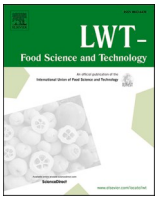
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Impacts of intense pulsed light (IPL) on reduction of indigenous microorganisms and quality changes in leafy vegetables

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ABSTRACT

The minimal processing applied to fresh vegetables raises concerns about microbial contamination. This study examined how intense pulsed light (IPL) treatment influences the microbial reduction and quality attributes in lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves. IPL treatments were applied at voltages of 800–2000 V for 1–4 min, and their effects on indigenous microorganisms and their physicochemical properties. The bactericidal efficacy varied with the color and surface characteristics of each vegetable, but in all cases IPL significantly reduced indigenous microorganisms. Increasing the energy fluence increased the degree of microbial inactivation, with maximum reductions in lettuce, perilla leaves, cabbage stems, and cabbage leaves of 1.74, 1.91, 2.27, and 1.51 log CFU/mL, respectively, for total aerobic bacteria, and of 1.19, 1.46, 1.34, and 1.42 log CFU/mL for yeasts and molds. No significant changes were observed in moisture content or pH immediately after treatment, while the total phenolic content varied significantly with the vegetable type. After 10 days of storage, IPL-treated samples maintained better microbial safety without significant deterioration in quality compared to untreated samples. These findings demonstrate a potential of IPL as a non-thermal sterilization method for preserving the safety and quality of fresh leafy vegetables.

1. Introduction

Increasing awareness about health issues is leading consumers to show stronger preferences for fresh and nutritious food choices. Fresh foods also contain various bioactive compounds such as lycopene, polyphenols, and plant extracts, have been reported to exert beneficial effects on both animal and human health including antioxidant, anti-inflammatory, and antimicrobial activities (Balgoon & Alghamdi, 2024; Elzaiat et al., 2024; Hammoud et al., 2025). Minimally processed raw vegetables retain more nutrients than their fully processed counterparts. However, the consumption of raw vegetables has been identified as a leading cause of infections caused by Shiga-toxin-producing *Escherichia coli* (*E. coli*) (STEC) (Feng, 2014). Numerous previous studies have emphasized the risk of STEC contamination in fresh produce, particularly in leafy greens such as romaine lettuce and spinach (Astill et al., 2020; Khalil & Gomaa, 2016; Söderqvist et al., 2019). Raw vegetables that are consumed without cooking are more susceptible to pathogen contamination and have a considerably higher risk of STEC infection. In addition to STEC, raw vegetables are at significant risks of contamination by other foodborne pathogens such as *Campylobacter*, *Salmonella*, and *Listeria* species (Qadri et al., 2015; Wadamori et al., 2017).

To ensure safe consumption, the World Health Organization and the Centers for Disease Control and Prevention recommend thorough washing and appropriate processing of raw vegetables. Recent studies have detected pesticide active ingredients in seasonal vegetables using Fourier Transform Infrared spectroscopy and this emphasizes the importance of food safety monitoring and public awareness (Mithu et al., 2025). Foodborne pathogens can be introduced into fresh produce from various sources, including contaminated soil, contaminated irrigation water, and unsanitary handling practices (Luna-Guevara et al., 2019). In addition, vegetables cultivated in smallholdings and peri-urban areas can also harbor considerable microbial contamination, so effective microbial decontamination from the production stage is required (Hassani et al., 2023). Although most fresh produce undergoes some level of processing to eliminate pathogens, contamination of

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vegetables with harmful microorganisms remains a major public health concern, emphasizing the need to ensure microbial safety (Shakeel et al., 2023). Fresh vegetables cannot be sterilized using heat or steam due to concerns about damaging their freshness and texture. This has prompted previous studies to apply γ-irradiation, cold plasma, electrolyzed water, ultrasound, and ultraviolet (UV) light as non-thermal methods for the microbial decontamination of fresh vegetables (Chen et al., 2020; Feliciano et al., 2017; Lee et al., 2021; Sethi et al., 2018; Zhang et al., 2021).

IPL is an additional non-thermal sterilization methodology that utilizes short pulses of broad-spectrum light, including in the UV, visible, and infrared (200–1100 nm) parts of the spectrum (Mandal et al., 2020). Previous studies have shown that IPL can effectively inactivate bacteria, yeasts, molds, and viruses in various food products (Abuagela et al., 2018; Ding et al., 2024; Fernández et al., 2020; Kramer et al., 2019; Pedrós-Garrido et al., 2018; Pérez-López et al., 2020; Woldemariam et al., 2022). Microorganisms are inactivated by IPL via photochemical, photothermal, and photophysical mechanisms, with the photochemical mechanism being the primary contributor and photothermal and photophysical mechanisms playing secondary roles (Mandal et al., 2020). The photochemical mechanism is primarily driven by UV-light absorption by DNA leading to the formation of dimers, particularly thymine-thymine dimers that disrupt DNA replication and transcription, ultimately resulting in irreversible molecular damage and microbial death (Cassar et al., 2022; Mandal et al., 2020). In addition, the photothermal mechanism involves localized heating induced by infrared light (Cassar et al., 2022). The photophysical mechanism, driven by visible light, occurs when light is absorbed by photosensitizers to generate reactive oxygen species that oxidize fatty acids in the cell membrane, ultimately resulting in the disruption of the cell membrane and the release of intracellular components (Kebbi et al., 2020).

Several studies have investigated the use of IPL treatment for microbial inactivation in fresh products. Avalos-Llano et al. (2016) treated fresh-cut apples with pulsed light at 4–16 J/cm^2 and achieved a maximum 1.55-log reduction in both mesophilic and psychrophilic bacteria. Jaiswal and Srivastava (2024) reported reductions of 2.55 and 1.87 log colony-forming units per gram (CFU/g) in *Salmonella* species and *Staphylococcus aureus*, respectively, for fresh-cut watermelon. Similarly, the application of IPL at 15.75 J/cm^2 to spinach resulted in an approximate 3.5-log reduction of *E. coli* O157:H7 (a pathogenic serotype of Shiga toxin–producing *E. coli*) (Mukhopadhyay et al., 2019). In strawberries, Duarte-Molina et al. (2016) observed 16–42 % reductions in mold decay in strawberries relative to control after 8 days of storage, following IPL treatment at energy fluences ranging from 2.4 to 47.8 J/cm^2. In summary, IPL has demonstrated significant microbial inactivation capabilities in a variety of fresh food products, and its effects have been observed not only immediately after treatment but also during subsequent storage. However, few studies have focused specifically on vegetables, and there remains a lack of research on the sterilization effects of IPL on indigenous, uninoculated microorganisms. In addition, sustainable food processing and agricultural practices increasingly rely on the transfer of knowledge and learning among producers and researchers to ensure both safety and long-term viability (Sule et al., 2025), so it is required to develop and optimize IPL nonthermal technologies to enhance microbial safety while maintaining food quality.

This study aimed to determine the efficacy of IPL treatment in inactivating indigenous microorganisms on various leafy vegetables: lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves. In addition to performing microbial analyses immediately after IPL treatment, physicochemical properties such as the moisture content, pH, and total polyphenol content (TPC) were measured to assess potential changes in quality attributes. Furthermore, the effects of IPL during subsequent storage were investigated by monitoring microbial survival, water loss, pH, and color changes over a 10-day refrigeration period.

2. Materials and methods

2.1. Sample preparation

The raw leafy vegetables used in this study were lettuce (*Lactuca sativa* var. *crispa*), perilla leaf (*Perilla frutescens* var. *Japonica* Hara), and napa cabbage (*Brassica rapa* subsp. *pekinensis*). The stems and leaf parts of napa cabbage exhibit significant differences in characteristics such as shape, color, thickness, and texture, and so experiments were conducted with these parts separately to obtain accurate and reliable results. All samples were purchased from a local market located in Seoul, South Korea. The samples were cut into squares (5 cm × 5 cm) obtained from the central part of the sample using sterilized scissors. The initial mean contamination levels of lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves were 5.49, 5.94, 5.56, and 6.03 log CFU/g, respectively, for total aerobic bacteria, and 4.42, 5.50, 4.86, and 4.75 log CFU/g for yeasts and molds. All samples were stored at 4 °C before IPL treatment.

2.2. IPL treatment

A custom-designed pilot-scale IPL device was used in this study (Fig. 1). Samples placed on sterile petri dishes were positioned 9 cm below a xenon lamp (type NL 9553, XAP series, Heraeus Noblelight, Cambridge, UK). To prevent overheating, cooling water ran down the outside of the lamp, and cooling fans were installed at the top and sides of the treatment chamber. Both sides of the samples were treated with IPL at voltages of 800, 1200, 1600, and 2000 V for 1, 2, and 4 min. The frequency and pulse duration were fixed at 2 Hz and 50 ms, respectively. The light spectrum was measured using a spectroradiometer (ILT 950 & 960, International Light Technologies, Peabody, MA, USA) by placing its sensor at the same location as the samples during treatment. The energy fluences per pulse at 800, 1200, 1600, and 2000 V were estimated to be 0.011, 0.015, 0.019, and 0.022 J/cm^2, respectively, from which the total energy fluences were calculated by multiplying by the frequency and treatment time, and these ranged from 1.37 to 10.51 J/cm^2.

2.3. Microbiological analysis

The reductions in the indigenous total aerobic bacteria and in the indigenous yeasts and molds in the leafy vegetable samples were investigated. After IPL treatment, each sample was placed inside a stomacher bag (B01196, Nasco Whirl–Pak, WI, USA) with sterile 0.85 % NaCl (physiological saline) solution in a 1:9 wt ratio. The samples were then homogenized using a stomacher (Tianjin Hengao Technology Development, Tianjin, China) at a speed of 6 strokes/s for 1 min. The homogenized solution was serially diluted with sterile 0.85 % NaCl solution. Then, 0.1 mL of the diluted solution was inoculated onto selective media: plate count agar for total aerobic bacteria and dichloran rose bengal chloramphenicol agar for yeasts and molds, and incubated at 37 °C for 48 h and 25 °C for 96 h, respectively. The microbial reductions were quantified as log-reduction values calculated using the formula log (N_0/N), where N_0 represents the initial number of microorganisms and N represents the number of microorganisms remaining after the treatment.

2.4. Quality attributes

Parameters that represent important quality attributes of leafy vegetables, such as their moisture content, pH, and TPC, were measured before and after IPL treatment. Additionally, samples that were either untreated or treated with IPL at 800 V for 4 min or at 2000 V for 2 min were sealed in adhesive polypropylene bags to prevent contamination and stored at 4 °C for 10 days. Weight loss, color, and pH were assessed every 2 days during the storage period.

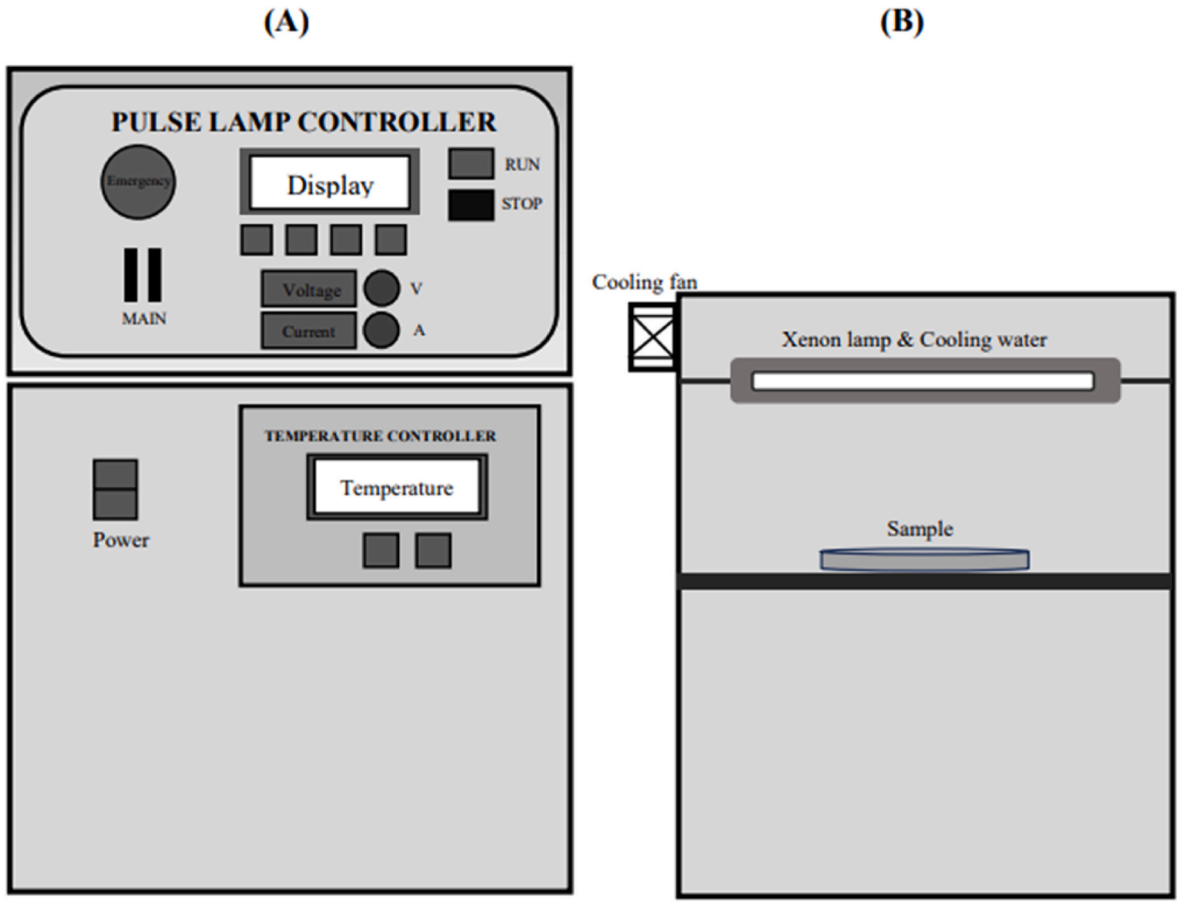


Fig. 1. Pilot-scale IPL device consisting of (A) a power supply (upper) and cooling system (lower), and (B) a treatment chamber.

2.4.1. Moisture content and water loss

The moisture content of samples was measured using an infrared moisture analyzer (FD-660, Kett Electric Laboratory, Tokyo, Japan). Water loss in each sample was estimated based on weight changes during the storage period. With the initial weight defined as W_i and the weight after storage defined as W_f, the percentage of water loss was calculated as follows:

$$\text{Weight loss}\ (\%) = \left(\left(W_i - W_f \right) \ / \ W_i \right) \times 100$$

2.4.2. pH

To measure pH, each sample was mixed with distilled water in a 1:3 ratio and then homogenized for 2 min. The pH values of the untreated and treated samples were determined using a pH/ion meter (Seven Compact S220, Mettler Toledo, Greifensee, Switzerland). The pH meter was calibrated prior to each measurement using standard buffer solutions to ensure measurement accuracy.

2.4.3. Total polyphenol content

The untreated and treated samples were stored at −85 °C to prevent degradation of polyphenols prior to analyzing the TPC. The frozen samples were freeze-dried at −85 °C for 48 h to effectively remove moisture without compromising the polyphenol content. The freeze-dried samples were ground into a uniformly fine powder using a mill to ensure consistency across the analyses. The extraction process required the preparation of two reagents to isolate polyphenols: 2 % sodium carbonate (Na_2CO_3) solution and 50 % Folin-Ciocalteu reagent. To produce the analytical samples, 0.2 g of freeze-dried powder was dissolved in 10 mL of acetone-water solution (1:1, v/v) and stored at room temperature for 15 h. Then, 0.1 mL of extract was dissolved in 2 mL of Na_2CO_3 solution and vortexed for 2 min. This suspension was treated with 0.1 mL of Folin-Ciocalteu reagent and incubated in the dark for 30 min to ensure the complete reaction of polyphenols.

According to the Folin-Ciocalteu method, the TPC of each sample was quantified using spectrophotometric analysis at an absorbance wavelength of 765 nm. A standard curve was determined by measuring the absorbance of gallic acid solutions with concentrations ranging from 100 to 1000 ppm. The TPC was converted to the milligrams of polyphenols per 100 g on a dry weight basis (DWB).

2.4.4. Color

Color measurements were performed using a spectrophotometer (Color Quest XE, HunterLab, Reston, VA, USA). Color is described using L^*, a^*, and b^* values in the Hunter color system, where L^* denotes lightness, with higher values indicating lighter shades and lower values indicating darker ones; a^* reflects the red–green axis (positive for red, negative for green); and b^* corresponds to the yellow–blue axis (positive for yellow, negative for blue). To quantify color changes over time, the color difference (ΔE) was calculated using the following formula:

$$\Delta E = \left((\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2 \right)^{1/2}$$

2.5. Statistical analyses

All experiments were conducted in triplicate. The results of each experiment were statistically analyzed using Microsoft Excel (Microsoft, Redmond, WA, USA) and expressed as mean ± standard-deviation values. Additionally, one-way analysis of variance was performed using standard analytical software (SPSS Statistics, version 22.0, SPSS, Chicago, IL, USA) to identify statistically significant differences, defined as $p < 0.05$.

3. Results and discussion

3.1. Inactivation of indigenous microorganisms

Fig. 2 presents the indigenous microbial inactivation results for the four sample types treated with IPL at different energy fluences. Increasing the energy fluence from 1.37 to 10.51 J/cm^2 resulted in 0.12- to 1.74-log reductions in total aerobic bacteria for lettuce, 0.15- to 1.91-log reductions for perilla leaves, 0.12- to 2.27-log reductions for napa cabbage stems, and 0.21- to 1.51-log reductions for napa cabbage leaves; the corresponding values for yeasts and molds were 0.06- to 1.19-log, 0.06- to 1.46-log, 0.06- to 1.34-log, and 0.13- to 1.42-log reductions, respectively. In several previous studies on leafy vegetables, the treatment effect on IPL was confirmed by inoculating the samples with microorganisms. Ferrario et al. (2015) reported that pulsed light treatments on inoculated produce achieved 0.3 to 2.6 log reductions for pathogens, while native microflora reductions were only 0.1 to 0.7 log. Kramer (2022) also achieved a reduction of up to 2.5 log for inoculated bacteria on ready-to-eat salad greens. Considering that this study targeted indigenous microorganisms present in four different vegetable matrices under realistic contamination conditions, the results of this study demonstrate that IPL treatment was highly effective and confirms practical research results.

Across all samples, an increase in energy fluence consistently resulted in greater reductions in total aerobic bacteria and in yeasts and molds. Additionally, the reductions achieved were generally higher for total aerobic bacteria than for yeasts and molds under the same treatment conditions. This suggests that the susceptibility to IPL treatment varies between different types of microorganisms, with total aerobic bacteria being more sensitive. Hwang et al. (2024) also reported that yeasts exhibited greater resistance to a high IPL fluence than did bacteria. Guerrero-Beltrán and Barbosa-Cánovas (2005) found that *E. coli* (aerobic bacteria) exhibited a greater reduction under UV-light treatment than did *Saccharomyces cerevisiae* (yeast) due to its lower resistance. Although inactivation tendencies have varied slightly among yeast strains, both *Pichia membranifaciens* and *Saccharomyces pastorianus* exhibited lower susceptibility than bacteria (Kim et al., 2017). Hakguder Taze et al. (2015) attributed the lower light sensitivity of yeasts and molds to their larger cells, lower content of pyrimidine bases, and differences in the composition and thickness of cell walls.

Furthermore, the different leafy vegetables investigated in this study exhibited different levels of total aerobic bacteria inactivation, in the following descending order: napa cabbage stems, perilla leaves, lettuce, and napa cabbage leaves. The shadow effect is influenced by surface roughness, and this was a key factor affecting the sterilization efficiency among the samples. Previous research has indicated that a higher surface roughness increases light absorption, thereby improving the sterilization efficiency (Niu et al., 2019). Studies of strawberries and raspberries have highlighted that the surface protrusions and irregularities of microorganisms result in them not being fully exposed to incident light, leading to only partial sterilization (Bialka & Demirci, 2008). Similarly, black pepper powder and red pepper powder exhibited lower inactivation than did sesame seeds due to their greater surface irregularity, which includes numerous holes, cracks, and crevices (Hwang et al., 2023). The smooth and relatively simple structure of the napa cabbage stems in the present study minimized the shadow effect, while samples with curved and irregular surfaces such as lettuce and napa cabbage leaves exhibited lower sterilization efficiency due to increased shadow effects. Incident light will fall on more-complex or irregular surfaces less evenly, increasing the likelihood of microorganism survival.

Surface color also plays a role in light absorption, in the following descending order: black, green, red, purple, yellow, pink, blue, and white (Paminto et al., 2021). Uemoto et al. (2010) investigated the correlation between paint color and spectral reflectance, and found that white reflects more than twice as much light as brown, while yellow reflects an intermediate amount. Darker vegetables, such as those that are black or green (e.g., perilla leaves), will absorb more light, which can increase the sterilization efficiency. In contrast, lighter-colored vegetables have lower light absorption, resulting in a lower sterilization efficiency. However, the sterilization efficiency cannot be determined by a single factor alone. In addition to morphological and color characteristics, both intrinsic properties including moisture content, nutritional properties, and pH and extrinsic properties including the initial microbial count and microorganism type also have significant impacts. Thus, the overall sterilization efficiency is impacted by a combination of

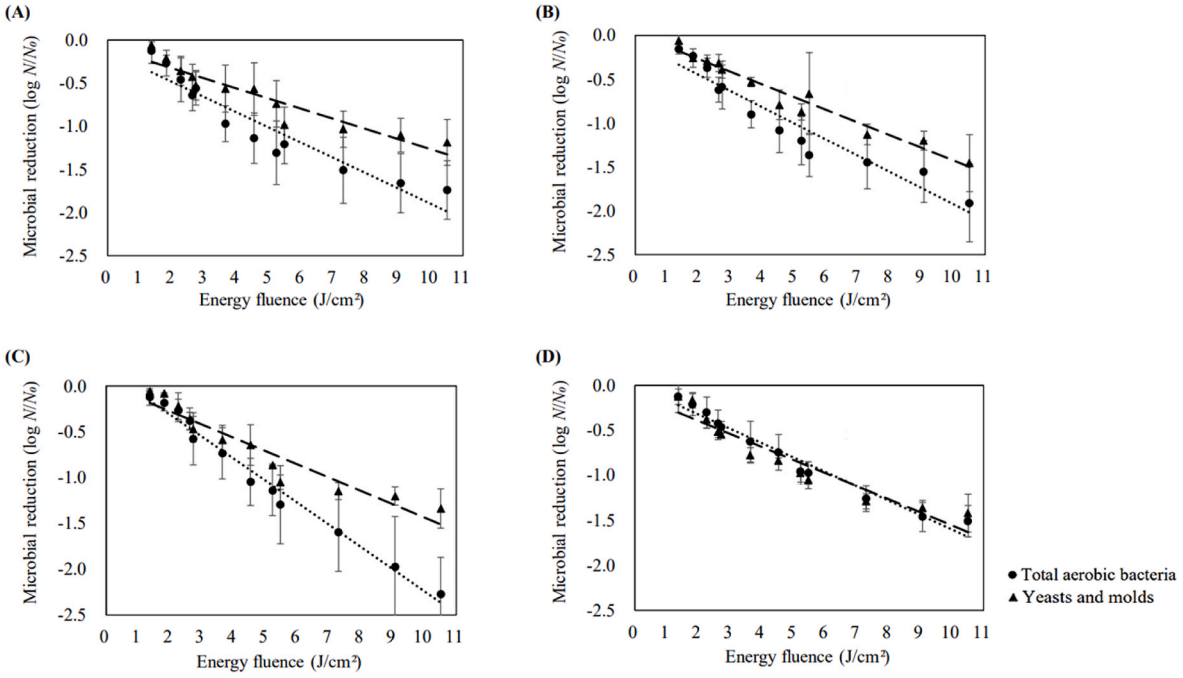


Fig. 2. Microbial reductions in (A) lettuce, (B) perilla leaves, (C) napa cabbage stems, and (D) napa cabbage leaves. The levels of total aerobic bacteria and of yeasts and molds were measured for each sample. Data mean and standard-deviation values.

various factors.

3.2. Quality attributes immediately after IPL treatment

3.2.1. Changes in moisture content

Moisture loss is likely to gradually damage the leaf tissue of leafy vegetables by causing wilting, reducing their elasticity and crispness and so ultimately lowering both the quality and market value of the food product (Ben-Yehoshua & Rodov, 2002; Vigneault et al., 2009). Additionally, moisture loss disrupts the metabolic balance of cells and increases the respiration rate, thereby accelerating senescence and promoting discoloration or decay (Chandra et al., 2021). Fig. 3 presents the changes in moisture content observed after IPL treatment. Across all samples there were only minimal changes in moisture content as a function of lamp voltage and duration. The moisture content ranged from 93.7 % to 94.7 % for lettuce; was initially 88.5 % and remained between 87.0 % and 88.7 % under most treatment conditions for perilla leaves; ranged from 95.2 % to 95.9 % for napa cabbage stems, with the untreated control having an initial moisture content of 95.7 %; and fluctuated between 93.7 % and 94.5 % for napa cabbage leaves, with the control sample initially at 94.1 %. The moisture content in each sample type did not differ significantly with the treatment condition. Reyes-Jurado et al. (2025) similarly observed no changes in the physicochemical properties of sunflower and quinoa seeds after IPL treatment, including in the moisture content. Keklik et al. (2019) also verified that IPL had no significant impact on moisture content in kashar cheese. Likewise, Gyawali et al. (2024) observed no significant changes in pecans, although a slight decrease was noted with longer processing times. These results together indicate that despite involving the application of large amounts of energy, IPL treatment does not substantially alter the moisture content of leafy vegetables.

3.2.2. Changes in pH

pH is an important indicator for tracking chemical changes during storage and for assessing the freshness and stability of leafy vegetables. Pigments such as chlorophylls, carotenoids, and anthocyanins are affected by the pH (Andrés-Bello et al., 2013); for example, chlorophyll breaks down at a low pH, causing green vegetables to turn yellow or brown (Chandra et al., 2021; Faisal et al., 2025). Vegetables generally maintain their freshness better under mildly acidic conditions, but tissue softening may occur at both high and low pH values (Brandt et al., 1984). Fig. 4 presents the pH changes observed after IPL treatment in the present study, indicating that the pH was affected minimally by treatment fluence in all samples, with only slight variations from the untreated samples under either low or high energy fluences. The pH values of untreated lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves were 5.67, 6.08, 5.84, and 6.07, respectively, and after IPL treatment these ranged from 5.58 to 5.66, from 6.06 to 6.24, from 5.70 to 5.99, and from 6.00 to 6.14, respectively. The pH of treated samples did not exhibit consistent changes according to the energy fluence. Furthermore, the statistical analyses revealed that the pH for each sample type did not differ significantly between treatment conditions, suggesting that the slight temperature increase and the light exposure during IPL treatment did not affect the pH values in the samples. Consistent with our results, Orlowska et al. (2013) observed that using a high-intensity pulsed xenon lamp at its lowest energy fluence did not impact the pH of a fructose solution. This was also in agreement with the results of Koh et al. (2016) and Maftei et al. (2014) for pulsed-light-treated cantaloupe and apple juice.

3.2.3. Changes in TPC

Polyphenols are antibacterial and antioxidant compounds found in vegetables that help to remove free radicals (Mandal & Domb, 2024). This makes them important factors influencing the antioxidant capacity and quality attributes of vegetables, including their sensory and nutritional properties (Fabela-Morón, 2024). As summarized in Fig. 5, the TPC values of untreated lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves were 327.57 ± 24.05, 408.12 ± 52.23, 76.20 ± 9.48, and 174.03 ± 5.47 mg/100 g (DWB), respectively, and after IPL treatment these ranged from 235.34 ± 35.51 to 352.14 ± 48.06, from 427.80 ± 10.93 to 541.54 ± 57.03, from 69.30 ± 8.03 to 99.20 ± 20.23, and from 139.20 ± 8.77 to 185.87 ± 13.93 mg/100 g (DWB), respectively. Although the TPC was affected when the energy fluence increased, only slight changes were observed relative to the untreated sample. Some previous studies found that TPC was not significantly affected by IPL, even in polyphenol-rich fruits such as strawberries (Avalos-Llano et al., 2018; Luksiene et al., 2013) and raspberries (Xu & Wu, 2016). However, other studies have also found increases or decreases in TPC associated with IPL treatment. Mandal et al. (2022) observed that the TPC of black tea changed significantly as the fluence was increased up to 3.82 J/cm^2, but found no significant difference at higher fluences. Chakraborty et al. (2020) found that the TPC of Indian gooseberry juice varied with the applied light energy, with a maximum increase of 10 % observed at 2727 J/cm^2 and a maximum decrease of 7 % observed at 3012 J/cm^2. Chakraborty et al. (2022) applied pulsed light to a mixed fruit beverage consisting of apple ber, carambola, and grape, and found that this reduced the TPC by 10–17 %, despite yielding superior results than for thermal pasteurization. Therefore, it can be concluded that the TPC of vegetables and fruits can be modified by IPL treatment.

We derived linear regression equations from the trend lines of TPC as a function of energy fluence (Fig. 5), whose slopes represent the effects of energy fluence on the TPC; these were 0.5738, 7.2985, 0.7563, and −1.8004 for lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves, respectively. Based on the absolute value of the slope, the magnitude of the TPC variation decreased in the following order: perilla leaves > napa cabbage leaves > napa cabbage stems > lettuce. The positive trends observed for lettuce, perilla leaves, and napa cabbage stems suggest that under certain conditions, IPL treatment can enhance the release or extraction of phenolic compounds (Dhar & Chakraborty, 2020). In contrast, the negative trend for napa cabbage leaves indicates the potential degradation of phenolic compounds at higher lamp voltages due to prolonged light exposure or thermal effects.

3.2.4. Quality attributes during storage

Changes in the quality attributes of leafy vegetables treated with IPL at 800 V for 4 min and 2000 V for 2 min (both corresponding to energy fluences of approximately 5 J/cm^2) were observed during a 10-day storage period at 4 °C following IPL treatment. This investigation focused on microbiological and physicochemical properties, including total aerobic bacteria, water loss, pH, and color. Fig. 6 presents the changes observed at the end of the storage period.

The initial populations of total aerobic bacteria were 5.50 ± 0.01, 5.54 ± 0.17, 5.91 ± 0.07, and 6.04 ± 0.04 log CFU/mL for lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves, respectively (data not shown). Following IPL treatment at 800 and 2000 V, the microbial population decreased by approximately 0.7–1.8 log units. However, bacterial proliferation was inevitable in both the treated and untreated samples during the subsequent storage period. By the end of the 10-day storage period, the mean bacterial counts in the untreated samples exceeded 6 log CFU/mL, whereas the treated samples showed mean bacterial levels around 5 log CFU/mL. It is essential to consider the possibility of viable but non-culturable (VBNC) cells, which are known to lose culturability under stress conditions but recover and resume growth in favorable environments. Several studies on UV and pulsed light treatments have reported that VBNC cells can exhibit abnormal regrowth or resuscitation during subsequent storage (Guo et al., 2019; Kramer, 2022). However, in the present study, IPL-treated samples consistently maintained approximately 1 log lower microbial counts than the untreated controls throughout the 10-day storage period without any sudden increase in population. This storage pattern is not compatible with VBNC-mediated recovery and it suggests that the

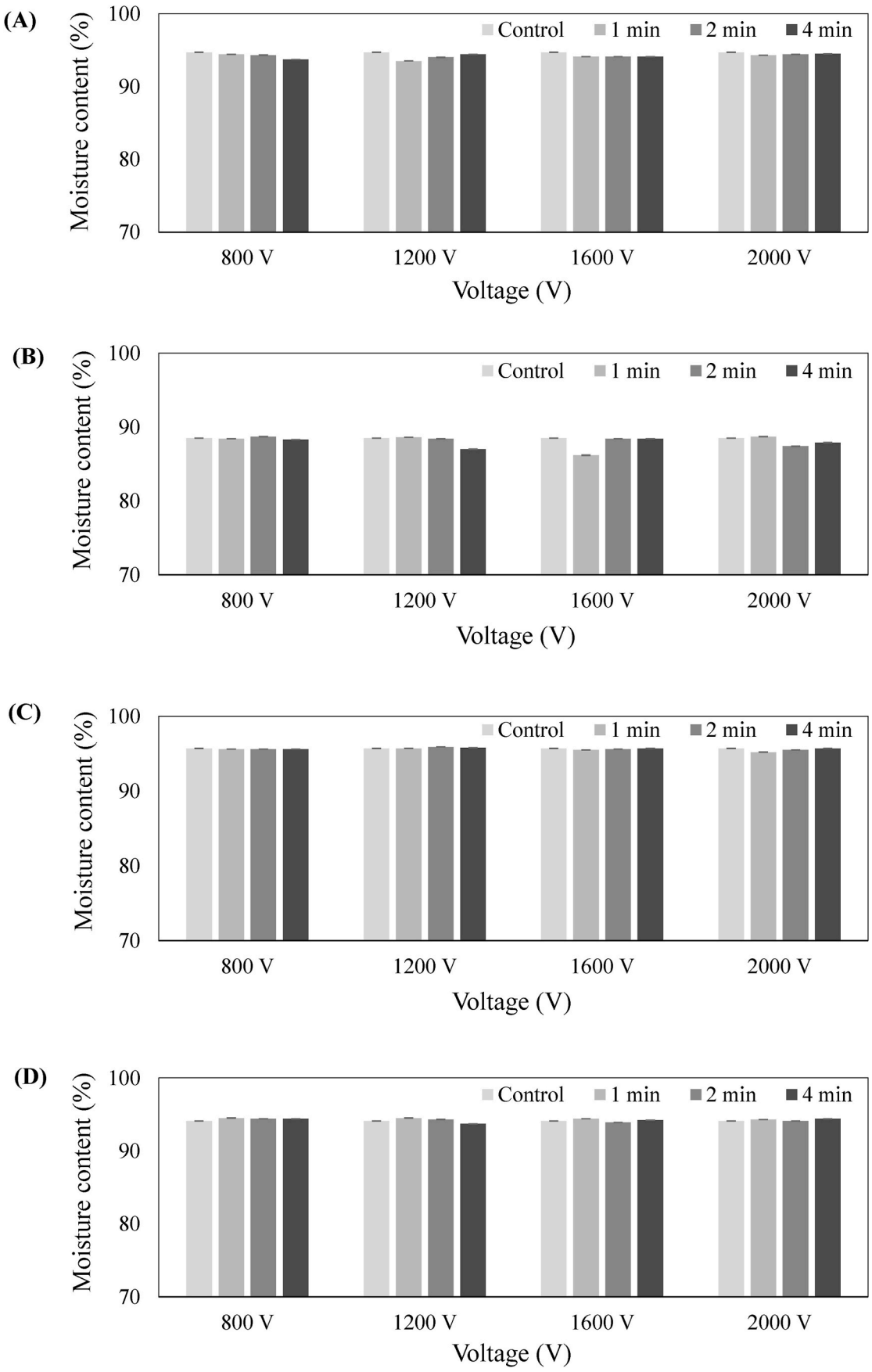


Fig. 3. Changes in moisture contents of (A) lettuce, (B) perilla leaves, (C) napa cabbage stems, and (D) napa cabbage leaves after IPL treatment. Data mean and standard-deviation values.

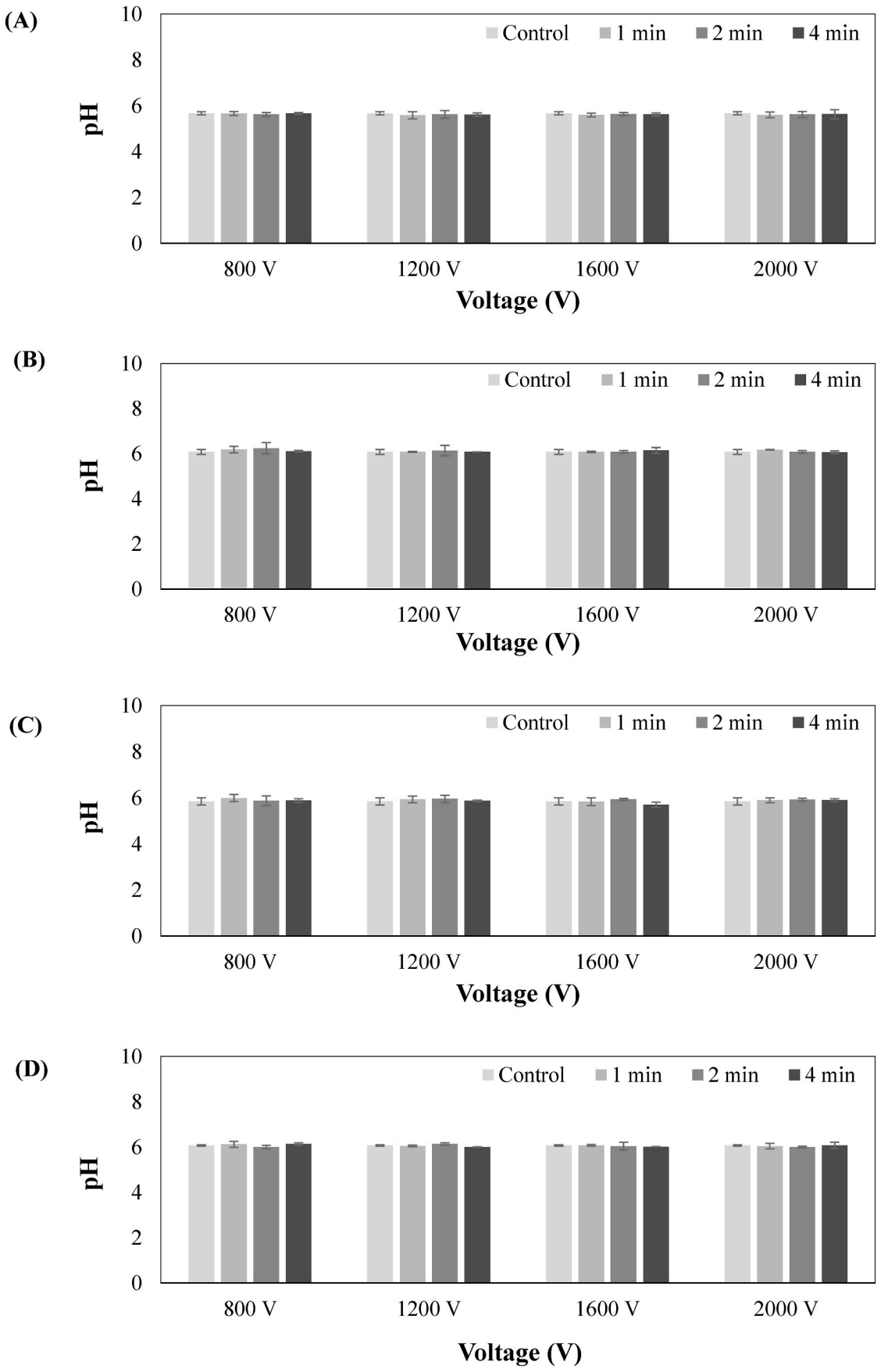


Fig. 4. Changes in pH of (A) lettuce, (B) perilla leaves, (C) napa cabbage stems, and (D) napa cabbage leaves after IPL treatment. Data mean and standard-deviation values.

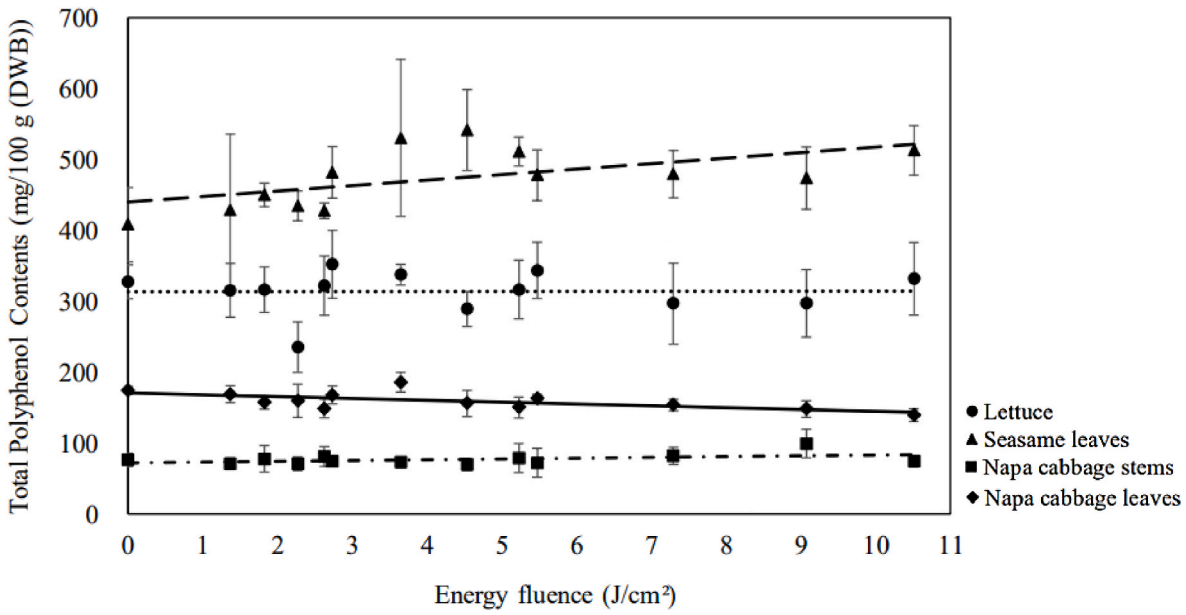


Fig. 5. TPC values of lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves after IPL treatment. Data mean and standard-deviation values.

observed microbial reductions resulted from irreversible inactivation rather than the induction of a dormant VBNC state. It was particularly notable that no statistically significant differences in bacterial counts were found between the 800 V and 2000 V treated samples, suggesting that these two treatments were comparably effective. These findings indicate that IPL treatment regardless of the energy fluence contributed to enhancing microbiological safety by suppressing bacterial growth relative to untreated controls.

Visually, the untreated lettuce, napa cabbage stems, and napa cabbage leaves exhibited fewer red or brown spots and discoloration, while shrinkage was observed for the perilla leaves (images not shown). Similar but more-pronounced changes were observed in samples treated with IPL at 2000 V, whereas fewer changes occurred in those treated with IPL at 800 V relative to the untreated samples. These qualitative observations were quantitatively supported by color analyses using the spectrophotometer. Immediately after IPL treatment there were no significant differences in the L^*, a^*, and b^* color parameters. However, all of the leafy vegetables (including untreated ones) exhibited changes in color during the storage period. These common changes were ascribed to the transformation of chlorophyll into pheophytin, as well as to enzymatic and non-enzymatic browning reactions involving pigments (Zhang et al., 2025). Furthermore, high lamp voltages induced severe discoloration, while treatment at lower lamp voltages resulted in less discoloration than for the untreated samples. The color difference (ΔE) values after 10 days of storage were similar in untreated and 800-V-treated samples, but significantly higher for 2000-V-treated samples. ΔE values can be categorized as follows: hardly noticeable (0–0.5), slightly noticeable (0.5–1.5), noticeable (1.5–3.0), appreciable (3.0–6.0), and very noticeable (6.0–12.0) (Choi et al., 2009; Cserhalmi et al., 2006; Yang et al., 2012). Based on these classification criteria, the color changes after storage for lettuce, perilla leaves, and napa cabbage leaves treated with IPL at 2000 V were clearly distinguishable from those for both untreated and 800-V-treated samples. Similar changes in fresh-cut lettuce were reported by Tao et al. (2019), who found that the highest fluence significantly affected L^* and a^* values during storage, while lower fluences maintained stable L^* values with minimal changes in a^* and b^*. Kramer et al. (2015) also observed lower b^* values and higher a^* values in mung bean sprouts and endive salads, respectively, at 1 and 4 days after IPL treatment. Although the energy fluence was similar, a higher lamp voltage led to greater cell membrane disruption, which increased the interaction between enzymes and substrates, thereby promoting more-severe enzymatic browning reactions (Gómez et al., 2012).

During storage, water in leafy vegetables evaporates through the stomata, resulting in weight loss, and so all of the leafy vegetables in the present study (including untreated samples) exhibited increased water loss as the storage period progressed. However, the untreated and 800-V-treated samples showed similar levels of water loss, whereas the 2000-V-treated samples exhibited a significantly larger weight reduction. In a related study, Valdivia-Nájar et al. (2018) reported that fresh-cut tomatoes weighed 9–10 % less after 20 days of IPL treatment, compared with 7 % less for untreated samples. Similar findings have been reported for Chinese bayberries, where a substantial increase in water loss was associated with IPL treatment (Wang et al., 2023). UV light has been reported to accelerate the chemical degradation of pectin by enhancing the activity of pectolytic enzymes (Kovács & Keresztes, 2002). Pectin is a key structural component of plant cell walls, and is enzymatically hydrolyzed by enzymes such as pectin methylesterase and polygalacturonase that are bound to the cell wall. The degradation of pectin during this process leads to the weakening of the cell wall, tissue softening, cell lysis, and ultimately the release of water retained by the pectic matrix (Rexová-Benková & Markovič, 1976). Therefore, it can be inferred that IPL treatments, including at UV wavelengths, may contribute to increased water loss in leafy vegetables by promoting cell wall degradation by activating pectolytic enzymes.

Regardless of the IPL treatment conditions, the pH values of napa cabbage stems and leaves remained comparable to those of the untreated samples throughout the storage period. Although statistically significant differences in pH were observed for lettuce and perilla leaves, the numerical values of these changes were minimal and unlikely to have any practical implications for product quality or stability. Similarly, tomatoes showed no significant change in pH following IPL treatment or during 21 days of storage (Pataro et al., 2015). Salinas-Roca et al. (2016) also reported that the pH of mango slices remained stable during 14 days of storage. These findings collectively suggest that the pH values of various vegetables and fruits are not substantially affected by IPL exposure, indicating that this treatment does not alter the intrinsic acidity of fresh produce.

It is necessary to optimize the light intensity during IPL treatment in order to minimize potential quality deterioration such as discoloration, browning, and loss of freshness while ensuring the microbial safety of leafy vegetables. Based on the findings presented here, treatment at a

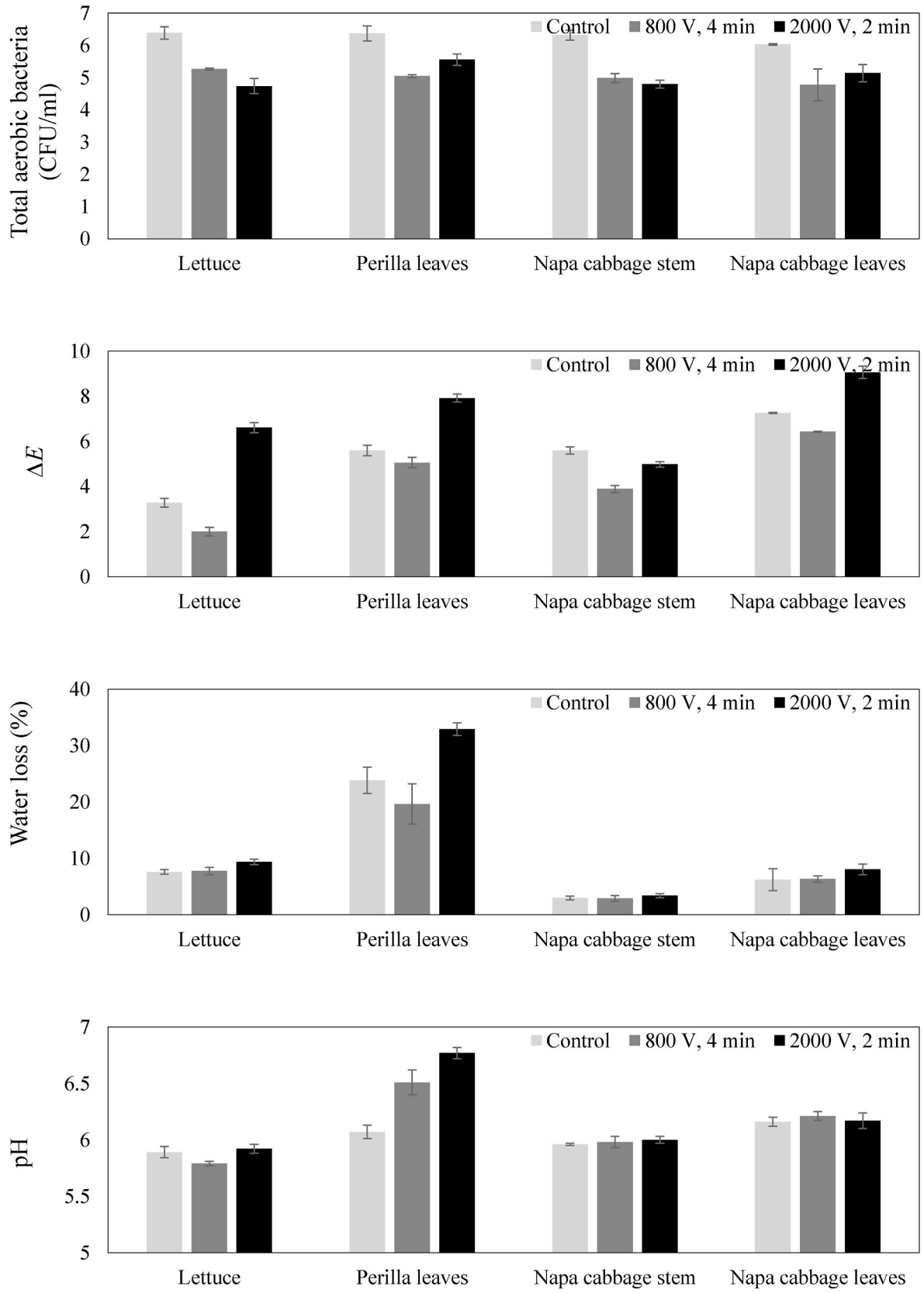


Fig. 6. Changes in (A) total aerobic bacteria, (B) ΔE, (C) water loss, and (D) pH of samples after 10 days of storage. Data mean and standard-deviation values.

lower lamp voltage (800 V) is recommended, since this minimized undesirable changes in color, water loss, and pH while still achieving a significant reduction in microbial populations during storage. Therefore, the application of lower-intensity IPL may serve as a more-balanced approach for simultaneously achieving favorable physicochemical properties and microbial safety.

4. Conclusions

This study evaluated the effects of IPL treatment on microbial inactivation, quality attributes, and the preservation of four types of leafy vegetables: lettuce, perilla leaves, napa cabbage stems, and napa cabbage leaves. The results demonstrated that IPL effectively reduced total aerobic bacteria as well as yeasts and molds, with the degree of microbial inactivation increasing with the energy fluence. Although the degree of microbial reduction varied among vegetable types, IPL proved to be an effective method for microbial control. The observed variations were attributed to surface characteristics, with smoother and structurally simpler vegetables exhibiting fewer shadowing effects and higher light transmittance. Additionally, darker vegetables absorbed more light, which enhanced microbial inactivation. Quality analyses revealed that IPL treatment had negligible effects on the moisture content and pH, while changes in TPC varied with the type of leafy vegetables. Regarding changes in quality attributes during storage, the physicochemical properties were better preserved for treatments with IPL at a lower lamp voltage but the same energy fluence than for treatments at higher lamp voltages, which resulted in chlorophyll degradation and increased water loss due to cell wall damage.

Beyond demonstrating the microbial and quality-related effects of IPL, this study provided important advances compared with previous research. Most earlier IPL studies relied on artificially inoculated pathogens or focused on single produce types, whereas our work uniquely evaluates IPL efficacy against naturally occurring indigenous microorganisms across four leafy vegetables with distinct structural and optical characteristics. This comparative approach clarified how surface roughness, tissue thickness, and color influence the efficiency of IPL inactivation, offering practical insights that have been largely absent from previous IPL literatures. These findings strengthen the applicability of IPL as a minimal processing technology for fresh vegetables encountered under real contamination conditions. Practical applications to fresh vegetables will require further research to identify the most suitable minimal processing conditions by evaluating the effects of a broader range of IPL treatment parameters on both physicochemical and nutritional properties.

CRediT authorship contribution statement

Chae-Rin Lee: Writing – original draft, Visualization, Validation. **Ji-Yoon Lee:** Writing – original draft, Methodology, Formal analysis, Data curation. **Hye-Jae Choi:** Writing – review & editing, Visualization, Validation, Methodology, Investigation, Formal analysis. **Hee-Jeong Hwang:** Writing – review & editing, Validation, Software, Methodology, Investigation, Formal analysis. **Myong-Soo Chung:** Writing – review & editing, Resources, Project administration, Funding acquisition.

Declaration of competing interest

The authors hereby declared no conflict of interest.

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Data availability

Data will be made available on request.

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