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Effects of *Boswellia Serrata* and Whey Protein Powders on Physicochemical Properties of Pork Patties

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Abstract: Processed meat products are prone to oxidative damage and quality decline during storage; however, these problems can be mitigated by the proper formulation of meat productions. This study evaluated the effects of natural anti-oxidants found in *Boswellia serrata* (B), whey protein powder (W), and their combination on pork patties during storage, exploring changes in textural properties and lipid oxidation susceptibility. The 2% whey-added group exhibited a higher crude protein content than the untreated control group. The highest water-holding capacity and lowest cooking losses were observed in mixed-additive groups (WB$_1$ (2% W/0.5% B) and WB$_2$ (2% W/1.0% B), and the highest sensory scores for overall acceptability were obtained for WB$_1$. Adding *B. serrata* can neutralize the hardness caused by whey powder, thereby improving palatability. From 7 d (days 7), the extents of lipid oxidation, determined using 2-thiobarbituric acid-reactive substances (TBARS) analysis, for the WB$_1$ and WB$_2$ groups were significantly lower than that of the control group. The WB$_1$ and WB$_2$ groups exhibited substantially suppressed total bacterial colony and *Escherichia coli* counts relative to the control group. Our findings suggest that the additive combination of *B. serrata* and whey protein powders can suppress lipid oxidation, improve storage stability, and enhance textural properties in the production of functional pork patties.

Keywords: *Boswellia serrata*; whey protein; sensory evaluation; texture; antioxidant; functional food

1. Introduction

Lipid oxidation is a principal factor affecting the quality and acceptability of meat products. Ground meat, in particular, is especially vulnerable to microbial contamination and lipid oxidation during processing and storage [1,2]. Such processes can produce off-flavors and toxic degradants, negatively affect sensory properties, lead to the loss of bioactive compounds and nutritional value, degrade color, and reduce shelf life and economic returns [3–5]. Uncontrolled oxidation and microbial contamination have now become one of the biggest economic problems in the processed meat industry [2].

Boswellia serrata and whey protein are natural substances that have antioxidant properties [1,6]. Consumers generally prefer natural antioxidants [7,8] over synthetic antioxidants such as butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), and tertiary-butylhydroquinone (TBHQ), considering the side effects of synthetic antioxidants. Furthermore, maintaining moisture, lipids, and food composition as well as improving flavor are necessary components in the applications of meat products. Non-meat protein additives such as whey protein are added to ground pork products

primarily to increase product yield, lower production cost, boost water-holding capacity (to enhance juiciness), and improve the textural and functional properties of the meat product [9–11].

Boswellia serrata is an important traditional medicinal plant grown in India, Northern Africa, and the Middle East [12,13]. Commonly known as Indian frankincense, the oleo-gum resin obtained from *B. serrata* possesses antioxidant, anti-inflammatory, antimicrobial, anticancer, and analgesic properties [14–16]. The main phytochemical components of the *B. serrata* resin are essential oils, gum, and resin, as well as various functional compounds such as terpenoids, β-boswellic acid, phenolic compounds, and polysaccharides [12,17,18]. Whey is a major by-product of cheese-making and has long been considered as waste [19]. It is composed of β-lactoglobulin, α-lactalbumin, immunoglobulin, bovine serum albumin, lactoferrin, lactoperoxidase, glycomacropeptide, and growth factors, as well as numerous biologically active factors and enzymes [20,21]. Because whey protein is a high-quality protein with an amino acid content and rapid digestibility that are biologically superior to most other proteins, much attention has been focused on it [22]. Recently, a number of studies have shown that whey protein and its related components have biological functions with specific health benefits, including antioxidant activity, immune regulation, and antibacterial and anticancer properties [23].

Although the effects of adding whey powder, tocopherol, spices, and phenolic compounds sourced from plant extracts on the functional properties of meat products have been studied [24–27], no researchers have considered the textural properties and antioxidation behaviors of pork patties supplemented with combinations of *B. serrata* and whey powders. This study aims to determine the effects of mixed *B. serrata* and whey powders on the quality characteristics of pork patties, including their impact on shelf life and oxidative stability.

2. Materials and Methods

2.1. Formulation and Preparation of Pork Patties

Pork patties containing *B. serrata* and whey powders were formulated as shown in Table 1. The *B. serrata* and whey powders were obtained from a local market and ESfood (Gyeonggi-do, Korea), respectively. A pork fillet and back fat were purchased from a local market (Seoul, Korea). All visible connective tissue and fat were removed from the pork fillet with a butcher's knife. The lean pork fillet and back fat were ground through a 5 mm plate, divided into five equal portions, and then mixed with the appropriate amounts of *B. serrata* and whey powders for 12 min using a stand mixer (SM 246, Poking Industrial Co., Ltd., Hong Kong, China). After thorough mixing, the pork mixtures were formed into identically sized patties (100 ± 1 g each) using a rectangular burger press (Spikomat Ltd., Nottingham, UK). The pork patties were vacuum packaged and stored at 4 °C for 0, 7, 14, or 21 d while the experiments were carried out in triplicate for each pork patty formulation.

Table 1. Formulation (wt%) of pork patties containing *Boswellia serrata* and whey powders.

Ingredients	Treatment [1]				
	CON	**W**	**B**	**WB$_1$**	**WB$_2$**
Pork fillet	75	75	75	75	75
Back fat	25	25	25	25	25
Salt	1	1	1	1	1
Whey	0	2	0	2	2
B. serrata	0	0	0.5	0.5	1
Total	101	103	101.5	103.5	104

[1] CON, control (no additive(s)); W, addition of 2% whey powder; B, addition of 0.5% *B. serrata* powder; WB$_1$, addition of 2% whey powder and 0.5% *B. serrata* powder; WB$_2$, addition of 2% whey powder and 1% *B. serrata* powder.

2.2. Proximate Composition

The moisture, ash, and crude protein and fat contents of the pork patties were analyzed using official methods (AOAC, 2012) [28]. The crude protein and crude fat contents were respectively determined by the Kjeldahl and Soxhlet methods. The moisture content was determined at 105 °C using a drying oven and the crude ash content was measured at 550 °C via the dry ashing method.

2.3. pH and Color

Following the homogenization of the pork patty samples (2 g) with distilled water (18 mL) for 60 s at $3220 \times g$ with a homogenizer (AM-1, Nihon Seiki Kaisha Co., Ltd., Nagoya, Japan), their pH values were measured with a pH meter (LAQUA F-71, Horiba Co., Kyoto, Japan).

The surface color of each uncooked pork patty was determined using a colorimeter (Minolta Chroma Meter CR-210, Japan) on day 0 and during specified storage intervals, measuring the CIE L^* (lightness), CIE a^* (redness), and CIE b^* (yellowness) values. The colorimeter was calibrated with a white standard plate (CIE L^* = +97.83, CIE a^* = −0.43, and CIE b^* = +1.98).

2.4. Water-Holding Capacity (WHC) and Cooking Loss

The water-holding capacity of each patty was determined according to the method of Akwetey and Yamoah [9]. A sample (5 g) was mixed thoroughly with distilled water (10 mL) in a tube and the mixture was centrifuged at 2000 rpm for 15 min at 15 °C. Then, the supernatant was carefully removed and the remaining sample was weighed. The WHC (%) was calculated as follows:

WHC (%) = (weight of sample after removing supernatant/weight of sample mixed with distilled water) × 100

The raw weight of each sample was obtained before and after cooking to an internal temperature of 75 °C; the patties were allowed cool at room temperature prior to weighing and texture profiling. The cooking loss from the patty samples was computed using the following equation by Murphy et al. [29]:

$$\text{Cooking loss (\%) = (raw patty weight−cooked patty weight)} \times 100 \tag{1}$$

2.5. Texture Profile Analysis (TPA)

The samples used in the cooking loss analysis were subjected to TPA (three replicates) using a texture analyzer (CT3-1000, Brookfield Engineering Laboratories, Inc., Middleboro, MA, USA). Samples were cut into $20 \times 20 \times 10$ mm^3 (length × width × height) portions. The operating conditions for the texture analyzer were set as follows: distance = 8.0 mm, pre-test speed = 5.0 mm/s, post-test speed = 2.0 mm/s, test speed = 2.0 mm/s, and force = 5.0 g. Textural attributes such as hardness (the peak force generated by the product on first compression (kg)), springiness (the ration of storage deformation to total deformation in the second loading cycle in texture profile analysis), cohesiveness (the ratio of active work to total work in the second loading cycle in the TPA), gumminess (hardness × cohesiveness (kg)), and chewiness (hardness × cohesiveness × springiness (kg)) were analyzed.

2.6. 2-Thiobarbituric Acid-Reactive Substances (TBARS)

Lipid oxidation was determined using the 2-thiobarbituric acid reactive substances (TBARS) method described by Buege and Aust [30]. Each sample (5 g) was homogenized with distilled water (15 mL) and BHT (100 μL, 6% in 100% ethanol) at 13,000 rpm for 1 min. After homogenization, a solution aliquot (2 mL) was transferred into a test tube, mixed with a trichloroacetic acid / 2-thiobarbituric acid reagent (4 mL), and heated in an 80 °C water bath for 15 min. After cooling in cold water, the mixture was centrifuged at $2000 \times g$ for 10 min at 25 °C, and then filtered through Whatman paper No. 4. Sample absorbance was measured at 531 nm using a spectrophotometer (UV/Vis Spectrophotometer, Mecasys, Daejeon, Korea). Results were expressed as 2-thiobarbituric acid-reactive substances in mg

malonaldehyde (MDA)/kg meat. A standard curve was prepared using 1,1,3,3-tetraethoxypropane (TEP) for calculations.

2.7. Volatile Basic Nitrogen (VBN)

The VBN contents of the samples were measured by Conway's microdiffusion method with a slight improvement [31]. A sample (5 g) was mixed with distilled water (15 mL) and homogenized at 10,000 rpm for 60 s. The homogenate was filtering and placed in the Conway's unit. The unit was then sealed and slowly agitated after added the Conway reagent. Titrated with 0.02 N sulfuric acid after incubation at 37 °C for 120 min.

2.8. Microbiological Analysis

Microbiological analysis of the pork patties was performed using 3M Petrifilm (3M, St. Paul, MN, USA). Patty samples (25 g) were homogenized with a 0.85% sterile saline solution (225 mL) in a side-filter bag for 90 s using a bag mixer (Bagmixer 400 W; Interscience, Woburn, MA, USA). The aerobic count plates were incubated at 37 ± 1 °C for 48 ± 2 h to determine the total bacterial counts. Coliform and *Escherichia coli*/coliform count plates were incubated at 37 ± 1 °C for 24–48 h to determine the *E. coli* and total coliforms. The colonies were counted, and results were expressed as log colony forming units per gram sample (CFU/g).

2.9. Sensory Evaluation

Sensory tests of the patties with or without the *B. serrata* and whey powders were performed at the Department of Food Science and Biotechnology of Animal Resources, Konkuk University. Panels for the sensory evaluation consisted of 20 randomly assigned trained researchers (8 men and 12 women with an average age of 27.8 years). Samples were cut into blocks ($1 \times 1 \times 1$ cm^3), placed on a white food plate at room temperature, and then served to the panelists at random. After eating one sample, panelists were asked to gargle with water and eat another sample one or two minutes later. The color (1 = undesirable, 9 = desirable), flavor (1 = undesirable, 9 = desirable), juiciness (1 = dry, 9 = juicy), tenderness (1 = tough, 9 = tender), and overall acceptability (1 = undesirable, 9 = desirable) of the cooked samples were evaluated using a nine-point hedonic scale.

2.10. Statistical Analysis

A total of 300 samples were analyzed ((20 patties $\times$ 5 batches $\times$ 3 (triplicate manufacture)) using SPSS 24.0 software (SPSS Inc., Chicago, IL, USA) using one-way analyses of variance and Tukey's test. p-values of less than 0.05 were considered significant.

3. Results and Discussion

3.1. Proximate Analysis, WHC, and Cooking Loss

The results of the proximate composition measurements, WHC, and cooking losses from the pork patties after adding the *B. serrata* and whey powders are shown in Table 2. The crude fat and ash contents of the pork patties do not change significantly with the addition of either powder or their combination. Adding *B. serrata* powder to the patties generates no difference from the control group in terms of moisture and crude protein contents, but the moisture content is significantly decreased with the addition of whey powder ($p < 0.05$). This may be because of the reduction in moisture content owing to the increase of dry matter in the formulation. A similar result was reported by Serdaroglu [32], in which the addition of whey powder reduced the moisture content in beef meatballs. Furthermore, the crude protein contents in all treatment groups containing whey powder exhibit significantly higher values than the control group ($p < 0.05$). This result is expected because whey powder is rich in protein, and its addition results in increased protein content, as was similarly described for chicken breast meat injected with whey protein [33].

Table 2. Proximate composition (%) analysis, WHC (%), cooking loss (%) of pork patties formulated with *Boswellia serrata* and whey powders.

Item [1]	CON	W	B	WB$_1$	WB$_2$	SEM [2]	*p*-Value
Ash	1.84 ± 0.01	1.90 ± 0.07	1.83 ± 0.02	1.90 ± 0.05	1.91 ± 0.02	0.03	0.14
Moisture	54.50 ± 0.37 [a]	53.09 ± 0.24 [b]	53.50 ± 0.37 [a,b]	53.02 ± 0.80 [b]	52.52 ± 0.20 [b]	0.37	*
Crude fat	26.02 ± 0.86	25.31 ± 0.44	25.63 ± 0.69	25.25 ± 0.35	25.13 ± 0.15	0.45	0.35
Crude protein	17.15 ± 0.85 [b]	18.89 ± 0.71 [a]	17.12 ± 0.36 [b]	18.79 ± 0.75 [a,b]	18.62 ± 0.36 [a,b]	0.52	*
WHC [3]	46.73 ± 0.27 [d]	48.35 ± 0.62 [b,c]	47.76 ± 0.07 [c]	48.82 ± 0.29 [a,b]	49.26 ± 0.10 [a]	0.27	***
Cooking loss	42.54 ± 0.31 [a]	39.80 ± 0.86 [b]	39.11 ± 0.69 [b]	38.53 ± 0.44 [b,c]	37.71 ± 0.71 [c]	0.45	***

[1] CON, control (no additive(s)); W, addition of 2% whey powder; B, addition of 0.5% *B. serrata* powder; WB$_1$, addition of 2% whey powder and 0.5% *B. serrata* powder; WB$_2$, addition of 2% whey powder and 1% *B. serrata* powder. [2] SEM: standard error of the mean. [a–d] Means within a row with different letters are significantly different ($p < 0.05$). * $p < 0.05$; *** $p < 0.001$. [3] WHC: water holding capacity

Cooking losses can influence both sensory properties and product quality. From Table 2, the highest WHC is observed for the WB$_1$ and WB$_2$ groups ($p < 0.05$) followed by the W and B samples, whereas the CON group has the lowest WHC. Furthermore, cooking losses are significantly lower for WB$_1$ and WB$_2$ than for the control group ($p < 0.05$), which reaffirms the water retention capabilities of the *B. serrata* and whey powders. Taylor and Walsh [34] reported that textured whey protein significantly increased water retention, and El-Magoli et al. [35] showed that the WHC depends on water-protein interactions. In agreement with those results, we can conclude that powders of *B. serrata* and whey have the ability to retain moisture and prevent patties from drying out, thereby improving palatability.

3.2. pH Analysis

The pH values of the uncooked pork patties during storage are presented in Figure 1. pH is a major factor affecting the quality of processed meat products. Changes in pH can affect properties such as freshness, texture, and color [36]. The pH values of all samples exhibit a slight upward trend with time until 7 days, which may be due to the growth of microorganisms and the enzymatic breakdown of proteins to produce alkaline substances [37]. During storage, the pH values of the W and CON groups are nearly identical, and no significant differences between groups B, WB$_1$, and WB$_2$ are observed until 14 days. However, after 7 days, the pH values in the *B. serrata* powder treatment groups (B, WB$_1$, and WB$_2$) are significantly lower compared to those in the control group ($p < 0.05$). Clearly, this change is mainly caused by the added *B. serrata* powder; specifically, it most likely results from the introduction of acidic components such as 11-keto-β-acetyl-β-boswellic acid (KBA), acetyl-11-keto-β-boswellic acid (AKBA), and acetyl-α-boswellic acid (AαBA) into the pork patties [17]. Similar decreasing pH trends have been observed upon the addition of edible seaweed that contains alginic acid to meat products [38]. As acids can effectively inhibit the growth of microorganisms, these findings could improve the storability of pork patties through the addition of *B. serrata*.
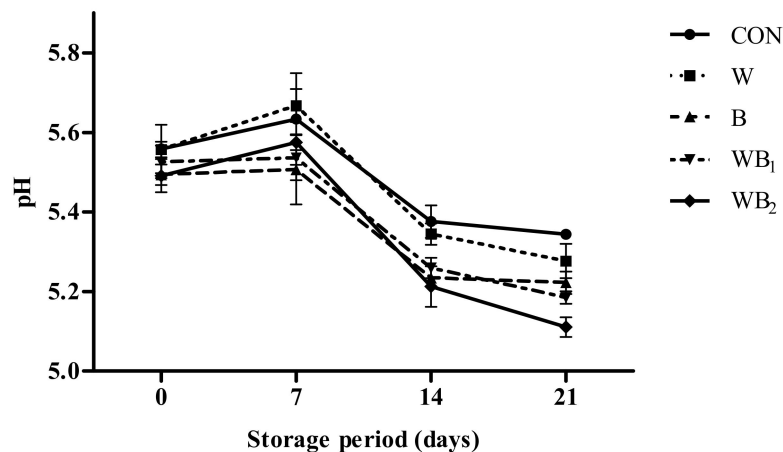
Figure 1. Changes in the pH values of pork patties with different treatments during refrigerated storage at 4 °C for 21 d. Error bars indicate standard deviations. CON (●), control (no additive(s)); W (■), addition of 2% whey powder; B (▲), addition of 0.5% *B. serrata* powder; WB$_1$ (▼), addition of 2% whey powder and 0.5% *B. serrata* powder; WB$_2$ (♦), addition of 2% whey powder and 1% *B. serrata* powder.

3.3. Color Measurements

Results obtained from the color measurements of the pork patties are presented in Table 3. The color of meat, which is associated with freshness and pH, will change over time depending on the concentrations of deoxymyoglobin, metmyoglobin, and oxymyoglobin [39]. Compared with the fresh patties (0 day), the L^* values of the 21 days samples decrease significantly for all treatment groups, and the a^* readings exhibit significant reductions in all groups from 7 to 14 days ($p < 0.05$). These changes originate from the metmyoglobin that is generated by pigment oxidation and reduces the lightness and redness values. Moreover, when the pH is low, the meat also becomes pale. The L^* values are higher for the B group than for CON at 0 and 14 days; furthermore, the L^* values are higher in WB$_2$ than in CON at 14 days ($p < 0.05$). For other storage periods, all treatment groups are lighter ($p < 0.05$) than the control group, which could be due to the prevention of pigment oxidation by the added *B. serrata* and whey powders. Similarly, previous reports have indicated that myoglobin oxidation and lipid oxidation are interrelated and that they affect the color of meat [40]. Our results agree with those obtained by Atughonu et al. [41], who found that non-meat proteins led to a higher value of L^* owing to their diluent effects on myoglobin pigments. The a^* values of CON significantly decrease ($p < 0.05$) from 14 to 21 days, but all treated groups' values remain stable. Similar results were observed by Zhang et al. and Ozer et al. [42,43]. Lipids change color from white to yellow as a result of oxidation. In most samples, the b^* values are similar at the beginning and end of storage. However, the three treatment groups, namely, WB$_1$, WB$_2$, and W display lower b^* values ($p < 0.05$) than the control group at 7 and 21 days, respectively. These results indicate that the additives employed in this study exhibit a protective effect with respect to discoloration of the meat product.

Table 3. Effects of *Boswellia serrata* and whey powders on the color of pork patties during storage.

L* (Lightness)	Treatment [1]					SEM [2]	p-Value	
Storage (Days)	CON	W	B	WB$_1$	WB$_2$		Treatment	Storage
0	65.32 ± 0.30 ^{bA}	65.55 ± 0.30 ^{bA}	66.85 ± 0.25 ^{aA}	65.11 ± 0.38 ^{bAB}	65.34 ± 0.43 ^{bA}	0.24	***	***
7	64.29 ± 0.87 ^{bAB}	65.38 ± 0.22 ^{aA}	65.76 ± 0.17 ^{aB}	65.34 ± 0.30 ^{aA}	65.16 ± 0.33 $_{abAB}$	0.32	**	**
14	63.45 ± 0.17 ^{cBC}	64.11 ± 0.58 ^{bcB}	66.38 ± 0.56 ^{aAB}	64.23 ± 0.51 ^{bcBC}	64.81 ± 0.38 $_{abAB}$	0.33	**	***
21	62.98 ± 0.44 ^{cC}	64.25 ± 0.56 ^{bB}	65.63 ± 0.49 ^{aB}	64.10 ± 0.63 ^{bC}	64.50 ± 0.12 ^{bB}	0.34	** *	***
A * (redness) Storage (days)	CON	W	B	WB$_1$	WB$_2$			
0	12.86 ± 0.18 ^{aA}	11.97 ± 0.52 ^{abA}	11.44 ± 0.47 ^{bA}	11.89 ± 0.32 ^{bA}	11.94 ± 0.49 ^{bA}	0.29		**
7	12.56 ± 0.13 ^{aA}	11.79 ± 0.26 ^{bA}	11.09 ± 0.15 ^{cA}	11.83 ± 0.03 ^{bA}	11.91 ± 0.24 ^{bA}	0.13		***
14	10.45 ± 0.39 ^{aB}	10.40 ± 0.15 ^{aB}	8.75 ± 0.11 ^{bB}	10.10 ± 0.38 ^{aB}	10.29 ± 0.25 ^{aB}	0.20	***	***
21	9.57 ± 0.11 ^{aC}	9.80 ± 0.36 ^{aB}	8.74 ± 0.13 ^{bB}	9.90 ± 0.18 ^{aB}	9.99 ± 0.36 ^{aB}	0.18		***
B * (yellowness) Storage (days)	CON	W	B	WB$_1$	WB$_2$			
0	7.75 ± 0.24	7.61 ± 0.13	7.52 ± 0.17	7.46 ± 0.21	7.43 ± 0.06 ^{A,B}	0.12	***	0.112
7	7.74 ± 0.05 ^{a}	7.54 ± 0.40 ^{ab}	7.75 ± 0.24 ^{a}	7.45 ± 0.15 ^{a,b}	7.20 ± 0.10 ^{b,B}	0.16	0.440	*
14	7.60 ± 0.07	7.48 ± 0.05	7.56 ± 0.22	7.42 ± 0.09	7.64 ± 0.07 ^{A}	0.08	0.395	0.098
21	7.79 ± 0.08 ^{a}	7.36 ± 0.13 ^{b}	7.59 ± 0.12 ^{ab}	7.41 ± 0.16 b	7.30 ± 0.24 ^{bB}	0.11	0.960 **	**

[1] CON, control (no additive(s)); W, addition of 2% whey powder; B, addition of 0.5% *B. serrata* powder; WB$_1$, addition of 2% whey powder and 0.5% *B. serrata* powder; WB$_2$, addition of 2% whey powder and 1% *B. serrata* powder. [2] SEM: standard error of the mean. $^{a–c}$ Means within a row with different letters are significantly different ($p < 0.05$). $^{A–C}$ Means within a column with different letters are significantly different ($p < 0.05$). * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$.

3.4. Texture Profile Analysis (TPA)

Table 4 shows the effects of the *B. serrata* and whey powders on the textural properties of cooked pork patties, in terms of attributes such as springiness, cohesiveness, chewiness, gumminess, and hardness. Pork patties with added whey powder exhibit higher ($p < 0.05$) values with regard to chewiness, gumminess, and hardness than the pork patties in the control group. No variations in springiness and cohesiveness are observed among the test and control groups. Lower values of hardness are observed in the B, WB_1, and WB_2 groups as compared to the CON and W groups ($p < 0.05$). Furthermore, owing to the reduced hardness, *B. serrata* treatment alone (group B) results in slightly lower chewiness in comparison to the WB_1, CON, and W groups ($p < 0.05$). Adding *B. serrata* can neutralize the hardness caused by whey powder, thereby improving palatability. Similar changes in texture profiles that increased the hardness and chewiness but did not affect the springiness and cohesiveness with added whey have been reported by Andic et al. [11]; furthermore, such changes were attributed to the β-lactoglobulin contained in whey powder. This substance has good thermogelation characteristics and can be denatured when heated in a usually thermally irreversible manner. The reduction in the hardness value can probably be attributed to the moisture-retention properties of the *B. serrata* powder. This is consistent with the results obtained by Wan Rosli et al. [44], who reported that addition of oyster mushroom results in higher water retention that contribute to hardness reduction in chicken patties. In addition, Verma et al. [45] found that, owing to its moisture-retention properties, sweet potato powder also reduced hardness values in low-fat formulated pork patties.

Table 4. Texture profile analysis of pork patties formulated with *Boswellia serrata* and whey powders [1].

Property	CON	W	B	WB_1	WB_2	SEM [2]	*p*-Value
Springiness (mm)	0.75 ± 0.02	0.75 ± 0.02	0.74 ± 0.02	0.75 ± 0.01	0.74 ± 0.01	0.01	0.197
Cohesiveness	0.21 ± 0.02	0.21 ± 0.02	0.23 ± 0.03	0.24 ± 0.04	0.24 ± 0.02	0.01	0.058
Chewiness (kg)	3.29 ± 0.08 [b]	3.49 ± 0.08 [a]	3.11 ± 0.09 [c]	3.26 ± 0.06 [b]	3.21 ± 0.13 [b,c]	0.04	***
Gumminess (kg)	4.39 ± 0.06 [b]	4.68 ± 0.09 [a]	4.21 ± 0.30 [b]	4.36 ± 0.04 [b]	4.36 ± 0.13 [b]	0.07	***
Hardness (kg)	20.53 ± 0.79 [b]	21.87 ± 0.54 [a]	18.07 ± 0.60 [c]	18.49 ± 0.55 [c]	18.19 ± 0.50 [c]	0.27	***

[1] CON, control (no addition); W, addition of 2% whey powder; B, addition of 0.5% *B. serrata* powder; WB_1, addition of 2% whey powder and 0.5% *B. serrata* powder; WB_2, addition of 2% whey powder and 1% *B. serrata* powder. [2] SEM: standard error of the mean. [a–d] Means within a row with different letters are significantly different ($p < 0.05$). *** $p < 0.001$

3.5. Storage Stability

The changes in the TBARS values during the storage of pork patties are shown in Figure 2. Although the values for the control group increase continuously over 21 days ($p < 0.05$), the other treatment groups exhibit no significant differences between 0 and 7 days, after which their TBARS values being to rise. From 7 days, the TBARS values of the WB_1 and WB_2 groups are significantly lower than that of the control, and the TBARS values of the WB_2 sample, in particular, are the lowest ($p < 0.05$). The detectable threshold of oxidized flavor in meat is observed when TBRAS values are 0.5–2.0 mg MDA/kg [46]. At day 14, the TBARS value of the CON group is 0.47, however the treatment groups are still lower than 0.5 until 21 days. According to other reports [17,23,47], this could be due to the addition of *B. serrata*, which contains terpenoids (KBA, AKBA, and AαBA), phenolic compounds, diterpene alcohols as well as the addition of whey, which can prevent lipid oxidation. Although there are no reports on meat products showing that *Boswellia* species display antioxidation effects, similar research has stated that rosemary contains phenol diterpenes (carnosic acid and rosmarinic acid) with antioxidant properties [48].

The VBN content of the pork patties tends to increase with storage time, with the B group showing the lowest ($p < 0.05$) value during days 7–21 (Figure 3). The VBN contents of the B and CON samples are significantly lower than those of the other groups that contain whey powder after 14 days. According to Korea Food Law (2002), the limit of allowable VBN content is 20 mg/100 g or less in meat. The value of CON group is close to 20 mg% after about 14 days of storage, however the B group take

approximately 21 days to reach the same value. This suggests that the addition of *B. serrata* may delay protein degradation, possibly owing to the antibacterial effects of the additive [47]. From our results, it is observed that samples treated with whey powder exhibit a higher VBN value compared to those in CON. During storage, meat products decompose, with proteins being degraded into amino acids and producing low molecular weight inorganic nitrogen. Therefore, it is speculated that an increase in the nitrogen content with the addition of whey powder results in the higher VBN values. These results agree with those obtained by Ha et al. [33], who found that the injection of whey protein in chicken breast meat produced higher VBN values compared to those of the control group.
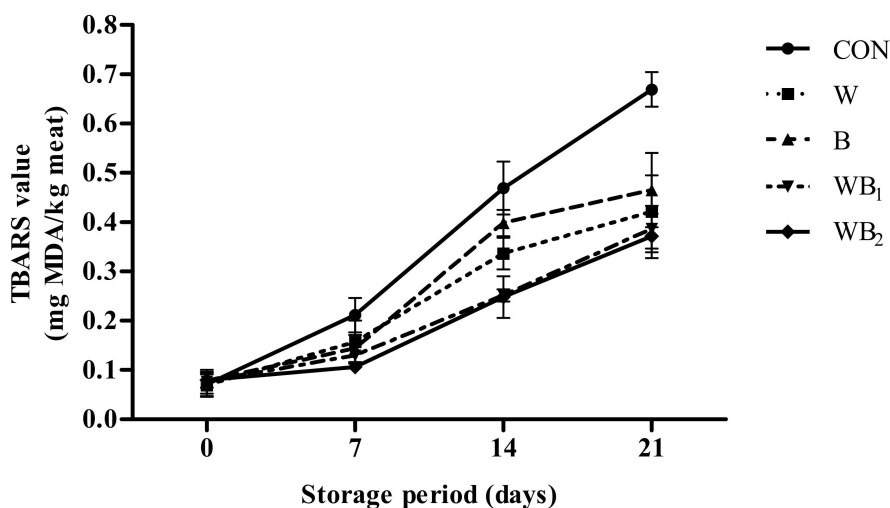
Figure 2. Changes in the 2-thiobarbituric acid reactive substances (TBARS) values of pork patties with different treatments during refrigerated storage at 4 °C for 21 days. Error bars indicate standard deviations. CON (•), control (no additive(s)); W (■), addition of 2% whey powder; B (▲), addition of 0.5% *B. serrata* powder; WB_1 (▼), addition of 2% whey powder and 0.5% *B. serrata* powder; WB_2 (♦), addition of 2% whey powder and 1% *B. serrata* powder.
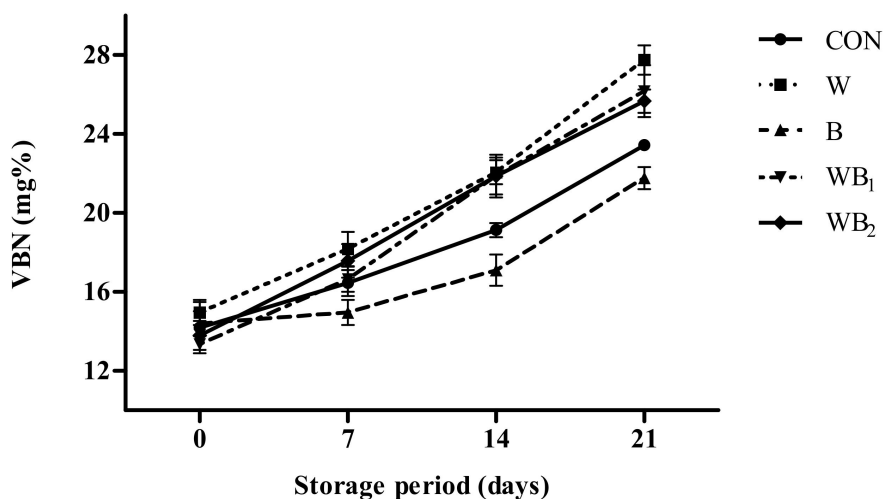
Figure 3. Changes in the volatile basic nitrogen (VBN) values of pork patties with different treatments during storage. Error bars indicate standard deviations. CON (•), control (no additive(s)); W (■), addition of 2% whey powder; B (▲), addition of 0.5% *B. serrata* powder; WB_1 (▼), addition of 2% whey powder and 0.5% *B. serrata* powder; WB_2 (♦), addition of 2% whey powder and 1% *B. serrata* powder.

The results of microbiological analyses of the pork patties during storage are shown in Figure 4. The total aerobic bacterial counts in all groups increase with storage time ($p < 0.05$). Initially, no significant differences are observed among the treatments containing *B. serrata*, although their microbe counts are slightly lower than those in W and CON. The rich terpenoid content in *B. serrata* may be an important factor with regard to its antibacterial activity [49]. The control group exhibits the highest bacterial counts from 7 to 21 days. The number of aerobic counts is significantly lower in WB_2 compared to the other treatments during 0–7 days; however, the B group shows the slowest growth rate and the lowest total bacterial count values from 14 days ($p < 0.05$). It may be that the AKBA in *B. serrata* can inhibit the formation of biofilms and lactoferrin in whey powder, thereby slowing bacterial growth, owing to their antibacterial capabilities [12,50]. Therefore, *B. serrata* and whey protein are capable of maintaining stability during storage, and *Boswellia* exhibits better antibacterial activity. Finally, neither *E. coli* nor other coliform bacteria were detected in any sample during storage in this study (data not shown).
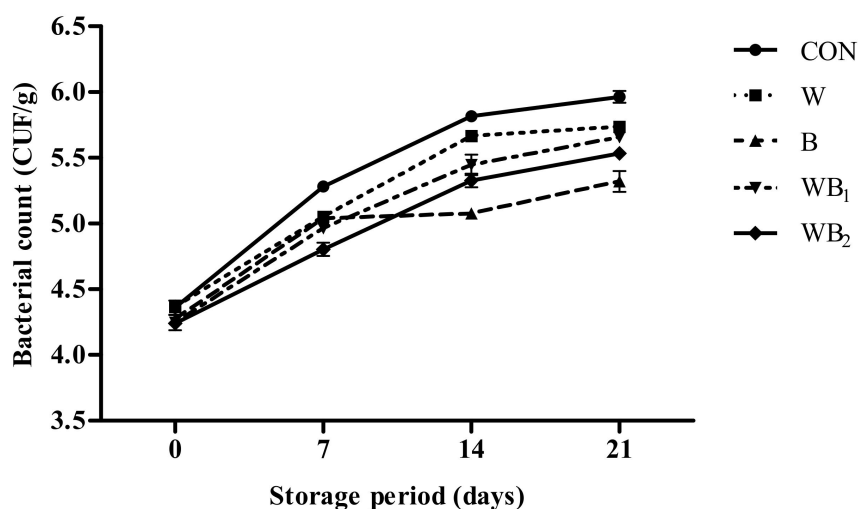

Figure 4. Effect of *B. serrata* and whey powders on the number of aerobic bacteria in pork patties during storage. Error bars indicate standard deviations. CON (●), control (no additive(s)); W (■), addition of 2% whey powder; B (▲), addition of 0.5% *B. serrata* powder; WB_1 (▼), addition of 2% whey powder and 0.5% *B. serrata* powder; WB_2 (♦), addition of 2% whey powder and 1% *B. serrata* powder.

3.6. Sensory Evaluation

Figure 5 shows the sensory evaluation of the differently treated pork patties. All products achieve similar color scores irrespective of the formulation. Compared to other groups, the WB_1 sample results in the highest sensory scores with regard to flavor, juiciness, and overall pork patty acceptability. This may be due to the aromatic components in *Boswellia*, which mainly comprise α-pinene, β-myrcene, linalool, and sesquiterpenoids that can convey a pleasant odor [47]. However, when added at 1%, the aroma, similar to spices, is too strong, such that WB_2 has the lowest score for flavor (although without a significant difference compared to the control and whey groups). The scores for juiciness and tenderness are not significantly different among the WB_1, WB_2, and B samples; however, they are significantly higher than those of the W group ($p < 0.05$). The lower cooking losses are a result of the greater water-holding capacity arising from the *Boswellia* in WB_1, WB_2, and B; therefore, higher juiciness and tenderness values are observed, consistent with the TPA results.
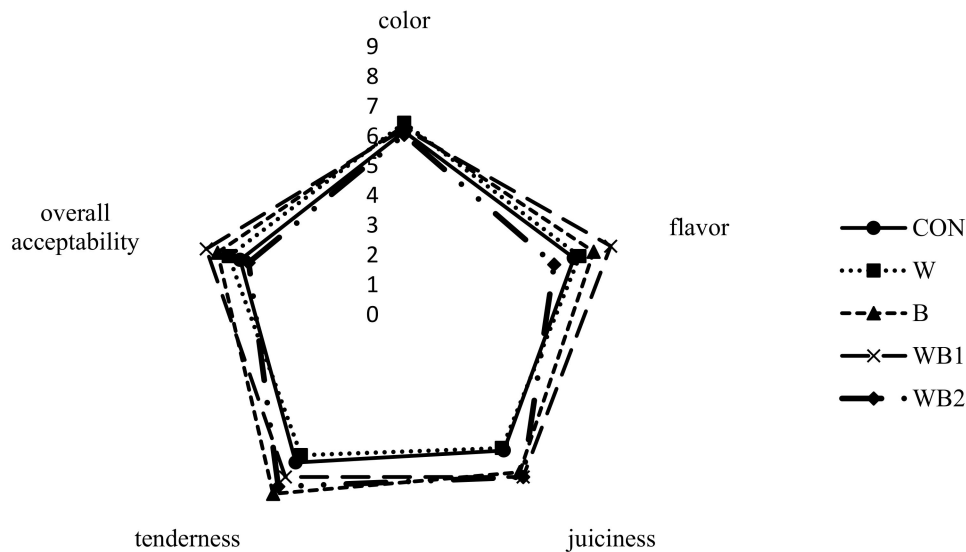
Figure 5. Effect of *Boswellia serrata* and whey powders on the sensory evaluation of pork patties. CON (●), control (no additive(s)); W (■), addition of 2% whey powder; B (▲), addition of 0.5% *B. serrata* powder; WB$_1$ (✕), addition of 2% whey powder and 0.5% *B. serrata* powder; WB$_2$ (♦), addition of 2% whey powder and 1% *B. serrata* powder.

4. Conclusions

This study shows that combined whey protein and *B. serrata* powders can extend the shelf life of pork patties by inhibiting lipid oxidation, reducing pH and bacterial growth, and effectively improving quality characteristics. Introducing additives into pork patties reduces the meat content and cost compared to ordinary patties. Furthermore, *B. serrata* and whey protein are natural antioxidants and can protect the product from discoloration and improve taste, which can increase consumer satisfaction. Further studies can apply these benefits to fermented meat products.

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