

Improvement of Lipid Oxidation Stability of Seasoned Pork and Formula Development of Seasoning Sauce for Pork *Bulgogi* with *Doenjang* and Onion Using Mixture Experimental Design

Hyun-Ju Oh, Chang-Soon Kim*, and Duk-Joon Chang¹

Department of Food and Nutrition, Changwon National University, Changwon, Gyeongnam 641-773, Korea

¹Department of Statistics, Changwon National University, Changwon, Gyeongnam 641-773, Korea

Abstract The antioxidative effect of *doenjang* (fermented soybean paste) and onion added in the seasoned pork, *bulgogi* was evaluated and the optimum mixture ratio of ingredients in seasoning sauce has been established using mixture experimental design (MED). When the seasoned pork, *bulgogi* was prepared with soy sauce 12% (control), *doenjang* 9% (in replacement of soy sauce 9%) added group (DG), and *doenjang* 9% and onion 33.4% (in replacement of water 33.4%) added group (DOG), and stored at -25°C for 6 months, the peroxide value (POV) and thiobarbituric acid (TBA) value of control, DG, and DOG were all lower than that of control from the initial stage of storage. The antioxidative effect of seasoned pork was found to increase with the replacement of *doenjang* in seasoning sauce for pork *bulgogi*. Moreover, antioxidative effects in DOG increased more synergistically. Therefore, the lipid oxidation stability of pork *bulgogi* was improved by the addition of *doenjang* and onion onto the seasoning sauce. The optimum mixture ratio of seasoning sauce for pork *bulgogi* followed up by the MED was found to be *doenjang* 7.10%, soy sauce 9.46%, onion 19.72%, and water 42.58% with excellent sensory quality.

Keywords: pork *bulgogi*, seasoning sauce, *doenjang*, onion, antioxidative effect, mixture experimental design (MED)

Introduction

The annual meat consumption amount in our nation is continuously increasing, and the consumption of pork meat per capita has been gradually increased from 14.8 kg in 1990 to 17.8 kg in 2005, and the trend of such increase is expected to be continued in a future (1). Compared to beef, the price of pork is cheap and its nutritional value is not inferior to beef, which makes the meat to be favored widely by public (2). The overconsumption of red meat foods lead to the problem of causing cardiovascular diseases since it is the source of animal fat and cholesterol (3).

With the increased acceptability of dining out consumption and convenient foods in recent years, the sales amount of seasoned meat in the year of 2006 was increased to 1,388.4 tons showing the 47.3% increase from the previous year (1), which suggested the increased consumption pattern of seasoning sauces for *bulgogi*, seasoned meat. In case of using pork as the source meat of seasoned pork, the disadvantages of having pork smell and off-flavor have been mitigated by using alcohol, spices such as ginger, and herbs (4). Modern people pursue new taste along with the interest on health. So, the development of new seasoned pork products distinguished from conventional seasoned pork product is essential by having the characteristic taste of *doenjang* (fermented soybean paste) and healthiness at the same time considering the fact that Koreans strongly favor the flavor of *doenjang*. Traditionally, *doenjang* has

been often cooked and set a table with *suwook* (boiled pork) (5). In such instance, it is well known that the flavor of *doenjang* and pork is well incorporated from our long term experiences of dietary habit. The characteristic basic taste of *doenjang* came from the saltiness, the sweetness by the hydrolysis of carbohydrates or dextrin, and from the savory taste by the hydrolysis of soy proteins. All these tastes are well incorporated to enhance the flavor of seasoned foods (6). It has been well known that the functional compounds found in *doenjang* exhibit antimutagenic, anticarcinogenic, antioxidative, cardiovascular disease preventing effects and other beneficial properties (7). As the study of lipid oxidation stability of meat products using traditional fermented soybean foods, Cheigh *et al.* (8) and Moon and Cheigh (9,10) reported the antioxidative effect of lipid tissue by the addition of high levels of *doenjang* and soy sauce (50-100% for 100 g of beef and fish meats) in model system. Other studies which reported the antioxidative property enhancement in sauces for seasoned meat by adding ginseng saponin (11), chitosan (12), and propolis (13) have been published. In the cooking of seasoned meat, the use of onion has been rapidly increased recently as the physiologically beneficial effects such as the antioxidative (14), antifatigue (15), and lipid metabolism improving effects (16) of onion have been known.

The response surface methodology (RSM) has been frequently applied in food processes and in developing optimum formula (17). Since the RSM is especially effective in pursuing the optimum condition in processing lines, it is utilized effectively in quality control. However, most of the products formulated by combined mixtures ingredients can cause interactions between composing ingredients, which can be difficult in achieving the optimization (17,18). In the case of foods mixed with

*Corresponding author: Tel: +82-55-213-3512; Fax: +82-55-281-7480
E-mail: cskim@changwon.ac.kr
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several ingredients, the response value is more highly affected by the mixing ratio than the mixing amount of each ingredient. The mixture experimental design (MED) fixes the mixture ratio of other ingredients which are not included within the limit of interest and selects the ingredients to be changed as the independent variables to locate the optimum mixing ratio of each independent variable under the assumption that the summation of mixture ratio is always uniform ($1=100\%$) (17-19). The MED that is widely introduced in domestic and overseas food development research includes the reports on coffee (20), rice cake (21,22), noodle (23), tortilla (24), cereal (25), and cheddar cheese (26).

The aim of this study was to evaluate the antioxidative improving effects of seasoned pork, *bulgogi* by the addition of *doenjang* and onion through the measurement of peroxide value (POV) and thiobarbituric acid (TBA) values, and to establish the optimum mixture ratio of seasoning sauce by applying MED for the development of pork *bulgogi* with addition of *doenjang* and onion with excellent sensory quality.

Materials and Methods

Samples and proximate analysis The fore-shank portions of individual porks were purchased at 24 hr post slaughter from a local supplier. The pork was trimmed of external fat and was sliced into pieces of 0.3 cm thickness. The sliced pork was stored in polyethylene bags at -80°C , and thawed at 4°C for 24 hr as it is needed. Traditional *doenjang* was provided by the Al-Ali food company and used while storing it at -25°C after grinding for homogenization. Onion, sugar, syrup, garlic, ginger, *soju* (alcohol), pepper, and soy sauce, used as seasonings in the formula, were purchased from a local market in Changwon, Korea. Proximate analyses of *doenjang* were measured according to the AOAC official method (27). The salt concentration and Brix of *doenjang* and soy sauce were measured using a salinity meter (TM-30D; Takemura Electric Works, Tokyo, Japan) and refractometer (N-50E; Atago, Tokyo, Japan). Proximate compositions of *doenjang* were 47.7% water, 13.9% crude protein, 7.5% crude lipid, 18.3% crude ash, and 13.5% salt. The salt concentration of soy sauce was 14.3%. The Brix of *doenjang* and soy sauce were 19.0 and 18.0°Bx.

Preparation of pork *bulgogi* For the 320 g of pork fore-shank (fat 19.6%), 180 g of basic seasoning sauce (sugar 9.96%, syrup 4.98%, garlic 2.49%, *soju* 3.23%, ginger 0.38%, pepper 0.10%, soy sauce 12.06%, and water 66.80%) was added to immerse the meat and packed it in a closed plastic container for the aging at refrigeration temperature (4) for 24 hr. In the case of adding *doenjang* and onion, the amount of soy sauce and water added onto the mixture was substituted in a certain ratio as shown in Table 1. The samples to be used for the measurement of lipid oxidation stability during the storage of the pork *bulgogi* were used while freeze storing (-25°C). The samples to be used for the sensory evaluation for the establishment of optimum mixture ratio of pork *bulgogi* have been prepared by the identical method as described in above except the freeze storing process and used for the test by cooking at 5 min in

strong fire and 2 min in medium fire after placing the seasoned pork in a preheated fry pan (stainless steel pan, Initiatives; Tefal, Seoul, Korea).

Measurement of lipid oxidation The raw pork, control of soy sauce 12% of seasoning sauce as the basic seasoning ratio as indicated in Table 1, *doenjang* 9% (in replacement of soy sauce 9%) added group (DG), and *doenjang* 9% and onion 33.4% (in replacement of water 33.4%) added group (DOG) were stored at -25°C . Before the experiments, samples were thawed at the refrigeration temperature for 24 hr and extracted the fat by using the Folch *et al.* (28) method to measure the POV and TBA value. The POV of samples was determined by the AOCS official method (29). TBA value of the samples was determined according to Tarladgis *et al.* (30) and expressed as mg of malonaldehyde (MA) per kg of fat.

Experimental design for optimization The optimization experiment of mixing ingredients for the manufacturing of pork *bulgogi* with addition of *doenjang* and onion applied the MED by using 4 independent variables of *doenjang*, soy sauce, onion, and water. Among the basic seasoning mixture ingredients (Table 1), the amount of other ingredients except the above stated 4 independent variables were kept unchanged. Namely, the total 38.05 g (21.14%) was remained from the total seasoning amount of 180 g (100%) after excluding the summated amount of 4 independent variables (141.95 g, 78.86%). The upper and lower limits of experimental range for the ingredients of *doenjang* (X_1), soy sauce (X_2), onion (X_3), and water (X_4) were limited as $0.02 \leq X_1 \leq 0.13$, $0.02 \leq X_2 \leq 0.12$, $0.25 \leq X_3 \leq 0.39$, and $0.41 \leq X_4 \leq 0.59$, respectively, based upon the preliminary sensory test results considering salinity, sweetness, and overall acceptability of pork *bulgogi*. The feasible space for a mixture experiment with 3 components is a triangle called

Table 1. Formula for seasoning sauce containing *doenjang* and onion (%)

Ingredient	Treatment ¹⁾		
	Control	DG	DOG
Sugar	9.96 (17.93)	9.96	9.96
Corn syrup	4.98 (8.96)	4.98	4.98
Garlic	2.49 (4.48)	2.49	2.49
<i>Soju</i> (alcohol)	3.23 (5.81)	3.23	3.23
Ginger	0.38 (0.69)	0.38	0.38
Black pepper	0.10 (0.18)	0.10	0.10
Subtotal	21.14 (38.05)	21.14	21.14
Soy sauce	12.06 (21.71)	3.06 (5.51)	3.06 (5.51)
<i>Doenjang</i>	-	9.00 (16.20)	9.00 (16.20)
Onion	-	-	33.40 (60.12)
Water	66.80 (120.24)	66.80 (120.24)	33.40 (60.12)
Subtotal	78.86 (141.95)	78.86 (141.95)	78.86 (141.95)
Total	100 (180)	100 (180)	100 (180)

¹⁾Control, soy sauce 12.06% added group; DG, *doenjang* 9% (in replacement of soy sauce 9%) added group; DOG, *doenjang* 9% and onion 33.4% (in replacement of water 33.4%) added group. The numbers in parenthesis indicate gram.

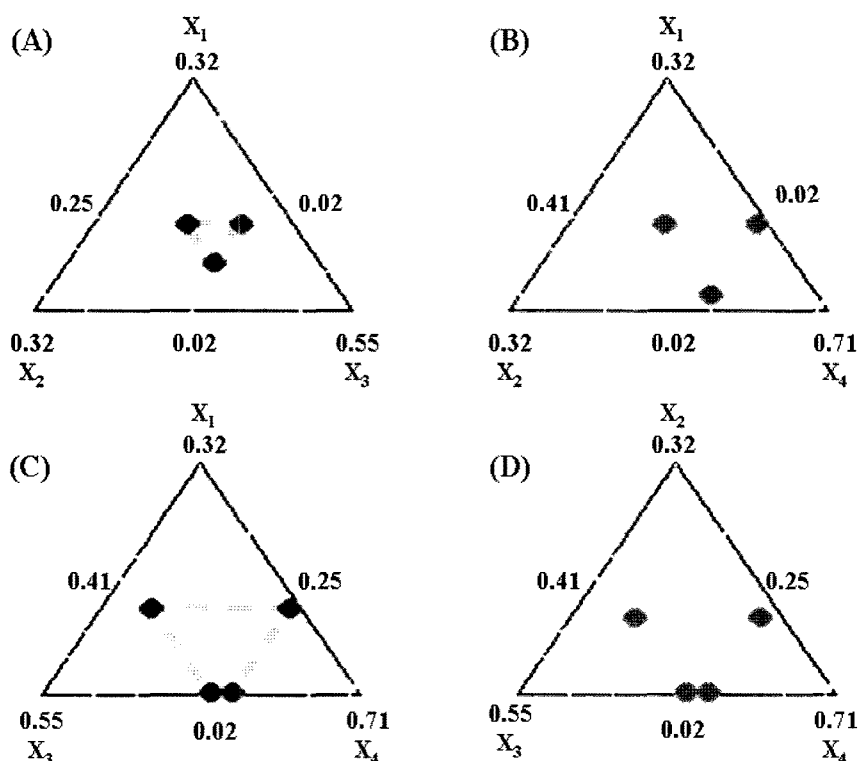


Fig. 1. Design plots in the 4 mixture variables. X_1 , Doenjang; X_2 , soy sauce; X_3 , onion; X_4 , water. The connecting lines limit the design region: (A) X_4 0.41, (B) X_3 0.25, (C) X_2 0.02, (D) X_1 0.02.

a simplex. The composition of each mixture varies depending on its position on the simplex region. The experimental points were formed as Fig. 1 in the dotted line due to limit of upper and/or lower bounds on a per-component. When the mixture ratio of water (X_4), onion (X_3), soy sauce (X_2), and doenjang (X_1) are 0.41, 0.25, 0.02, and 0.02, respectively, Fig. 1 (A) is simplex design plot expressed the 2-dimension experimental regions of 3 components (X_3 , X_2 , X_1), except fixed X_4 , for design of extreme vertical design which are satisfied with $X_1 + X_2 + X_3 + X_4 = 1$. Also, (B), (C), and (D) of Fig. 1 are simplex design plots expressed in regard to 3 components, except fixed X_3 , X_2 , and X_1 , respectively. The 25 experiment points for the extreme vertical design are shown in Table 2.

Sensory analysis Composed of university students and faculty members, 20 panelists were selected for sensory evaluation using a preference test. Three samples of pork *bulgogi* (50 g) were presented in white plates coded with 3-digit numbers, and samples were randomly evaluated. Unsalted crackers and water were given to cleanse the palate between samples. Each panelist evaluated the sample by a structured 9-point hedonic scale of 1 (disliked extremely) to 9 (liked extremely). The overall acceptability and taste preference were evaluated.

Statistical analysis The POV and TBA value were analyzed with analysis of variance using SPSS 12.0 statistical program, and the level of significance among samples was tested at $p < 0.05$ with Duncan's multiple range test. Experimental design and data for optimization

of pork *bulgogi* were subjected to statistical analysis using the MINITAB (Release 14, Minitab Inc., State College, PA, USA) and SAS statistical program (ver. 9.13). The response surface plots (SAS/GRAPH) were constructed to see response of components in models.

Results and Discussion

Effect of doenjang and onion on lipid oxidation stability of seasoned pork The results of changes in the POV and TBA value during the storage at -25°C for 6 months are shown in Fig. 2. The POV results at 0 day in raw pork, control of seasoned pork, DG, and DOG revealed to show 4.33 ± 0.58 , 2.67 ± 0.58 , 2.33 ± 0.58 , and 1.33 ± 0.58 meq/kg, respectively. The POV value in all types of seasoned pork showed a constantly lower than the raw pork. Ingredients such as garlic, ginger, and black pepper in seasoning sauce may also contribute to inhibition of lipid oxidation of *bulgogi* (31). Park *et al.* (32) reported that when the POV of meat products reached more than 25–30 meq/kg, their acceptability as a food was limited. In the point of view, the POV value in the raw pork was already reached 45.76 ± 1.58 meq/kg at the 3rd month, but all types of seasoned pork maintained lower than 30 meq/kg even at the 4th month demonstrating their excellent lipid oxidation stability. DG maintained lower POV during the storage period and its maximum value of POV was 25.55 ± 0.50 meq/kg at the 5th month, which was significantly lower than the control group of seasoned pork that showed 38.77 ± 0.38 meq/kg at the same month. Therefore, the antioxidative property was found to be enhanced by

Table 2. Levels of factors in seasoning sauce with addition of *doenjang*, soy sauce, onion, and water by using the mixture experimental design

Treatment No.	Pseudo component ¹⁾				Actual component (g)			
	X ₁	X ₂	X ₃	X ₄	<i>Doenjang</i>	Soy sauce	Onion	Water
1	0.1300	0.0700	0.3900	0.4100	18.45	9.94	55.36	58.2
2	0.1300	0.0200	0.3900	0.4600	18.45	2.84	55.36	65.3
3	0.1058	0.0725	0.3592	0.4625	15.02	10.29	50.99	65.65
4	0.1058	0.0975	0.3342	0.4625	15.02	13.84	47.44	65.65
5	0.0400	0.1200	0.2500	0.5900	5.68	17.03	35.49	83.75
6	0.0508	0.0975	0.3592	0.4925	7.22	13.84	50.98	69.91
7	0.0200	0.1200	0.2700	0.5900	2.84	17.03	38.33	83.75
8	0.0800	0.1200	0.3900	0.4100	11.36	17.03	55.36	58.2
9	0.0508	0.0475	0.3492	0.5525	7.22	6.74	49.56	78.43
10	0.1300	0.0300	0.2500	0.5900	18.45	4.26	35.49	83.75
11	0.0200	0.0200	0.3700	0.5900	2.84	2.84	52.52	83.75
12	0.1058	0.0525	0.2892	0.5525	15.02	7.45	41.05	78.43
13	0.0508	0.0475	0.3592	0.5425	7.22	6.74	50.98	77.01
14	0.0808	0.0975	0.3592	0.4625	11.48	13.84	50.98	65.65
15	0.0200	0.0200	0.3900	0.5700	2.84	2.84	55.36	80.91
16	0.1058	0.0475	0.3592	0.4875	15.02	6.74	50.99	69.2
17	0.0508	0.0975	0.2992	0.5525	7.21	13.84	42.47	78.43
18	0.1300	0.1200	0.2500	0.5000	18.45	17.03	35.49	70.98
19	0.0200	0.1200	0.3900	0.4700	2.84	17.03	55.36	66.72
20	0.0608	0.0975	0.2892	0.5525	8.63	13.84	41.05	78.43
21	0.1300	0.0200	0.2600	0.5900	18.45	2.84	36.91	83.75
22	0.1300	0.1200	0.3400	0.4100	18.46	17.03	48.26	58.2
23	0.1058	0.0975	0.2892	0.5075	15.02	13.84	41.05	72.04
24	0.0817	0.0750	0.3283	0.5150	11.59	10.65	46.61	73.1
25	0.1058	0.0475	0.2942	0.5525	15.02	6.74	41.76	78.43

¹⁾X₁, *Doenjang*; X₂, soy sauce; X₃, onion; X₄, water. X₁+X₂+X₃+X₄=1 (141.95 g).

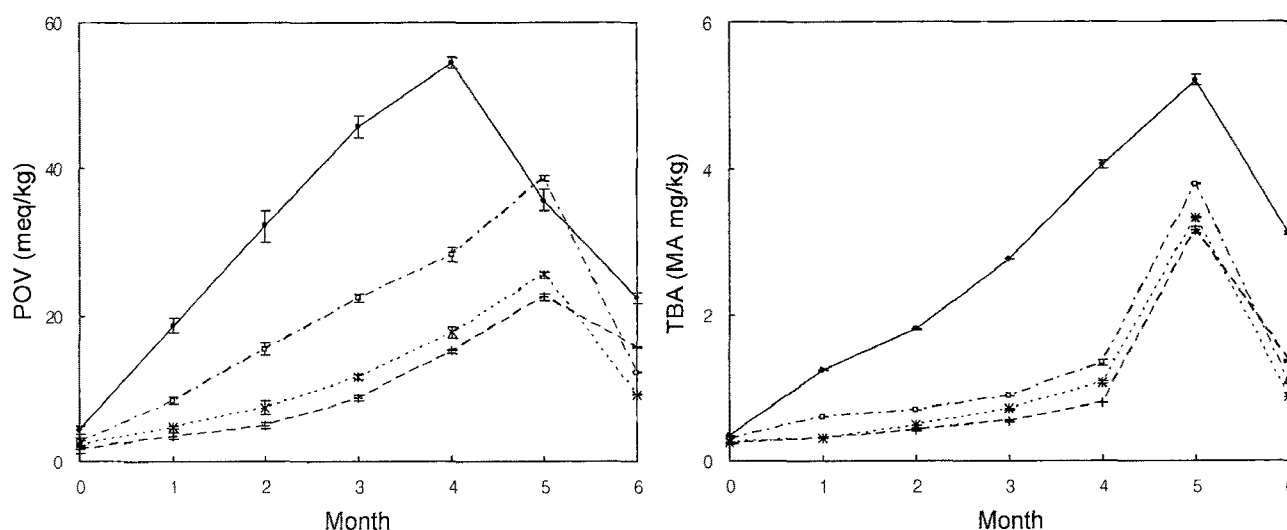


Fig. 2. Changes in peroxide value (POV) and thiobarbituric acid value (TBA) of seasoned pork samples during the storage at -25°C. —•— Raw pork; -□- control (soy sauce 12.06% added group); ...*... DG (*doenjang* 9% added group); ---+--- DOG (*doenjang* 9 and onion 33.4% added group). Values are means±SD, n=3.

doenjang instead of using soy sauce. As the similar trend as POV, the TBA value revealed significantly lower value in all types of seasoned pork than in the raw pork. The TBA value of raw pork at the 2nd month was 1.81±0.009

MA mg/kg and acutely increased up to 5.21±0.064 MA mg/kg at the 5th month. The control of seasoned pork, DG and DOG smooth increased at the 4th month by showing 1.34±0.036, 1.08±0.023, and 0.80±0.005 MA mg/kg,

respectively. However, the values were later decreased after showing the increase to 3.78 ± 0.116 , 3.34 ± 0.006 , and 3.16 ± 0.033 MA mg/kg, respectively, at the 5th month. The cause of POV and TBA value reduction is due to the initiation of peroxide decomposition into the secondary oxidation products (aldehydes, ketones, epoxydes, and carbonyl compounds) after 5th month as the oxidation proceeded (33,34). It was thought that inhibition of lipid oxidation in control, DG, and DOG was caused by not only the antioxidative products in soybean itself, but also the increase of Maillard reaction products (MRPs), amino acids, peptides, and aglycone of isoflavone through the fermentation procedures of soy sauce and *doenjang* (31, 35,36). By replacing some of the soy sauce with *doenjang*, the antioxidative capacity of the seasoned pork had strengthened more than before. It seemed that antioxidant substances of *doenjang* due to long-term fermentation and

ripening more produce than that of soy sauce (35). Moreover, replacing some of the water with onions could have stronger antioxidative synergistic effect than in case of seasoning sauce that was only added with *doenjang*. The present experiment results are supported by the report of Lee *et al.* (37) which cited the increase of antioxidative effect when the conventional antioxidants of butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA), and ascorbic acid were used together with soybean product extracts and by the report of Jurdi *et al.* (38) which reported the effective suppression of lipid oxidation during the refrigeration and freeze storage period of lamb patty by using onion juice (20%). Although the increase of added amount of *doenjang* and onion could increase the antioxidative capacity, the overuse of these ingredients could cause deterioration of sensory quality. So, the control of using the appropriate amount is necessary.

Table 3. Response of dependent variables to reactions for independent variables

Treatment No.	Independent variable ¹⁾				Dependent variable	
	X ₁	X ₂	X ₃	X ₄	Overall acceptability (Y ₁)	Taste preference (Y ₂)
1	0.1300	0.0700	0.3900	0.4100	5.70	5.90
2	0.1300	0.0200	0.3900	0.4600	5.85	5.80
3	0.1058	0.0725	0.3592	0.4625	6.60	6.80
4	0.1058	0.0975	0.3342	0.4625	6.25	6.10
5	0.0400	0.1200	0.2500	0.5900	6.70	6.35
6	0.0508	0.0975	0.3592	0.4925	5.35	5.50
7	0.0200	0.1200	0.2700	0.5900	5.35	5.45
8	0.0800	0.1200	0.3900	0.4100	7.40	7.10
9	0.0508	0.0475	0.3492	0.5525	5.00	5.30
10	0.1300	0.0300	0.2500	0.5900	5.75	5.50
11	0.0200	0.0200	0.3700	0.5900	2.90	3.50
12	0.1058	0.0525	0.2892	0.5525	6.00	5.95
13	0.0508	0.0475	0.3592	0.5425	5.40	4.80
14	0.0808	0.0975	0.3592	0.4625	5.85	5.60
15	0.0200	0.0200	0.3900	0.5700	4.80	4.20
16	0.1058	0.0475	0.3592	0.4875	5.70	5.80
17	0.0508	0.0975	0.2992	0.5525	5.20	5.75
18	0.1300	0.1200	0.2500	0.5000	6.30	6.20
19	0.0200	0.1200	0.3900	0.4700	6.30	6.50
20	0.0608	0.0975	0.2892	0.5525	7.20	7.10
21	0.1300	0.0200	0.2600	0.5900	5.30	5.00
22	0.1300	0.1200	0.3400	0.4100	5.95	5.85
23	0.1058	0.0975	0.2892	0.5075	7.35	7.25
24	0.0817	0.0750	0.3283	0.5150	6.80	6.25
25	0.1058	0.0475	0.2942	0.5525	3.10	3.10

¹⁾X₁, *Doenjang*; X₂, soy sauce; X₃, onion; X₄, water.

Table 4. Canonical equation for seasoned pork, *bulgogi* with addition of *doenjang*, soy sauce, onion, and water¹⁾

Attribute	Predicted model	R ²
Overall acceptability	$Y_1 = -295.04X_1 + 71.78X_2 + 96.12X_3 + 14.08X_4 + 280.82X_1X_2 + 158.51X_1X_3 + 419.56X_1X_4 - 277.31X_2X_3 + 3.01X_2X_4 - 183.96X_3X_4$	0.9860***
Taste preference	$Y_2 = -167.29X_1 - 16.79X_2 + 68.61X_3 + 8.46X_4 + 196.67X_1X_2 + 68.53X_1X_3 + 257.13X_1X_4 - 115.08X_2X_3 + 101.03X_2X_4 - 124.40X_3X_4$	0.9881***

¹⁾***Significant at $p < 0.001$, R² = coefficient of determination.

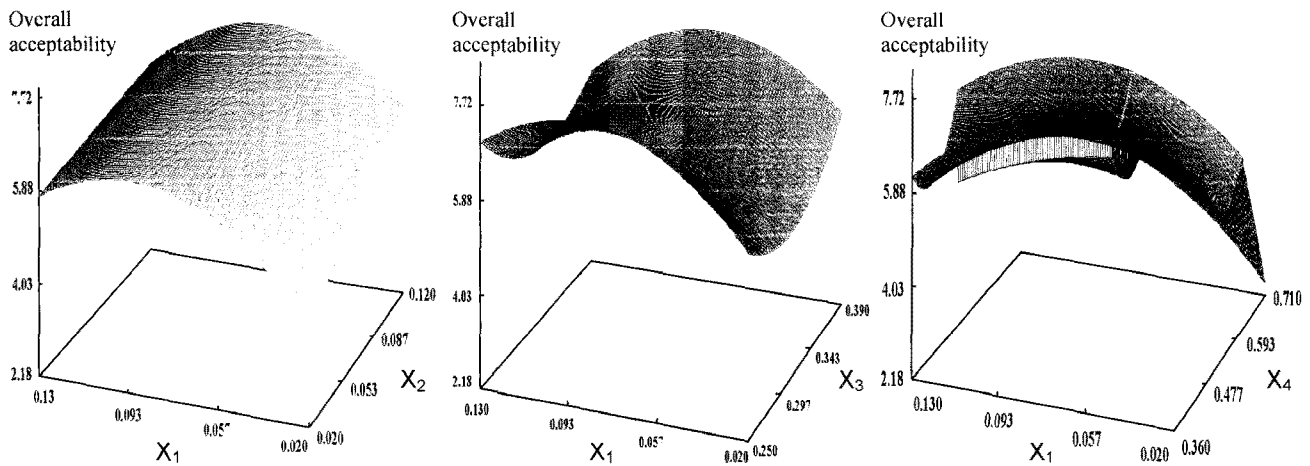


Fig. 3. Response surface plots for the effects of individual factors on the overall acceptability. X_1 , Doenjang; X_2 , soy sauce; X_3 , onion; X_4 , water.

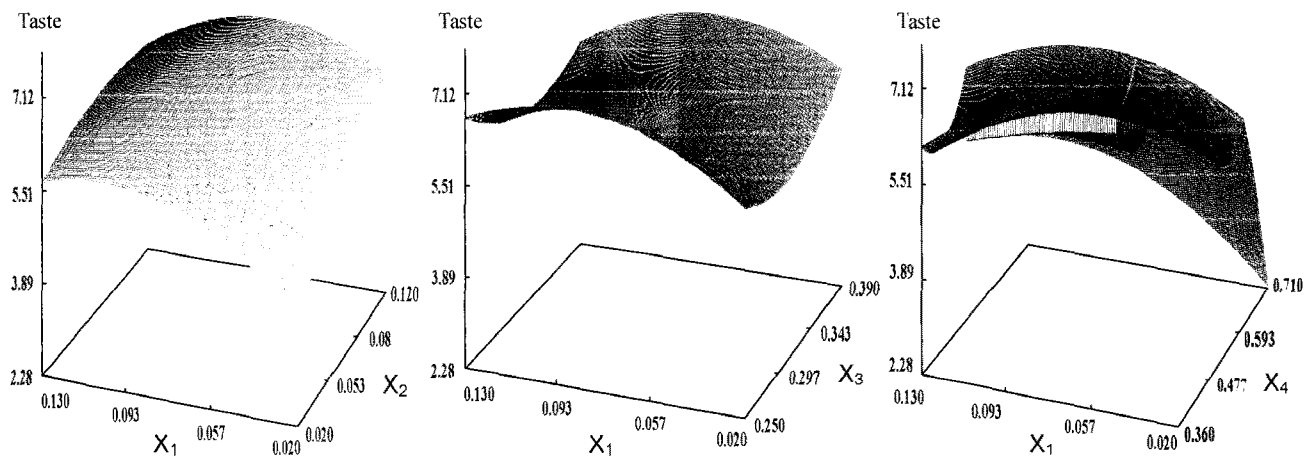


Fig. 4. Response surface plots for the effects of individual factors on the taste preference. X_1 , Doenjang; X_2 , soy sauce; X_3 , onion; X_4 , water.

Development of seasoning sauce for pork bulgogi using MED In the MED for following up the optimum mixture ratio of pork bulgogi by adding doenjang and onion, the response values designed in 25 experiment points could be listed as in Table 3. The scores of overall acceptability (Y_1) were ranged 2.90-7.40 and the taste preferences (Y_2) were ranged 3.10-7.10. The secondary regression model that fits to the overall acceptability and to the taste preference could be listed as in Table 4. The determination coefficients (R^2) for overall acceptability (Y_1) and taste preference (Y_2) resulted 0.9860 and 0.9881, respectively. The secondary regression model could be told to be well explained by the 4 experimental factors of doenjang (X_1), soy sauce (X_2), onion (X_3), and water (X_4). Based upon the appropriate canonical equation of Table 4, the response values for all available observation values were calculated by using the SAS/GRAPH program and the 3 dimensional response surface plots using fitted second order model were represented in Fig. 3 and 4. Seeing the contribution of ingredient mixture ratio in sensory evaluation from the response surface plot, both of overall acceptability and taste preference were increased as the mixture ratios of soy sauce (X_2) and onion (X_3) were increased, and the increased use of doenjang (X_1) eventually resulted to decrease the score after reaching the maximum score.

Namely, the mixture ratio of doenjang (X_1) and onion (X_3) are far more sensitive to taste preference and overall acceptability. Since the trend of taste preference and overall acceptability was similar, it is considered that the taste preference must have been largely affected on the overall acceptability.

Optimization of seasoning sauce The optimization region of ingredient mixture ratio is represented as in Fig. 5 and the white portion (overlap) of the plot is the optimum response region that simultaneously considered the both response values of overall acceptability and taste preference. Among the overlapped regions, the current study followed up the region that scored more than 7.2 in overall acceptability and more than 6.7 in taste preference, and the consequent result is listed as in Table 5. The optimum manufacturing conditions of seasoning sauce with addition of doenjang and onion which was acquired by the mixture experimental analyses of overall acceptability and taste preference of pork bulgogi was represented in Table 6. Namely, among the appropriate response values listed in Table 5, the optimum response condition of doenjang 0.09 (12.78 g), soy sauce 0.12 (17.03 g), onion 0.25 (35.49 g), and water 0.54 (76.65 g) was selected by showing the highest scores of overall acceptability and taste preference

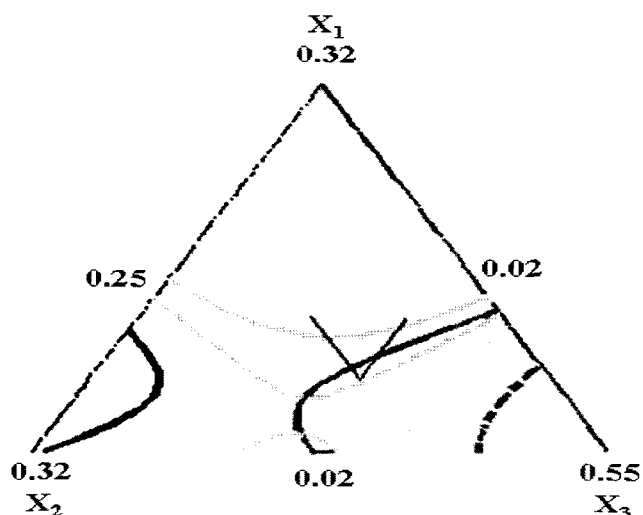


Fig. 5. Overlapping contour plot for overall acceptability and taste preference for blends containing *doenjang* (X_1), soy sauce (X_2), onion (X_3), and water (X_4). Overall acceptability; ----- 6.3, 7.5, taste preference; ——— 6.8, 9.0. Fixed value; $X_4=0.41$.

Table 5. Observation of response conditions for the optimization mixture¹⁾

X_1	X_2	X_3	X_4	Overall acceptability (Y_1)	Taste preference (Y_2)
0.08	0.10	0.25	0.57	7.20	6.74
0.08	0.10	0.39	0.43	7.20	6.89
0.08	0.11	0.25	0.56	7.45	6.93
0.08	0.11	0.39	0.42	7.32	6.95
0.08	0.12	0.25	0.55	7.69	7.10
0.08	0.12	0.26	0.54	7.44	6.93
0.08	0.12	0.27	0.53	7.21	6.78
0.08	0.12	0.38	0.42	7.21	6.83
0.08	0.12	0.39	0.41	7.43	6.99
0.09	0.10	0.25	0.56	7.26	6.80
0.09	0.11	0.25	0.55	7.49	6.97
0.09	0.11	0.26	0.54	7.23	6.80
0.09	0.11	0.39	0.41	7.25	6.90
0.09	0.12	0.25	0.54	7.72	7.12
0.09	0.12	0.26	0.53	7.46	6.94
0.09	0.12	0.27	0.52	7.23	6.79
0.09	0.12	0.38	0.41	7.35	6.92
0.10	0.10	0.25	0.55	7.23	6.80
0.10	0.11	0.25	0.54	7.45	6.95
0.10	0.12	0.25	0.53	7.67	7.09
0.10	0.12	0.26	0.52	7.39	6.91

¹⁾ X_1 , *Doenjang*; X_2 , soy sauce; X_3 , onion; X_4 , water.

by recording 7.72 and 7.12, respectively. From the basic mixture ratio, the follow up result (Table 7) of applying the above optimum seasoning sauce mixture ratio was found to be *doenjang* 7.10, soy sauce 9.46, onion 19.72, and water 42.58%. The actual values measured by conducting the acceptability tests for pork *bulgogi* that was manufactured by adopting the above optimum mixture ratio earned from

Table 6. Optimum conditions of seasoning sauce with addition of *doenjang*, soy sauce, onion, and water by using mixture experimental design

Component	Optimum proportion	Optimum amount (g)
<i>Doenjang</i> (X_1)	0.09	12.78
Soy sauce (X_2)	0.12	17.03
Onion (X_3)	0.25	35.49
Water (X_4)	0.54	76.65
Sum	1.00	141.95

Table 7. Optimum formula for seasoning sauce for pork *bulgogi* by using the mixture experimental design

Ingredient	Optimum level (%)
Sugar	9.96
Corn syrup	4.98
Garlic	2.49
<i>Soju</i> (alcohol)	3.23
Ginger	0.38
Black pepper	0.10
<i>Doenjang</i>	7.10
Soy sauce	9.46
Onion	19.72
Water	42.58

Table 8. Sensory evaluation of seasoned pork with addition of *doenjang*, soy sauce, onion, and water by mixture experimental design

	Predicted value ¹⁾	Actual value ²⁾
Overall acceptability	7.72	8.11±0.27
Taste preference	7.12	7.52±0.32

¹⁾Predicted score from mixture design.

²⁾Actual score obtained at optimum conditions from mixture design (n=20).

the MED were compared to the predicted values calculated by using canonical equation of Table 4, and the overall acceptability and taste preference of the actual values scored 8.11 and 7.52, respectively (Table 8). The scores were higher than the predicted values of 7.72 and 7.12, which enabled us to confirm that the above suggested mixture ratio of *doenjang*, soy sauce, onion, and water could be applied to the application in the seasoning sauce of pork *bulgogi* and the ratio was the appropriate mixture ratio.

In conclusion, the lipid oxidation stability of pork *bulgogi* was improved by the addition of *doenjang* and onion onto the seasoning sauce. The optimum mixture ratio of seasoning sauce for pork *bulgogi* followed up by the MED was found to be *doenjang* 7.10%, soy sauce 9.46%, onion 19.72%, and water 42.58%.

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References

- MAF. 2005 Statistics. Ministry of Agriculture and Forestry, Seoul, Korea. p. 330 (2006)
- Hah KH, Ahn CN, Joo ST, Park GB, Park KH, Kim IS. Effects of aging at low temperature on storage stability of seasoned pork. Korean J. Food Sci. Anim. Resour. 26: 85-91 (2006)
- Sung NK. Formation of cholesterol oxidation products in foods. Food Ind. Nutr. 5: 10-20 (2000)
- Park JG, Her JH, Li SY, Cho SH, Youn SK, Choi JS, Park SM, Ahn DH. Study on the improvement of storage property and quality in the traditional seasoning beef containing medicinal herb extracts. J. Korean Soc. Food Sci. Nutr. 34: 113-119 (2005)
- Jung IC, Moon YH, Kang SJ. Effects of addition of mugwort powder on the physicochemical and sensory characteristics of boiled pork. Korean J. Food Sci. Anim. Resour. 24: 15-22 (2004)
- Park JS, Lee MY, Lee TS. Compositions of sugars and fatty acids in soybean paste (*doenjang*) prepared with different microbial sources. J. Korean Soc. Food Nutr. 24: 917-924 (1995)
- Kim SH. New trends of studying on potential activities of *doenjang*, fibrinolytic activity. Korea Soybean Digest 15: 8-15 (1998)
- Cheigh HS, Park KS, Moon GS, Park KY. Antioxidative characteristics of fermented soybean paste and its extracts on the lipid oxidation. J. Korean Soc. Food Sci. Nutr. 19: 163-167 (1990)
- Moon GS, Cheigh HS. Antioxidative effect of soybean sauce on the lipid oxidation of cooked meat. Korean J. Food Sci. Technol. 18: 313-318 (1986)
- Moon GS, Cheigh HS. Antioxidative characteristics of soybean sauce in lipid oxidation process. Korean J. Food Sci. Technol. 19: 537-542 (1987)
- Cho SH, Park YM, Yoo YM, Chae HS, Wyi JJ, Ahn CN, Kim JH, Lee JM, Kim YK, Yun SG. Physico-chemical and sensory characteristics of pork *bulgogi* containing ginseng saponin. Korean J. Food Sci. Anim. Resour. 22: 30-36 (2002)
- Youn SK, Choi JS, Park SM, Ahn DH. Studies on the improvement of shelf-life and quality of vacuum-packaged seasoned pork meat by added chitosan during storage. J. Anim. Sci. Technol. Korea 46: 1024-1030 (2004)
- Han GJ, Shin DS, Kim JS, Cho YS, Jeong KS. Effects of propolis addition on quality characteristics of oriental medicinal seasoning pork. Korean J. Food Sci. Tech. 38: 75-81 (2006)
- Hertog MSL, Feskens FJM, Hollman PCM, Kormhout D. Dietary antioxidant flavonoids and risk of coronary heart disease. The Zutphen elderly study. Lancet 342: 1007-1011 (1993)
- Chung DO, Park YK. The study of soft drinks production and functional food in onions. Korean J. Soc. Food Sci. 15: 158-162 (1999)
- Woo HS, Aan BJ, Bae JH, Kim S, Choi HJ, Han HS, Choi C. Effect of biologically active fractions from onion on physiological activity and lipid metabolism. J. Korean Soc. Food Sci. Nutr. 32: 119-123 (2003)
- Mendes LC, Menezes HC, Aparecida M, Silva AP. Optimization of the roasting of robusta coffee (*C. canephora conillon*) using acceptability tests and RSM. Food Qual. Prefer. 12: 153-162 (2001)
- Nardi JV, Acchar W, Hotza D. Enhancing the properties of ceramic products through mixture design and response surface analysis. J. Eur. Ceram. Soc. 24: 375-379 (2004)
- Jop SCF, Silva RSF, Beleia AP. Formulation and evaluation of dry dessert mix containing sweetener combinations using mixture response methodology. Food Chem. 66: 167-171 (1999)
- Lee JS, Moon JW. A case study on the optimum formulation of coffee by a mixture experiment design. J. Industrial Tech. Kangwon National University. Korea. 22: 83-87 (2002)
- Jang MS, Park JE. Optimization of ingredient mixing ratio for preparation of *seolgitteok* with saltwort (*Salicornia herbacea* L.). J. Korean Soc. Food Sci. Nutr. 35: 641-648 (2006)
- Park HY, Jang MS. Ingredient mixing ratio optimization for the preparation of *seolgitteok* with barley (*Hordeum vulgare* L.) sprout powder. Korean J. Food Cook. Sci. 23: 551-560 (2007)
- Kim SS, Kim BY, Hahm YT, Shin DH. Least cost and optimum mixing programming by *yulmu* mixture noodle. Korean J. Food Sci. Technol. 31: 385-390 (1999)
- Yoo JH, Han GH. Analysis of optimal mixing ratios in tortilla preparations with rice and wheat flour. Korean J. Food Nutr. 20: 1-8 (2007)
- Silvia D, Dutcosky M, Victória E, Grossmann RS, Silva SF, Welsch AK. Combined sensory optimization of prebiotic cereal product using multicomponent mixture experiments. Food Chem. 98: 630-638 (2006)
- Yang B, Vickers Z. Optimization of cheddar cheese taste in model cheese systems. J. Food Sci. 69: S229-S236 (2004)
- AOAC. Official Methods of Analysis of AOAC Intl. 16th ed. Method 967.12, 969.16, 945.46. Association of Official Analytical Communities, Arlington, VA, USA (1990)
- Folch I, Lee M, Statney GHS. A simple method for the isolation and purification of total lipids from animal tissues. J. Biol. Chem. 226: 497-509 (1957)
- AOCS. Official Methods and Recommended Practices of the AOCS. 4th ed. Method Cd 8-53. American Oil Chemists' Society, Champaign, IL, USA (1960)
- Tarladgis BG, Watts BM, Younathan MT, Dugan L. A distillation method for the quantitative determination malonaldehyde in rancid foods. J. Am. Oil Chem. Soc. 37: 44-48 (1960)
- Shin DK, Lee YO. Antioxidative effect of seasoning on the lipid oxidation of *bulgogi* cooked meat. Korean Oil Chem. Soc. 7: 75-81 (1990)
- Park SY, Yoo JH, Uh JH, Eun JB, Lee HC, Kim YJ, Chin KB. Evaluation of lipid oxidation and oxidative products as affected by pork meat cut, packaging method, and storage time during frozen storage (-10°C). J. Food. Sci. 72: c114-c119 (2007)
- Tironi VA, Tomas MC, Anon MC. Structural and functional changes in myofibrillar proteins of sea salmon (*Pseudoperca semifasciata*) by interaction with malon aldehyde (RI). J. Food. Sci. 67: 930-935 (2002)
- Gokalp HY, Ockerman HW, Plimpton RF, Harper WJ. Fatty acids of neutral and phospholipids, rancidity scores and TBA values as influenced by packaging and storage. J. Food Sci. 48: 829-834 (1983)
- Park KY, Jung KO. Fermented soybean products as functional foods: Functional properties of *doenjang* (fermented soybean paste). pp. 555-596. In: Asian Functional Foods. CRC Press, Inc., Boca Raton, FL, USA (2005)
- Kim HJ, Sohn KH, Chae SH, Kwak TK, Yim SK. Brown color characteristics and antioxidizing activity of *doenjang* extracts. Korean J. Soc. Food Cook. Sci. 18: 644-654 (2002)
- Lee CH, Moon SY, Lee JC, Lee JY. Study on the antioxidant activity of soybean products extracts for application of animal products. Korean J. Food Sci. Anim. Resour. 24: 405-410 (2004)
- Jurdi HD, Mac-Neil JH, Yared DM. Antioxidant activity of onion and garlic juices in stored cooked ground lamb. J. Food Protect. 50: 411-413 (1987)