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# Effect of deep-sea mineral water on growth performance, water intake, blood characteristics and serum immunoglobulins in the growing-finishing pigs

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**Abstract**

Brine mineral water (BMW) is groundwater near the deep sea, and the mineral component of the BMW is different from the water of the deep sea because the components of BMW are derived from the unique geographical features surrounding it. Recently, BMW has attracted attention due to the unique health-related minerals it possesses; however, the influence of BMW on physiological function has not yet been determined in domestic animals. Therefore, this experiment investigated the influence of BMW on the growth performance, water intake, blood properties, and immunoglobulin (Ig) levels of serum in growing-finishing pigs. A total of 64 pig barrows (Landrace × Yorkshire × Duroc) with an average initial weight of 40.56 ± 0.17 kg were used in the experiment, and 0%, 2%, 3%, and 5% samples of BMW diluted with freshwater were provided to experimental animals during the 56 days. We found that the gain/feed ratio in the 3% BMW group was significantly higher than that in the 5% BMW group of growing-finishing pigs ($p < 0.05$). The water intake was significantly increased in the 5% BMW group compared with the other groups ($p < 0.05$) of growing-finishing pigs. Additionally, the concentrations of red blood cells (RBCs), hemoglobin (HGB), and hematocrit (HCT) were significantly higher in the 3% BMW group than in the control group. The level of high-density lipoprotein cholesterol was higher in the 3% BMW group than in the 5% BMW group ($p < 0.05$). Furthermore, IgG and IgM levels in the serum were significantly higher in the 2% and 3% BMW groups than in the control group ($p < 0.05$). These results suggest that a dilution of 3% BMW in the intake water could improve the levels of RBCs and serum Igs in growing-finishing pigs.

**Keywords:** Brine mineral water, Growth performance, Red blood cells, Immunoglobulin, Growing-finishing pigs

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**Availability of data and material**
Upon reasonable request, the datasets of this study can be available from the corresponding author.

**Authors' contributions**
Conceptualization: Park CK.
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Formal analysis: Lee SH.
Methodology: Lee SH, Park CK.
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Investigation: Lee SH, Park CK.
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**Ethics approval and consent to participate**
All procedures that involved the use of animals were approved by the Kangwon National University Institutional Animal Care and Use Committee (KW-210112-1).

# INTRODUCTION

Deep ocean water below 200 meters has characteristics such as lower temperature, higher mineral nutrient content, and more microelements compared to other seawater [1]. Brine mineral water (BMW), which is derived from deep ocean water and has recently attracted attention due to ionized unique minerals, differs from general deep seawater because its contents depend on the surrounding geographical features [1,2]. For this reason, BMW contains several unique minerals, such as natural selenium (Se), germanium (Ge), molybdenum (Mo), and vanadium (V); furthermore, it has a beneficial influence in terms of animal physiology because its ratio of calcium (Ca) to magnesium (Mg) ions is optimal for absorption in mammals [3,4].

Pigs have a dietary requirement for certain minerals, including macro- and microminerals [5]. The functions of these minerals range from structural components in some tissues and enzymes to the regulation of metabolism in growing pigs [5]. In particular, BMW contains Ca, chlorine (Cl), Mg, and potassium (K), which are classified as macrominerals in terms of dietetics for growing pigs [3,4]. These macrominerals play a role in the development and maintenance of the skeletal system, components of extracellular anions, regulation of enzymatic metabolism, and cofactors of many enzyme systems [5]. Furthermore, microminerals, which include cobalt (Co), copper (Cu), and Se of the BMW, are components of vitamin $B_{12}$ and the enzyme glutathione peroxidase and are involved in the synthesis of HGB [3–5]. Therefore, from the swine dietetics perspective, BMW has the potential to regulate enzymatic metabolism and provide components for the development of tissues, skeletal systems, and enzymes. In a previous study, we determined that the growth performances and factors regarding RBCs of weaning pigs were improved by intake of diluted BMW, which contains macro- and microminerals [3]. However, the influence of BMW on growth performance and blood characteristics has not yet been studied in growing pigs.

Because the usage of antibiotics causes bacterial antibiotic resistance in domestic animals, the addition of antibiotics to feed has become illegal; consequently, disease has become more prevalent in domestic animals. Therefore, more studies on antibiotic substitution [6], enzyme supplements [7], minerals [8], and probiotics [9] have been conducted to enhance the immunity and growth of domestic animals. Furthermore, interest in environmentally friendly farming has been rapidly increasing, with a focus on the effect of natural substances on the growth and immunity of livestock [10,11]. In this context, BMW can be a valuable source of additive feeding for livestock in terms of environmental friendliness and the intake of natural minerals. In practice, some studies have reported that BMW suppresses cancer cells [4], may be useful for recovering from physical fatigue [2], and may increase the viability of reproductive cells [12].

BMW is groundwater that is geographical near the sea, and the concentration of sodium chloride (NaCl) is higher than that of freshwater since BMW is generated from the ocean [3]. From the animal nutrition perspective, salty water and growth performances are negatively correlated for domestic animals, so scientific experiments on diluted BMW intake should be conducted regarding the usage of the feeding system in domestic animals. To solve this task, we investigated the osmotic pressure of diluted BMW with freshwater, and the optimal dilution concentration of BMW for the animal experiment was determined using porcine reproductive cells in a previous study [12]. However, the physiological functions of BMW have not yet been investigated in growing-finishing pigs; therefore, we investigated the effect of BMW on the growth performance, water intake, blood cell composition, serum cholesterol, and Ig of growing-finishing pigs.

# MATERIALS AND METHODS

## Preparation of the brine mineral water

The BMW was collected from approximately 1,100 meters underground around a coastal peninsula at 500 meters near the coast (37.65541534867342, 129.0508611658692; Geumjin-ri, Okgye-myun, Gangneung-si, Gangwon-do, Korea) using a water pump system (HA-1688, Han-il, Seoul, Korea). The collected BMW was laid to sink colloid particles for 48 h at room temperature, the supernatant was filtered through a 1.0 μm pore size membrane filter (Advantec, Tokyo, Japan). The mineral composition analysis and information were referred to in the previous study [13] and the mineral composition of the BMW is shown in Table 1.

## Animals and experimental design

All procedures that involved the use of animals were followed by the Kangwon National University Institutional Animal Care and Use Committee (KW-210112-1). The BMW was diluted with freshwater until 0%, 2%, 3%, and 5% in 300 liters water tanks (Nwel, Seoul, Korea) before intake by experimental animals through a water supply pipe [3]. A total of 64 castrated pigs (Landrace × Yorkshire × Duroc) with initial body weight (BW) of 40.56 ± 0.17 kg were randomly divided into four groups (n = 4 per group, 4 replicates per group) based on their BW. The treatment groups were divided into four as 0% BMW (freshwater), 2% BMW, 3% BMW, and 5% BMW which were utilized in the experiment. The experiment was conducted during the 56 days and was divided into the growing phase (0 to 27 days) and the finishing phase (28 to 56 days). Animals were provided free access to BMW through nipple waterers from water tanks throughout the experiment. All diets were provided in meal form during the experiment. Table 2 lists the ingredient composition of the basal diets. The diets were formulated to meet or exceed the requirements for all nutrients for the growing-finishing pigs [5]. At the beginning and the end of the experiment, BW and feed intake were measured to determine the average daily gain (ADG), average daily feed intake (ADFI), and

**Table 1. Components of the brine mineral water**

| Components | Concentration |
|---|---|
| Ca (mg/L) | 1,657 |
| Mg (mg/L) | 995 |
| K (mg/L) | 205 |
| Cl (mg/L) | 19,393 |
| Na (mg/L) | 9,092 |
| $SO_4$ (mg/L) | 4,402 |
| Sr (mg/L) | 30.40 |
| SiO2 (mg/L) | 13.10 |
| Zn (mg/L) | 8.46 |
| F (mg/L) | 2.60 |
| I (mg/L) | 0.11 |
| Ti (mg/L) | 0.62 |
| Se (μg/L) | 480.00 |
| V (μg/L) | 305.00 |
| Mn (μg/L) | 16.00 |
| Cu (μg/L) | 12.00 |
| Co (μg/L) | 2.80 |
| Ge (μg/L) | 1.40 |

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Table 2. Formulae and chemical composition of experimental diets for the growing-finishing pigs

| Ingredients (%) | Growing phase (0 to 27 days) | Finishing phase (28 to 56 days) |
|---|---|---|
| Total | 100.00 | 100.00 |
| Corn | 46.26 | 67.86 |
| Soybean meal | 34.00 | 18.76 |
| Animal fat | 4.50 | 3.50 |
| Wheat | 11.00 | 3.69 |
| DCP | 0.80 | 0.80 |
| Limestone | 0.60 | 0.85 |
| Salt | 0.30 | 0.30 |
| DL-Methionine | 0.32 | 0.21 |
| L-Lysine | 0.90 | 1.71 |
| Mineral premix[1)] | 0.15 | 0.15 |
| Vitamin premix[2)] | 0.15 | 0.15 |
| Choline chloride | 0.02 | 0.02 |
| Calculated values | | |
| ME (kcal/kg) | 3,300 | 3,300 |
| Crude protein (%) | 20.00 | 18.00 |
| Calcium (%) | 0.30 | 0.25 |
| Lysine (%) | 1.00 | 1.00 |
| Methionine and cysteine (%) | 0.59 | 0.48 |

[1)]Mineral premix, supplied per kilogram of diet: 45 mg Fe, 0.25 mg Co, 50 mg Cu, 15 mg Mn, 25 mg Zn, 0.35 mg I, 0.13 mg Se;
[2)]Vitamin premix, supplied per kilogram of diet: 16,000 IU vitamin A, 3,000 IU vitamin $D_3$, 40 IU vitamin E, 5.0mg vitamin $K_3$, 5.0 mg vitamin $B_1$, 20 mg vitamin $B_2$, 4 mg vitamin $B_6$, 0.08 mg vitamin $B_{12}$, 40 mg pantothenic acid, 45 mg niacin, 0.15 mg biotin, 0.65 mg folic acid, 12 mg antioxidant.
DCP, digestible crude protein.

gain/feed ratio (G/F). The average daily water intake (L/days) of the pigs is strongly related to ADFI, and it was calculated as (3.053 × ADFI, kg) + 0.149 according to the previous study [14]. The average daily intake of the minerals from BMW was calculated based on Table 1.

## Complete blood corpuscle count

At the end of the experiment, blood from two pigs in each pen was collected from the cervical vein using 18-gauge needle syringes (Hwagin, Seoul, Korea). The blood samples were stored in $K_2$EDTA micro trainer tubes (Becton–Dickson, Franklin Lakes, NJ, USA) for complete blood corpuscle count (CBC) analysis. Anti-coagulated blood samples in $K_2$EDTA vacuum tubes were used to measure total RBC, HGB, HCT, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin (MCHC), white blood cell (WBC) count, and platelet (PLT) levels; all blood cell parameters were assessed using an automatic CBC analyzer (ForCyte, Oxford Science, Oxford, CT, USA).

## Analysis of biochemical factors and immunoglobulins in the serum

The blood was also collected from two pigs in each group and collected blood samples for analysis of biochemical factors, IgG, and IgM were stored in vacutainer serum tubes (Becton–Dickinson). Then, the samples were centrifuged at 4°C and 1,000×g for 20 min, the serum was derived from the supernatant of centrifuged blood. The glucose (GLU), total cholesterol (CHOL), high-density lipoprotein (HDL), low-density lipoprotein (LDL), and triglyceride (TG) were analyzed using each parameter strips (Arkray, Kyoto, Japan) and the level of the parameter strip were measured by an automated chemistry analyzer (Spotchem EZ SP-4430, Arkray). Also, the level of IgG and IgM were detected using the IgG (CSB-E06804p, CUSABIO, Wuhan, China) and IgM (CSB-E06805p, CUSABIO) ELISA kit, and all process were followed the manufacturer's instructions. The IgG and IgM density values were read at 450 nm using spectrophotometry.

### Statistical analysis

The average of each group (cage) was used for each experiment unit when the ADG, ADFI, G/F, and BW, and intake of minerals from the BMW during water intake were analyzed. Also, the average of the two pigs in each group was used for each experiment unit when the CBC analysis, biochemical analysis, and Ig levels were analyzed. The BMW level was considered as fixed effect to used to do statistical analysis in various parameters such as growth performance, mineral intake, and blood characteristics. Then, the collected data were analyzed through general linear mean (GLM) by statistical analysis system software (SAS version 9.2, SAS Institute, Cary, NC, USA). Differences were considered to be significant when their probability of occurring by chance was less than 5% ($p < 0.05$).

## RESULTS

### Growth performance

The influence of BMW on pig growth during the 56-day experiment is shown in Table 3. The ADG, ADFI, and G/F levels did not significantly differ among the treatment groups, whereas the final BW was lower in the 5% BMW group than in the other treatment groups at the growing phase ($p < 0.05$). Of these results, the G/F was higher in the 3% BMW group ($p = 0.391$) than in the other treatment groups at the growing phase. The G/F was also higher in the 3% BMW group than in the other BMW groups ($p = 0.067$) at the finishing phase. The final BW was significantly

**Table 3. Effects of brine mineral water (BMW) on the growth performance in the growing-finishing pigs**

| Items | Diluted BMW in intake water (%) | | | | SEM | *p*-value |
|---|---|---|---|---|---|---|
| | 0 | 2 | 3 | 5 | | |
| Growing phase | | | | | | |
| Initial BW (kg) | 40.55 | 40.98 | 40.59 | 40.12 | 1.499 | 0.989 |
| ADG (g) | 827 | 817 | 862 | 815 | 12.964 | 0.084 |
| ADFI (g) | 2,123 | 2,074 | 2,044 | 2,047 | 46.936 | 0.768 |
| G/F | 0.394 | 0.398 | 0.424 | 0.406 | 0.012 | 0.391 |
| Final BW (kg) | 70.50[a] | 70.83[a] | 69.67[a] | 65.86[b] | 0.996 | 0.026 |
| Finishing phase | | | | | | |
| ADG (g) | 1,034 | 1,050 | 1,084 | 945 | 36.728 | 0.132 |
| ADFI (g) | 3,304[b] | 3,418[b] | 3,391[b] | 3,539[a] | 48.966 | 0.042 |
| G/F | 0.314 | 0.307 | 0.320 | 0.267 | 0.013 | 0.067 |
| Final BW (kg) | 99.39[a] | 99.61[a] | 99.78[a] | 92.77[b] | 1.092 | 0.002 |
| Total phase | | | | | | |
| ADG (g) | 930[ab] | 934[ab] | 973[a] | 880[b] | 18.153 | 0.037 |
| ADFI (g) | 2,713 | 2,746 | 2,717 | 2,793 | 41.482 | 0.625 |
| G/F | 0.354 | 0.353 | 0.372 | 0.336 | 0.011 | 0.205 |

[a,b]Means in the same row with different superscripts differ ($p < 0.05$).

Growing phase, 0 to 27 days; Finishing phase, 28 to 56 days; Total phase, 0 to 56 days.

BW, body weight; ADG, average daily gain; ADFI, average daily feed intake; G/F, gain/feed ratio.

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**Table 4.** Effect of brine mineral water (BMW) on the daily water intake and intake of minerals of the BMW in the growing-finishing pigs

| Items | Diluted BMW in intake water (%) | | | | SEM | *p*-value |
|---|---|---|---|---|---|---|
| | 0 | 2 | 3 | 5 | | |
| Growing phase | | | | | | |
| Water (L/day) | 6.631 | 6.482 | 6.389 | 6.399 | 0.143 | 0.767 |
| Na (g/day) | 0.000$^{d}$ | 1.179$^{c}$ | 1.743$^{b}$ | 2.909$^{a}$ | 0.050 | 0.001 |
| Cl (g/day) | 0.000$^{d}$ | 2.514$^{c}$ | 3.717$^{b}$ | 6.205$^{a}$ | 0.107 | 0.001 |
| Ca (g/day) | 0.000$^{d}$ | 0.215$^{c}$ | 0.318$^{b}$ | 0.530$^{a}$ | 0.009 | 0.001 |
| Mg (g/day) | 0.000$^{d}$ | 0.129$^{c}$ | 0.191$^{b}$ | 0.318$^{a}$ | 0.005 | 0.001 |
| Se (mg/day) | 0.000$^{d}$ | 0.062$^{c}$ | 0.092$^{b}$ | 0.154$^{a}$ | 0.003 | 0.001 |
| Finishing phase | | | | | | |
| Water (L/day) | 10.235$^{b}$ | 10.585$^{ab}$ | 10.502$^{ab}$ | 10.953$^{a}$ | 0.149 | 0.042 |
| Na (g/day) | 0.000$^{d}$ | 1.925$^{c}$ | 2.865$^{b}$ | 4.979$^{a}$ | 0.031 | 0.001 |
| Cl (g/day) | 0.000$^{d}$ | 4.106$^{c}$ | 6.110$^{b}$ | 10.621$^{a}$ | 0.065 | 0.001 |
| Ca (g/day) | 0.000$^{d}$ | 0.351$^{c}$ | 0.522$^{b}$ | 0.907$^{a}$ | 0.006 | 0.001 |
| Mg (g/day) | 0.000$^{d}$ | 0.211$^{c}$ | 0.313$^{b}$ | 0.545$^{a}$ | 0.003 | 0.001 |
| Se (mg/day) | 0.000$^{d}$ | 0.102$^{c}$ | 0.151$^{b}$ | 0.263$^{a}$ | 0.002 | 0.001 |
| Total phase | | | | | | |
| Water (L/day) | 8.433$^{b}$ | 8.534$^{ab}$ | 8.446$^{ab}$ | 8.676$^{a}$ | 0.127 | 0.042 |
| Na (g/day) | 0.000$^{d}$ | 1.552$^{c}$ | 2.304$^{b}$ | 3.944$^{a}$ | 0.035 | 0.001 |
| Cl (g/day) | 0.000$^{d}$ | 3.310$^{c}$ | 4.914$^{b}$ | 8.413$^{a}$ | 0.075 | 0.001 |
| Ca (g/day) | 0.000$^{d}$ | 0.283$^{c}$ | 0.420$^{b}$ | 0.719$^{a}$ | 0.006 | 0.001 |
| Mg (g/day) | 0.000$^{d}$ | 0.170$^{c}$ | 0.252$^{b}$ | 0.432$^{a}$ | 0.004 | 0.001 |
| Se (mg/day) | 0.000$^{d}$ | 0.082$^{c}$ | 0.122$^{b}$ | 0.208$^{a}$ | 0.004 | 0.001 |

$^{a–d}$Means in the same row with different superscripts differ ($p < 0.05$).

Growing phase, 0 to 27 days; Finishing phase, 28 to 56 days; Total phase, 0 to 56 days.

higher in the 3% BMW group than in the 5% BMW group ($p < 0.05$). In particular, the ADG had significantly decreased in the 5% BMW group compared to the other treatment groups during the growing-finishing phase ($p < 0.05$). There was no significant difference between the ADFI and G/F among the treatment groups in the growing-finishing phase.

## Intake of water and additional minerals from brine mineral water

The average daily water and mineral intake from the BMW are shown in Table 4. The water intake was not different among the groups at the growing phase but it significantly increased in the 5% BMW group at the finishing phase ($p < 0.05$). The quantities of ingested Na, Cl, Ca, Mg, and Se from BMW were significantly higher in all the BMW groups than in the control group ($p < 0.05$).

## Complete blood corpuscle count

The RBC, HGB, and HCT levels were significantly higher in the 3% BMW group than in the control group ($p < 0.05$) in growing-finishing pigs (Table 5). However, the MCV, MCH, and MCHC levels did not significantly differ among the groups. Substances in the RBCs without MCV increased in the BMW groups compared with the control group, but there was no significant difference in WBC and PLT among the treatment groups (Table 5). None of the CBC levels exceeded the normal range in growing-finishing pigs.

**Table 5.** **Effects of brine mineral water (BMW) on the complete blood corpuscle count of blood, biochemical analysis, and immune response of the serum in the growing-finishing pigs (0 to 56 days)**

| Items | Diluted BMW in intake water (%) | | | | SEM | *p*-value |
|---|---|---|---|---|---|---|
| | 0 | 2 | 3 | 5 | | |
| Complete blood corpuscle count | | | | | | |
| RBC (× $10^6$ cell/μL) | $6.963^c$ | $7.216^{bc}$ | $7.593^a$ | $7.296^b$ | 0.085 | 0.006 |
| HGB (g/dL) | $13.033^b$ | $13.872^{ab}$ | $14.561^a$ | $14.172^a$ | 0.273 | 0.033 |
| HCT (%) | $38.100^b$ | $39.311^{ab}$ | $41.539^a$ | $38.606^{ab}$ | 0.859 | 0.139 |
| MCV (fL) | 54.689 | 54.494 | 55.183 | 55.222 | 0.724 | 0.899 |
| MCH (pg) | 18.767 | 19.317 | 19.294 | 19.322 | 0.423 | 0.765 |
| MCHC (g/dL) | 34.344 | 35.417 | 34.989 | 35.172 | 0.511 | 0.602 |
| WBC (× $10^6$ cell/μL) | 16.639 | 15.206 | 18.533 | 13.433 | 1.359 | 0.135 |
| PLT (× $10^6$ cell/μL) | 195.333 | 195.333 | 160.056 | 100.778 | 29.048 | 0.187 |
| Biochemical analysis | | | | | | |
| GLU (mg/dL) | 95.389 | 96.333 | 103.527 | 95.167 | 2.473 | 0.181 |
| CHOL (mg/dL) | 93.444 | 85.111 | 98.833 | 96.111 | 4.071 | 0.196 |
| HDL (mg/dL) | $35.333^a$ | $33.667^{ab}$ | $36.000^a$ | $28.333^b$ | 1.563 | 0.054 |
| LDL (mg/dL) | 38.889 | 38.167 | 39.611 | 38.667 | 0.898 | 0.798 |
| TG (mg/dL) | 47.667 | 38.556 | 46.000 | 31.667 | 9.902 | 0.455 |
| Serum immunoglobulins | | | | | | |
| IgG (mg/dL) | $383.167^b$ | $437.111^a$ | $429.556^a$ | $409.500^{ab}$ | 8.096 | 0.017 |
| IgM (mg/dL) | $102.889^c$ | $138.778^a$ | $121.778^b$ | $126.722^{ab}$ | 3.348 | 0.001 |

$^{a-c}$Means in the same row with different superscripts differ ($p < 0.05$).

RBC, red blood cell; HGB, hemoglobin; HCT, hematocrit; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; WBC, white blood cell; PLT, platelet; GLU, glucose; CHOL, total cholesterol; HDL, high-density lipoprotein cholesterol; LDL, low-density lipoprotein cholesterol; TG, triglyceride; IgG, immunoglobulin G; IgM, immunoglobulin M.

### Biochemical factors, immunoglobulin G, and immunoglobulin M

Table 5 shows the effects of BMW on the GLU, CHOL, HDL, LDL, and TG levels during the 56-day experiment. The GLU, CHOL, LDL, and TG levels in the porcine serum did not significantly differ among the groups for up to 56 days; however, the HDL levels were significantly lower in the 5% BMW group than in the control group ($p < 0.05$). The IgG levels were also higher in the BMW groups than in the control group, particularly in the 2% and 3% BMW groups (Table 5). The IgM levels were also significantly higher in the BMW groups than in the control group, and the level in the 2% BMW group was significantly ($p < 0.01$) higher than those in the 3% and 5% BMW groups in growing-finishing pigs (Table 5).

## DISCUSSION

Natural substances are typically more physically and chemically stable than artificial mixtures, such as chemical products and high molecular substances [15]. In this context, BMW is useful for the addition of multiple complexes to feed because it is composed of valuable minerals that are advantageous for mammalian growth. In practice, BMW has antidiabetic influences in mice [16]; moreover, a study of weaning pigs also found that using BMW improved the feed efficiency and hematological values in weaning pigs [3]. Our study also showed that supplementation with BMW improved the growth performance and RBC levels in growing-finishing pigs, but over 5% BMW in the intake water negatively affected growth performance in growing-finishing pigs. Thus, we suggest that diluting intake water with two to three percentage of BMW is optimal for the growth

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performance in growing-finishing pigs. Eventually, we determined that BMW was beneficial for the growth performance of growing-finishing pigs, but we confirmed that supplementation with BMW in the form of water was inefficient from an animal management perspective. Therefore, we strongly suggest that further studies should be conducted on the effect of extracted BMW powder in feed additives on the growth performance of pigs.

An appropriate level of sodium and chloride is essential for growth performance and feed efficiency, but excessive amounts cause sodium toxicity and side effects such as nervousness, weakness, and epilepsy, which can cause death [17,18]. Some studies report that deep-ocean seas and mineral-rich deep-ocean seas contain various beneficial minerals [19,20]; however, the sodium and chloride concentrations of seawater should be considered when being used by animals and humans because excessive NaCl causes dehydration and sodium toxicity [21]. With intake of over 5% BMW, the viability of male reproductive cells decreased and the abnormal morphology increased; in particular, most cells shrank in cell culture medium containing 10% BMW [12]. In our study, the water intake was increased in the BMW groups compared to the control group in finishing pigs; however, there were no differences among the 0%, 2%, and 3% BMW groups in growing-finishing pigs. We suggest that 2% to 3% BMW may be the optimal concentration for the intake of beneficial minerals from BMW for the management of growing-finishing pigs.

Dehydration and acute contraction of the spleen occur when the RBC concentrations are increased, whereas low RBC levels cause hemorrhagic anemia, hemolytic anemia, and oligemia in mammals [21]. Furthermore, high levels of HGB cause hemolytic anemia, HGB-free disease, and lipidemia, whereas microcytosis is caused by low HGB. In this study, all BMW groups showed increased RBC levels, but none of the parameter levels exceeded the normal range in growing-finishing pigs. These results indicate that BMW is a stable feed additive in growing-finishing pigs. Additionally, Se is an essential mineral in mammals that protects the membrane of RBCs from peroxides and supports oxygen transport in blood vessels [22]. Thus, we suggest that the total RBC, HGB, and HCT levels of the treatment groups were increased due to Se in the BMW. Based on these results, we will conduct a future study on the effects of the extracted mineral powder derived from BMW on the growth performance, blood characteristics, and immune response in pigs and apply it to study reproductive performance in sows.

WBCs are classified as granulocytes and agranulocytes; granulocytes are composed of neutrophils, eosinophils, and basophils for the performance phagocytosis, neutralization of poison, transportation of heparin, and storage of peroxidase. Lymphocytes and monocytes in agranulocytes play roles in the production of antibodies, the differentiation of T and B cells, and phagocytosis. In this study, there were no significant differences in WBC parameters between the control and BMW groups; however, the WBC parameters did not exceed the normal range in growing-finishing pigs. Based on this result, we suggest that BMW does not influence the immune system of pigs through granulocytes and agranulocytes.

Cholesterol metabolism is directly related to internal lipids, which influence the growth performance of pigs [5]. LDL and HDL are types of cholesterol; HDL plays an anti-inflammatory and antioxidant role in mammals [23]. HDL is increased by Se, which has been shown to reduce CHOL in rats [24]. Furthermore, Cu and Se supplementation depletes lipid contents in rats [25]. In this study, although the concentration of HDL in porcine serum was significantly lower in the 5% BMW group than in the control group, the HDL levels of the 2% and 3% BMW groups did not significantly differ from that of the control group. We suggest that the Se of BMW positively regulates the HDL levels of growing-finishing pigs.

When WBCs are exposed to antigens from external materials, such as microorganisms and viruses, IgM forms pentomic molecule chains, which produce many antibodies [26]. A diet of fermented liquid feed increased IgG concentrations in weaning pigs [27], and the IgG and IgM levels were higher in the BMW groups of this study. Supplementation with BMW increased Ig, which is directly related to the immune system and may protect growing-finishing pigs from diseases; these results indicate that the addition of BMW diluted with water is beneficial to the growth, RBC activity, cholesterol metabolism, and immunity of growing-finishing pigs. Previous studies have reported that supplementation with BMW could improve the growth performance and RBC levels of weaning pigs [3]; thus, the continuous intake of BMW could increase growth performance and immunity.

## CONCLUSION

The addition of BMW improved the feed efficiency, total RBC, HGB, HCT, HDL cholesterol, IgG, and IgM levels during the growth phase in pigs. The results of this experiment suggest that the addition of 2 to 3% BMW to water improved the growth and immunity of growing-finishing pigs. In conclusion, BMW has the potential to be used as a feed additive for enhancing the immunity and feed efficiency of growing-finishing pig diets.

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