat-sensitive nutritional components in food, no damage to the food texture, and no chances of the formation of any toxic compound in food due to heat. Thus, with non-thermal treatments, consumers get fresh processed food with high nutrition and good color and flavor. But there are two sides of the coin: with advantages come some disadvantages as well. If food is exposed for a longer period or treated at a higher intensity, these non-thermal technologies may lead to some undesirable changes in food, such as oxidation of lipids and loss of color and flavor. But these technologies have many advantages compared to thermal processing. Additionally, the development of equipment to process food in bulk using non-thermal technology, understanding the proper mechanisms, development of processing standards using non-thermal treatments, and clarifying consumer myths and misunderstanding about these technologies will be helpful in the promotion of non-thermal technologies in the food sector. Once these limitations are properly overcome in a planned manner, non-thermal technologies will have a broader scope for development and commercialization in food processing industries, delivering safe and nutritious food with good color and mouthfeel to consumers.

AUTHOR CONTRIBUTIONS

RD: conceptualization, skeleton of the manuscript, and reviewing/editing. UA: correction of draft and editing. HJ: preparation of draft and making corrections as per suggestions. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Hernández-Hernández HM, Moreno-Vilet L, Villanueva-Rodríguez SJ. Current status of emerging food processing technologies in Latin America: novel non-thermal processing. *Innov Food Sci Emerg Technol.* (2019) 58:102233. doi: 10.1016/j.ifset.2019.102233
2. Pereira RN, Vicente AA. Environmental impact of novel thermal and non-thermal technologies in food processing. *Food Res Int.* (2010) 43:1936–43. doi: 10.1016/j.foodres.2009.09.013
3. Iqbal A, Murtaza A, Hu W, Ahmad I, Ahmed A, Xu X. Activation and inactivation mechanisms of polyphenol oxidase during thermal and non-thermal methods of food processing. *Food Bioprod Process.* (2019) 117:170–82. doi: 10.1016/j.fbp.2019.07.006
4. Oz E. Effects of smoke flavoring using different wood chips and barbecuing on the formation of polycyclic aromatic hydrocarbons and heterocyclic aromatic amines in salmon fillets. *PLoS ONE.* (2020) 15:e0227508. doi: 10.1371/journal.pone.0227508
5. Oz F, Kizil M, Çelik T. Effects of different cooking methods on the formation of heterocyclic aromatic amines in goose meat. *J Food Process Preserv.* (2016) 40:1047–53. doi: 10.1111/jfpp.12685
6. Babaoglu AS, Karakaya M, Öz F. Formation of polycyclic aromatic hydrocarbons in beef and lamb kokorec: effects of different animal fats. *Int J Food Prop.* (2017) 20:1960–70. doi: 10.1080/10942912.2016.1225761
7. Oz E. The impact of fat content and charcoal types on quality and the development of carcinogenic polycyclic aromatic hydrocarbons and heterocyclic aromatic amines formation of barbecued fish. *Int J Food Sci Technol.* (2021) 56:954–64. doi: 10.1111/ijfs.14748
8. Birmpa A, Sfika V, Vantarakis A. Ultraviolet light and Ultrasound as non-thermal treatments for the inactivation of microorganisms in fresh ready-to-eat foods. *Int J Food Microbiol.* (2013) 167:96–102. doi: 10.1016/j.ijfoodmicro.2013.06.005
9. Beyrer M, Pina-Perez MC, Martinet D, Andlauer W. Cold plasma processing of powdered Spirulina algae for spore inactivation and preservation of bioactive compounds. *Food Control.* (2020) 118:107378. doi: 10.1016/j.foodcont.2020.107378
10. Pizarro-Oteíza S, Briones-Labarca V, Pérez-Won M, Uribe E, Lemus-Mondaca R, Cañas-Sarazúa R, et al. Enzymatic impregnation by high hydrostatic pressure as pretreatment for the tenderization process of Chilean abalone (Concholepas concholepas). *Innov Food Sci Emerg Technol.* (2020) 65:102451. doi: 10.1016/j.ifset.2020.102451
11. Frewer LJ, Bergmann K, Brennan M, Lion R, Meertens R, Rowe G, et al. Consumer response to novel agri-food technologies: implications for predicting consumer acceptance of emerging food technologies. *Trends Food Sci Technol.* (2011) 22:442–56. doi: 10.1016/j.tifs.2011.05.005
12. Jan A, Sood M, Sofi SA, Norzom T. Non-thermal processing in food applications: a review. *Int J Food Sci Nutr.* (2017) 2:171–80.
13. Jadhav H, Annapure U. Greener route for intensified synthesis of Tricaprylin using Amberlyst-15. *J Chem Sci.* (2021) 133:1. doi: 10.1007/s12039-020-01869-z
14. Choi MS, Jeon EB, Kim JY, Choi EH, Lim JS, Choi J, et al. Impact of non-thermal dielectric barrier discharge plasma on *Staphylococcus aureus* and *Bacillus cereus* and quality of dried blackmouth angler (Lophiomus setigerus). *J Food Eng.* (2020) 278:109952. doi: 10.1016/j.jfoodeng.2020.109952
15. Choudhary R, Bandla S. Ultraviolet pasteurization for food industry. *Int J Food Sci Nutr Eng.* (2012) 2:12–5. doi: 10.5923/j.food.20120201.03
16. Thirumdas R, Sarangapani C, Annapure US. Cold plasma: a novel non-thermal technology for food processing. *Food Biophys.* (2014) 10:1–11. doi: 10.1007/s11483-014-9382-z
17. Thirumdas R, Sarangapani C, Barba FJ. Pulsed electric field applications for the extraction of compounds and fractions (fruit juices, winery, oils, by-products, etc.). In: Barba FJ, Parniakov O, Wiktor A, editors. *Pulsed Electric Fields to Obtain Healthier and Sustainable Food for Tomorrow*. Spain: INC (2020). p. 227–46. doi: 10.1016/B978-0-12-816402-0.00010-0
18. Vorobiev E, Lebovka N. Pulsed electric field in green processing and preservation of food products. In: Chemat F, Vorobiev E, editors. *Green Food Processing Techniques*. France: Elsevier Inc. (2019). p. 403–30. doi: 10.1016/B978-0-12-815353-6.00015-X
19. Niu D, Zeng XA, Ren EF, Xu FY, Li J, Wang MS, et al. Review of the application of pulsed electric fields (PEF) technology for food processing in China. *Food Res Int.* (2020) 137:109715. doi: 10.1016/j.foodres.2020.109715
20. Thirumdas R, Saragapani C, Ajinkya MT, Deshmukh RR, Annapure US. Influence of low pressure cold plasma on cooking and textural properties of brown rice. *Innovat Food Sci Emerg Technol.* (2016)37:53–60. doi: 10.1016/j.ifset.2016.08.009
21. Thirumdas R, Trimukhe A, Deshmukh RR, Annapure US. Functional and rheological properties of cold plasma treated rice starch. *Carbohydr Polym.* (2017) 157:1723–31. doi: 10.1016/j.carbpol.2016.11.050
22. Cui H, Yang X, Abdel-Samie MA, Lin L. Cold plasma treated phlorotannin/Momordica charantia polysaccharide nanofiber for active food packaging. *Carbohydr Polym.* (2020) 239:116214. doi: 10.1016/j.carbpol.2020.116214
23. Bulbul VJ, Bhushette PR, Zambare RS, Deshmukh RR, Annapure US. Effect of cold plasma treatment on Xanthan gum properties. *Polym Test.* (2019) 79:106056. doi: 10.1016/j.polymertesting.2019.106056
24. Deng LZ, Tao Y, Mujumdar AS, Pan Z, Chen C, Yang XH, et al. Recent advances in non-thermal decontamination technologies for microorganisms and mycotoxins in low-moisture foods. *Trends Food Sci Technol.* (2020) 106:104–12. doi: 10.1016/j.tifs.2020.10.012
25. Natarajan S, Ponnusamy V. A review on the applications of ultrasound in food processing. *Mater Today Proc.* (2020) 10:1–4. doi: 10.1016/j.matpr.2020.09.516
26. Bhargava N, Mor RS, Kumar K, Sharanagat VS. Advances in application of ultrasound in food processing: a review. *Ultrason Sonochem.* (2021) 70:105293. doi: 10.1016/j.ultsonch.2020.105293
27. Tsevdou M, Gogou E, Taoukis P. High hydrostatic pressure processing of foods. In: Taoukis P, editor. *Green Food Processing Techniques*. France: Elsevier Inc. (2019). p. 87–137. doi: 10.1016/B978-0-12-815353-6.00004-5
28. Roberts PB. Irradiation of foods | processing technology. *Encycl Food Sci Nutr.* (2003) 19:3390–6. doi: 10.1016/B0-12-227055-X/00655-6
29. Ohlsson T, Bengtsson N. Minimal processing of foods with non-thermal methods. In: Bengtsson N, editor. *Minimal Processing Technologies in the Food Industries*. Cambridge: Woodhead Publishing Limited (2002). p. 34–60. doi: 10.1533/9781855736795.24
30. Chen P, Deng S, Cheng Y, Lin X, Metzger L, Ruan R. Non-thermal food pasteurization processes: an introduction. In: Doona CJ, Kustin K, Feeherry FE, editor. *Case Studies in Novel Food Processing Technologies*. Cambridge: Woodhead Publishing Limited (2010). p. 1–18. doi: 10.1533/9780857090713.1
31. Rifna EJ, Singh SK, Chakraborty S, Dwivedi M. Effect of thermal and non-thermal techniques for microbial safety in food powder: recent advances. *Food Res Int.* (2019) 126:108654. doi: 10.1016/j.foodres.2019.108654
32. Hati S, Patel M, Yadav D. Food bioprocessing by non-thermal plasma technology. *Curr Opin Food Sci.* (2018) 19:85–91. doi: 10.1016/j.cofs.2018.03.011
33. Terefe NS, Janakievski F, Glagovskaia O, Stockmann R. Forward osmosis: an emerging non-thermal concentration technology for liquid foods. In: Stockmann R, editor. *Reference Module in Food Science*. Cambridge: Elsevier (2019). p. 1–8.
34. Mazzutti S, Pedrosa RC, Salvador Ferreira SR. Green processes in foodomics. In: Salvador Ferreira SR, editor. *Supercritical Fluid Extraction of Bioactives*. *Reference Module in Food Science*. Cambridge: Elsevier (2020). p. 1–19. doi: 10.1016/B978-0-08-100596-5.22816-3
35. Brunner G. Supercritical fluids: technology and application to food processing. *J Food Eng.* (2005) 67:21–33. doi: 10.1016/j.jfoodeng.2004.05.060
36. Chemat F, Zill-E-Huma, Khan MK. Applications of ultrasound in food technology: Processing, preservation and extraction. *Ultrason Sonochem.* (2011) 18:813–35. doi: 10.1016/j.ultsonch.2010.11.023
37. Mason TJ, Cintas P. Sonochemistry. *Handb Green Chem Technol.* (2007) 2021:372–96. doi: 10.1002/9780470988305.ch16
38. Bhangu SK, Ashokkumar M. Theory of sonochemistry. *Top Curr Chem.* (2016) 374:1–28. doi: 10.1007/978-3-319-54271-3_1

39. Mason TJ, Chemat F, Ashokkumar M. Power ultrasonics for food processing. In: Ashokkumar M, editor. *Power Ultrasonics: Applications of High-Intensity Ultrasound*. Cambridge: Elsevier Ltd. (2015). p. 815–43. doi: 10.1016/B978-1-78242-028-6.00027-2
40. Delmas H, Barthe L. Ultrasonic mixing, homogenization, and emulsification in food processing and other applications. In: Gallego J, Karl F, Juan A, editors. *Power Ultrasonics: Applications of High-Intensity Ultrasound*. Cambridge: Elsevier Ltd. (2015). p. 757–91. doi: 10.1016/B978-1-78242-028-6.00025-9
41. Li W, Gamlath CJ, Pathak R, Martin GJO, Ashokkumar M. Ultrasound – the physical and chemical effects integral to food processing. In: Knoerzer K, Juliano P, Smithers G, editors. *Innovative Food Processing Technologies*. Cambridge (2021). p. 329–58. doi: 10.1016/B978-0-08-100596-5.22679-6
42. Jadhav HB, Gogate PR, Waghmare JT, Annapure US. Intensified synthesis of palm olein designer lipids using sonication. *Ultrason Sonochem.* (2021) 73:105478. doi: 10.1016/j.ultsonch.2021.105478
43. Jadhav HB, Annapure U. Process intensification for synthesis of triglycerides of capric acid using green approaches. *J Indian Chem Soc.* (2021) 98:100030. doi: 10.1016/j.jics.2021.100030
44. Sun X, Zhang W, Zhang L, Tian S, Chen F. Molecular and emulsifying properties of arachin and conarachin of peanut protein isolate from ultrasound-assisted extraction. *Lwt.* (2020) 132:109790. doi: 10.1016/j.lwt.2020.109790
45. Cheila CB, dos Anjos GL, Nóbrega RSA, da S. Magaton A, de Miranda FM, Dias F. Greener ultrasound-assisted extraction of bioactive phenolic compounds in Croton heliotropiifolius Kunth leaves. *Microchem J.* (2020) 159:105525. doi: 10.1016/j.microc.2020.105525
46. Cavallo C, Carlucci D, Carfora V, Caso D, Cicia G, Clodoveo ML, et al. Innovation in traditional foods: a laboratory experiment on consumers' acceptance of extra-virgin olive oil extracted through ultrasounds. *NJAS - Wageningen J Life Sci.* (2020) 92:100336. doi: 10.1016/j.njas.2020.100336
47. Juliano P, Bainczyk F, Swiergon P, Supriyatna MIM, Guillaume C, Ravetti L, et al. Extraction of olive oil assisted by high-frequency ultrasound standing waves. *Ultrason Sonochem.* (2017) 38:104–14. doi: 10.1016/j.ultsonch.2017.02.038
48. Iftikhar M, Zhang H, Iftikhar A, Raza A, Begum N, Tahamina A, et al. Study on optimization of ultrasonic assisted extraction of phenolic compounds from rye bran. *Lwt.* (2020) 134:110243. doi: 10.1016/j.lwt.2020.110243
49. Qin L, Yu J, Zhu J, Kong B, Chen Q. Ultrasonic-assisted extraction of polyphenol from the seeds of Allium senescens L. and its antioxidative role in Harbin dry sausage. *Meat Sci.* (2021) 172:108351. doi: 10.1016/j.meatsci.2020.108351
50. Wu L, Li L, Chen S, Wang L, Lin X. Deep eutectic solvent-based ultrasonic-assisted extraction of phenolic compounds from Moringa oleifera L. leaves: optimization, comparison and antioxidant activity. *Sep Purif Technol.* (2020) 247:117014. doi: 10.1016/j.seppur.2020.117014
51. Martínez-Ramos T, Benedito-Fort J, Watson NJ, Ruiz-López II, Che-Galicia G, Corona-Jiménez E. Effect of solvent composition and its interaction with ultrasonic energy on the ultrasound-assisted extraction of phenolic compounds from Mango peels (Mangifera indica L.). *Food Bioprod Process.* (2020) 122:41–54. doi: 10.1016/j.fbp.2020.03.011
52. Saxena A, Tripathi BP, Kumar M, Shahi VK. Membrane-based techniques for the separation and purification of proteins: an overview. *Adv Colloid Interface Sci.* (2009) 145:1–22. doi: 10.1016/j.cis.2008.07.004
53. Chow R, Blindt R, Chivers R, Povey M. A study on the primary and secondary nucleation of ice by power ultrasound. *Ultrasonics.* (2005) 43:227–30. doi: 10.1016/j.ultras.2004.06.006
54. Miles CA, Morley MJ, Rendell M. High power ultrasonic thawing of frozen foods. *J Food Eng.* (1999) 39:151–9. doi: 10.1016/S0260-8774(98)00155-1
55. Cheng XF, Zhang M, Adhikari B. Effect of ultrasonically induced nucleation on the drying kinetics and physical properties of freeze-dried strawberry. *Dry Technol.* (2014) 32:1857–64. doi: 10.1080/07373937.2014.952741
56. Mothibe KJ, Zhang M, Mujumdar AS, Wang YC, Cheng X. Effects of ultrasound and microwave pretreatments of apple before spouted bed drying on rate of dehydration and physical properties. *Dry Technol.* (2014) 32:1848–56. doi: 10.1080/07373937.2014.952381
57. Tian Y, Zhao Y, Huang J, Zeng H, Zheng B. Effects of different drying methods on the product quality and volatile compounds of whole shiitake mushrooms. *Food Chem.* (2016) 197:714–22. doi: 10.1016/j.foodchem.2015.11.029
58. Tao Y, Han M, Gao X, Han Y, Show PL, Liu C, et al. Applications of water blanching, surface contacting ultrasound-assisted air drying, and their combination for dehydration of white cabbage: drying mechanism, bioactive profile, color and rehydration property. *Ultrason Sonochem.* (2019) 53:192–201. doi: 10.1016/j.ultsonch.2019.01.003
59. Ricce C, Rojas ML, Miano AC, Siche R, Augusto PED. Ultrasound pre-treatment enhances the carrot drying and rehydration. *Food Res Int.* (2016) 89:701–8. doi: 10.1016/j.foodres.2016.09.030
60. Szadzińska J, Łechtańska J, Kowalski SJ, Stasiak M. The effect of high power airborne ultrasound and microwaves on convective drying effectiveness and quality of green pepper. *Ultrason Sonochem.* (2017) 34:531–9. doi: 10.1016/j.ultsonch.2016.06.030
61. Rojas ML, Augusto PED, Cárcel JA. Ethanol pre-treatment to ultrasound-assisted convective drying of apple. *Innov Food Sci Emerg Technol.* (2020) 61:102328. doi: 10.1016/j.ifset.2020.102328
62. Prosapio V, Norton I. Simultaneous application of ultrasounds and firming agents to improve the quality properties of osmotic + freeze-dried foods. *Lwt.* (2018) 96:402–10. doi: 10.1016/j.lwt.2018.05.068
63. Cárcel JA, Benedito J, Bon J, Mulet A. High intensity ultrasound effects on meat brining. *Meat Sci.* (2007) 76:611–9. doi: 10.1016/j.meatsci.2007.01.022
64. Irving L. Oscillations in ionized gases. *Sci. Am*. (1948) 178:50–3. doi: 10.1038/scientificamerican0552-50
65. Luo QZ, D'Angelo N, Merlino RL. Shock formation in a negative ion plasma. *Phys Plasmas.* (1998) 5:2868–70. doi: 10.1063/1.873007
66. Niemira BA. Cold plasma decontamination of foods *. *Annu Rev Food Sci Technol.* (2012) 3:125–42. doi: 10.1146/annurev-food-022811-101132
67. Alves Filho EG, de Brito ES, Rodrigues S. Effects of cold plasma processing in food components. In: Bermudez-Aguirre D, editor. *Advances in Cold Plasma Applications for Food Safety and Preservation*. Washington, DC: Elsevier Inc. (2019). p. 253–68. doi: 10.1016/B978-0-12-814921-8.00008-6
68. Keener KM, Misra NN. Future of cold plasma in food processing. In: Cullen PJ, Schluter O, editors. *Cold Plasma in Food and Agriculture: Fundamentals and Applications*. Washington, DC: Elsevier Inc. (2016). p. 343–60. doi: 10.1016/B978-0-12-801365-6.00014-7
69. Misra NN, Roopesh MS. Cold plasma for sustainable food production and processing. In: Vorobiev E, Chemat F, editors. *Green Food Processing Techniques*. France: Elsevier Inc. (2019). p. 431–53. doi: 10.1016/B978-0-12-815353-6.00016-1
70. Roth JR, Nourgostar S, Bonds TA. The one atmosphere uniform glow discharge plasma (OAUGDP) - a platform technology for the 21st century. *IEEE Trans Plasma Sci.* (2007) 35:233–50. doi: 10.1109/TPS.2007.892711
71. Chizoba Ekezie FG, Sun DW, Cheng JH. A review on recent advances in cold plasma technology for the food industry: current applications and future trends. *Trends Food Sci Technol.* (2017) 69:46–58. doi: 10.1016/j.tifs.2017.08.007
72. Phan KTK, Phan HT, Brennan CS, Phimolsiripol Y. Nonthermal plasma for pesticide and microbial elimination on fruits and vegetables: an overview. *Int J Food Sci Technol.* (2017) 52:2127–37. doi: 10.1111/ijfs.13509
73. Lin L, Liao X, Li C, Abdel-Samie MA, Cui H. Inhibitory effect of cold nitrogen plasma on Salmonella Typhimurium biofilm and its application on poultry egg preservation. *Lwt.* (2020) 126:109340. doi: 10.1016/j.lwt.2020.109340
74. Devi Y, Thirumdas R, Sarangapani C, Deshmukh RR, Annapure US. Influence of cold plasma on fungal growth and aflatoxins production on groundnuts. *Food Control.* (2017) 77:187–91. doi: 10.1016/j.foodcont.2017.02.019
75. Bang IH, Lee ES, Lee HS, Min SC. Microbial decontamination system combining antimicrobial solution washing and atmospheric dielectric barrier discharge cold plasma treatment for preservation of mandarins. *Postharvest Biol Technol.* (2020) 162:111102. doi: 10.1016/j.postharvbio.2019.111102
76. Liao X, Su Y, Liu D, Chen S, Hu Y, Ye X, et al. Application of atmospheric cold plasma-activated water (PAW) ice for preservation of shrimps (Metapenaeus ensis). *Food Control.* (2018) 94:307–14. doi: 10.1016/j.foodcont.2018.07.026
77. Gan Z, Feng X, Hou Y, Sun A, Wang R. Cold plasma jet with dielectric barrier configuration: investigating its effect on the cell membrane of *E. coli* and

S. cerevisiae and its impact on the quality of chokeberry juice. *Lwt.* (2021) 136:110223. doi: 10.1016/j.lwt.2020.110223
78. Shah U, Ranieri P, Zhou Y, Schauer CL, Miller V, Fridman G, et al. Effects of cold plasma treatments on spot-inoculated *Escherichia coli* O157:H7 and quality of baby kale (Brassica oleracea) leaves. *Innov Food Sci Emerg Technol.* (2019) 57:102104. doi: 10.1016/j.ifset.2018.12.010
79. Hou Y, Wang R, Gan Z, Shao T, Zhang X, He M, et al. Effect of cold plasma on blueberry juice quality. *Food Chem.* (2019) 290:79–86. doi: 10.1016/j.foodchem.2019.03.123
80. Starek A, Pawłat J, Chudzik B, Kwiatkowski M, Terebun P, Sagan A, et al. Evaluation of selected microbial and physicochemical parameters of fresh tomato juice after cold atmospheric pressure plasma treatment during refrigerated storage. *Sci Rep.* (2019) 9:8407. doi: 10.1038/s41598-019-44946-1
81. Illera AE, Chaple S, Sanz MT, Ng S, Lu P, Jones J, et al. Effect of cold plasma on polyphenol oxidase inactivation in cloudy apple juice and on the quality parameters of the juice during storage. *Food Chem X.* (2019) 3:100049. doi: 10.1016/j.fochx.2019.100049
82. Dasan BG, Boyaci IH. Effect of cold atmospheric plasma on inactivation of *Escherichia coli* and physicochemical properties of apple, orange, tomato juices, and sour cherry nectar. *Food Bioprocess Technol.* (2018) 11:334–43. doi: 10.1007/s11947-017-2014-0
83. Amaral GV, Silva EK, Costa ALR, Alvarenga VO, Cavalcanti RN, Esmerino EA, et al. Whey-grape juice drink processed by supercritical carbon dioxide technology: Physical properties and sensory acceptance. *Lwt.* (2018) 92:80–6. doi: 10.1016/j.lwt.2018.02.005
84. Roh SH, Oh YJ, Lee SY, Kang JH, Min SC. Inactivation of *Escherichia coli* O157:H7, Salmonella, Listeria monocytogenes, and Tulane virus in processed chicken breast via atmospheric in-package cold plasma treatment. *Lwt.* (2020) 127:109429. doi: 10.1016/j.lwt.2020.109429
85. Roh SH, Lee SY, Park HH, Lee ES, Min SC. Effects of the treatment parameters on the efficacy of the inactivation of Salmonella contaminating boiled chicken breast by in-package atmospheric cold plasma treatment. *Int J Food Microbiol.* (2019) 293:24–33. doi: 10.1016/j.ijfoodmicro.2018.12.016
86. Moutiq R, Misra NN, Mendonça A, Keener K. In-package decontamination of chicken breast using cold plasma technology: microbial, quality and storage studies. *Meat Sci.* (2020) 159:107942. doi: 10.1016/j.meatsci.2019.107942
87. Lin HM, Zhang S, Zheng RS, Miao JY, Deng SG. Effect of atmospheric cold plasma treatment on ready-to-eat wine-pickled Bullacta exarata. *Lwt.* (2020) 120:108953. doi: 10.1016/j.lwt.2019.108953
88. Jahromi M, Niakousari M, Golmakani MT, Ajalloueian F, Khalesi M. Effect of dielectric barrier discharge atmospheric cold plasma treatment on structural, thermal and techno-functional characteristics of sodium caseinate. *Innov Food Sci Emerg Technol.* (2020) 66:102542. doi: 10.1016/j.ifset.2020.102542
89. Sharma S, Singh RK. Cold plasma treatment of dairy proteins in relation to functionality enhancement. *Trends Food Sci Technol.* (2020) 102:30–6. doi: 10.1016/j.tifs.2020.05.013
90. Gao Y, Zhuang H, Yeh HY, Bowker B, Zhang J. Effect of rosemary extract on microbial growth, pH, color, and lipid oxidation in cold plasma-processed ground chicken patties. *Innov Food Sci Emerg Technol.* (2019) 57:102168. doi: 10.1016/j.ifset.2019.05.007
91. Sarangapani C, Ryan Keogh D, Dunne J, Bourke P, Cullen PJ. Characterisation of cold plasma treated beef and dairy lipids using spectroscopic and chromatographic methods. *Food Chem.* (2017) 235:324–33. doi: 10.1016/j.foodchem.2017.05.016
92. Gavahian M, Chu YH, Mousavi Khaneghah A, Barba FJ, Misra NN. A critical analysis of the cold plasma induced lipid oxidation in foods. *Trends Food Sci Technol.* (2018) 77:32–41. doi: 10.1016/j.tifs.2018.04.009
93. Temelli F, Saldaña MDA, Comin L. Application of supercritical fluid extraction in food processing. In: Pawliszyn J, editor. *Comprehensive Sampling and Sample Preparation*. Vol. 4. Washington: Elsevier. (2012). p. 415–40. doi: 10.1016/B978-0-12-381373-2.00142-3
94. Deotale SM, Dutta S, Moses JA, Anandharamakrishnan C. Advances in supercritical carbon dioxide assisted sterilization of biological matrices. In: Knoerzer K, Juliano P, Smithers G, editors. *Innovative Food Processing Technologies.* Cambridge (2021). p. 660–77. doi: 10.1016/B978-0-08-100596-5.22932-6
95. Lefebvre T, Destandau E, Lesellier E. Sequential extraction of carnosic acid, rosmarinic acid and pigments (carotenoids and chlorophylls) from Rosemary by online supercritical fluid extraction-supercritical fluid chromatography. *J Chromatogr A.* (2020) 1639:461709. doi: 10.1016/j.chroma.2020.461709
96. Santos PH, Kammers JC, Silva AP, Vladimir J. Antioxidant and antibacterial compounds from feijoa leaf extracts obtained by pressurized liquid extraction and supercritical fluid extraction. *Food Chem.* (2020) 344:128620. doi: 10.1016/j.foodchem.2020.128620
97. Molino A, Mehariya S, Di Sanzo G, Larocca V, Martino M, Leone GP, et al. Recent developments in supercritical fluid extraction of bioactive compounds from microalgae: role of key parameters, technological achievements and challenges. *J CO2 Util.* (2020) 36:196–209. doi: 10.1016/j.jcou.2019.11.014
98. Pavlić B, Pezo L, Marić B, Tukuljac LP, Zeković Z, Solarov MB, et al. Supercritical fluid extraction of raspberry seed oil: experiments and modelling. *J Supercrit Fluids.* (2020) 157:104687. doi: 10.1016/j.supflu.2019.104687
99. Priyanka, Khanam S. Selection of suitable model for the supercritical fluid extraction of carrot seed oil: a parametric study. *Lwt.* (2020) 119:108815. doi: 10.1016/j.lwt.2019.108815
100. Santos OV, Lorenzo ND, Souza ALG, Costa CEF, Conceição LRV, Lannes SC da S, et al. CO2 supercritical fluid extraction of pulp and nut oils from *Terminalia catappa* fruits: thermogravimetric behavior, spectroscopic and fatty acid profiles. *Food Res Int.* (2020) 139:109814. doi: 10.1016/j.foodres.2020.109814
101. Ferrentino G, Giampiccolo S, Morozova K, Haman N, Spilimbergo S, Scampicchio M. Supercritical fluid extraction of oils from apple seeds: process optimization, chemical characterization and comparison with a conventional solvent extraction. *Innov Food Sci Emerg Technol.* (2020) 64:102428. doi: 10.1016/j.ifset.2020.102428
102. Al-Otoom A, Al-Asheh S, Allawzi M, Mahshi K, Alzenati N, Banat B, et al. Extraction of oil from uncrushed olives using supercritical fluid extraction method. *J Supercrit Fluids.* (2014) 95:512–8. doi: 10.1016/j.supflu.2014.10.023
103. Salea R, Veriansyah B, Tjandrawinata RR. Optimization and scale-up process for supercritical fluids extraction of ginger oil from Zingiber officinale var. *Amarum*. *J Supercrit Fluids.* (2017) 120:285–94. doi: 10.1016/j.supflu.2016.05.035
104. Rebolleda S, Rubio N, Beltrán S, Sanz MT, González-Sanjosé ML. Supercritical fluid extraction of corn germ oil: study of the influence of process parameters on the extraction yield and oil quality. *J Supercrit Fluids.* (2012) 72:270–7. doi: 10.1016/j.supflu.2012.10.001
105. De Oliveira PMA, De Almeida RH, De Oliveira NA, Bostyn S, Gonçalves CB, De Oliveira AL. Enrichment of diterpenes in green coffee oil using supercritical fluid extraction - characterization and comparison with green coffee oil from pressing. *J Supercrit Fluids.* (2014) 95:137–45. doi: 10.1016/j.supflu.2014.08.016
106. Priyanka, Khanam S. Influence of operating parameters on supercritical fluid extraction of essential oil from turmeric root. *J Clean Prod.* (2018) 188:816–24. doi: 10.1016/j.jclepro.2018.04.052
107. Fornari T, Vicente G, Vázquez E, García-Risco MR, Reglero G. Isolation of essential oil from different plants and herbs by supercritical fluid extraction. *J Chromatogr A.* (2012) 1250:34–48. doi: 10.1016/j.chroma.2012.04.051
108. Pinto D, De La Luz Cádiz-Gurrea M, Sut S, Ferreira AS, Leyva-Jimenez FJ, Dall'acqua S, et al. Valorisation of underexploited *Castanea sativa* shells bioactive compounds recovered by supercritical fluid extraction with CO2: a response surface methodology approach. *J CO2 Util.* (2020) 40:101194. doi: 10.1016/j.jcou.2020.101194
109. Gallego R, Bueno M, Herrero M. Sub- and supercritical fluid extraction of bioactive compounds from plants, food-by-products, seaweeds and microalgae – an update. *TrAC - Trends Anal Chem.* (2019) 116:198–213. doi: 10.1016/j.trac.2019.04.030
110. Torres-Ossandón MJ, Vega-Gálvez A, López J, Stucken K, Romero J, Di Scala K. Effects of high hydrostatic pressure processing and supercritical fluid extraction on bioactive compounds and antioxidant capacity of Cape gooseberry pulp (Physalis peruviana L.). *J Supercrit Fluids.* (2018) 138:215–20. doi: 10.1016/j.supflu.2018.05.005

111. Koubaa M, Mhemdi H, Fages J. Recovery of valuable components and inactivating microorganisms in the agro-food industry with ultrasound-assisted supercritical fluid technology. *J Supercrit Fluids.* (2018) 134:71–9. doi: 10.1016/j.supflu.2017.12.012
112. Spilimbergo S, Bertucco A. Non-thermal bacteria inactivation with dense CO2. *Biotechnol Bioeng.* (2003) 84:627–38. doi: 10.1002/bit.10783
113. Silva EK, Meireles MAA, Saldaña MDA. Supercritical carbon dioxide technology: a promising technique for the non-thermal processing of freshly fruit and vegetable juices. *Trends Food Sci Technol.* (2020) 97:381–90. doi: 10.1016/j.tifs.2020.01.025
114. Bertolini FM, Morbiato G, Facco P, Marszałek K, Pérez-Esteve É, Benedito J, et al. Optimization of the supercritical CO2 pasteurization process for the preservation of high nutritional value of pomegranate juice. *J Supercrit Fluids.* (2020) 164:1–11. doi: 10.1016/j.supflu.2020.104914
115. Cappelletti M, Ferrentino G, Spilimbergo S. Supercritical carbon dioxide combined with high power ultrasound: an effective method for the pasteurization of coconut water. *J Supercrit Fluids.* (2014) 92:257–63. doi: 10.1016/j.supflu.2014.06.010
116. Cappelletti M, Ferrentino G, Endrizzi I, Aprea E, Betta E, Corollaro ML, et al. High pressure carbon dioxide pasteurization of coconut water: a sport drink with high nutritional and sensory quality. *J Food Eng.* (2015) 145:73–81. doi: 10.1016/j.jfoodeng.2014.08.012
117. Smigic N, Djekic I, Tomic N, Udovicki B, Rajkovic A. The potential of foods treated with supercritical carbon dioxide (sc-CO 2) as novel foods. *Br Food J.* (2019) 121:815–34. doi: 10.1108/BFJ-03-2018-0168
118. Yu T, Iwahashi H. Conversion of waste meat to resources by enzymatic reaction under high pressure carbon dioxide conditions. *High Press Res.* (2019) 39:367–73. doi: 10.1080/08957959.2019.1593406
119. Farkas J. Irradiation for better foods. *Trends Food Sci Technol.* (2006) 17:148–52. doi: 10.1016/j.tifs.2005.12.003
120. Bashir K, Jan K, Kamble DB, Maurya VK, Jan S, Swer TL. History, status and regulatory aspects of gamma irradiation for food processing. In: Knoerzer K, Juliano P, Smithers G, editors. *Innovative Food Processing Technologies.* Cambridge (2021). p. 101–7. doi: 10.1016/B978-0-08-100596-5.23051-5
121. Jan K, Bashir K, Maurya VK. Gamma irradiation and food properties. In: Knoerzer K, Juliano P, Smithers G, editors. *Innovative Food Processing Technologies.* Cambridge (2021). p. 41–60. doi: 10.1016/B978-0-08-100596-5.23052-7
122. Castell-Perez ME, Moreira RG. Irradiation and consumers acceptance. In: Knoerzer K, Juliano P, Smithers G, editors. *Innovative Food Processing Technologies.* Cambridge (2021). p. 122–35. doi: 10.1016/B978-0-12-815781-7.00015-9
123. Robichaud V, Bagheri L, Aguilar-Uscanga BR, Millette M, Lacroix M. Effect of ϒ-irradiation on the microbial inactivation, nutritional value, and antioxidant activities of infant formula. *Lwt.* (2020) 125:109211. doi: 10.1016/j.lwt.2020.109211
124. Gaougaou G, Shankar S, Liot Q, Constant P, Déziel E, Lacroix M. Gamma irradiation triggers a global stress response in *Escherichia coli* O157:H7 including base and nucleotides excision repair pathways. *Microb Pathog.* (2020) 149:104342. doi: 10.1016/j.micpath.2020.104342
125. Fallah AA, Siavash Saei-Dehkordi S, Rahnama M. Enhancement of microbial quality and inactivation of pathogenic bacteria by gamma irradiation of ready-to-cook Iranian barbecued chicken. *Radiat Phys Chem.* (2010) 79:1073–8. doi: 10.1016/j.radphyschem.2010.04.015
126. Shalaby AR, Anwar MM, Sallam EM, Emam WH. Quality and safety of irradiated food regarding biogenic amines: Ras cheese. *Int J Food Sci Technol.* (2016) 51:1048–54. doi: 10.1111/ijfs.13058
127. Li C, He L, Jin G, Ma S, Wu W, Gai L. Effect of different irradiation dose treatment on the lipid oxidation, instrumental color and volatiles of fresh pork and their changes during storage. *Meat Sci.* (2017) 128:68–76. doi: 10.1016/j.meatsci.2017.02.009
128. Bashir K, Jan K, Aggarwal M. Thermo-rheological and functional properties of gamma-irradiated wholewheat flour. *Int J Food Sci Technol.* (2017) 52:927–35. doi: 10.1111/ijfs.13356
129. Ghabraie M, Vu KD, Tnani S, Lacroix M. Antibacterial effects of 16 formulations and irradiation against Clostridium sporogenes in a sausage model. *Food Control.* (2016) 63:21–7. doi: 10.1016/j.foodcont.2015.11.019
130. Cassares M, Sakotani NL, Kunigk L, Vasquez PAS, Jurkiewicz C. Effect of gamma irradiation on shelf life extension of fresh pasta. *Radiat Phys Chem.* (2020) 174:108940. doi: 10.1016/j.radphyschem.2020.108940
131. Bhat NA, Wani IA, Hamdani AM, Masoodi FA. Effect of gamma-irradiation on the thermal, rheological and antioxidant properties of three wheat cultivars grown in temperate Indian climate. *Radiat Phys Chem.* (2020) 176:108953. doi: 10.1016/j.radphyschem.2020.108953
132. Sharma P, Sharma SR, Dhall RK, Mittal TC, Bhatia S. Physio-chemical behavior of γ-irradiated garlic bulbs under ambient storage conditions. *J Stored Prod Res.* (2020) 87:101629. doi: 10.1016/j.jspr.2020.101629
133. Carvalho Mesquita T, Evangelista Vasconcelos Schiassi MC, Maria Teixeira Lago A, Careli-Gondim Í, Mesquita Silva L, de Azevedo Lira N, et al. Grape juice blends treated with gamma irradiation evaluated during storage. *Radiat Phys Chem.* (2020) 168:108570. doi: 10.1016/j.radphyschem.2019.108570
134. Syauqi A, Dadang D, Harahap IS, Indarwatmi M. Gamma irradiation against mealybug Dysmicoccus lepelleyi (Betrem) (Hemiptera: Pseudococcidae) on mangosteen fruit (Garcinia mangostana L.) as a quarantine treatment. *Radiat Phys Chem.* (2020) 179:108954. doi: 10.1016/j.radphyschem.2020.108954
135. Lim JS, Ha JW. Effect of acid adaptation on the resistance of *Escherichia coli* O157:H7 and Salmonella enterica serovar Typhimurium to X-ray irradiation in apple juice. *Food Control.* (2021) 120:107489. doi: 10.1016/j.foodcont.2020.107489
136. Arshad RN, Abdul-Malek Z, Munir A, Buntat Z, Ahmad MH, Jusoh YMM, et al. Electrical systems for pulsed electric field applications in the food industry: an engineering perspective. *Trends Food Sci Technol.* (2020) 104:1–13. doi: 10.1016/j.tifs.2020.07.008
137. Aadil RM, Zeng XA, Ali A, Zeng F, Farooq MA, Han Z, et al. Influence of different pulsed electric field strengths on the quality of the grapefruit juice. *Int J Food Sci Technol.* (2015) 50:2290–6. doi: 10.1111/ijfs.12891
138. Barba FJ, Parniakov O, Pereira SA, Wiktor A, Grimi N, Boussetta N, et al. Current applications and new opportunities for the use of pulsed electric fields in food science and industry. *Food Res Int.* (2015) 77:773–98. doi: 10.1016/j.foodres.2015.09.015
139. Preetha P, Pandiselvam R, Varadharaju N, Kennedy ZJ, Balakrishnan M, Kothakota A. Effect of pulsed light treatment on inactivation kinetics of *Escherichia coli* (MTCC 433) in fruit juices. *Food Control.* (2021) 121:107547. doi: 10.1016/j.foodcont.2020.107547
140. Timmermans RAH, Mastwijk HC, Berendsen LBJM, Nederhoff AL, Matser AM, Van Boekel MAJS, et al. Moderate intensity Pulsed Electric Fields (PEF) as alternative mild preservation technology for fruit juice. *Int J Food Microbiol.* (2019) 298:63–73. doi: 10.1016/j.ijfoodmicro.2019.02.015
141. Toepfl S, Heinz V, Knorr D. High intensity pulsed electric fields applied for food preservation. *Chem Eng Process Process Intensif.* (2007) 46:537–46. doi: 10.1016/j.cep.2006.07.011
142. Mannozzi C, Rompoonpol K, Fauster T, Tylewicz U, Romani S, Rosa MD, et al. Influence of pulsed electric field and ohmic heating pretreatments on enzyme and antioxidant activity of fruit and vegetable juices. *Foods.* (2019) 8:247. doi: 10.3390/foods8070247
143. Liang R, Zhang Z, Lin S. Effects of pulsed electric field on intracellular antioxidant activity and antioxidant enzyme regulating capacities of pine nut (Pinus koraiensis) peptide QDHCH in HepG2 cells. *Food Chem.* (2017) 237:793–802. doi: 10.1016/j.foodchem.2017.05.144
144. López-Gámez G, Elez-Martínez P, Martín-Belloso O, Soliva-Fortuny R. Pulsed electric fields affect endogenous enzyme activities, respiration and biosynthesis of phenolic compounds in carrots. *Postharvest Biol Technol.* (2020) 168:111284. doi: 10.1016/j.postharvbio.2020.111284
145. Käferböck A, Smetana S, de Vos R, Schwarz C, Toepfl S, Parniakov O. Sustainable extraction of valuable components from Spirulina assisted by pulsed electric fields technology. *Algal Res.* (2020) 48:101914. doi: 10.1016/j.algal.2020.101914
146. Gateau H, Blanckaert V, Veidl B, Burlet-Schiltz O, Pichereaux C, Gargaros A, et al. Application of pulsed electric fields for the biocompatible extraction of proteins from the microalga *Haematococcus pluvialis*. *Bioelectrochemistry.* (2021) 137:107588. doi: 10.1016/j.bioelechem.2020.107588
147. Wang L, Boussetta N, Lebovka N, Vorobiev E. Cell disintegration of apple peels induced by pulsed electric field and efficiency of bio-compound extraction. *Food Bioprod Process.* (2020) 122:13–21. doi: 10.1016/j.fbp.2020.03.004

148. Chittapun S, Jonjaroen V, Khumrangsee K, Charoenrat T. C-phycocyanin extraction from two freshwater cyanobacteria by freeze thaw and pulsed electric field techniques to improve extraction efficiency and purity. *Algal Res.* (2020) 46:101789. doi: 10.1016/j.algal.2020.101789
149. Pataro G, Carullo D, Falcone M, Ferrari G. Recovery of lycopene from industrially derived tomato processing by-products by pulsed electric fields-assisted extraction. *Innov Food Sci Emerg Technol.* (2020) 63:102369. doi: 10.1016/j.ifset.2020.102369
150. Pashazadeh B, Elhamirad AH, Hajnajari H, Sharayei P, Armin M. Optimization of the pulsed electric field -assisted extraction of functional compounds from cinnamon. *Biocatal Agric Biotechnol.* (2020) 23:101461. doi: 10.1016/j.bcab.2019.101461
151. Liu C, Pirozzi A, Ferrari G, Vorobiev E, Grimi N. Impact of pulsed electric fields on vacuum drying kinetics and physicochemical properties of carrot. *Food Res Int.* (2020) 137:109658. doi: 10.1016/j.foodres.2020.109658
152. Rybak K, Samborska K, Jedlinska A, Parniakov O, Nowacka M, Witrowa-Rajchert D, et al. The impact of pulsed electric field pretreatment of bell pepper on the selected properties of spray dried juice. *Innov Food Sci Emerg Technol.* (2020) 65:102446. doi: 10.1016/j.ifset.2020.102446
153. Li J, Shi J, Huang X, Zou X, Li Z, Zhang D, et al. Effects of pulsed electric field on freeze-thaw quality of Atlantic salmon. *Innov Food Sci Emerg Technol.* (2020) 65:102454. doi: 10.1016/j.ifset.2020.102454
154. Zhang F, Tian M, Du M, Fang T. Enhancing the activity of pectinase using pulsed electric field (PEF) treatment. *J Food Eng.* (2017) 205:56–63. doi: 10.1016/j.jfoodeng.2017.02.023
155. Dong M, Xu Y, Zhang Y, Han M, Wang P, Xu X, et al. Physicochemical and structural properties of myofibrillar proteins isolated from pale, soft, exudative (PSE)-like chicken breast meat: effects of pulsed electric field (PEF). *Innov Food Sci Emerg Technol.* (2020) 59:102277. doi: 10.1016/j.ifset.2019.102277
156. Zhu F. Modifications of starch by electric field based techniques. *Trends Food Sci Technol.* (2018) 75:158–69. doi: 10.1016/j.tifs.2018.03.011
157. Chen BR, Wen QH, Zeng XA, Abdul R, Roobab U, Xu FY. Pulsed electric field assisted modification of octenyl succinylated potato starch and its influence on pasting properties. *Carbohydr Polym.* (2020) 254:117294. doi: 10.1016/j.carbpol.2020.117294
158. Duque SMM, Leong SY, Agyei D, Singh J, Larsen N, Oey I. Understanding the impact of Pulsed Electric Fields treatment on the thermal and pasting properties of raw and thermally processed oat flours. *Food Res Int.* (2020) 129:108839. doi: 10.1016/j.foodres.2019.108839
159. Lin ZR, Zeng XA, Yu SJ, Sun DW. Enhancement of ethanol-acetic acid esterification under room temperature and non-catalytic condition via pulsed electric field application. *Food Bioprocess Technol.* (2012) 5:2637–45. doi: 10.1007/s11947-011-0678-4
160. Zhang ZH, Han Z, Zeng XA, Wang MS. The preparation of Fe-glycine complexes by a novel method (pulsed electric fields). *Food Chem.* (2017) 219:468–76. doi: 10.1016/j.foodchem.2016.09.129
161. Van Loey IA, Smout C, Hendrickx M. High hydrostatic pressure technology in food preservation. In: Zeuthen P, Bogh-Sorensen L, editors. *Food Preservation Techniques.* Cambridge (2003). p. 428–48. doi: 10.1533/9781855737143.3.428
162. Huang HW, Hsu CP, Wang CY. Healthy expectations of high hydrostatic pressure treatment in food processing industry. *J Food Drug Anal.* (2020) 28:1–13. doi: 10.1016/j.jfda.2019.10.002
163. Rendueles E, Omer MK, Alvseike O, Alonso-Calleja C, Capita R, Prieto M. Microbiological food safety assessment of high hydrostatic pressure processing: a review. *LWT - Food Sci Technol.* (2011) 44:1251–60. doi: 10.1016/j.lwt.2010.11.001
164. González-Cebrino F, Durán R, Delgado-Adámez J, Contador R, Ramírez R. Changes after high-pressure processing on physicochemical parameters, bioactive compounds, and polyphenol oxidase activity of red flesh and peel plum purée. *Innov Food Sci Emerg Technol.* (2013) 20:34–41. doi: 10.1016/j.ifset.2013.07.008
165. Daher D, Le Gourrierec S, Pérez-Lamela C. Effect of high pressure processing on the microbial inactivation in fruit preparations and other vegetable based beverages. *Agric.* (2017) 7:1–18. doi: 10.3390/agriculture7090072
166. Bulut S, Karatzas KAG. Inactivation of *Escherichia coli* K12 in phosphate buffer saline and orange juice by high hydrostatic pressure processing combined with freezing. *Lwt.* (2021) 136:110313. doi: 10.1016/j.lwt.2020.110313
167. Cap M, Paredes PF, Fernández D, Mozgovoj M, Vaudagna SR, Rodriguez A. Effect of high hydrostatic pressure on Salmonella spp inactivation and meat-quality of frozen chicken breast. *Lwt.* (2020) 118:108873. doi: 10.1016/j.lwt.2019.108873
168. Cava R, García-Parra J, Ladero L. Effect of high hydrostatic pressure processing and storage temperature on food safety, microbial counts, colour and oxidative changes of a traditional dry-cured sausage. *Lwt.* (2020) 128:109462. doi: 10.1016/j.lwt.2020.109462
169. de Jesus ALT, Cristianini M, dos Santos NM, Maróstica Júnior MR. Effects of high hydrostatic pressure on the microbial inactivation and extraction of bioactive compounds from açaí (Euterpe oleracea Martius) pulp. *Food Res Int.* (2020) 130:108856. doi: 10.1016/j.foodres.2019.108856
170. Ninčević Grassino A, Ostojić J, Miletić V, Djaković S, Bosiljkov T, Zorić Z, et al. Application of high hydrostatic pressure and ultrasound-assisted extractions as a novel approach for pectin and polyphenols recovery from tomato peel waste. *Innov Food Sci Emerg Technol.* (2020) 64:102424. doi: 10.1016/j.ifset.2020.102424
171. Cascaes Teles AS, Hidalgo Chávez DW, Zarur Coelho MA, Rosenthal A, Fortes Gottschalk LM, Tonon RV. Combination of enzyme-assisted extraction and high hydrostatic pressure for phenolic compounds recovery from grape pomace. *J Food Eng.* (2020) 288:110128. doi: 10.1016/j.jfoodeng.2020.110128
172. Suwal S, Perreault V, Marciniak A, Tamigneaux É, Deslandes É, Bazinet L, et al. Effects of high hydrostatic pressure and polysaccharidases on the extraction of antioxidant compounds from red macroalgae, *Palmaria palmata* and *Solieria chordalis*. *J Food Eng.* (2019) 252:53–9. doi: 10.1016/j.jfoodeng.2019.02.014
173. Naderi N, Pouliot Y, House JD, Doyen A. High hydrostatic pressure effect in extraction of 5-methyltetrahydrofolate (5-MTHF) from egg yolk and granule fractions. *Innov Food Sci Emerg Technol.* (2017) 43:191–200. doi: 10.1016/j.ifset.2017.08.009
174. Rios-Corripio G, Welti-Chanes J, Rodríguez-Martínez V, Guerrero-Beltrán JÁ. Influence of high hydrostatic pressure processing on physicochemical characteristics of a fermented pomegranate (Punica granatum L.) beverage. *Innov Food Sci Emerg Technol.* (2020) 59:102249. doi: 10.1016/j.ifset.2019.102249
175. Malinowska-Pańczyk E. Can high hydrostatic pressure processing be the best way to preserve human milk? *Trends Food Sci Technol.* (2020) 101:133–8. doi: 10.1016/j.tifs.2020.05.009
176. Rocha-Pimienta J, Martillanes S, Ramírez R, Garcia-Parra J, Delgado-Adamez J. *Bacillus cereus* spores and *Staphylococcus aureus* sub. *aureus* vegetative cells inactivation in human milk by high-pressure processing. *Food Control.* (2020) 113:107212. doi: 10.1016/j.foodcont.2020.107212
177. Carullo D, Barbosa-Cánovas GV, Ferrari G. Changes of structural and techno-functional properties of high hydrostatic pressure (HHP) treated whey protein isolate over refrigerated storage. *Lwt.* (2020) 137:110436. doi: 10.1016/j.lwt.2020.110436
178. Popović V, Koutchma T, Pagan J. Emerging applications of ultraviolet light-emitting diodes for foods and beverages. In: Knoerzer K, Muthukumarappan K, editors. *Innovative Food Processing Technologies.* Voctoria (2021). p. 335–44. doi: 10.1016/B978-0-08-100596-5.22667-X
179. Guerrero-Beltrán JÁ, Ochoa-Velasco CE. Ultraviolet-C light technology and systems for preservation of fruit juices and beverages. In: Knoerzer K, Muthukumarappan K, editors. *Innovative Food Processing Technologies.* Voctoria (2021). p. 210–26. doi: 10.1016/B978-0-08-100596-5.22937-5
180. Koutchma T, Bissonnette S, Popović V. An update on research, development and implementation of UV and pulsed light technologies for nonthermal preservation of milk and dairy products. In: Knoerzer K, Muthukumarappan K, editors. *Innovative Food Processing Technologies.* Voctoria (2021). p. 256–76. doi: 10.1016/B978-0-08-100596-5.22680-2

181. Fenoglio D, Ferrario M, Schenk M, Guerrero S. Effect of pilot-scale UV-C light treatment assisted by mild heat on *E. coli, L. plantarum and S. cerevisiae* inactivation in clear and turbid fruit juices. storage study of surviving populations. *Int J Food Microbiol.* (2020) 332:108767. doi: 10.1016/j.ijfoodmicro.2020.108767
182. Xiang Q, Fan L, Zhang R, Ma Y, Liu S, Bai Y. Effect of UVC light-emitting diodes on apple juice: inactivation of Zygosaccharomyces rouxii and determination of quality. *Food Control.* (2020) 111:107082. doi: 10.1016/j.foodcont.2019.107082
183. Ferreira TV, Mizuta AG, Menezes JL de, Dutra TV, Bonin E, Castro JC, et al. Effect of ultraviolet treatment (UV–C) combined with nisin on industrialized orange juice in *Alicyclobacillus acidoterrestris* spores. *Lwt.* (2020) 133:109911. doi: 10.1016/j.lwt.2020.109911
184. Fundo JF, Miller FA, Mandro GF, Tremarin A, Brandão TRS, Silva CLM. UV-C light processing of Cantaloupe melon juice: evaluation of the impact on microbiological, and some quality characteristics, during refrigerated storage. *Lwt.* (2019) 103:247–52. doi: 10.1016/j.lwt.2019.01.025
185. Delorme MM, Guimarães JT, Coutinho NM, Balthazar CF, Rocha RS, Silva R, et al. Ultraviolet radiation: an interesting technology to preserve quality and safety of milk and dairy foods. *Trends Food Sci Technol.* (2020) 102:146–54. doi: 10.1016/j.tifs.2020.06.001
186. Kumar A, Rani P, Purohit SR, Rao PS. Effect of ultraviolet irradiation on wheat (Triticum aestivum) flour: study on protein modification and changes in quality attributes. *J Cereal Sci.* (2020) 96:103094. doi: 10.1016/j.jcs.2020.103094
187. Dyshlyuk L, Babich O, Prosekov A, Ivanova S, Pavsky V, Chaplygina T. The effect of postharvest ultraviolet irradiation on the content of antioxidant compounds and the activity of antioxidant enzymes in tomato. *Heliyon.* (2020) 6:e03288. doi: 10.1016/j.heliyon.2020.e03288
188. Zhu Y, Koutchma T. UV light technology for mycotoxins reduction in foods and beverages. In: Knoerzer K, Muthukumarappan K, editors. *Innovative Food Processing Technologies.* Voctoria (2021). p. 398–415. doi: 10.1016/B978-0-08-100596-5.22686-3
189. Orlowska M, Koutchma T, Grapperhaus M, Gallagher J, Schaefer R, Defelice C. Continuous and pulsed ultraviolet light for nonthermal treatment of liquid foods. Part 1: effects on quality of fructose solution, apple juice, and milk. *Food Bioprocess Technol.* (2013) 6:1580–92. doi: 10.1007/s11947-012-0779-8
190. Jeon MJ, Ha JW. Inactivating foodborne pathogens in apple juice by combined treatment with fumaric acid and ultraviolet-A light, and mechanisms of their synergistic bactericidal action. *Food Microbiol.* (2020) 87:103387. doi: 10.1016/j.fm.2019.103387
191. Mehta D, Sharma N, Bansal V, Sangwan RS, Yadav SK. Impact of ultrasonication, ultraviolet and atmospheric cold plasma processing on quality parameters of tomato-based beverage in comparison with thermal processing. *Innov Food Sci Emerg Technol.* (2019) 52:343–9. doi: 10.1016/j.ifset.2019.01.015
192. Oner ME, Demirci A. Ozone for food decontamination: theory and applications. In: Lelieveld H, Gabric D, Holah J, editors. *Handbook of Hygiene Control in the Food Industry: Second Edition*. Cambridge: Elsevier Ltd (2016). p. 491–501. doi: 10.1016/B978-0-08-100155-4.00033-9
193. Tiwari BK, Brennan CS, Curran T, Gallagher E, Cullen PJ, O' Donnell CP. Application of ozone in grain processing. *J Cereal Sci.* (2010) 51:248–55. doi: 10.1016/j.jcs.2010.01.007
194. Giménez B, Graiver N, Giannuzzi L, Zaritzky N. Treatment of beef with gaseous ozone: physicochemical aspects and antimicrobial effects on heterotrophic microflora and listeria monocytogenes. *Food Control.* (2021) 121:1–9. doi: 10.1016/j.foodcont.2020.107602
195. Mohammad Z, Kalbasi-Ashtari A, Riskowski G, Juneja V, Castillo A. Inactivation of Salmonella and Shiga toxin-producing *Escherichia coli* (STEC) from the surface of alfalfa seeds and sprouts by combined antimicrobial treatments using ozone and electrolyzed water. *Food Res Int.* (2020) 136:109488. doi: 10.1016/j.foodres.2020.109488
196. Taiye Mustapha A, Zhou C, Wahia H, Amanor-Atiemoh R, Otu P, Qudus A, et al. Sonozonation: enhancing the antimicrobial efficiency of aqueous ozone washing techniques on cherry tomato. *Ultrason Sonochem.* (2020) 64:105059. doi: 10.1016/j.ultsonch.2020.105059
197. Pinto L, Palma A, Cefola M, Pace B, D'Aquino S, Carboni C, et al. Effect of modified atmosphere packaging (MAP) and gaseous ozone pre-packaging treatment on the physico-chemical, microbiological and sensory quality of small berry fruit. *Food Packag Shelf Life.* (2020) 26:100573. doi: 10.1016/j.fpsl.2020.100573
198. Patil S, Valdramidis VP, Cullen PJ, Frias J, Bourke P. Inactivation of *Escherichia coli* by ozone treatment of apple juice at different pH levels. *Food Microbiol.* (2010) 27:835–40. doi: 10.1016/j.fm.2010.05.002
199. Choi MR, Liu Q, Lee SY, Jin JH, Ryu S, Kang DH. Inactivation of *Escherichia coli* O157:H7, Salmonella typhimurium and Listeria monocytogenes in apple juice with gaseous ozone. *Food Microbiol.* (2012) 32:191–5. doi: 10.1016/j.fm.2012.03.002
200. Porto E, Alves Filho EG, Silva LMA, Fonteles TV, do Nascimento RBR, Fernandes FAN, et al. Ozone and plasma processing effect on green coconut water. *Food Res Int.* (2020) 131:109000. doi: 10.1016/j.foodres.2020.109000

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Shelf-Life Extension of Refrigerated Turbot (*Scophthalmus maximus*) by Using Weakly Acidic Electrolyzed Water and Active Coatings Containing Daphnetin Emulsions

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This research was to investigate the effect of weakly acidic electrolytic water (WAEW) treatments combining with the locust bean gum (LBG) and sodium alginate (SA) active coatings, containing daphnetin emulsions on microbiological, physicochemical, and sensory changes of turbot (*Scophthalmus maximus*) during refrigerated storage at 4°C for 24 days. Results showed that WAEW, together with LBG-SA coatings containing daphnetin emulsions treatments, could significantly lower the total viable count (TVC), H_2S-producing bacteria, pseudomonas spp., and psychrotrophic bacteria counts, and inhibit the productions of off-flavor compounds, including the total volatile basic nitrogen (TVB-N), inosine (HxR), and hypoxanthine (Hx). Furthermore, the treatments also prevented textural deterioration, delayed water migration, and had higher organoleptic evaluation results. Therefore, WAEW, together with LBG-SA coatings, containing daphnetin emulsions treatments, had the potential to improve the quality of turbot during refrigerated storage.

Keywords: active coating, daphnetin, turbot, quality, shelf-life

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INTRODUCTION

Turbot (*Scophthalmusmaximus*) has high economic value and nutritional value and is widely cultivated in China (1). The flavor and high glial protein contents of turbot make it an economically important fish in high demand (2). However, fresh turbot is perishable due to chemical and biological changes, and its organoleptic properties have easily deteriorated during refrigerated storage (3).

The quality deterioration of fish after death results from the microbiological spoilage and biochemical reactions. The specifics spoilage organisms (SSOs) are considered to play a key role in the fish spoilage process (4). Weak acid electrolytic water (WAEW) is produced by the electrolysis of dilute sodium chloride or hydrochloric acid solution and exhibits strong antibacterial activities against SSOs, which has been considered as a new sanitizer (5). WAEW kills the SSOs physically, and it does not generate resistance (6). The antimicrobial mechanism of WAEW may be related to

the damage of the microbial cell protective barrier, the changes of the cell membrane permeability, the leakage of inclusions, and the inactivation of some key enzymes (7). WAEW has been tested against the main foodborne pathogens, including *Escherichia coli*, *Listeria monocytogenes*, *salmonella*, and *staphylococcus aureus* (8). Palotás et al. (9) reported that a common carp (*Cyprinus carpio*) treated with WAEW (100-mg/kg chloride ion concentration for 5 min) had additional bactericidal efficacy on the surface of the carp fillets and increased the shelf life of the samples, causing 2.4-lg CFU/g decrease, compared with the control by the end of the 7-day storage at 2°C. Khazandi et al. (10) stated that WAEW (either 45 or 150 mg/kg of free chlorine) significantly reduced the total bacterial load and SSOs on King George whiting and Tasmanian Atlantic salmon fillets (about 1–2 lg CFU/g) during storage at 4°C and significantly extended the shelf life of the fillets by 2 and 4 days, respectively.

Active coatings could retard the chemical and microbiological deteriorations of foods by serving as both gas barriers and carriers of food antioxidant or antimicrobial agents (11). The sources of proteins, lipids, or polysaccharides have been researched to meet the demand for novel and environmentally sustainable biomaterials for active coatings (12). Plant-based gums are promising materials for fish preservation because they can keep good quality and prolong the shelf life of foods by increasing a water barrier, reducing microbial contamination, maintaining the flavor, retarding reducing the degree of shrinkage distortion, and preventing fat oxidation (13). However, the active coatings have limitations in antimicrobial and antioxidative properties. The combination of antimicrobial or antioxidative agents with active coatings for fish preservation is of great interest (11). Among natural preservatives, plant polyphenols possess multiple biological functions in inhibiting lipid oxidation, enzymatic reactions, and food spoilage by microorganisms (14). Bazargani-Gilani and Pajohi-Alamoti (15) reported that sodium alginate (SA) active coating containing resveratrol inhibited the increase of pH, peroxide, and K values of rainbow trout fillets and extended the shelf life during refrigerated storage. Cao et al. (16) showed chitosan coating containing chlorogenic acid could inhibit lipid and protein oxidation in snakehead fish fillets stored at 2°C for 5 months. Nie et al. (17) reported the pectin coating infused with gallic acid had lower levels of total volatile basic nitrogen (TVB-N), lipid oxidation, and total sulfhydryls of Japanese sea bass fillets stored at 4°C. Besides, active coatings have been successfully combined, within a hurdle strategy, with other technologies as, for example, WAEW. Luan et al. (18) reported that the chitosan active coating combined with WAEW treatments could retard hairtail spoilage and slow down the protein deterioration of a hairtail (*Trichiutus haumela*) fillet during cold storage at −3°C, which extended the shelf life of hairtail to 6–7 days. Feng et al. (19) stated that active coatings containing epsilon-polylysine hydrochloride and rosemary extract combined with WAEW could effectively inhibit microbial growth and delay the increase in TVB-N, thiobarbituric acid and metmyoglobin value in puffer fish (*Takifugu obscurus*) during refrigerated storage at 4°C, which extended the shelf life to 14 days.

Daphnetin (7, 8-dihydroxycoumarin) is a dihydroxylated derivative of coumarin derived from plants and has been reported to possess antimicrobial, antioxidant, antimalarial, anticoagulation, and immunomodulating activities (20). Despite the fact that there were some studies on daphnetin extending the shelf life of fish during refrigerated storage (21), little is known about the effect of WAEW combining with active coatings containing daphnetin emulsions on fish preservation. Therefore, the objective of the present study was to evaluate the preservative effects of WAEW combining with locust bean gum (LBG) and SA-based active coatings containing different concentrations of daphnetin emulsions (0.16, 0.32, and 0.64 g/L, respectively) on turbot during refrigerated storage at 4°C in the aspects of microbiological analysis, TVB-N,K-values, free amino acids (FAAs), water distribution and migration, texture profile analysis (TPA), and organoleptic evaluation.

MATERIALS AND METHODS

Preparation of Turbot Samples

WAEW was generated by electrolyzing hydrochloric acid solution (3%) in a chamber, using a continuous electrolysis generator (FX-SWS100, Fangxin Water Treatment Equipment Co. Ltd., Yantai, China). The parameters of the WAEW were as follows: the concentration of hypochlorous acid was 30 mg/kg; the oxidation-reduction potential was 1,100 mV, and the pH value was 6.5. The concentrations of daphnetin emulsions used in the present study were 0.16, 0.32, and 0.64 g/L, respectively, and prepared according to our previous research (21). The final active coating solutions were marked as LBG-SA-0.16D, LBG-SA-0.32D, and LBG-SA-0.64D, respectively.

Fresh turbot weighing 500 ± 50 g was purchased from a local market in Luchao Port and randomly divided into five batches: (i) washed with deionized water (CK); (ii) immersed in WAEW for 10 min, followed by a coating with an LBG-SA-active coating (0D); (iii) immersed in WAEW for 10 min, followed by a coating with an LBG-SA-0.16D-active coating (0.16D); (iv) immersed in WAEW for 10 min, followed by a coating with an LBG-SA-0.32D-active coating (0.32D); (vi) and immersed in WAEW for 10 min, followed by a coating with an LBG-SA-0.64D-active coating (0.64D). Then the treated turbot samples were individually packed in a sterile polyethylene bag and stored at 4°C. Turbot samples were randomly sampled for quality analysis on 0, 4, 8, 12, 16, 20, and 24th days, respectively.

Microbiological Analysis

About 5-g turbot samples and 45-ml normal saline were fully homogenized and subjected to serial dilutions. The microbiological analyses were carried out (22): (i) determination of total viable counts (TVC) on a plate count agar medium was cultivated at 30°C for 48 h; (ii) determination of H_2S-producing bacteria on an iron agar medium was cultivated at 30°C for 48 h; (iii) determination of *Pseudomonas* spp. on a *Pseudomonas* CFC selective agar medium was cultivated at 30°C for 48 h; (iv) determination of psychrophilic bacteria on a plate count agar medium was cultivated at 4°C for 7 days. The final calculation

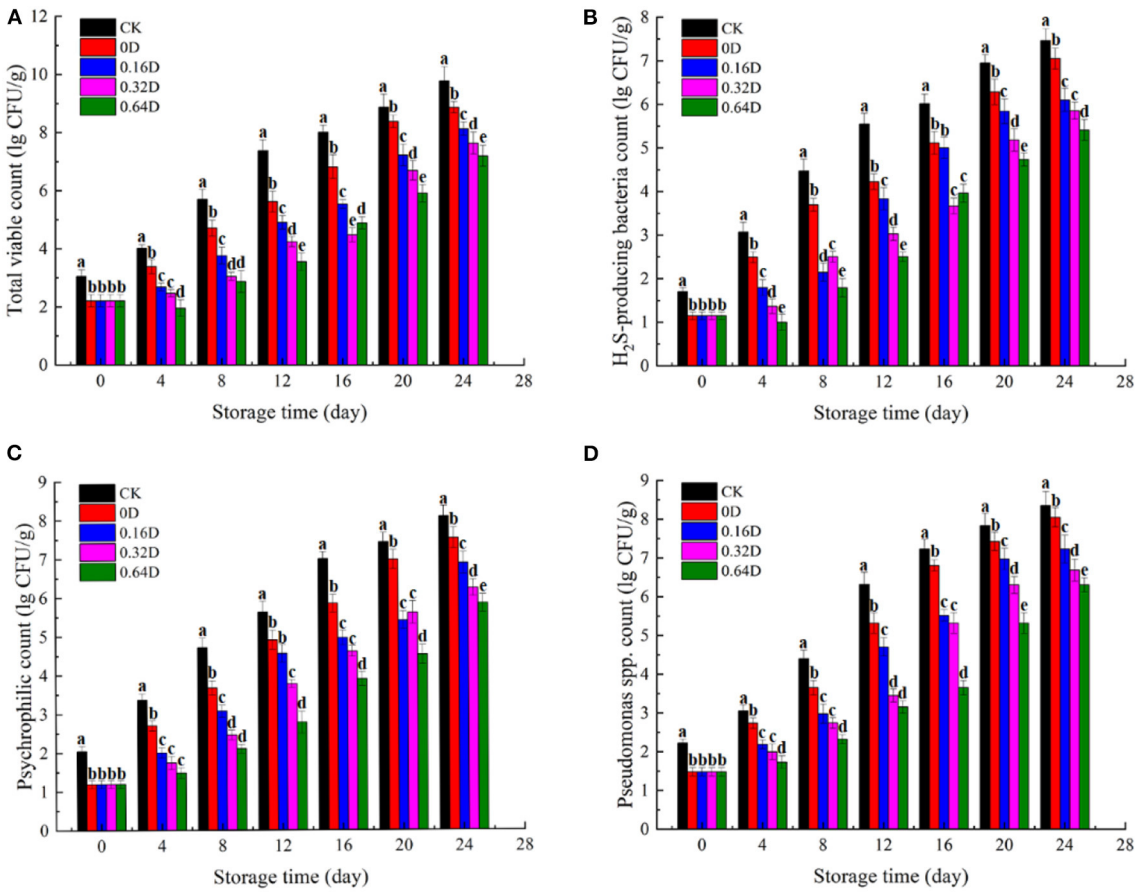

FIGURE 1 | Changes in **(A)** total viable counts, **(B)** H_2S-producing bacteria counts, **(C)** psychrophilic counts, and **(D)** *Pseudomonas* spp. counts of turbot samples during refrigerated storage [CK, washed with deionized water; 0D, immersed in weakly acidic electrolytic water (WAEW) for 10 min, followed by a coating with a locust bean gum–sodium alginate (LBG-SA)-active coating; 0.16D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.16 g/L daphnetin; 0.32D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.32 g/L daphnetin; 0.64D: immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.64 g/L daphnetin].

result was the logarithm of the mean of colony forming units (CFU) on the culture medium with 30–300 colonies.

Determination of TVB-N

TVB-N determination was performed with the method of Zhuang et al. (23). In brief, 5.0-g minced turbot flesh was homogenized with 45-ml deionized water. The homogenate was stirred at 25°C for 30 min, and then centrifuged at 3,040 × g for 5 min. Subsequently, 5.0 ml of the supernatant was taken to determine the TVB-N content, using the Kjeldahl nitrogen-determination instrument (Kjeltec 8,400, Foss, Denmark). TVB-N content was expressed as a mg N/100-g turbot sample.

Determination of K Value

The ATP-related compounds were measured by HPLC (Waters 2,695, Milford, CT, USA) according to Cao et al. (24). The K value was determined according to the following concentration ratio:

$$K\text{value}\% = \frac{\text{HxR} + \text{Hx}}{\text{ATP} + \text{ADP} + \text{AMP} + \text{IMP} + \text{HxR} + \text{Hx}} \times 100$$

Determination of FAAs

FAAs were performed as described by Yu et al. (25) using, an amino acid analyzer (Hitachi L-8800, Tokyo, Japan). The FAAs identification and quantification were completed by the retention time and peak area with reference to FAAs standards (Sigma Chemical Co. St Louis, MO).

Low-Field Nuclear Magnetic Resonance Analysis

The distribution and migration of water in turbot samples were evaluated through proton relaxation experiments, using an LF-NMR analyzer (NiumagMesoMR23-60H.I, Suzhou, China), with a proton resonance frequency of 21 MHz (corresponding to the pulse sequence of Carr–Purcell–Meiboom–Gill) (26). Thedorsal muscle of turbot samples were cut into 3 × 2 × 1.5 cm (about 5 g) and wrapped with polyethylene film. For each measurement, 16 scans were performed with 3,000 echoes.

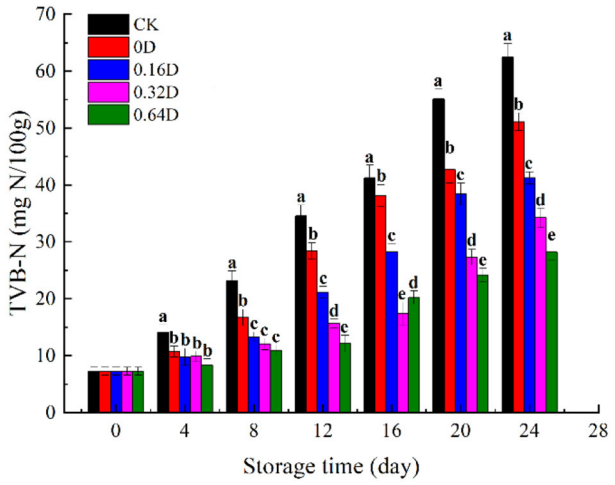

FIGURE 2 | Changes in total volatile basic nitrogen values (TVB-N) of turbot samples during refrigerated storage [CK, washed with deionized water; 0D, immersed in weakly acidic electrolytic water (WAEW) for 10 min, followed by a coating with a locust bean gum–sodium alginate (LBG-SA)-active coating; 0.16D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.16 g/L daphnetin; 0.32D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.32 g/L daphnetin; 0.64D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.64 g/L daphnetin].

Magnetic Resonance Imaging Analysis

The proton density-weighted images were obtained by MRI experiments on all turbot samples, also using the above-mentioned LF-NMR analyzer. The slice width was 1.4 mm, time of repetition was 500 ms, and time of echo was 20 ms.

Determination of TPA

TPA was performed, using a texture analyzer (TA.XT Plus; Stable Micro Systems, Ltd., Godalming, Surrey, UK), equipped with a cylindrical probe (P/5). The 3 × 2 × 1.5 cm (about 5 g) dorsal muscle was tested with a constant test speed of 1 m/s and sample deformation of 50% to obtain the parameters of hardness, springiness, chewiness, and cohesiveness. Each experiment was repeated six times.

Organoleptic Evaluation

For organoleptic evaluation, the quality index method (QIM) developed by Meral et al. (27) was mentioned. Ten trained professional panelists participated in the organoleptic evaluation. The odor, color, mucus, elasticity, and muscle tissue of the turbot samples were scored at each sampling time. Number 10 indicates the best quality, while a lower score indicates poor quality. The participants were asked to state whether the turbot sample was acceptable or not to determine the shelf life.

Statistical Analysis

The one-way ANOVA-Duncan test program in SPSS 22.0 software was used for multiple comparisons, and the results were expressed as means ± SD.

RESULTS AND DISCUSSIONS

Microbiological Results

The changes in microbial communities (lg CFU/g) of all turbot samples during refrigerated storage at 4°C were shown in **Figure 1**. The TVC count of CK sample on 0 day was 3.1 lg CFU/g, and WAEW treatment reduced the TVC count to 2.2 lg CFU/g. The TVC counts of all turbot samples increased during refrigerated storage, and the WAEW and daphnetin-treated samples had lower TVC counts in each sampling time, compared with the CK sample. The TVC count of CK sample on 12th day was 7.4 lg CFU/g, which exceeded the "shelf-life" limit of 7.0 lg CFU/g for marine fish (28). H_2S-producing bacteria (mainly *Shewanella putrefaciens*) and *Pseudomonas* spp. are the common SSOs in some marine fish and fish products during refrigerated storage (29, 30). The population of the two microbial communities increased with storage time in all samples (**Figures 1B,C**), which showed similar trends as that of TVC. At the beginning, the counts of H_2S-producing bacteria of the CK and WAEW-treated samples were 2.7 and 1.1 lg CFU/g, respectively, and the counts increased in all the samples during refrigerated storage. The H_2S-producing bacteria counts were reached to 7.5, 7.1, 6.1, 5.9, and 5.4 lg CFU/g for CK, 0, 0.16, 0.32, and 0.64D, respectively, at the end of the storage. The initial counts of *Pseudomonas* spp. treated with/without WAEW were 2 and 1.1 lg CFU/g and increased to over 7 lg CFU/g on 16 and 20th days for both CK and 0D samples, respectively. Other samples were still under 7 lg CFU/g at the end of the storage. Psychrotrophic bacteria could cause deterioration in odor, texture and flavor through the production of metabolic compounds, such as aldehydes, ketones, biogenic amines, and volatile sulfides (31). In the current study, the counts of psychrotrophic bacteria treated with/without WAEW were 2.1 and 1.2 lg CFU/g and increased in all the turbot samples during refrigerated storage. The CK and 0D samples exceeded the upper acceptable limit of 6 lg CFU/g on the 16th day; however, the daphnetin-treated turbot samples were still below the upper limit at the end of the storage.

TVB-N Results

The TVB-N quantifies the presence of nitrogenous compounds (ammonia, dimethyl amine, and trimethyl amine) in marine fish, revealing the degree of freshness (32). Its increase during storage is related to the activity of spoilage bacteria and endogenous enzymes (33). The TVB-N values of turbot samples during refrigerated storage are presented in **Figure 2**. The initial TVB-N value was 7.26 mg N/100 g, indicating good quality and increased with storage time for all the samples; furthermore, the increase was faster after the middle of refrigerated storage because of the increased bacterial activity, endogenous enzymes, storage conditions, and hygienic practices (34). The CK sample had higher TVB-N value throughout the refrigerated storage, and its fits with that TVC had a sharp increase. The CK and 0D samples exhibited a higher increase rate, reaching to 34.58 and 38.61 mg N/100g on 12 and 16th days, exceeded the maximum allowable value (30–35 mg N/100 g) of TVB-N in marine fish (35). The usage of WAEW and daphnetin treatments showed

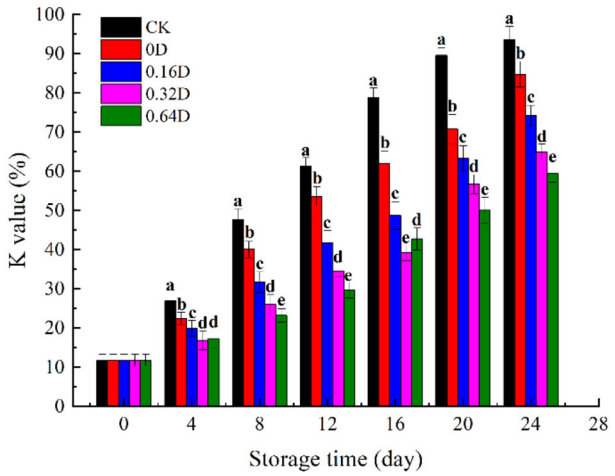

FIGURE 3 | Changes in K value of turbot samples during refrigerated storage [CK, washed with deionized water; 0D, immersed in weakly acidic electrolytic water (WAEW) for 10 min, followed by a coating with a locust bean gum–sodium alginate (LBG-SA)-active coating; 0.16D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.16 g/L daphnetin; 0.32D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.32 g/L daphnetin; 0.64D, immersed in WAEW for 10 min, followed by a coating with an LBG-SA coating with 0.64 g/L daphnetin].

a significant inhibiting effect on the TVB-N increase of turbot samples during the refrigerated storage ($p < 0.05$), compared with the CK sample. The 0.64D sample was still acceptable at the end of the storage, which indicated that WAEW and daphnetin treatments could effectively inhibit the growth of microorganisms and slow down the production of nitrogen and amine substances to delay the deterioration of turbot. Some studies showed that the microbial changes of fish were correlated with TVB-N values (36, 37). Similarly, the TVB-N value of turbot increased with the microbial growth during refrigerated storage, indicating that volatile alkaline compounds were mainly produced by the metabolic activities of SSOs (38). WAEW and daphnetin treatments inhibited the microbial growth in turbot during refrigerated storage, protecting toward protein degradation and producing less ammonia and amine compounds, thus having lower TVB-N values, compared with the CK sample (39).

K Value Results

The K value has been useful for determining quality as it negatively correlates with the freshness of fish, and the increase in K-value is related to the degradation of ATP (40). The degradation of ATP is an autolytic change, which is accompanied by muscle softening during the stiffness process and results in a decrease in fish freshness. The sequence of ATP degradation to Hx is as follows: ATP→ ADP→ AMP→ IMP→ HxR→ Hx (41). The initial K value was found to be 11.72% (**Figure 3**), and the muscle of turbot is fresh, which was similar to the previously reported value of turbot (42). The K value increased in all samples during refrigerated storage, and the CK and 0D samples increased significantly faster than that of the daphnetin-treated sample ($p < 0.05$). The K value of CK 0, 0.16, 0.32, and 0.64D samples at the end of storage were 93.61%, 84.72%, 74.16%, 64.85%, and 59.36%, respectively, where the K values of the 0.64D-treated samples were significantly lower than the other samples. Only the 0.64D sample was still acceptable (lower than 60%), and other samples were categorized as fish spoilage (>60%) (43). WAEW and daphnetin treatments could effectively reduce the degradation of ATP, which can be attributed to the inhibitory effect of WAEW and daphnetin on nucleotide-degrading enzymes and bacteria responsible for nucleotide degradation (44). As reported by Jung et al. (45), the brown sole (*Pleuronectes herzensteini*) treated with WAEW could delay the degradation of nucleic acids-related substances in the muscle and retain the freshness of the fish. According to Alasalvar et al. (46), the conversion of ATP to IMP was a totally autolytic process within 1–2-day storage; however, the subsequent breakdown of IMP to Hx was caused by both fish and microbial enzymes. In the present research, the decreased degradation of ATP to IMP in WAEW and daphnetin-treated samples might result from inactivation of microorganisms by both WAEW and LBG-SA active coatings, containing daphnetin emulsions.

FAAs Results

FAAs are the main contributors to the development of taste sensations in fish, including umami, bitterness, and sweetness, as well as being precursors to biogenic amines, such as histamine derived from histidine *via* microbial metabolism (47), which could affect the flavor and taste of aquatic products during processing or storage. Alanine and glycine have a sweet taste (48), while glutamic acid and aspartic acid have the typical "freshness" of aquatic products (3). The concentrations of FAAs in turbot samples on 0, 12, and 24th days are shown in **Table 1**. In the present research, most of FAAs and total FAAs showed upward trends in all the samples during the refrigerated storage. The main FAAs in the turbot samples were glutamic acid, glycine, alanine, valine, phenylalanine, and lysine, accounting for 49.23–55.49% of the total FAAs contents. The glycine content in the CK samples increased from 5.12 mg/100 g on 0 day to 14.56 mg/100 g at the end of storage. The changes of glycine content in the WAEW and daphnetin-treated samples were similar to that of the CK samples; however, their final contents were significantly ($p < 0.05$) higher. The glutamic acid and alanine contents of WAEW and daphnetin-treated turbot samples also increased significantly during the refrigerated storage due to the short peptides were broken down by proteolytic enzymes, resulting in an increase in the FAAs contents. Histidine, as an off-flavor amino acid, accounted for 1.22–11.99% of the total FAAs contents. The histidine content in the CK samples increased from 1.57 mg/100 g on 0 day to 32.02 mg/100 g at the end of storage; however, the histidine contents of 0.16, 0.32, and 0.64D samples were 16.46, 10.48, and 14.50 mg/100 g at that time, respectively. The lower histidine concentration resulted in a reduction of bitterness in the turbot samples during the refrigerated storage (47). The flavor deterioration is associated with the reduction of some umami and sweet flavor enhancing amino acids and the accumulation of off-flavor amino acids. WAEW and LBG-SA active coatings,

TABLE 1 | Changes in the free amino acid content of turbot during the refrigerated storage at 4°C.

Storage Time	Samples	FAAs								
		Asp	**Thr**	**Ser**	**Glu**	**Gly**	**Ala**	**Cys**	**Val**	**Met**
0d	-	0.46 ± 0.01	1.99 ± 0.05	4.40 ± 0.13	3.70 ± 0.11	5.12 ± 0.18	8.31 ± 0.25	1.46 ± 0.02	5.11 ± 0.15	2.27 ± 0.10
12d	CK	1.87 ± 0.03^{b}	3.75 ± 0.02^{b}	2.73 ± 0.09^{c}	5.38 ± 0.08^{d}	6.46 ± 0.08^{c}	18.37 ± 0.23^{c}	1.70 ± 0.01^{a}	7.33 ± 0.12^{b}	3.32 ± 0.08^{b}
	0D	1.76 ± 0.02^{c}	3.77 ± 0.01^{b}	5.79 ± 0.16^{b}	7.89 ± 0.14^{b}	5.96 ± 0.05^{d}	19.10 ± 0.05^{b}	1.54 ± 0.01^{c}	6.62 ± 0.01^{c}	2.89 ± 0.12^{c}
	0.16D	1.28 ± 0.02^{d}	3.62 ± 0.05^{bc}	5.85 ± 0.15^{b}	6.66 ± 0.16^{c}	7.44 ± 0.18^{a}	13.89 ± 0.27^{e}	1.57 ± 0.01^{bc}	6.31 ± 0.14^{cd}	2.38 ± 0.05^{d}
	0.32D	0.98 ± 0.01^{e}	3.38 ± 0.10^{c}	5.59 ± 0.03^{b}	7.94 ± 0.07^{b}	5.31 ± 0.03^{e}	16.26 ± 0.06^{d}	1.60 ± 0.01^{b}	6.06 ± 0.04^{d}	3.34 ± 0.01^{b}
	0.64D	2.66 ± 0.02^{a}	5.57 ± 0.06^{a}	6.47 ± 0.06^{a}	10.42 ± 0.19^{a}	7.00 ± 0.27^{b}	20.48 ± 0.10^{a}	-	8.84 ± 0.03^{a}	5.29 ± 0.02^{a}
24d	CK	1.99 ± 0.01^{e}	2.62 ± 0.01^{e}	-	9.75 ± 0.10^{e}	14.56 ± 0.03^{e}	32.48 ± 0.28^{d}	1.34 ± 0.01^{e}	25.33 ± 0.19^{c}	6.45 ± 0.23^{e}
	0D	2.75 ± 0.01^{c}	11.04 ± 0.02^{c}	-	17.56 ± 0.01^{b}	21.86 ± 0.02^{b}	46.30 ± 0.06^{a}	3.76 ± 0.01^{b}	35.32 ± 0.04^{a}	26.45 ± 0.05^{c}
	0.16D	2.24 ± 0.02^{d}	22.69 ± 0.06^{a}	-	15.44 ± 0.03^{d}	17.84 ± 0.04^{d}	43.34 ± 0.09^{b}	3.02 ± 0.01^{c}	33.81 ± 0.07^{b}	28.30 ± 0.06^{b}
	0.32D	2.84 ± 0.06^{b}	3.06 ± 0.02^{d}	1.21 ± 0.03	22.22 ± 0.03^{a}	24.31 ± 0.05^{a}	38.61 ± 0.12^{c}	9.92 ± 0.06^{a}	34.93 ± 0.18^{a}	31.88 ± 0.17^{a}
	0.64D	4.73 ± 0.04^{a}	19.95 ± 0.28^{b}	-	15.88 ± 0.18^{c}	20.99 ± 0.23^{c}	43.97 ± 0.48^{b}	2.32 ± 0.03^{d}	33.65 ± 0.31^{b}	23.27 ± 0.14^{d}
		Ile	Leu	Tyr	Phe	Lys	His	Arg	Pro	Total
0d	-	2.47 ± 0.07	3.74 ± 0.10	1.82 ± 0.02	2.33 ± 0.03	0.78 ± 0.04	1.57 ± 0.07	2.15 ± 0.08	3.06 ± 0.14	50.73 ± 1.50
12d	CK	3.04 ± 0.04^{b}	4.55 ± 0.06^{bc}	3.16 ± 0.06^{b}	3.11 ± 0.03^{b}	0.78 ± 0.01^{a}	2.32 ± 0.04^{a}	2.85 ± 0.05^{d}	4.54 ± 0.10^{b}	75.26 ± 1.09^{b}
	0D	3.13 ± 0.12^{b}	4.32 ± 0.08^{c}	3.18 ± 0.07^{b}	2.25 ± 0.07^{b}	0.27 ± 0.03^{b}	2.39 ± 0.06^{a}	3.03 ± 0.06^{c}	4.92 ± 0.09^{b}	78.84 ± 1.30^{b}
	0.16D	3.05 ± 0.06^{b}	4.50 ± 0.08^{bc}	2.69 ± 0.03^{bc}	3.10 ± 0.04^{b}	0.29 ± 0.01^{b}	2.19 ± 0.05^{a}	3.23 ± 0.10^{b}	8.50 ± 3.31^{a}	76.56 ± 2.94^{b}
	0.32D	3.31 ± 0.03^{b}	4.96 ± 0.05^{b}	2.69 ± 0.03^{bc}	3.24 ± 0.03^{b}	1.00 ± 0.15^{a}	2.11 ± 0.08^{a}	3.60 ± 0.04^{a}	8.02 ± 3.47^{a}	79.38 ± 3.90^{b}
	0.64D	5.34 ± 0.01^{a}	8.46 ± 0.03^{a}	7.09 ± 0.13^{a}	7.65 ± 0.12^{a}	0.85 ± 0.01^{a}	1.25 ± 0.04^{b}	1.44 ± 0.03^{e}	3.66 ± 0.05^{c}	102.46 ± 0.50^{a}
24d	CK	17.06 ± 0.18^{d}	31.31 ± 0.20^{e}	26.09 ± 0.21^{e}	40.41 ± 0.38^{d}	21.63 ± 0.23^{e}	32.02 ± 0.48^{a}	1.16 ± 0.01^{e}	2.93 ± 0.01^{c}	267.13 ± 2.24^{e}
	0D	22.02 ± 0.01^{b}	40.77 ± 0.05^{c}	29.12 ± 0.80^{d}	41.83 ± 0.62^{c}	43.45 ± 0.09^{b}	22.48 ± 0.02^{b}	1.82 ± 0.07^{c}	5.33 ± 0.17^{b}	371.83 ± 1.52^{c}
	0.16D	21.68 ± 0.07^{c}	42.35 ± 0.16^{b}	48.16 ± 0.28^{b}	60.76 ± 0.33^{b}	44.13 ± 0.24^{a}	16.46 ± 0.65^{c}	2.05 ± 0.01^{b}	6.76 ± 3.38^{a}	409.03 ± 2.26^{a}
	0.32D	22.54 ± 0.11^{a}	43.57 ± 0.22^{a}	60.74 ± 0.70^{a}	62.58 ± 0.57^{a}	23.86 ± 0.20^{c}	10.48 ± 0.01^{e}	1.34 ± 0.01^{d}	2.63 ± 0.10^{c}	396.72 ± 2.34^{b}
	0.64D	22.01 ± 0.25^{b}	39.01 ± 0.42^{d}	34.37 ± 0.51^{c}	38.83 ± 0.45^{e}	22.91 ± 0.23^{d}	14.50 ± 0.17^{d}	2.57 ± 0.03^{a}	5.24 ± 0.13^{b}	344.20 ± 3.45^{d}

"-" is not detected. Different lowercase letters in different groups from the same day indicate significant differences ($p < 0.05$).

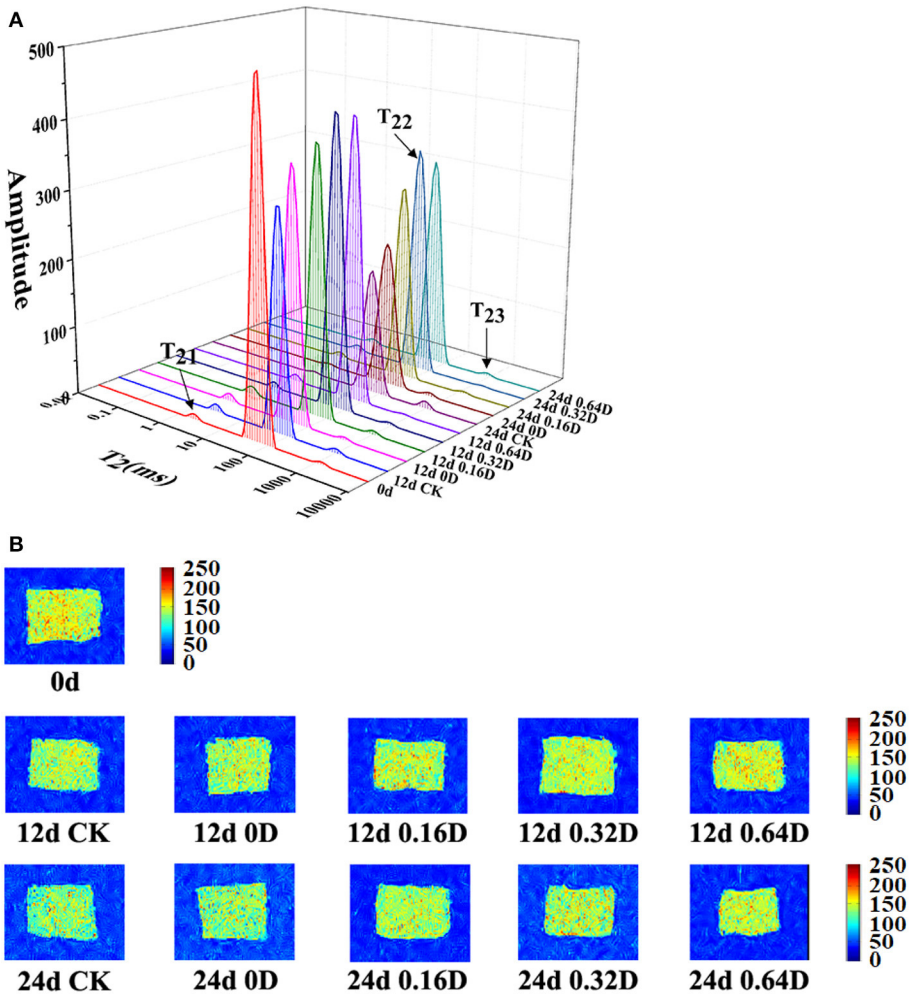

FIGURE 4 | Changes in **(A)** water distribution and **(B)** magnetic resonance imaging of turbot samples during the refrigerated storage [CK, washed with deionized water; 0D, immersed in weakly acidic electrolytic water (WAEW) for 10 min, followed by a coating with a locust bean gum–sodium alginate (LBG-SA)-active coating; 0.16D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.16 g/L daphnetin; 0.32D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.32 g/L daphnetin; 0.64D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.64 g/L daphnetin].

containing daphnetin emulsions treatments, were effective in delaying the flavor deterioration process and maintaining the flavor of turbot samples during the refrigerated storage.

Water Distribution Results

LF-NMR can be used to describe the changes of water distribution and transfer in fish and fish products by measuring the proton relaxation (49). **Figure 4A** shows the distribution of T_2 transverse relaxation times of turbot samples on 0, 12, and 24th days during the refrigerated storage. There are three peaks corresponding to the three relaxation components, known as T_{21} (<10 ms), T_{22} (20–400 ms) and T_{23} (>1,000 ms) and stand for bound water, immobilized water, and free water, respectively. The pT_{21}, pT_{22}, and pT_{23} corresponded to the areas of T_{21}, T_{22}, and T_{23} (50). pT21 varied, ranging from.83 to 1.12% during the refrigerated storage. There was no significant difference among WAEW and daphnetin-treated samples ($p > 0.05$) in the pT21 during storage, indicating T_{21} could not be affected by WAEW and daphnetin treatments as well as storage time, which was due to the water entrapped within highly organized myofibril structures (51). pT_{22} diminished progressively, and pT_{23} increased constantly during the storage ($p < 0.05$). In the present study, The CK samples had significantly lower immobilized water contents than that of WAEW and daphnetin-treated samples during the refrigerated storage. About 0.32 and 0.64D samples had higher contents of immobilized water on 12 and 24th days, probably owing to the WAEW, and LBG-SA active coatings, containing 0.32 or 0.64 g/L daphnetin emulsions treatments, could effectively suppress the changes of the immobilized water into free water. Some research reported that water located within myofibrillar macromolecules released or translated to free water due to the destruction of myofibril structures

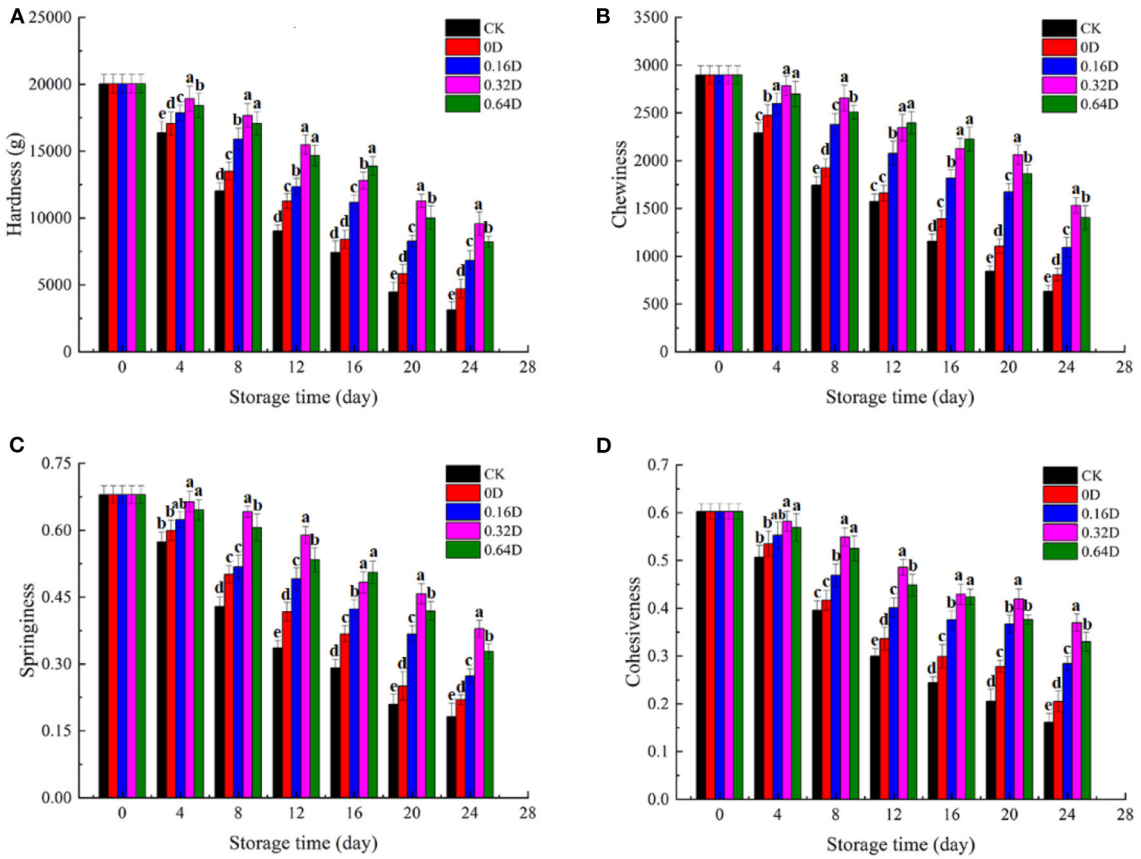

FIGURE 5 | Changes in **(A)** hardness, **(B)** chewiness, **(C)** springiness, and **(D)** cohesiveness of turbot samples during the refrigerated storage [CK, washed with deionized water; 0D, immersed in weakly acidic electrolytic water (WAEW) for 10 min, followed by coating with locust bean gum–sodium alginate (LBG-SA)-active coating; 0.16D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.16-g/L daphnetin; 0.32D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.32 g/L daphnetin; 0.64D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.64-g/L daphnetin].

(38, 52). Besides, this process of water migration was also well reflected in a followed phenomenon that WAEW and daphnetin treatments retarded the change rates of T_{22} and T_{23}. WAEW and daphnetin treatments could effectively delay the water located within myofibrillar macromolecules to release or translate to free water based on the destruction of myofibril structures.

MRI can highlight the signal of different phases of water in the organization. The proton density map is much brighter, and the pseudo-color image is redder (53). The sample was brighter and redder in images on 0 day (**Figure 4**), and the brightness was darker and bluer with the refrigerated storage time increasing. The color of CK samples on 12 and 24th days was bluer and darker than that of WAEW and daphnetin-treated samples. The results demonstrated that the density of water protons in CK was the lowest compared with the other samples, and the tissue structure was seriously damaged, resulting in water loss. However, there was less structural disruption of myofibrils in WAEW and daphnetin-treated samples during the refrigerated storage (54). The brightness of 0.32D and 0.64D samples was lighter compared with the other samples, which indicated that the WAEW and LBG-SA active coatings, containing 0.32 or 0.64-g/L daphnetin emulsions treatments could be more suitable for quality maintenance of turbot samples during refrigerated storage, and the result was consistent with the variation of LF-NMR transverse relaxation.

TPA Results

Texture is considered as a valuable quality attribute to evaluate the influence of preservation (55). The physical properties (hardness, chewiness, springiness, and cohesiveness) for textural evaluation of turbot samples during the refrigerated storage are shown in **Figure 5**. The texture properties of all the turbot samples were decreased during the refrigerated storage, indicating the Argo turbot samples lost good quality and freshness (5). The hardness of the fresh sample was 2.0×10^4 g and decreased significantly ($p < 0.05$), with increasing storage time, regardless of the storage conditions. The muscles became softer and less elastic with increasing storage time, resulting from the activity of the autolytic enzyme, the degradation of myofibrils, and the destruction of the connective tissue (56). The chewiness showed similar behaviors with that of hardness as a decrease in these two parameters was observed during the refrigerated storage, which is consistent with the findings of Liu et al. (57).

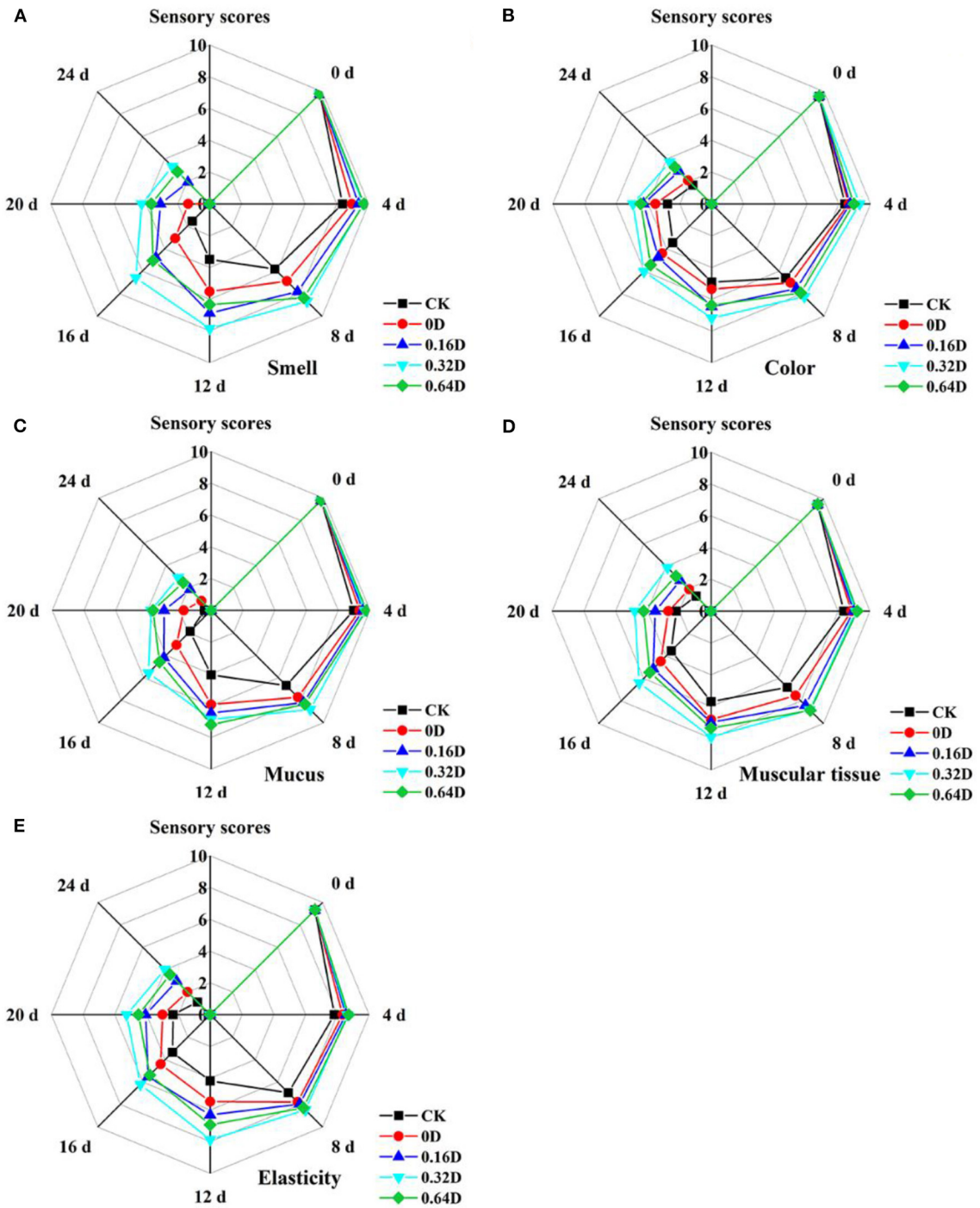

FIGURE 6 | Changes in **(A)** smell, **(B)** color, **(C)** mucus, **(D)** muscular tissue, and **(E)** elasticity of the turbot samples during the refrigerated storage [CK, washed with deionized water; 0D, immersed in weakly acidic electrolytic water (WAEW) for 10 min, followed by coating with locust bean gum–sodium alginate (LBG-SA)-active coating; 0.16D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.16-g/L daphnetin; 0.32D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.32 g/L daphnetin; 0.64D, immersed in WAEW for 10 min, followed by coating with LBG-SA coating with 0.64-g/L daphnetin].

WAEW and daphnetin-treated samples had significant higher chewiness values ($p < 0.05$) compared with the CK samples during the storage. Springiness is used to describe the ability of muscles to return to their original form after the removal of deformation force, as well as their resistance to subsequent deformation (58). The average value of springiness of fresh turbot

was about.67 and decreased significantly ($p < 0.05$) by 73.19, 67.60, 59.80, 44.23, and 51.68% for CK, 0, 0.16, 0.32, and 0.64D turbot samples at the end of storage, respectively, indicating the occurrence of deterioration in texture. Cohesiveness reflects the interior adhesive force of samples. The cohesiveness of turbot also showed a decreasing trend during the refrigerated storage. The initial values of cohesiveness were about 0.60 and decreased to 0.16, 0.20, 0.28, 0.37, and 0.33 for CK, 0D, 0.16D, 0.32D, and 0.64D turbot samples at the end of the storage, respectively. Decrease in springiness and cohesiveness may be attributed to the destruction of covalent cross-linking structure between proteins, the decomposition of proteins, and the reduced force between muscle fiber as the microbial growth and enzymatic carried by the fish itself (59). Some studies pointed out that texture deterioration might be caused by the degradation and disintegration of myofibril structure, affecting the acceptance of consumers (60). The combined effect of WAEW and daphnetin could inhibit the growth of microorganisms and delay the degradation of muscle fibers to protect the muscle texture of turbot. Therefore, WAEW and daphnetin treatments were effective in minimizing changes in turbot muscle tissues during the refrigerated storage.

Organoleptic Evaluation Results

The organoleptic evaluation results, including smell, color, mucus, muscular tissue and elasticity of turbot samples during the refrigerated storage, are presented in **Figure 6**. At the beginning, all the turbot samples had high organoleptic scores, demonstrating excellent quality. The organoleptic scores of all the turbot samples decreased significantly ($p < 0.05$) during the refrigerated storage; however, WAEW and daphnetin-treated samples had significant higher scores ($p < 0.05$) than that of CK samples. On the 12th day, the score of CK was lower than the limit value of 5, which was considered as unacceptable for turbot in the present research. However, 0.16, 0.32, and 0.64D exceeded the limitation on the 16, 20, and 20th days, respectively. WAEW and daphnetin treatments helped the turbot samples maintain a relatively better organoleptic quality during the refrigerated storage; thus, the way of WAEW and daphnetin treatments could be an effective way to delay the quality deterioration and maintain the organoleptic quality of turbot samples. However, the organoleptic evaluation showed some mismatching in the microbiological and chemical results of turbot samples, which may be probably due to the spoilage caused by microbial growth on the surface of the turbot samples. Therefore, it is appropriate to evaluate the storage quality of the turbot samples by the comprehensive analysis of physicochemical, microbiological, and organoleptic evaluation.

CONCLUSIONS

This research investigated the effect of WAEW combined with LBG-SA-active coatings, containing daphnetin emulsions (0.16, 0.32, and 0.64 mg/mL, respectively) treatments on the quality of the turbot samples during the refrigerated storage at 4°C. The microbiological analysis showed that WAEW and daphnetin treatments effectively inhibit the growth of spoilage microorganisms, attributing to the sterilizate activity of WAEW and the antibacterial activity of an active coating, containing daphnetin emulsions. WAEW and daphnetin treatments were highly efficient in maintaining lower TVB-N and K values and higher contents of aromatic amino acids, such as glutamic acid, glycine, and alanine in the refrigerated turbot samples. Moreover, TPA and LN-NMR results also stated that the presence of WAEW combined with daphnetin treatments showed positive effects on retarding the degradation of myofibril structure. WAEW, together with LBG-SA active coatings, containing 0.32 or 0.64-mg/mL daphnetin emulsions treatments had similar effects on quality maintenance of the turbot samples during the refrigerated storage. Therefore, 0.32-mg/mL daphnetin addition was determined to maintain the quality of turbot for economic consideration and the principle of using as few food additives as possible.

DATA AVAILABILITY STATEMENT

The original contributions presented in the study are included in the article/supplementary material, further inquiries can be directed to the corresponding author/s.

AUTHOR CONTRIBUTIONS

WL and JM: conceptualization, investigation, and writing–original draft. WL: data curation, formal analysis, and software. QW: Revising and editing the article. JX: funding acquisition and validation. WL, JM, and JX: methodology. JM and JX: project administration and writing–review & editing. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Lv X, Ma H, Sun M, LinY, Bai F, Li J, et al. A novel bacteriocin dy4-2 produced by lactobacillus plantarum from cutlassfish and its application as bio-preservative for the control of pseudomonas fluorescens in fresh turbot (Scophthalmus maximus) fillets. *Food Control.* (2018) 89:22–31. doi: 10.1016/j.foodcont.2018.02.002
2. Nie X, Zhang F, Wang T, Zheng X, Li Y, Huang B, et al. Physiological and morphological changes in turbot (*Psetta maxima*) gill tissue during waterless storage. *Aquaculture.* (2019) 508:30–35. doi: 10.1016/j.aquaculture.2019.04.060

3. Mei J, Liu F, Fang SY, Lan WQ, Xie J. High-CO_2 modified atmosphere packaging with superchilling (−1.3 °C) inhibit biochemical and flavor changes in turbot (*Scophthalmus maximus*) during storage. *Molecules.* (2020) 25:2826. doi: 10.3390/molecules25122826
4. Li T, Sun X, Chen H, He B, Mei Y, Wang D, et al. Effect of the combination of vanillin and chitosan coating on the microbial diversity and shelf-life of refrigerated turbot (*Scophthalmus maximus*) filets. *Front Microbiol.* (2020) 11:462. doi: 10.3389/fmicb.2020.00462
5. Li P, Chen Z, Tan M, Mei J, Xie J. Evaluation of weakly acidic electrolyzed water and modified atmosphere packaging on the shelf life and quality of farmed puffer fish (*Takifugu obscurus*) during cold storage. *J Food Safety.* (2020) 40(3):e12773. doi: 10.1111/jfs.12773
6. Orejel JRC, Cano-Buendia JA. Applications of electrolyzed water as a sanitizer in the food and animal-by products industry. *Processes.* (2020) 8:19. doi: 10.3390/pr8050534
7. Tang J, Chen H, Lin H, Hung Y-C, Xie H, Chen Y. Acidic electrolyzed water treatment delayed fruit disease development of harvested longans through inducing the disease resistance and maintaining the ROS metabolism systems. *Postharvest Biol Tec.* (2021) 171:111349. doi: 10.1016/j.postharvbio.2020.111349
8. Jiang Y, Ai C, Liao X, Liu D, Ding T. Effect of slightly acidic electrolyzed water (SAEW) and ultraviolet light illumination pretreatment on microflora inactivation of coriander. *LWT-Food Sci Technol.* (2020) 132:109898. doi: 10.1016/j.lwt.2020.109898
9. Palotás P, Palotás P, Jónás G, Lehel J, Friedrich L. Preservative effect of novel combined treatment with electrolyzed active water and lysozyme enzyme to increase the storage life of vacuum-packaged carp. *J Food Quality.* (2020) 2020:4861471. doi: 10.1155/2020/4861471
10. Khazandi M, Deo P, Ferro S, Venter H, Pi H, Crabb S, et al. Efficacy evaluation of a new water sanitizer for increasing the shelf life of Southern Australian King George Whiting and Tasmanian Atlantic Salmon fillets. *Food Microbiol.* (2017) 68:51–60. doi: 10.1016/j.fm.2017.06.008
11. Chaijan S, Panpipat W, Panya A, Cheong L-Z, Chaijan M. Preservation of chilled Asian sea bass (*Lates calcarifer*) steak by whey protein isolate coating containing polyphenol extract from ginger, lemongrass, or green t*ea. Food Control.* (2020) 118:107400. doi: 10.1016/j.foodcont.2020.107400
12. Ma X, Zhou Q, Qiu W, Mei J, Xie J. An active gelatin coating containing eugenol and vacuum delays the decay of Chinese seabass (*Lateolabrax maculatus*) fillets during cold storage: a microbiome perspective. *Coatings.* (2021) 11(2):147. doi: 10.3390/coatings11020147
13. Song Y, Liu L, Shen H, You J, Luo Y. Effect of sodium alginate-based edible coating containing different anti-oxidants on quality and shelf life of refrigerated bream (*Megalobrama amblycephala*). *Food Control.* (2011) 22:608–15. doi: 10.1016/j.foodcont.2010.10.012
14. Yong H, Liu J. Active packaging films and edible coatings based on polyphenol-rich propolis extract: a review. *Compr Rev Food Sci S.* (2021) 20:2106–45. doi: 10.1111/1541-4337.12697
15. Bazargani-Gilani B, Pajohi-Alamoti M. The effects of incorporated resveratrol in edible coating based on sodium alginate on the refrigerated trout (*Oncorhynchus mykiss*) fillets' sensorial and physicochemical features. *Food Sci Biotechnol.* (2020) 29:207–16. doi: 10.1007/s10068-019-00661-1
16. Cao XH, Islam MN, Chitrakar B, Duan ZH, Xu WX, Zhong SY. Effect of combined chlorogenic acid and chitosan coating on antioxidant, antimicrobial, and sensory properties of snakehead fish in cold storage. *Food Sci Nutr.* (2020) 8:973–81. doi: 10.1002/fsn3.1378
17. Nie XB, Gao Z, Ren XR, Jiang QN, Li SL, Jiang CX, et al. Effect of pectin coating infused with gallic acid on the quality and shelf life of Japanese sea bass (*Lateolabrax japonicas*) fillets. *Food Bioprocess Tech*. (2020) 13:300–07. doi: 10.1007/s11947-019-02396-7
18. Luan L, Wu C, Wang L, Li Y, Ishimura G, Yuan C, et al. Protein denaturation and oxidation in chilled hairtail (*Trichiutus haumela*) as affected by electrolyzed oxidizing water and chitosan treatment. *Int J Food Prop.* (2018) 20:S2696–707. doi: 10.1080/10942912.2017.1397693
19. Feng HJ, Lan WQ, Sun XH, Xie J. Effects of slightly acidic electrolyzed water pretreatment combined with biopreservatives on the shelf life of refrigerated obscure pufferfish (*Takifugu obscurus*). *J Food Sci.* (2021) 86:484–94. doi: 10.1111/1750-3841.15596
20. Liu W, Mei J, Xie J. Elucidating Antibacterial activity and mechanism of daphnetin against Pseudomonas fluorescens and Shewanella putrefaciens. *J Food Qual.* (2020) 2020:6622355. doi: 10.1155/2020/6622355
21. Liu W, Mei J, Xie J: Effect of locust bean gum-sodium alginate coatings incorporated with daphnetin emulsions on the quality of Scophthalmus maximus at refrigerated condition. *Int J Biol Macromol.* (2021) 170:129–39. doi: 10.1016/j.ijbiomac.2020.12.089
22. Li N, Mei J, Shen Y, Xie J. Quality improvement of half-smooth tongue sole (*Cynoglossus semilaevis*) fillets by chitosan coatings containing rosmarinic acid during storage. *CyTA-J Food.* (2018) 16:1018–29. doi: 10.1080/19476337.2018.1518344
23. Zhuang S, Li Y, Hong H, Liu Y, Shu R, Luo Y. Effects of ethyl lauroyl arginate hydrochloride on microbiota, quality and biochemical changes of container-cultured largemouth bass (*Micropterus salmonides*) fillets during storage at 4 °C. *Food Chem.* (2020) 324:126886. doi: 10.1016/j.foodchem.2020.126886
24. Cao J, Wang Q, Ma T, Bao K, Yu X, Duan Z, et al. Effect of EGCG-gelatin biofilm on the quality and microbial composition of tilapia fillets during chilled storage. *Food Chem.* (2020) 305:125454. doi: 10.1016/j.foodchem.2019.125454
25. Yu D, Jing D, Yang F, Gao P, Jiang Q, Xu Y, et al. The factors influencing the flavor characteristics of frozen obscure pufferfish (*Takifugu Obscurus*) during storage: ice crystals, endogenous proteolysis and oxidation. *Int J Refrig.* (2021) 122:147–55. doi: 10.1016/j.ijrefrig.2020.10.028
26. Yu YJ, Yang SP, Lin T, Qian YF, Xie J, Hu C. Effect of cold chain logistic interruptions on lipid oxidation and volatile organic compounds of salmon (*Salmo salar*) and their correlations with water dynamics. *Front Nutr.* (2020) 7:155. doi: 10.3389/fnut.2020.00155
27. Meral R, Alav A, Karakas C, Dertli E, Yilmaz MT, Ceylan Z. Effect of electrospun nisin and curcumin loaded nanomats on the microbial quality, hardness and sensory characteristics of rainbow trout fillet. *LWT-Food Sci Technol.* (2019) 113:108292. doi: 10.1016/j.lwt.2019.108292
28. Aghaei Z, Ghorani B, Emadzadeh B, Kadkhodaee R, Tucker N. Protein-based halochromic electrospun nanosensor for monitoring trout fish freshness. *Food Control.* (2020) 111:107065. doi: 10.1016/j.foodcont.2019.107065
29. Sterniša M, Bucar F, Kunert O, Smole MoŽina S. Targeting fish spoilers Pseudomonas and Shewanella with oregano and nettle extracts. *Int J Food Microbiol.* (2020) 328:108664. doi: 10.1016/j.ijfoodmicro.2020.108664
30. da Rocha M, Alemán A, Romani VP, López-Caballero ME, Gómez-Guillén MC, Montero P, et al. Effects of agar films incorporated with fish protein hydrolysate or clove essential oil on flounder (*Paralichthys orbignyanus*) fillets shelf-life. *Food Hydrocolloids.* (2018) 81:351–63. doi: 10.1016/j.foodhyd.2018.03.017
31. Singh S, Lee M, Gaikwad KK, Lee YS. Antibacterial and amine scavenging properties of silver-silica composite for post-harvest storage of fresh fish. *Food Bioprod Process.* (2018) 107:61–9. doi: 10.1016/j.fbp.2017.10.009
32. Socaciu MI, Fogarasi M, Simon EL, Semeniuc CA, Socaci SA, Podar AS, et al. Effects of whey protein isolate-based film incorporated with tarragon essential oil on the quality and shelf-life of refrigerated brook trout. *Foods.* (2021) 10:401. doi: 10.3390/foods10020401
33. Sun X, Guo X, Ji M, Wu J, Zhu W, Wang J, et al. Preservative effects of fish gelatin coating enriched with CUR/βCD emulsion on grass carp (*Ctenopharyngodon idellus*) fillets during storage at 4 °C. *Food Chem.* (2019) 272:643–52. doi: 10.1016/j.foodchem.2018.08.040
34. Zhuang S, Li Y, Jia S, Hong H, Liu Y, Luo Y. Effects of pomegranate peel extract on quality and microbiota composition of bighead carp (*Aristichthys nobilis*) fillets during chilled storage. *Food Microbiol.* (2019) 82:445–54. doi: 10.1016/j.fm.2019.03.019
35. Sarika AR, Lipton AP, Aishwarya MS. Biopreservative efficacy of bacteriocin GP1 of Lactobacillus rhamnosus GP1 on stored fish filets. *Front Nutr.* (2019) 6:29. doi: 10.3389/fnut.2019.00029
36. Huang H, Sun W, Xiong G, Shi L, Jiao C, Wu W, et al. Effects of HVEF treatment on microbial communities and physicochemical properties of catfish fillets during chilled storage. *LWT-Food Sci Technol.* (2020) 131:109667. doi: 10.1016/j.lwt.2020.109667
37. Otero L, Pérez-Mateos M, López-Caballero ME: Hyperbaric cold storage versus conventional refrigeration for extending the shelf-life of hake

loins. *Innov Food Sci Emerg.* (2017) 41:19–25. doi: 10.1016/j.ifset.2017.01.003
38. Li P, Peng Y, Mei J, Xie J. Effects of microencapsulated eugenol emulsions on microbiological, chemical and organoleptic qualities of farmed Japanese sea bass (*Lateolabrax japonicus*) during cold storage. *LWT-Food Sci Technol.* (2020) 118:108831. doi: 10.1016/j.lwt.2019.108831
39. Xuan XT, Fan YF, Ling JG, Hu YQ, Liu DH, Chen SG, et al. Preservation of squid by slightly acidic electrolyzed water ice. *Food Control.* (2017) 73:1483–9. doi: 10.1016/j.foodcont.2016.11.013
40. Kang T, Shafel T, Lee D, Lee CJ, Lee SH, Jun S. Quality retention of fresh tuna stored using supercooling technology. *Foods.* (2020) 9:1356. doi: 10.3390/foods9101356
41. Logotheti M, Theochari K, Kostakis M, Pasias IN, Thomaidis NS. Development and validation of a HILIC-UV method for the determination of nucleotides in fish samples. *Food Chem.* (2018) 248:70–7. doi: 10.1016/j.foodchem.2017.12.040
42. Li X, Chen Y, Cai L, Xu Y, Yi S, Zhu W, et al. Freshness assessment of turbot (*Scophthalmus maximus*) by Quality Index Method (QIM), biochemical, and proteomic methods. *LWT-Food Sci. Technol.*(2017) 78:172–180. doi: 10.1016/j.lwt.2016.12.037
43. Zhou Q, Li P, Fang S, Liu W, Mei J, Xie J. Preservative effects of gelatin active coating enriched with eugenol emulsion on Chinese seabass (*Lateolabrax maculatus*) during superchilling (−0.9 °C) storage. *Coatings.* (2019) 9:489. doi: 10.3390/coatings9080489
44. Mohan CO, Ravishankar CN, Ashok Kumar K, Srinivasa Gopal TK. Biogenic amines and nucleotide breakdown products of sodium acetate, sodium lactate, and sodium citrate treated seer fish (*Scomberomorus commerson*) during iced storage. *J Food Safety.* (2019) 39:e12633. doi: 10.1111/jfs.12633
45. Jung S, Ko BS, Jang H-J, Park HJ, Oh S-W. Effects of slightly acidic electrolyzed water ice and grapefruit seed extract ice on shelf life of brown sole (*Pleuronectes herzensteini*). *Food Sci Biotechnol.* (2018) 27:261–7. doi: 10.1007/s10068-017-0198-8
46. Alasalvar C, Taylor KDA, ÖKsüz A, Shahidi F, Alexis M. Comparison of freshness quality of cultured and wild sea bass (*Dicentrarchus labrax*). *J Food Sci.* (2002) 67:3220–6. doi: 10.1111/j.1365-2621.2002.tb09569.x
47. Yu D, Xu Y, Regenstein JM, Xia W, Yang F, Jiang Q, et al. The effects of edible chitosan-based coatings on flavor quality of raw grass carp (*Ctenopharyngodon idellus*) fillets during refrigerated storage. *Food Chem.* (2018) 242:412–20. doi: 10.1016/j.foodchem.2017.09.037
48. Ruiz-Capillas C, Moral A. Free amino acids in muscle of Norway lobster (*Nephrops novergicus* (L.)) in controlled and modified atmospheres during chilled storage. *Food Chem.* (2004) 86:85–91. doi: 10.1016/j.foodchem.2003.08.019
49. Lan W, Hu X, Sun X, Zhang X, Xie J. Effect of the number of freeze-thaw cycles number on the quality of Pacific white shrimp (*Litopenaeus vannamei*): an emphasis on moisture migration and microstructure by LF-NMR and SEM. *Aquaculture Fish.* (2020) 5:193–200. doi: 10.1016/j.aaf.2019.05.007
50. Li DY, Huang Y, Wang KX, Dong XP, Yu D, Ge LH, et al. Microstructural characteristics of turbot (*Scophthalmus maximus*) muscle: effect of salting and processing. *Int J Food Prop.* (2018) 21:1291–302. doi: 10.1080/10942912.2018.1460758
51. Zhou Q, Li P, Fang S, Mei J, Xie J. Preservative effects of gelatin active coating containing eugenol and higher CO_2 concentration modified atmosphere packaging on Chinese sea bass (*Lateolabrax maculatus*) during superchilling (0.9 °C) storage. *Molecules.* (2020) 25:871. doi: 10.3390/molecules25040871
52. Albertos I, Martin-Diana AB, Cullen PJ, Tiwari BK, Ojha SK, Bourke P, et al. Shelf-life extension of herring (*Clupea harengus*) using in-package atmospheric plasma technology. *Innov Food Sci Emerg.* (2019) 53:85–91. doi: 10.1016/j.ifset.2017.09.010
53. Wang X-Y, Xie J. Evaluation of water dynamics and protein changes in bigeye tuna (*Thunnus obesus*) during cold storage. *LWT-Food Sci Technol.* (2019) 108:289–96. doi: 10.1016/j.lwt.2019.03.076
54. Li N, Liu W, Shen Y, Mei J, Xie J. Coating effects of ε-polylysine and rosmarinic acid combined with chitosan on the storage quality of fresh half-smooth tongue sole (*Cynoglossus semilaevis* Günther) fillets. *Coatings*(2019) 9(4):273. doi: 10.3390/coatings9040273
55. Nisar T, Yang X, Alim A, Iqbal M, Wang Z-C, Guo Y. Physicochemical responses and microbiological changes of bream (*Megalobrama ambycephala*) to pectin based coatings enriched with clove essential oil during refrigeration. *Int J Biol Macromol.* (2019) 124:1156–66. doi: 10.1016/j.ijbiomac.2018.12.005
56. Li P, Mei J, Xie J. Chitosan-sodium alginate bioactive coatings containing ε-polylysine combined with high CO_2 modified atmosphere packaging inhibit myofibril oxidation and degradation of farmed pufferfish (*Takifugu obscurus*) during cold storage. *LWT-Food Sci Technol.* (2021) 140:110652. doi: 10.1016/j.lwt.2020.110652
57. Liu B, Liu Z, Li D, Yu M, Liu Y, Qin L, et al. Action of endogenous proteases on texture deterioration of the bay scallop (*Argopecten irradians*) adductor muscle during cold storage and its mechanism. *Food Chem.* (2020) 323:126790. doi: 10.1016/j.foodchem.2020.126790
58. Xu D, Sun L, Li C, Wang Y, Ye R. Inhibitory effect of glucose oxidase from *Bacillus* sp. CAMT22370 on the quality deterioration of Pacific white shrimp during cold storage. *LWT-Food Sci Technol.* (2018) 92:339–346. doi: 10.1016/j.lwt.2018.02.025
59. Li T, Jiang Y, Jin G, Zhao Q, Li J. Effects of fish-derived biological preservatives on cold storage of grass carp (*Ctenopharyngodon idellus*) fillets. *J Food Prot.* (2016) 79:1707–16. doi: 10.4315/0362-028X.JFP-15-571
60. Pornrat S, Sumate T, Rommanee S, Sumolaya K, Kerr WL. Changes in the ultrastructure and texture of prawn muscle (*Macrobrachuim rosenbergii*) during cold storage. *LWT - Food Sci Technol.* (2007) 40:1747–54. doi: 10.1016/j.lwt.2007.01.006

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