



Effects of Spent Composts of Selenium-enriched Mushroom and Sodium Selenite on Plasma Glutathione Peroxidase Activity and Selenium Deposition in Finishing Hanwoo Steers

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ABSTRACT : Effects of spent composts of selenium-enriched mushroom (Se-SMC) on plasma glutathione peroxidase (GSH-Px) activity and selenium (Se) deposition in finishing Hanwoo (*Bos taurus coreanae*) steers were investigated. Twenty-five Hanwoo steers (average body weight = 613 kg, average age = 22 months) were allotted to treatments in five groups of five steers per pen for 12 weeks preceding slaughter. Treatments were SMC alone (CON; 0.1 ppm Se), 0.3 ppm (0.3 Se-SMC), 0.6 ppm (0.6 Se-SMC), 0.9 ppm (0.9 Se-SMC), and 0.9 ppm (sodium selenite; SENI) Se. During the experimental period, blood samples were taken to analyze Se concentrations and GSH-Px activities. Muscle and liver samples were collected for analyses of Se contents after slaughter. Dry matter intake and body weight gain were not affected by Se-SMC or sodium selenite supplementation. Selenium concentration in the whole blood and GSH-Px activity in plasma were linearly increased ($p < 0.01$) with increasing levels of Se-SMC. The whole blood Se concentration of SENI treatment was significantly higher ($p < 0.05$) than that of CON treatment from 4 weeks, whereas there was no significant difference in GSH-Px activities between both treatments at 8 and 12 weeks. Selenium content in the hind leg and liver increased linearly ($p < 0.05$) with increasing levels of Se-SMC, but those of SENI treatments were not significantly different from CON treatments. These results suggested that Se in the Se-SMC was highly bioavailable to blood and tissues of ruminants, especially compared with Se in the sodium selenite. Therefore, Se-SMC might be used not only as an inexpensive way of providing Se for ruminants but also as another way of producing Se-fortified beef. (**Key Words :** Spent Composts of Se-Enriched Mushroom, Sodium Selenite, Glutathione Peroxidase, Selenium Deposition, Hanwoo Steers)

INTRODUCTION

It has been well known that selenium (Se) plays an important role in cellular antioxidant defense system of living tissues (Gerloff, 1992), in which the Se-containing antioxidant enzyme glutathione peroxidase (GSH-Px) is responsible for catalyzing the decomposition of lipid hydroperoxides into less-reactive products (Rotruck et al., 1973; Han et al., 2004). More recently, it has been reported that adequate or supranutritional levels of Se intake could

not only decrease incidences of tumors in humans (Clark et al., 1996), but also alleviate risks of many diseases associated with Se (Rayman, 2000). For these reasons, some animal nutritionists have made attempts to accumulate Se into animal products such as meat, milk and egg by supplementing dietary Se sources in the feed (Ortman and Pehrson, 1999; Lawler et al., 2004; Payne et al., 2005). In their studies, supplementation of inorganic Se such as sodium selenite or sodium selenate to animal feed showed considerable limitations in the intestinal absorption and retention of the animal tissues. In contrast, organic Se has been found to be an effective form to enhance Se contents in animal products (Mahan and Parrett, 1996; Ortman and Pehrson, 1999). Selenized yeasts generally used as an organic Se source are efficient in increasing Se contents in the animal products, but price is the limitation since they are relatively expensive.

In previous studies, by utilizing the Se-accumulating property of mushroom (Van Elteren et al., 1998), Se-

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Table 1. Chemical compositions of spent composts of mushrooms

Item	Moisture (g)	CP ¹ (g)	CF ² (g)	EE ³ (g)	CA ⁴ (g)	NFE ⁵ (g)	Se (mg)
	----- kg ⁻¹ DM ⁸ -----						
Se-SMC ⁶	507.14	86.02	270.44	35.91	69.18	538.45	5.03
SMC ⁷	509.91	84.88	296.27	33.67	70.80	514.38	0.16

¹ Crude protein. ² Crude fiber. ³ Ether extract. ⁴ Crude ash. ⁵ Nitrogen-free extract.

⁶ Spent mushroom composts from Se-enriched mushroom. ⁷ Spent mushroom composts from normal mushroom. ⁸ Dry matter.

Table 2. Ingredients and chemical compositions of the experimental diets

Item	Treatments				
	CON	0.3 Se-SMC	0.6 Se-SMC	0.9 Se-SMC	SENI
Ingredient composition (DM basis)					
Se-SMC ¹ (%)	-	3.77	9.79	15.84	-
SMC ¹ (%)	15.76	12.01	6.02	-	15.76
Corn grain (%)	62.45	62.43	62.41	62.39	62.45
Barley grain (%)	6.19	6.19	6.19	6.19	6.19
Corn gluten meal (%)	2.23	2.23	2.23	2.23	2.23
Tall fescue straw (%)	5.61	5.61	5.60	5.60	5.61
Barley bran (%)	2.14	2.14	2.14	2.14	2.14
Molasses sugarcane (%)	5.51	5.51	5.51	5.50	5.51
Sodium selenite ² (mg/kg)	-	-	-	-	1.83
Vitamin/mineral mix ³ (%)	0.11	0.11	0.11	0.11	0.11
Chemical composition (DM basis)					
Dry matter (%)	75.69	75.42	75.74	75.51	75.23
Crude protein (%)	11.66	11.82	11.49	11.19	11.38
Crude fiber (%)	12.27	11.62	13.18	13.76	12.71
Ether extract (%)	3.46	3.02	3.85	3.92	3.65
Crude ash (%)	2.28	2.19	2.32	2.33	2.29
Nitrogen-free extract (%)	70.33	71.35	69.16	68.80	69.97
Calcium (%)	0.84	0.87	0.85	0.83	0.88
Phosphorus (%)	0.28	0.31	0.30	0.29	0.32
Selenium (mg/kg)	0.083	0.299	0.623	0.897	0.902
TDN ⁴ (%)	74.72	74.75	74.53	74.32	74.68

¹ See Table 1. ² Sodium selenite was purchased from sigma chemicals.

³ Consisted of Ca 15%, P 6.8%, Mg 7.0%, Na 7.8%, Zn 5,000 mg/kg, Mn 4,000 mg/kg, Cu 500 mg/kg, I 300 mg/kg, Co 20 mg/kg, Se 0 mg/kg, vitamin A 400,000 IU/kg, vitamin D₃ 75,000 IU/kg, and vitamin E 500 mg/kg.

⁴ TDN value was calculated according to the regression equation described by Wardeh (1981).

enriched mushroom and spent mushroom composts (SMC) were produced when sodium selenite as an inorganic Se source was added to mushroom composts. It was proven that spent mushroom composts (Se-SMC) contained considerable amounts of Se mainly as the organic form (Lee et al., 2005). Therefore, it was hypothesized that the supplement of Se-SMC as a nonconventional feed would not only produce Se-fortified beef but also reduce feed cost. In the present study, effects of Se-SMC in comparison with sodium selenite on plasma GSH-Px activity and Se deposition in finishing Hanwoo (*Bos taurus coreanae*) steers were determined.

MATERIALS AND METHODS

Preparation of spent mushroom composts and experimental design

The SMC was obtained from two different mushroom farms where Se-enriched and normal mushrooms of the

same species (*Flammulina velutipes*) were cultivated under the same growth condition. Selenium-enriched mushroom is produced by supplementing inorganic Se as sodium selenite (2 mg Se per kg composts on the fresh basis) in the composts, while normal mushroom is produced by the conventional method without the supplement of sodium selenite. After about a 60-d period of mushroom growth, both the SMCs were transported into Hanwoo cattle farm (located at Chonnam province of Korea) to be utilized as a feed ingredient of experimental diets. Chemical compositions of each SMC from Se-enriched and normal mushrooms are presented in Table 1.

Steers were assigned randomly to one of five treatments (n = 5 per treatment): SMC alone (CON; 0.1 ppm Se), 0.3 ppm (0.3 Se-SMC), 0.6 ppm (0.6 Se-SMC), 0.9 ppm (0.9 Se-SMC) Se, and 0.9 ppm (sodium selenite; SENI). The total of 25 finishing Hanwoo steers (average body weight = 613 kg, average age = 22 months), as similar as possible in age and body weight, were allotted to treatments in five

Table 3. Effects of Se-SMC and sodium selenite on dry matter intake and body weight of Hanwoo steers

Item	Treatments					SE ¹	Significance
	CON	0.3 Se-SMC	0.6 Se-SMC	0.9 Se-SMC	SENI		
Period of feeding	----- DM intake (kg/head/d) -----						
1 to 30 d	9.05	9.04	9.12	9.08	9.07	1.28	NS
31 to 60 d	9.38	9.55	9.47	9.33	9.43	1.30	NS
61 to 90 d	9.53	9.39	9.54	9.58	9.51	1.35	NS
Overall	9.32	9.33	9.38	9.33	9.34	1.27	NS
	----- (BW gain) -----						
Total BW gain (kg)	65.78	66.45	65.57	66.32	66.01	9.57	NS
ADG ² (g)	730.87	738.36	728.50	736.83	733.44	106.33	NS

NS: Not significant. ¹ Pooled standard error. ² Average daily gain.

groups of five steers per pen. Each diet was fed to steers in an individual gate feeding system.

Experimental diets, feeding, and management

Ingredients and chemical compositions of the experimental diets are presented in Table 2. Except for the sodium selenite treatment, other experimental diets were formulated to combine SMC with Se-SMC in order to adjust the levels of Se in the diets. For the sodium selenite treatment, sodium selenite was dissolved in distilled water (0.9 mg Se/kg feed, DM basis) and then added to control diet (CON). Selenium contents in the treatment diets were within the range of expected concentrations, and CP and TDN contents (on DM basis) were similar among treatments as shown in Table 2. Thus, diets were isocaloric and isonitrogenous among treatments. The nutritional levels of experimental diets were determined on the basis of the official Korean feeding standard for Hanwoo beef cattle (MAF and NLRI, 2002). Due to the high concentration of moisture for SMC, the diets were formulated on the cycle of 2 weeks period to prevent unfavorable fermentation. The experimental diets were stored in the polyethylene vinyl envelope with 0.7×1.5 m size in order to keep anaerobic condition until fed to animals. Diets were analyzed for Se content to make sure that they contained the appropriate Se levels for each treatment after formulated and packed. All steers were conformed to the experimental environment and experimental diets for 2 weeks period in which steers were gradually switched from a conventional diet to the experimental diet and the main feeding trial was subsequently performed for 12 weeks. Treatment diets were provided for *ad libitum* intake twice daily at 07:00 and 19:00 h, and water was allowed to be accessible freely through the automatic water provider. The water contained undetectable concentration of Se (<2 ng/ml). Daily dry matter intake was recorded by the difference between the supply and ort amounts, and initial and final body weights for all animals were measured to observe body weight gain on the daily basis throughout the experimental period. At the end of the experimental period, all animals were slaughtered in the slaughterhouse (National Agricultural

Cooperative Federation, Chonnam, Korea) for collection of tissues from hind leg (*m. triceps surae*) as a skeletal muscle and liver as a high metabolic organ for Se.

Sample collection and analytical methods

Each treatment diet was sampled after manufactured and analyzed for nutritional components according to AOAC (1995). Blood samples were collected from the jugular vein into 10-ml heparinized tubes (Vacutainer tube, Becton-Dickinson, Inc., NJ, USA) at 2, 4, 8 and 12 weeks after feeding for the measurements of whole blood Se concentration and GSH-Px activity in plasma. Blood samples for whole blood Se analyses were frozen at -75°C and freeze-dried. Blood samples for GSH-Px activity were immediately centrifuged (1,500×g for 15 min) to obtain plasma, which was stored at -75°C until analyses. A cut of about 1 kg from the hind leg and liver taken from slaughtered animals was thoroughly minced, freeze-dried, and kept at -75°C until analyses. With the hind leg tissue, subcutaneous adipose tissues were removed to analyze total Se content.

Selenium analyses for all samples (diets, whole blood and tissues) were determined by the fluorometric method of AOAC (1995). For more detail procedure of Se analysis, all samples lyophilized were ignited and digested in the oxygen-saturated combustion flask (ELJEE glass manufacturer, Seoul, Korea) with nitric acid (70%, Sigma Aldrich, USA). The pH for digested samples was adjusted to 2±0.2 using ammonium hydroxide (Sigma Aldrich, USA). After the addition of 2,3-diaminonaphthalene (Sigma Aldrich, USA), the samples were placed at room temperature for about 2 h to allow the formation of piarselenol complex. Placed samples were transferred into a separator funnel and the piarselenol complex in samples was extracted by shaking after the addition of cyclohexane (Sigma Aldrich, USA). The fluorescence of the extracted complex was measured with the excitation wavelength set at 375 nm and emission wavelength at 520 nm. The samples completed the analytical preparation were assayed in a fluorescence spectrophotometer (Perkin-Elmer Model 204, USA) equipped with xenon power supply (Perkin-Elmer

Table 4. Effects of Se-SMC and sodium selenite on the plasma GSH-Px activity of Hanwoo steers

Item	Treatments					SE ¹	Significance
	CON	0.3 Se-SMC	0.6 Se-SMC	0.9 Se-SMC	SENI		
----- GSH-Px activity (unit) ² -----							
2 weeks	0.527 ^c	0.658 ^b	0.931 ^a	0.939 ^a	0.690 ^b	0.0584	***
4 weeks	0.734 ^b	0.772 ^b	1.045 ^a	1.165 ^a	1.028 ^a	0.1141	**
8 weeks	0.697 ^b	0.740 ^b	1.060 ^a	1.168 ^a	0.708 ^b	0.0904	***
12 weeks	0.622 ^b	0.670 ^b	1.364 ^a	1.457 ^a	0.689 ^b	0.1194	***

** p<0.01 and *** p<0.001.

^{a, b, c} Means in a row with different superscripts differ significantly (p<0.05).

¹ Pooled standard error.

² One unit of glutathione peroxidase (GSH-Px) activity equals 1 μ mol of NADPH oxidized per min/ml of plasma.

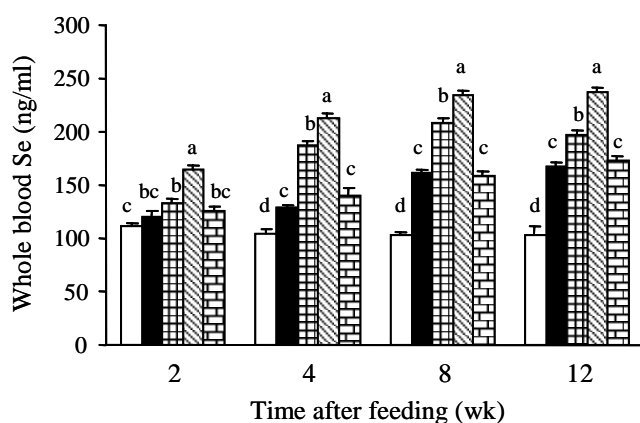


Figure 1. Effects of Se-SMC and sodium selenite supplement on the whole blood Se concentrations in Hanwoo steers. Legend: CON (open bar), 0.3 Se-SMC (solid bar), 0.6 Se-SMC (crosshatched bar), 0.9 Se-SMC (diagonal bar), and SENI (block bar). ^{a, b, c, d} Within each feeding time, bars with different letters are significantly different (p<0.05). Vertical bars represent standard error.

Model 150, USA).

The GSH-Px activity in plasma was assayed by the coupled enzymatic method of Lawrence and Burk (1976) by utilizing hydrogen peroxide and cumene hydroperoxide as substrates. The medium mixture for GSH-Px assay consisted of 10 mM GSH, 2 mM NADPH, 10 mM EDTA, 10 mM sodium azide, glutathione reductase (10 unit per milliliter), 500 mM potassium phosphate and distilled water. The mixture was incubated at 25°C for 5 min before the addition of 500 times diluted plasma. After the addition of substrate and diluted plasma to the mixture, GSH-Px activity was assayed in the spectrophotometer (Shimadzu, Japan) with the kinetic function. The absorbance at 340 nm was recorded for 3 min at 10 sec intervals. The activity was defined as micromoles of NADPH oxidized per minute and per milliliter of plasma.

Statistical analysis

Statistical analysis for all dependent variables was performed as a completely randomized design using the general linear model of SAS program (Version 8.1; SAS

Inst. Inc., Cary, NC, 2000). Dependent variables were dry matter intake, performances (body weight gains), Se concentration in whole blood, plasma GSH-Px activity, and Se contents of tissues. The model included treatments (df = 4) and steers within the treatment (df = 4). Steers within the treatment were used as error terms to test effects of treatments. Significant differences among treatments were determined by Duncan's multiple range test at a level of p<0.05 (Steel and Torrie, 1980).

RESULTS AND DISCUSSION

Daily dry matter intake and body weight gain

Either Se-SMC or sodium selenite supplementation to the diets did not affect DM intakes for Hanwoo steers throughout the trial (Table 3). The present result agreed with Rock et al. (2001) and Gunter et al. (2003), who reported that organic (0.3 and 26 ppm, respectively) or inorganic (0.3 and 26 ppm, respectively) Se supplementation in the diets of gestational ewes or beef cattle did not affect feed intakes.

Dietary Se levels applied for the present study did not result in any sign of selenosis, such as hair loss, separation of the hoof, respiratory failure and so on, as described in NRC (1996). Even though it has been known to be toxic in the dietary Se level from 5 to 20 ppm in swine, resulting in decreased ADG and feed intake (Kim and Mahan, 2001), the toxic level of dietary Se for cattle has not been well defined until now. Hintze et al. (2002) reported that a ration containing more than 10 ppm of Se for beef steers did not influence feed intake and show any toxic symptoms. It can be assumed that ruminants might be more tolerable for Se toxicity than monogastrics. Therefore, the highest level of Se (0.9 ppm) in the Se-SMC and SENI treatments of the present study was relatively safe in terms of steer health.

In addition, total BW gain and ADG of Hanwoo steers during the experimental period were not affected by Se-SMC or SENI (Table 3). Numerous studies investigated to determine effects of dietary Se sources or levels on the Se transfer to tissue for fortification and prevention of Se deficiency in the animals. Of those studies, Lawler et al. (2004) reported that when beef steers were provided diets

with 0.38 or 2.8 ppm of Se using the high-Se wheat and sodium selenate, the high levels of dietary Se did not affect ADG, feed efficiency, and DMI. Likewise, Hintze et al. (2002) reported no differences in performances (feed intake, ADG and final body weight) between steers finished (105 d) on either 0.62 or 11.9 ppm of Se in the diet of high-Se hay and wheat mix. Those studies were in accordance with results from the present study, showing that the highest level of dietary Se (0.9 ppm) did not adversely affect the performances of finishing beef steers.

Blood measurements

Whole blood Se concentrations of Hanwoo steers were significantly increased with enhancing levels of Se-SMC and the supplement of SENI in the diets ($p < 0.01$; Figure 1). The significant differences of whole blood Se concentrations across the treatments were clearly noted with increasing levels of Se-SMC. Between the two types (Se-SMC and sodium selenite) of treatments with 0.9 ppm Se, the whole blood Se concentration of Se-SMC was significantly higher than that of the sodium selenite group ($p < 0.05$). As the supplement period lasted, Se concentrations of the Se-SMC groups were remarkably increased, whereas that of the sodium selenite group was less remarkable. The increase of whole blood Se concentration by increasing levels of Se from Se-SMC was accomplished within 2 weeks and subsequently reached a plateau after 4 weeks. These results were similar to those from Ortman and Pehrson (1999), who reported that the supplement of different Se sources in the diet of dairy cows elevated the whole blood Se levels within 2 weeks, in which selenized yeast consisting of mainly selenomethionine as the organic Se form showed a much higher level for whole blood Se than the inorganic Se, such as sodium selenite and selenate.

Thus, it can be assumed that Se present in Se-SMC is excellent in the intestinal absorption and the main chemical form of Se present in Se-SMC is likely to be a molecular form of Se organically bound to the mycelium protein, showing that the whole blood Se concentration of Se-SMC treatment was much higher than that of sodium selenite treatment at the same Se level in the diet. In the present study, lower Se concentration in the whole blood of sodium selenite treatment seemed that most of the sodium selenite fed by steers was excreted into feces. In ruminants, organically bound Se in the diet is known to be more effective in the intestinal absorption and Se accumulation in tissues than inorganic Se sources (van Ryssen et al., 1989). Previous studies have found that a considerable amount of inorganic Se is formed as insoluble selenide in the rumen, subsequently excreted into feces (Peterson and Spedding, 1963; Wright and Bell, 1966).

Meanwhile, Lee et al. (2005) reported that

approximately 70% of Se present in Se-SMC from Se-enriched mushrooms used in this experiment was organic Se. Stefánka et al. (2001) also demonstrated that inorganic Se added to mushroom compost was converted to organic Se and their predominant form of Se was mostly selenocystine. However, Se distribution among different molecular forms for mushroom species and its SMC used in this experiment has not been determined.

In the present study, even though GSH-Px activities in plasma were less altered compared with the changes of whole blood Se concentrations, GSH-Px activities for groups supplemented with Se-SMC were generally higher than those of CON treatments ($p < 0.01$; Table 4). Significant differences in the plasma GSH-Px activities among treatments began to be detected within 2 weeks. After the supplementation period of 4 weeks, steers fed diet containing 0.3 ppm Se (0.3 Se-SMC) showed slightly higher GSH-Px activities compared to those of CON treatments, but did not significantly differ. On the other hand, GSH-Px activities from 0.6 and 0.9 Se-SMC groups were much higher than those from the CON and 0.3 Se-SMC groups ($p < 0.05$), but significant difference for GSH-Px activity was not observed between 0.6 and 0.9 Se-SMC treatments. Overall, whole pattern for GSH-Px moved upward as dietary Se levels increased, indicating that plasma GSH-Px activities were controlled by the dietary Se content. Even though GSH-Px activity of SENI treatment was significantly increased compared to that of the CON treatment until 4 weeks, its significance between both treatments was not observed in the period of 8 and 12 weeks.

Glutathione peroxidase activities, a physiologically functional form of Se, have been proposed as the best estimate of Se status for human or animal (Stevens et al., 1985; Smith et al., 1988). Gunter et al. (2003) reported that supplement of Se-enriched yeast or sodium selenite increased GSH-Px activity of cows compared with no Se supplemented group. Similarly, Hintze et al. (2002) fed beef steers with a different background (seleniferous or nonseleniferous areas) either 11.9 or 0.62 mg of Se/kg of diet as high-Se hay and wheat mix, and they found that the increased GSH-Px activity was shown in the steers fed the high-Se diet throughout the feeding trial, regardless of the background. Therefore, proportional increases for GSH-Px activities of Se-SMC treatments observed in these experiments seem to be closely related with the increased blood Se concentration by supplementing Se-SMC containing a high proportion of organic Se. However, in spite of the significant increase for whole blood Se concentration of SENI treatment compared to that of CON treatment at 8 and 12 weeks, no significant difference regarding GSH-Px activities between CON and SENI was found. The present results were similar to those of Enjalbert

Table 5. Effects of Se-SMC and sodium selenite on selenium deposition in muscular and hepatic tissues

Item	Treatments					SE ¹	Significance
	CON	0.3 Se-SMC	0.6 Se-SMC	0.9 Se-SMC	SENI		
	----- Tissue Se ($\mu\text{g/g}$ of dry weight) -----						
Hind leg	0.273 ^c	0.368 ^{ab}	0.398 ^{ab}	0.457 ^a	0.318 ^{bc}	0.0505	*
Liver	0.789 ^c	1.401 ^c	2.392 ^b	3.096 ^a	1.258 ^c	0.3543	***

* $p < 0.05$ and *** $p < 0.001$.

^{a, b, c} Means in a row with different superscripts differ significantly ($p < 0.05$). ¹ Pooled standard error.

et al. (1999) and Rowntree et al. (2004), who reported that supplement of inorganic Se as sodium selenite increased plasma Se concentration, but plasma GSH-Px activity was not affected.

On the other hand, it could be explained that the significant difference for GSH-Px activity between CON and SENI treatments at 2 and 4 weeks might be attributable to the increase of whole blood Se concentration in SENI treatment compared to CON treatment. Therefore, one cannot rule out the possibility that the selenite supplement may temporarily increase plasma GSH-Px activity with increasing whole blood Se concentration during the early phase of selenite supplement. However, since plasma GSH-Px is a short-term indicator of Se status and 98% of GSH-Px activity is associated with erythrocytes (Scholz and Hutchinson, 1979), GSH-Px activity of erythrocytes might be further elucidated.

Meanwhile, the possible reason for no significant difference for the activity of GSH-Px between CON and 0.3 Se-SMC after 4 weeks seems to be due to marginal blood Se concentration in order to express the adequate activity of GSH-Px. As shown in Figure 1, the whole blood Se concentrations for 0.6 and 0.9 Se-SMC treatments started from 4 weeks were maintained more than 180 ng/ml which is known to be an adequate level (160 to 1,200 ng/ml) for optimal GSH-Px activity and immune function (Puls, 1989), whereas blood Se concentrations in CON and 0.3 Se-SMC treatments showed marginal levels (60 to 150 ng Se/mL; Puls, 1989). Thus, it can be conceived that in the present study, marginal points of whole blood Se concentration acted as a limiting factor for the expression of adequate GSH-Px activity. However, in an investigation with swine by Mahan et al. (1999), a narrow range of Se concentrations with 0.05, 0.1, 0.2 or 0.3 ppm in the diet even increased GSH-Px activity quadratically compared with the basal diet over a whole period. One cannot rule out the possibility that such a result might be due to species specificity between ruminants and monogastrics.

Even though it could not be shown increases of GSH-Px activities at a low Se level (CON vs. 0.3 Se-SMC), possibly due to the low Se level in plasma and/or species specificity, it was clearly shown that GSH-Px activities at the higher levels of Se (0.6 and 0.9 Se-SMC) were increased, possibly resulting in beneficial effects on animal health.

Se deposition in tissues

As the level of Se-SMC was increased, Se contents in the hind leg and liver were linearly increased ($p < 0.05$), but those of SENI treatments were not significantly different compared with CON treatments (Table 5). Selenium contents in the hind leg and liver were the highest in the 0.9 Se-SMC group, followed by those in 0.6 Se-SMC, 0.3 Se-SMC, SENI, and CON treatments ($p < 0.05$). These results showed that Se deposition in the tissue increased with enhancing levels of Se as the form of Se-SMC. However, taken account into Se intakes among treatments, the rate of Se retention in the skeletal muscle was lower than expected.

Hintze et al. (2001) reported that in ruminants, skeletal muscle Se had a stronger association ($r = 0.66$) with whole blood Se, thus Se concentration of whole blood was the best predictor for that of skeletal muscle. Similarly, results from the present study showed that muscular Se concentration was proportionally increased with enhancing Se levels in the diet as the form of Se-SMC. Many studies have reported that the feeding of organic sources such as Se-enriched yeasts and some forages or grains from seleniferous regions during the defined period enabled animals to increase blood Se concentration and to accumulate Se in their tissues (van Ryssen et al., 1989; Kim and Mahan, 2001; Lawler et al., 2004). The Se speciation for Se-SMC used in the present study was not determined. However, a high proportion of organic Se in Se-SMC as described in the previous report (Lee et al., 2005) could explain the significant increase of Se deposition in the skeletal muscle of Hanwoo beef steers. Selenium concentration in the muscle was similar to the result of Hintze et al. (2002) utilized the same Se level (approximately 0.6 ppm) in the diet as the present study. On the other hand, Se concentration in the liver remarkably increased with supplementing Se-SMC. These results were similar to the data provided by Combs and Combs (1986), which reported that Se concentration in the liver was around four times higher than that in the skeletal muscle. The present data indicated that Se concentration in the liver was up to maximally four folds increased compared with the CON treatment. The increasing concentration of Se in the liver by supplemental Se-SMC level showed the same pattern as skeletal muscle. High Se concentration in the liver compared with muscle might result from the fact that liver acts as a major pool of Se in the body.

In contrast, Se increment in the muscle by feeding Se-SMC was relatively low as minimum 35% to maximum 68% compared with Se-SMC unsupplemented group. Lawler et al. (2004) reported that when high-Se hay or wheat diet with supranutritional level of Se was fed to beef steers, Se concentration in the muscle showed tremendously increased value compared with the control or sodium selenate groups, suggesting that dietary level and chemical form of Se are crucial factors to produce high Se beef. However, Hintze et al. (2002) reported that the transfer of dietary Se to muscle was inefficient as maximum of 2.6%.

Meanwhile, the World Health Organization (1996) reported that the recommended daily allowance (RDA) of Se for adults was 40 µg. In the present study, it was calculated that, on the fresh basis of the highest Se content (0.9 Se-SMC treatment) for the hind leg (30.88% DM), 100 g of the beef contained approximately 14.11 µg Se, providing approximately 35.3% Se of the RDA.

The above results showed that incorporation of Se-SMC into the diet could be easy and inexpensive way to increase Se concentration in the body of beef steers, but sodium selenite was inefficient in the view of intestinal absorption and deposition of Se. In conclusion, based on the health benefits of Se by the reports of Clark et al. (1996) and Rayman (2000), it would be expected that Se concentrations increased in beef by supplementing Se-SMC to Hanwoo steers could contribute to human health by providing more Se.

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REFERENCES

- AOAC. 1995. Official Methods of Analysis. 16th ed. Association of Official Analytical Chemists, Washington, DC.
- Clark, L. C., G. F. Combs Jr., B. W. Turnbull, E. H. Slate, D. K. Chalker, J. Chow, L. S. Davis, R. A. Glover, G. F. Graham, E. G. Gross, A. Krongrad, J. L. Leshner Jr., H. K. Park, B. B. Sanders Jr., C. L. Smith and J. R. Taylor. 1996. Effects of selenium supplementation for cancer prevention in patients with carcinoma of the skin. *J. Am. Med. Assoc.* 276:1957-1963.
- Combs, G. F. and S. B. Combs. 1986. *The Role of Selenium in Nutrition*. Academic Press, Inc., New York, NY.
- Enjalbert, F., P. Lebreton, O. Salat and F. Schelcher. 1999. Effects of pre- or postpartum selenium supplementation on selenium status in beef cows and their calves. *J. Anim. Sci.* 77:223-229.
- Gerloff, B. J. 1992. Effect of Se supplementation on dairy cattle. *J. Anim. Sci.* 70:3934-3940.
- Gunter, S. A., P. A. Beck and J. M. Phillips. 2003. Effects of supplementary selenium source on the performance and blood measurements in beef cows and their calves. *J. Anim. Sci.* 81:856-864.
- Han, B., S. Yoon, J. Su, H. R. Han, M. Wang, W. Qu and D. Zhong. 2004. Effects of selenium, copper and magnesium on antioxidant enzymes and lipid peroxidation in bovine fluorosis. *Asian-Aust. J. Anim. Sci.* 17:1695-1699.
- Hintze, K. J., G. P. Lardy, M. J. Marchello and J. W. Finley. 2001. Areas with high concentrations of selenium in the soil and forage produce beef with enhanced concentrations of selenium. *J. Agric. Food Chem.* 49:1062-1067.
- Hintze, K. J., G. P. Lardy, M. J. Marchello and J. W. Finley. 2002. Selenium accumulation in beef: Effect of dietary selenium and geographical area of animal origin. *J. Agric. Food Chem.* 50:3938-3942.
- Kim, Y. Y. and D. C. Mahan. 2001. Comparative effects of high dietary levels of organic and inorganic selenium on selenium toxicity of growing-finishing pigs. *J. Anim. Sci.* 79:942-948.
- Lawler, T. L., J. B. Taylor, J. W. Finley and J. S. Caton. 2004. Effect of supranutritional and organically bound selenium on performance, carcass characteristics, and selenium distribution in finishing beef steers. *J. Anim. Sci.* 82:1488-1493.
- Lawrence, R. A. and R. F. Burk. 1976. Glutathione peroxidase activity in selenium-deficient rat liver. *Biochem. Biophys. Res. Commun.* 71:952-958.
- Lee, S. H., W. S. Kwak and W. Y. Kim. 2005. Studies on the selenium type and metabolism of selenium accumulation in the selenium-enriched mushroom, *Flammulina velutipes*, and its spent mushroom composts. *J. Anim. Sci. Technol. (Kor.)* 47:305-316.
- Mahan, D. C., T. R. Cline and B. Richert. 1999. Effects of dietary levels of selenium-enriched yeast and sodium selenite as selenium sources fed to growing-finishing pigs on performance, tissue selenium, serum glutathione peroxidase activity, carcass characteristics, and loin quality. *J. Anim. Sci.* 77:2172-2179.
- Mahan, D. C. and N. A. Parrett. 1996. Evaluating the efficacy of selenium-enriched yeast and sodium selenite on tissue selenium retention and serum glutathione peroxidase activity in grower and finisher swine. *J. Anim. Sci.* 74:2967-2974.
- Ministry of Agriculture and Forestry (MAF) and National Livestock Research Institute (NLRI). 2002. *Korean Feeding Standard for Korean Cattle (Hanwoo)*, Korea.
- National Research Council (NRC). 1996. *Nutrient Requirements of Beef Cattle*. 7th Ed. National Academy Press, Washington, DC.
- Ortman, K. and B. Pehrson. 1999. Effect of selenate as a feed supplement to dairy cows in comparison to selenite and selenium yeast. *J. Anim. Sci.* 77:3365-3370.
- Payne, R. L., T. K. Lavergne and L. L. Southern. 2005. Effect of inorganic versus organic selenium on hen production and egg selenium concentration. *Poult. Sci.* 84:232-237.
- Peterson, P. J. and D. J. Spedding. 1963. The excretion by sheep of ⁷⁵selenium incorporated into red clover (*Trifolium pratense* L.): The chemical nature of the excreted selenium and its uptake by three plant species. *N. Z. J. Agric. Res.* 6:13-23.
- Puls, R. 1989. *Mineral Levels in Animal Health: Diagnostic Data*. Sherpa Int., Clearbrook, British Columbia, Canada.
- Rayman, M. P. 2000. The importance of selenium to human health.

- The Lancet 356:233-241.
- Rock, M. J., R. L. Kincaid and G. E. Carstens. 2001. Effects of prenatal source and level of dietary selenium on passive immunity and thermometabolism of newborn lambs. *Small Rumin. Res.* 40:129-138.
- Rotruck, J. T., A. L. Pope, H. E. Ganther, D. G. Hafeman, A. B. Swanson and W. G. Hoekstra. 1973. Selenium: Biochemical role as a component of glutathione peroxidase. *Science* 179:588-590.
- Rowntree, J. E., G. M. Hillm, D. R. Hawkins, J. E. Link, M. J. Rincker, G. W. Bednar and R. A. Kreft Jr. 2004. Effect of Se on selenoprotein activity and thyroid hormone metabolism in beef and dairy cows and calves. *J. Anim. Sci.* 82:2995-3005.
- SAS Institute Inc. 2000. SAS/STAT® User's Guide (Release 8.1 ed.). Statistics, SAS Inst, Inc., Cary, NC.
- Scholz, R. W. and L. J. Hutchinson. 1979. Distribution of glutathione peroxidase activity and selenium in the blood of dairy cows. *Am. J. Vet. Res.* 40:245-249.
- Smith, K. L., J. S. Hogan and H. R. Conrad. 1988. Selenium in dairy cattle: Its role in disease resistance. *Vet. Med.* 83:72-78.
- Steel, R. G. D. and J. H. Torrie. 1980. *Principles and Procedures of Statistics: A Biometrical Approach* (2nd Ed.). McGraw-Hill Book Co., New York.
- Stefánka, Z., I. Ipolyi, M. Dernovics and P. Fodor. 2001. Comparison of sample preparation methods based on proteolytic enzymatic processes for Se-speciation of edible mushroom (*Agaricus bisporus*) samples. *Talanta* 55:437-447.
- Stevens, J. B., W. G. Olson, R. Kraemer and J. Archambeau. 1985. Serum selenium concentrations and glutathione peroxidase activity in cattle grazing forages of various selenium concentrations. *Am. J. Vet. Res.* 46:1556-1560.
- Van Elteren, J. T., U. D. Woroniecka and K. J. Kroon. 1998. Accumulation and distribution of selenium and cesium in the cultivated mushroom *agaricus bisporus* - A radiotracer - aided study. *Chemosphere* 36:1787-1798.
- Van Ryssen, J. B. J., J. T. Deagen, M. A. Beilstein and P. D. Whanger. 1989. Comparative metabolism of organic and inorganic selenium by sheep. *J. Agric. Food Chem.* 37:1358-1363.
- Wardeh, M. F. 1981. Models for estimating energy and protein utilization for feeds. Ph.D. Dissertation; Utah State Univ., Logan.
- World Health Organization. 1996. Selenium. In: *Trace Elements in Human Nutrition and Health*. Geneva WHO. pp. 105-122.
- Wright, P. L. and M. C. Bell. 1966. Comparative metabolism of selenium and tellurium in sheep and swine. *Am. J. Physiol.* 211:6-10.