Effect of pomegranate (Punica granatum L.) peel extract on the quality and storage stability of dakgalbi prepared from the breast meat of Korean native chicken

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Abstract

Purpose

This study assessed whether pomegranate (*Punica granatum* L.) peel extract (PPE) improves the quality and refrigerated storage stability of soy sauce–marinated dakgalbi prepared from Woorimatdag No. 1 chicken breast.

Methods

Dakgalbi was produced with three formulations: control (CON), 0.14% PPE, and 0.18% PPE, vacuum-packaged, and stored at 4°C for 16 days. Antioxidant/antimicrobial activities of PPE were characterized, and product pH, water-holding capacity, thiobarbituric acid reactive substances (TBARS), total volatile basic nitrogen (VBN), color, total aerobic bacteria, and coliforms were measured on days 1, 8, 12, and 16. Sensory attributes were evaluated by a trained 15-member panel on days 1, 8, and 12.

Results

PPE showed high phenolic content and antimicrobial activity (dose-dependent except against *Listeria monocytogenes*). In dakgalbi, PPE lowered VBN on day 16 and reduced total aerobic bacteria (from day 12) and coliforms (day 16) versus CON. PPE improved water-holding capacity at day 16, while TBARS did not differ among treatments. Appearance scores decreased in PPE treatments after day 8 ($p < 0.05$), but flavor and off-flavor remained stable, and overall acceptability was maintained through day 12.

Conclusion

PPE enhances refrigerated stability of soy sauce–marinated dakgalbi mainly by suppressing microbial growth and proteolysis. Sensory quality remains acceptable through day 12, with the exception of appearance, and 0.14% PPE appears sufficient.

Introduction

Chicken meat is a nutritious and affordable protein source, and growing consumer emphasis on product quality has increased interest in native chicken breeds with distinctive flavor profiles [1, 2]. However, conventional native chickens exhibit slow growth and limited productivity, resulting in unstable supply [3]. To overcome these limitations, improved native breeds such as Woorimatdag have been developed. Woorimatdag No. 1, a three-way crossbreed, shortens the rearing period while retaining the characteristic flavor of traditional native chickens, making it a promising raw material for value-added chicken products [4]. Nevertheless, studies evaluating its application in processed products remain limited.

Dakgalbi is a popular Korean chicken dish and is considered nutritionally favorable due to its use of lean chicken meat and vegetables. Despite its popularity, dakgalbi has a short shelf life, which restricts its competitiveness in the expanding home meal replacement market [5]. Owing to their susceptibility to microbial proliferation and oxidative rancidity, meat products have traditionally relied on synthetic antioxidants; however, safety concerns associated with synthetic additives have driven demand for natural alternatives [6, 7].

Fruit peels are rich in phenolic compounds with strong antioxidant activity, and pomegranate (*Punica granatum* L.) peel in particular contains abundant flavonoids and hydrolysable tannins such as punicalagin [8]. Pomegranate peel extract has been shown to enhance various attributes of food products [9, 10]; however, its application to Korean chicken products remains poorly studied.

Therefore, this study aimed to examine the effect of pomegranate (*Punica granatum* L.) peel extract on the quality and storage stability of dakgalbi prepared from the breast meat of Woorimatdag no. 1 chickens.

Materials and Methods

Preparation of pomegranate peel extract and determination of total phenolic content (TPC)

Fresh pomegranates were sourced from a local online vendor (Vmart, Coupang, Korea). After washing and sterilizing the surface with 70% ethanol, the peels were separated from the arils. The peels were then ground, dried by lyophilization, and subjected to aqueous extraction (1:50 w/v ratio) at 20 ℃ with continuous stirring for 4 h. The resulting solution was passed through Whatman No. 4 filter paper and then freeze-dried to obtain pomegranate peel extract (PPE) powder.

TPC was quantified following the Folin–Ciocalteu method with reference to [11] with slight modifications. Briefly, diluted PPE (0.5 mL) was combined with distilled water (5 mL), followed by the addition of Folin–Ciocalteu reagent. After a 3-min reaction, 1 N sodium carbonate was introduced. The mixture was incubated in the dark (90 min, 25 ℃). A SpectraMax M2 spectrophotometer (Molecular Devices, San Jose, CA, USA) was used to measure absorbance at 760 nm. A gallic acid standard curve was used to derive TPC, which was reported as mg GAE/g DM.

Antioxidant activity of pomegranate peel extract

2,2'-azinobis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) assay

The ABTS assay was conducted according to [11]. ABTS radical cation formation was initiated by mixing ABTS (7 mM) with potassium persulfate (2.45 mM) and incubating the mixture in the dark for 12–16 h. The prepared solution was adjusted with solvent until its absorbance reached 0.700 ± 0.02 at 735 nm. The extract (50 μL) was reacted with the ABTS solution (950 μL) in the dark (30 min, 30°C), and absorbance was read at 735 nm.

Ferric reducing antioxidant power (FRAP)

The FRAP assay was carried out following [11]. The FRAP assay utilized a working reagent consisting of 300 mM acetate buffer, 20 mM $FeCl_3 \cdot 6H_2O$ and 10 mM TPTZ in 40 mM HCl (1:1:10 ratio). The sample (25 μL) was blended with the FRAP reagent (175 μL) and incubated (30 min, 37°C). Absorbance was monitored at 590 nm.

Oxygen radical absorbance capacity (ORAC)

ORAC analysis was performed using a method reported previously [11]. The extract (25 μL, 60 μg/mL) was combined with fluorescein (150 μL, 80 nM) and incubated (15 min, 37°C) prior to initiating the reaction with AAPH (25 μL, 150 mM). Fluorescence was monitored at 1-min intervals at 37°C (Ex 480 nm, Em 520 nm) using a SpectraMax M2 spectrophotometer. Antioxidant capacity was determined relative to Trolox and presented as mmol TE per g of sample.

Antimicrobial activity of pomegranate peel extract

Bacterial strains and culture conditions

The antimicrobial efficacy of PPE was tested against *Staphylococcus aureus* (KCCM 12256), *Listeria monocytogenes* (KCCM 40307), *Salmonella Enteritidis* (CCARM 8260), and *Escherichia coli* (KCCM 11234). Cultures were grown on Mueller–Hinton agar (MHA) or broth (MHB) at 37°C (*L. monocytogenes* at 30°C).

Paper disc diffusion assay

To evaluate antimicrobial effects, inhibition zones were determined using the disc diffusion approach described in [11]. PPE stock solutions (1.25, 2.5, 5, and 10 mg/disc) were prepared in DMSO and filtered (0.45 μm). Bacterial suspensions (5–6 log CFU/mL) were spread on MHA plates. Agar plates were overlaid with sterile paper discs (8 mm), each receiving 50 μL of PPE solution. DMSO served as the negative control, whereas streptomycin was used as the positive control. Inhibition zone diameters were recorded after 24 h of incubation.

Preparation and packaging of dakgalbi

Breast meat from Woorimatdag No. 1 was skinned, diced into approximately 1 cm cubes, and mixed with the designated sauce (Table 1) at a 4:1 meat-to-sauce ratio. Based on preliminary sensory screening conducted to select acceptable addition levels (data not shown), PPE was incorporated into the sauce at 0.14% and 0.18%. Each treatment was prepared in three independent production batches (n = 3). For each batch, portions were vacuum-sealed in BPA-free film (0.08 mm thickness) using a Lovero SBV-400 TS machine (Sambo Tech, Gimpo, Korea) and stored at 4°C. Separate packages were analyzed after 1, 8, 12, and 16 days of refrigerated storage.

Proximate composition and pH

Proximate analysis followed AOAC standards [12]. A homogenate was prepared by combining dakgalbi (3 g) with 27 mL of distilled water. The pH of the resulting homogenate was recorded using an Orion 230A pH meter (Thermo Electron Corporation, Beverly, MA, USA).

Water-holding capacity (WHC)

WHC was determined by heating 0.5 g of the sample (20 min, 80°C) and subsequently cooled. The sample was subjected to centrifugation at 2,000 × g (20 min, 4°C). WHC was calculated as the percentage of moisture retained relative to the total moisture content, accounting for fat content [11].

WHC (%) = [(total moisture content – centrifugal water loss) / total moisture content] × 100

Centrifugal water loss = [(pre-centrifugation weight – post-centrifugation weight) / (initial sample weight × fat content correction factor)] × 100

Fat content correction factor = 1 – (crude fat / 100)

Thiobarbituric acid reactive substances (TBARS)

Lipid oxidation was evaluated by TBARS as described previously [1]. Dakgalbi (5 g) was mixed with distilled water (15 mL) and 7.2% BHA (50 μL) for 30 s using a PolyTron® PT-2500E. The homogenate (1 mL) was mixed with TBA solution (2 mL, 20 mM thiobarbituric acid (in 15% trichloroacetic acid)), heated (15 min, 90°C), cooled (10 min, room temperature), and centrifuged (2,000 × g, 10 min, 4°C). The absorbance of the collected supernatant was recorded at 531 nm with a SpectraMax M2 spectrophotometer. TBARS values were expressed as milligrams of malondialdehyde (MDA) per kilogram of sample:

TBARS (mg MDA/kg sample) = (Asample – Ablank) × 5.88

Total volatile basic nitrogen (VBN)

A Conway microdiffusion cell was used to quantify VBN, following the procedure in [1]. After homogenization of 10 g sample with 50 mL distilled water (30 min, 4°C), the mixture was filtered using Whatman No. 1 paper. Inside the cell, H_2SO_4 (0.01 N, 1 mL) was placed in the inner well, while 1 mL of the filtrate and saturated K_2CO_3 (1 mL) were added to the outer well. Following sealing and reaction (60 min, 25°C), the inner well solution was analyzed by NaOH (0.01 N) neutralization with Brunswick reagent (10 μL) to determine VBN (mg/100 g).

Instrumental color

Surface color was recorded using a CR-400 colorimeter (Minolta, Japan) equipped with an 8 mm aperture and illuminant C. Samples were removed from vacuum packaging, allowed to bloom for 10 min, and measured at multiple surface locations, including outer and cross-sectional areas.

Microbiological analysis

Microbiological analysis of total aerobic bacteria (TAB), coliforms, and *E. coli* was performed with Petrifilm plates (Aerobic Plate Count and Coliform/*E. coli* Count Plates; 3M, St. Paul, MN, USA). Dakgalbi (3 g) was blended with sterile saline (27 mL) using a stomacher (BagMixer 400; Interscience, France), serially diluted, and inoculated (1 mL). After incubation (48 h, 37°C), microbial counts were recorded by log CFU/g.

Sensory evaluation

A panel of 15 trained judges from Kangwon National University assessed the sensory qualities. Pan-grilled samples were evaluated for appearance, aroma, taste, flavor, and overall acceptability using a 9-point hedonic scale (1 = extremely undesirable; 9 = extremely desirable). Tenderness (1 = very tough; 9 = very tender), juiciness (1 = very dry; 9 = very juicy), and off-flavor intensity (1 = very strong; 9 = very weak) were also assessed. The Institutional Review Board of Kangwon National University granted ethical approval, approval number KWNUIRB-2021-05-004-001, and all participants provided written consent prior to participation.

Statistical analysis

Statistical analyses were performed using the SAS (version 9.2). One-way ANOVA was used to compare treatments within each storage day and to compare storage days within each treatment. Means were compared using Tukey's method whenever statistical significance was observed at $p < 0.05$. Data represent three independent production batches per treatment at each storage day ($n = 3$).

Results and Discussion

Antioxidant capacity and antimicrobial activity of pomegranate peel extract (PPE)

The antioxidant activity of PPE is shown in Fig. 1. The TPC of water-soluble PPE was 179.16 ± 3.83 mg GAE/g DM (data not shown), consistent with values previously reported in the literature [13].

Antioxidant capacity assessed by ABTS, FRAP, and ORAC assays was 2735.67 ± 65.04, 2285.85 ± 4.07, and 1063.82 ± 118.38 mol Trolox equivalents (TE)/g DM, respectively. Consistent with previous reports, ORAC values were lower than ABTS values [14], and FRAP results fell within the range reported for pomegranate peel extracts from different cultivars [15]. These results indicate that PPE possesses strong antioxidant potential and is potentially suitable as a naturally derived antioxidant for enhancing storage stability and improving the quality of meat products such as dakgalbi.

The antimicrobial activity of PPE was evaluated using a paper disc diffusion assay against Gram-positive (*Staphylococcus aureus, Listeria monocytogenes*) and Gram-negative bacteria (*Salmonella Enteritidis, Escherichia coli*) (Table 2). PPE (1.25–10 mg/disc) generated measurable inhibition zones against all tested strains, and the inhibitory effect increased as the applied dose increased ($p < 0.05$). Among the tested microorganisms, *S. Enteritidis* exhibited the highest sensitivity to PPE, while *L. monocytogenes* showed relatively weaker susceptibility. Similar antimicrobial effects of PPE against foodborne pathogens have been reported previously [16]. The antimicrobial effects observed may be attributed to the ability of phenolic constituents to disrupt bacterial cell membranes [9]. Notably, PPE produced inhibition zones exceeding 10 mm even at 1.25 mg/disc, indicating strong antimicrobial potential [17].

Proximate composition

The proximate composition of soy sauce–marinated dakgalbi supplemented with PPE is shown in Table 3. The proximate composition (moisture, crude fat, crude protein, and crude ash) did not differ significantly among treatments. Similarly, pomegranate seed extract was reported to have no effect on the proximate composition of low-fat emulsion-type pork sausages during storage [18]. Collectively, these results indicate that PPE supplementation does not affect the basic nutritional composition of soy sauce dakgalbi prepared from Woorimatdag breast meat, supporting its use without compromising key nutritional attributes.

Table 1. Formulation of soy sauce for the marinated Woorimatdag No. 1 chicken

Ingredients (%)	Treatment		
	Control	0.14PPE	0.18PPE
Soy sauce	40.00	40.00	40.00
Starch syrup	12.00	12.00	12.00
Sugar	12.00	12.00	12.00
Oyster sauce	5.00	5.00	5.00
Garlic	12.00	12.00	12.00
Ginger	2.00	2.00	2.00
Onion	8.00	8.00	8.00
Pepper	1.00	1.00	1.00
Cooking rice wine	6.00	6.00	6.00
Sesame oil	2.00	2.00	2.00
Pomegranate peel extract	0.00	0.14	0.18

0.14PPE, dakgalbi with 0.14% pomegranate peel extract addition; 0.18PPE, dakgalbi with 0.18% pomegranate peel extract addition.

Table 2. The antimicrobial effect of pomegranate peel extract, using the paper disc diffusion assay method (unit: mm)

mg/disc	*E. coli*	*S. enteritidis*	*L. monocytogense*	*S. aureus*
PC	16.67^{B}	13.28^{D}	15.57	12.63^{E}
1.25	13.54^{C}	14.54^{D}	11.94	14.01^{D}
2.5	15.47^{BC}	16.44^{C}	12.89	16.23^{C}
5	19.24^{A}	19.57^{B}	14.57	19.12^{B}
10	21.17^{A}	22.34^{A}	14.35	21.76^{A}
SEM	0.563	0.377	5.653	0.218

Diameter of paper disc (8mm) was included in the value; PC, positive control (Streptomycin, 0.01 mg/disc for *E. coli, L. monocytogenes, and S. aureus*, 0.20mg/disc for *S. Enteritidis*.).

$^{A\text{-}E}$ Means denoted by different superscripts within the same column indicate a significant difference ($p < 0.05$).

Table 3
Proximate composition of the dakgalbi with pomegranate peel extract

Treatment	Proximate composition (%)			
	Moisture	**Crude protein**	**Crude fat**	**Crude ash**
Control	71.49	20.31	0.89	1.88
0.14PPE	71.65	20.20	0.83	1.88
0.18PPE	71.45	20.73	0.91	2.01
SEM	0.154	1.926	0.006	0.015

0.14PPE, dakgalbi with 0.14% pomegranate peel extract addition; 0.18PPE, dakgalbi with 0.18% pomegranate peel extract addition.

pH

Changes in pH of soy sauce–marinated dakgalbi supplemented with PPE are shown in Table 4. At the initial storage point, pH values ranged from 5.59 to 5.64 across all treatments and were lower than those reported for gochujang-marinated dakgalbi [19], likely reflecting the influence of the soy sauce–based marinade. A similar reduction in initial pH has been reported in soy sauce–treated, vacuum-packed pork compared with gochujang- or soybean paste–marinated samples [20]. During storage, pH in the control and 0.18PPE increased on day 8 and decreased after day 12 ($p < 0.05$), whereas pH in 0.14PPE remained unchanged throughout storage ($p > 0.05$), consistent with previous findings in dakgalbi products [5]. The pH decrease after day 12 may be attributed to lactic acid bacteria proliferation in vacuum-packaged meat [4]. Notably, the control group exhibited lower pH than the PPE-treated groups after day 12, suggesting that PPE may have moderated acidification during storage. According to [11], chicken meat with pH values of 5.5–6.2 is considered microbiologically acceptable. In the present study, pH values remained within this range through day 16; however, shelf-life evaluation should be supported by microbiological and chemical spoilage indicators.

Table 4
Changes in the physicochemical properties of the dakgalbi with pomegranate peel extract during storage at 4°C

Trait	Treatment	Storage day				
		1	8	12	16	SEM
pH	Control	5.61^{b}	5.68^{a}	5.53^{Bc}	5.05^{Bd}	0.003
	0.14PPE	5.64^{a}	5.69^{a}	5.66^{Aa}	5.20^{Ab}	0.006
	0.18PPE	5.59^{c}	5.68^{a}	5.63^{Ab}	5.19^{Ad}	0.001
	SEM	0.001	0.001	0.001	0.001	
Water holding capacity (%)	Control	56.63^{ab}	58.89^{a}	53.96^{Bb}	46.10^{Cc}	2.959
	0.14PPE	56.13^{ab}	60.40^{ab}	62.23^{Aa}	51.46^{Ab}	4.529
	0.18PPE	55.92^{a}	58.42^{a}	53.19^{Bab}	49.18^{Bb}	4.942
	SEM	3.373	7.206	4.339	0.333	
TBARS (mg MDA/kg)	Control	0.98^{b}	1.15^{ab}	1.29^{a}	1.12^{ab}	0.006
	0.14PPE	0.92^{b}	1.07^{a}	1.17^{a}	1.13^{a}	0.002
	0.18PPE	0.97	1.09	1.12	1.11	0.004
	SEM	0.003	0.004	0.009	0.001	
VBN (mg%)	Control	18.51^{c}	22.61^{b}	24.36^{b}	32.22^{Aa}	0.607
	0.14PPE	19.19^{c}	21.76^{bc}	22.66^{b}	27.46^{Ba}	0.610
	0.18PPE	18.09^{c}	22.07^{b}	22.85^{b}	28.66^{Ba}	0.544
	SEM	0.748	0.324	0.702	0.475	

0.14PPE, dakgalbi with 0.14% pomegranate peel extract addition; 0.18PPE, dakgalbi with 0.18% pomegranate peel extract addition; TBARS, 2-thiobarbituric acid reactive substances; VBN, volatile basic nitrogen; SEM, standard error of mean.

[A-C]Means denoted by different superscripts within the same column indicate a significant difference ($p < 0.05$).

[a-d]Means denoted by different superscripts within the same row indicate a significant difference ($p < 0.05$).

Water-holding capacity (WHC)

Changes in WHC of soy sauce–marinated dakgalbi supplemented with PPE are shown in Table 4. In the control group, WHC increased slightly from 56.63% (day 1) to 58.89% (day 8) ($p > 0.05$) and then decreased to 46.10% by day 16 ($p < 0.05$). In the 0.14PPE group, WHC increased from 56.13% to 62.23% on day 12 ($p > 0.05$) and then decreased to 51.46% on day 16 ($p < 0.05$). Similarly, WHC in the 0.18PPE group increased from 55.92% (day 1) to 58.42% (day 8) ($p > 0.05$) and decreased to 49.18% (day 16) ($p < 0.05$). A transient increase in WHC during storage has also been reported for dakgalbi supplemented with calamansi extract [11], potentially due to proteolysis that generates additional water-binding sites. On day 16, WHC was significantly higher in PPE-treated groups—especially 0.14PPE—than in the control. Because WHC is directly associated with processing yield and sensory attributes of raw and processed meat [21], these findings suggest that PPE supplementation may improve WHC in dakgalbi during extended storage.

Thiobarbituric acid reactive substances (TBARS)

As shown in Table 4, TBARS values in soy sauce–marinated dakgalbi were monitored following PPE supplementation. Initial TBARS values were similar among treatments (0.92–0.98 mg MDA/kg). Over the storage period, TBARS levels rose in the control and 0.14PPE treatments ($p < 0.05$), while remaining statistically unchanged in the 0.18PPE group ($p > 0.05$), reflecting the progression of lipid oxidation under refrigerated conditions. A comparable increase in TBARS over storage has been reported for raw chicken meat treated with spice extracts [22]. However, no statistically significant differences in TBARS values were observed across treatments throughout the storage period. Thus, under the conditions of this study, PPE did not effectively inhibit lipid oxidation in soy sauce–marinated dakgalbi prepared from Woorimatdag No. 1 breast meat. This result is consistent with [11], who reported that calamansi extract suppressed oxidation in thigh-meat dakgalbi but not in breast-meat dakgalbi.

Total volatile basic nitrogen (VBN)

Changes in VBN values of soy sauce–marinated dakgalbi supplemented with PPE are shown in Table 4. At the beginning of storage, VBN values ranged from 18.09 to 19.19 mg% across treatments and exceeded those reported for fresh chicken breast [1], which may be attributed to the soy sauce–based marinade. During storage, VBN values increased in parallel with total aerobic bacterial counts (Fig. 2), reflecting protein degradation associated with microbial and enzymatic activity [23]. At day 16, the control group showed a significantly higher VBN value (32.22 mg%) than the 0.14PPE (27.46 mg%) and 0.18PPE (28.66 mg%) groups, indicating that PPE supplementation attenuated protein degradation during storage. Although VBN values above 20 mg% are generally considered indicative of spoilage in fresh meat [24], elevated VBN levels in processed meat products do not necessarily reflect spoilage to the same extent [25].

Instrumental color

Changes in instrumental color of soy sauce–marinated dakgalbi supplemented with PPE are shown in Table 5. For CIE L* (lightness), no significant change was observed in the control during storage, whereas PPE-treated samples showed a significant increase on day 16. Lightness was consistently higher in the control than in PPE-treated groups, likely due to the darker appearance imparted by dried PPE, as processing conditions such as drying can markedly affect color development [26]. For CIE a* (redness), the 0.14PPE group exhibited a higher value on day 12, but otherwise no significant within- or between-treatment differences were observed, indicating stable redness during storage. Redness values in this study (3.80–5.19) were higher than those reported for raw broiler and Korean native chicken breast meat [1], likely due to pigments from dakgalbi seasonings. This is consistent with [27], who reported that pomegranate peel extract darkened chicken patties without affecting redness. For CIE b* (yellowness), values in the control and 0.14PPE groups decreased initially and then increased during storage ($p < 0.05$), whereas no significant change in CIE b* was observed in the 0.18PPE group across storage days ($p > 0.05$). Across storage periods, PPE-treated groups consistently exhibited lower yellowness than the control, in agreement with previous findings in frankfurters supplemented with pomegranate peel extract [28].

Table 5
Changes in the surface color of the dakgalbi with pomegranate peel extract during storage at 4°C

Color	Treatment	Storage day				
		1	8	12	16	SEM
CIE L*	Control	45.24^{A}	43.72^{A}	44.08^{A}	46.19^{A}	2.426
	0.14PPE	37.62^{Bb}	37.16^{Bb}	40.28^{Ba}	41.34^{Ba}	0.734
	0.18PPE	38.62^{Bb}	38.75^{Bb}	39.21^{Bb}	40.72^{Ba}	0.256
	SEM	3.262	0.610	0.351	0.332	
CIE a*	Control	4.26	4.78	4.58	4.04	0.183
	0.14PPE	3.80^{b}	4.19^{b}	5.19^{a}	4.24^{b}	0.047
	0.18PPE	4.30	4.55	4.71	4.44	0.171
	SEM	0.169	0.095	0.098	0.172	
CIE b*	Control	19.92^{Aa}	16.90^{Ac}	18.36^{Ab}	17.12^{Ac}	0.144
	0.14PPE	15.18^{Cb}	13.73^{Bc}	16.82^{Ba}	15.98^{Bab}	0.183
	0.18PPE	16.19^{Ba}	14.75^{Bb}	15.11^{Cab}	15.87^{Bab}	0.299
	SEM	0.128	0.415	0.175	0.117	

0.14PPE, dakgalbi with 0.14% pomegranate peel extract addition; 0.18PPE, dakgalbi with 0.18% pomegranate peel extract addition; SEM, standard error of mean.

$^{A\text{-}C}$Means denoted by different superscripts within the same column indicate a significant difference ($p < 0.05$).

$^{a\text{-}c}$Means denoted by different superscripts within the same row indicate a significant difference ($p < 0.05$).

Microbiological analysis

Changes in microbial counts of soy sauce–marinated dakgalbi supplemented with PPE are shown in Fig. 2. Total aerobic bacteria (TAB), *E. coli*, and coliforms were monitored during 16 days of refrigerated storage (4°C). TAB populations rose significantly during storage in all treatments ($p < 0.05$), although no treatment-related differences were detected up to day 8. From day 12 onward, the control exhibited a marked increase in TAB counts compared with samples receiving PPE treatment. On day 16, TAB counts were 6.27, 5.96, and 5.89 log CFU/g for the control, 0.14PPE, and 0.18PPE groups, respectively, all below the MFDS limit (6.70 log CFU/g), although the control group was closer to the threshold ($p < 0.05$). These results indicate that PPE supplementation suppressed TAB growth during storage. Coliform counts increased markedly by day 16. No treatment differences were observed through day 12, except for slightly higher counts in the 0.14PPE group on day 1. At day 16, coliform levels were significantly greater in the control compared with PPE-treated samples ($p < 0.05$), indicating that PPE exerted an inhibitory effect. No detectable levels of *E. coli* were observed in any treatment throughout storage. In line with prior studies, pomegranate peel extract has been associated with reduced bacterial growth in meat products [28]. The antimicrobial activity of PPE is primarily attributed to phenolic compounds and flavonoids, which are known to disrupt bacterial cell membranes [9].

Sensory evaluation

Sensory evaluation results of soy sauce–marinated dakgalbi supplemented with PPE are presented in Table 6. Sensory evaluation serves as a critical factor in determining the acceptability of food products and provides indicators for the potential success of future products in the market [29]. Sensory attributes were evaluated over 12 days of storage. With the exception of appearance, sensory scores were largely comparable across all samples. Appearance ratings for PPE-containing samples declined after the eighth day of storage relative to untreated samples ($p < 0.05$), suggesting that PPE influenced visual attributes during storage. In the control group, flavor and off-flavor scores deteriorated significantly by day 12, whereas PPE-treated groups maintained these attributes without significant changes throughout the evaluation period. Overall, PPE supplementation did not negatively affect sensory quality or consumer acceptability of soy sauce–marinated dakgalbi through day 12. Sensory testing at day 16 was not performed due to panel safety considerations for samples stored for an extended period. These findings are consistent with previous reports showing no adverse effects of PPE on sensory attributes of cooked chicken patties [10] and support the potential use of PPE as a natural preservative in this product.

Table 6
Changes in the sensory properties of the dakgalbi with pomegranate peel extract during storage at 4°C

Trait	Treatment	Storage day			
		1	8	12	SEM
Appearance	Control	7.58	7.50^{A}	7.17^{A}	0.775
	0.14PPE	6.92	6.50^{B}	6.25^{AB}	1.399
	0.18PPE	7.17^{a}	6.00^{Bb}	6.08^{Bb}	1.169
	SEM	1.500	0.789	1.056	
Aroma	Control	7.08	7.50	6.92	0.753
	0.14PPE	7.33	7.25	6.83	1.048
	0.18PPE	7.67^{a}	7.00^{ab}	6.75^{b}	0.816
	SEM	0.674	0.705	1.237	
Taste	Control	7.17	7.42	6.83	0.795
	0.14PPE	7.42	7.08	6.83	1.379
	0.18PPE	7.33	7.08	6.92	0.803
	SEM	1.008	0.932	1.038	
Flavor	Control	7.17^{a}	7.42^{a}	6.25^{b}	0.753
	0.14PPE	7.33	7.00	6.50	1.384
	0.18PPE	7.08	6.83	6.67	1.068
	SEM	0.826	0.745	1.634	
Off-flavor	Control	7.33^{ab}	7.58^{a}	6.58^{b}	0.924
	0.14PPE	7.42	7.42	6.92	1.477
	0.18PPE	7.25	7.00	7.08	1.247
	SEM	0.965	0.783	1.902	
Juiciness	Control	5.50	6.17	6.00	1.414
	0.14PPE	5.67	6.00	6.58	1.624
	0.18PPE	6.00	5.83	6.25	2.119
	SEM	1.626	2.586	0.944	

Tenderness	Control	6.33	6.00	6.25	1.725
	0.14PPE	6.42	5.92	6.92	2.205
	0.18PPE	7.00	5.83	6.42	2.018
	SEM	1.139	3.472	1.336	
Overall acceptability	Control	6.92	7.25	6.58	0.912
	0.14PPE	7.17	6.58	6.67	1.311
	0.18PPE	7.00	6.33	6.67	1.071
	SEM	0.866	1.328	1.098	

Appearance, 1=extremely undesirable, 9=extremely desirable; Aroma, 1=extremely undesirable, 9=extremely desirable; Taste, 1=extremely undesirable, 9=extremely desirable; Flavor, 1=extremely undesirable, 9=extremely desirable; Off-flavor, 1=very strong, 9=very weak; Juiciness, 1=very dry, 9=very juicy; Tenderness, 1=very tough, 9=very tender; Overall acceptability, 1=extremely undesirable, 9=extremely desirable.

0.14PPE, dakgalbi with 0.14% pomegranate peel extract addition; 0.18PPE, dakgalbi with 0.18% pomegranate peel extract addition; SEM, standard error of mean.

^{A,B}Means denoted by different superscripts within the same column indicate a significant difference ($p < 0.05$).

^{a,b}Means denoted by different superscripts within the same row indicate a significant difference ($p < 0.05$).

Conclusions

Pomegranate peel extract (PPE) enhanced the refrigerated storage stability of soy sauce–marinated dakgalbi prepared from Woorimatdag No. 1 breast meat, primarily by suppressing microbial proliferation (total aerobic bacteria and coliforms) and attenuating proteolysis, as indicated by reduced VBN, while improving water-holding capacity and maintaining proximate composition and overall sensory acceptability. However, PPE did not significantly reduce TBARS, and potential appearance/color penalties were observed. Further work is warranted to verify mechanisms (microbial community shifts and active-compound standardization) and to confirm performance across broader processing and product conditions.

Declarations

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Ethics Approval

The sensory evaluation received ethical approval from the Institutional Review Board of Kangwon National University (KWNUIRB-2021-05-004-001).

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Figures

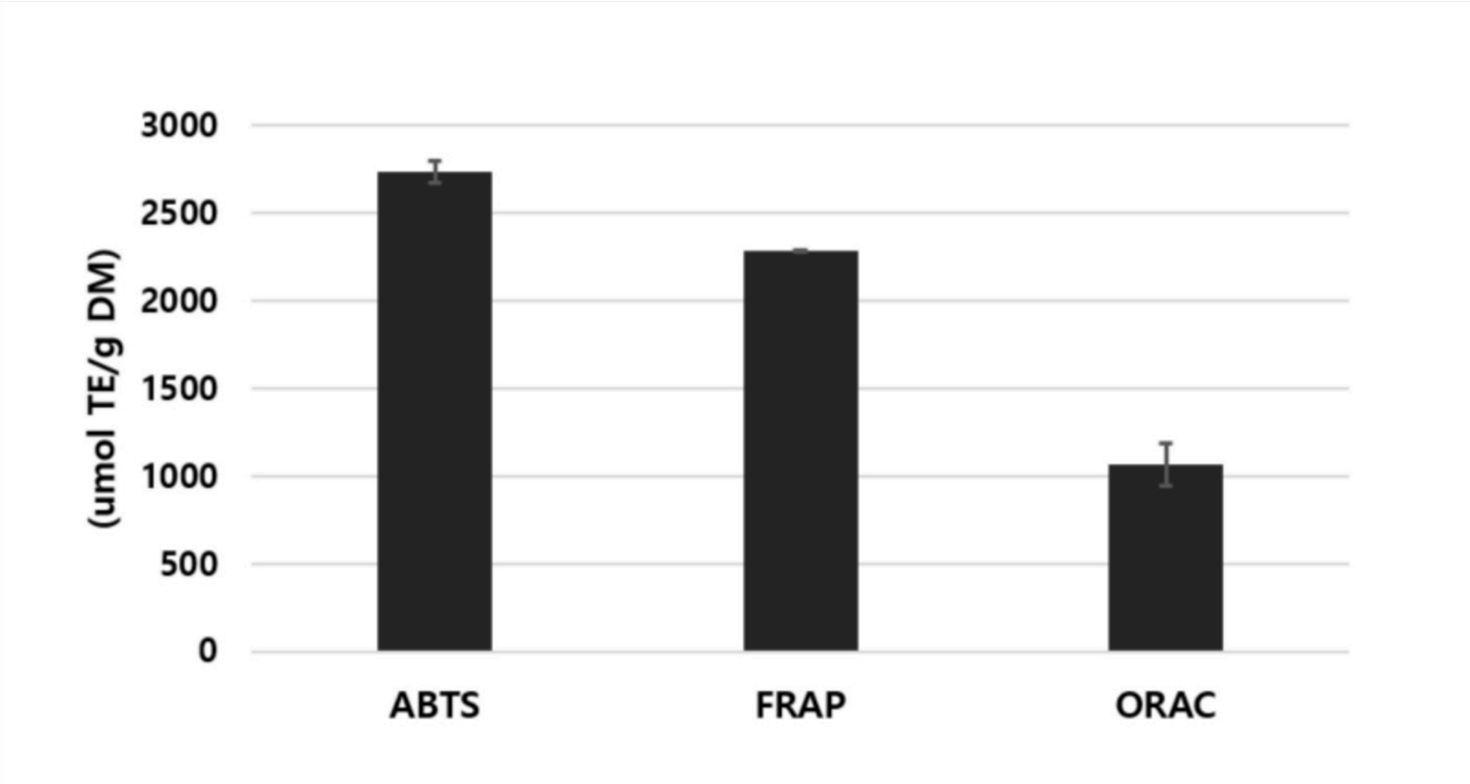

Figure 1

Antioxidant activity of pomegranate peel extract.

ABTS, 2,2'-azinobis-3-ethylbenzothiazoline-6-sulfonic acid; FRAP, ferric reducing antioxidant power; ORAC, oxygen radical absorption capacity; TE, trolox equivalent; DM, dry matter.

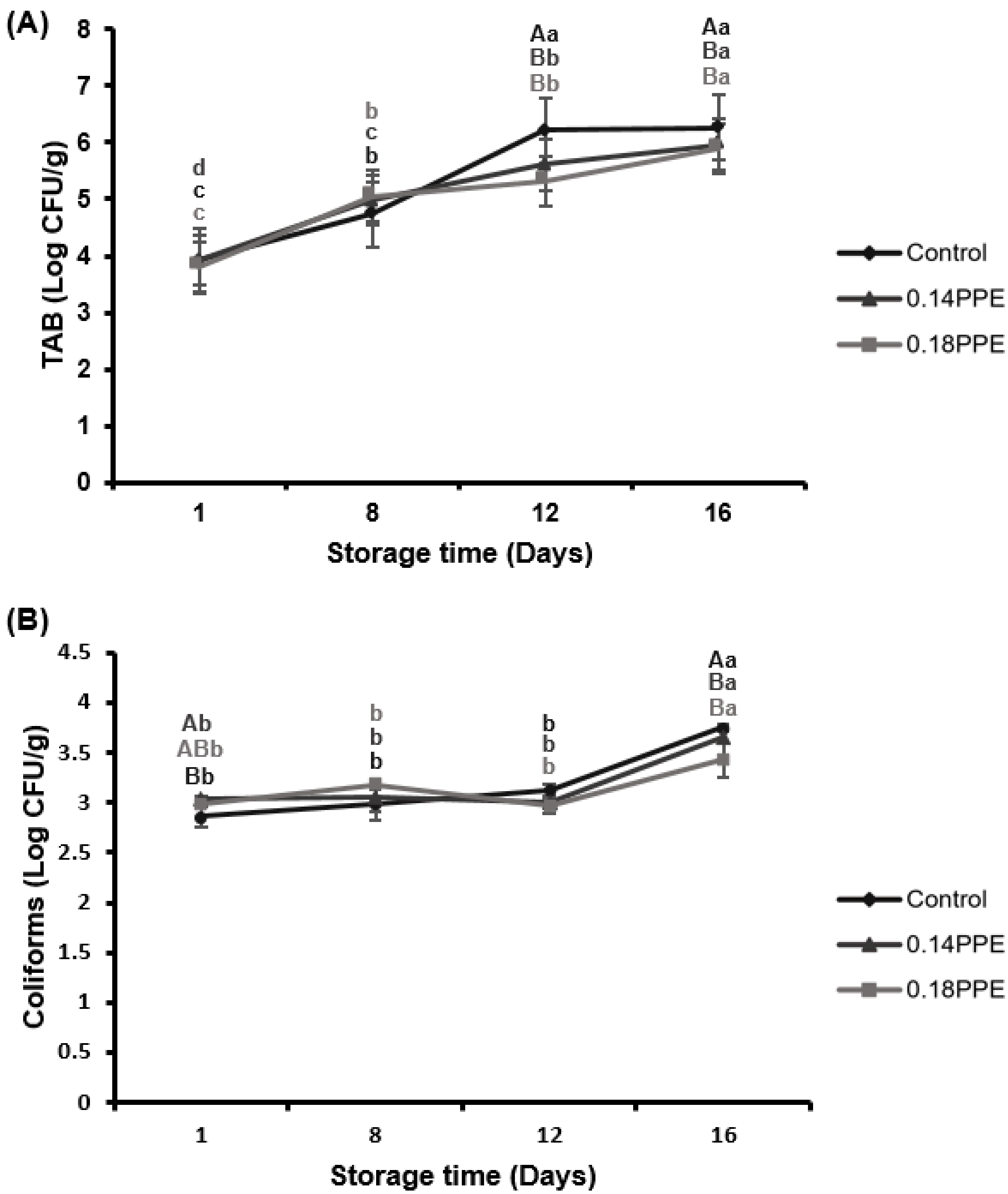

Figure 2

Changes in the TAB (A) and coliforms (B) of the dakgalbi with pomegranate peel extract during storage at 4°C.

0.14PPE, dakgalbi with 0.14% pomegranate peel extract addition; 0.18PPE, dakgalbi with 0.18% pomegranate peel extract addition; TAB, Total aerobic bacteria; [A,B]Means denoted by different superscripts among treatments differ significantly ($p < 0.05$); [a-d]Means denoted by different superscripts between storage days differ significantly ($p < 0.05$).