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Quality Characteristics of *Kochujang* Prepared with Different *Meju* Fermented with *Aspergillus* sp. and *Bacillus subtilis*

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Abstract For preparation of high quality *kochujang* by the traditional fermentation method, 4 types of *kochujang* were prepared with brick- or grain-shaped *meju* fermented with different strains (*Aspergillus sojae, Aspergillus oryzae+Bacillus subtilis*). After 100 days of fermentation at 25°C, the moisture, pH, salt, and ethanol content of *kochujang* were 40.52-43.20%, 4.71-4.82, 8.7-9.1%, and 0.75-0.94%, respectively, showing slight differences according to the strains and shapes of *meju*. Titratable acidities were slightly increased for up to 60 days of fermentation. The amino-type nitrogen content of *kochujang* prepared with brick-shaped *meju* (*A. oryzae+B. subtilis*) was the highest (164.20 mg%) among all of the *kochujang* types. The redness (a) value of *kochujang* prepared with brick-shaped *meju* (*A. sojae*) were higher (19.08) than those of other treatments (18.37-18.59). Sensory evaluation of *kochujang* prepared with grain-shaped *meju* (*A. sojae*) showed the highest scores for color and overall acceptability, at 6.43±1.87 and 6.29±1.44, respectively. It was estimated that high quality *kochujang* could be made by using *meju* fermented with selected strains.

Keywords: *kochujang, Aspergillus sojae, Aspergillus oryzae, Bacillus subtilis, meju* shape

Introduction

Kochujang is a hot, sweet, salty, and savory flavored condiment that has been used in Korean homes for a long time. Its main ingredients are soybean, *meju* powder (Korean-style soybean *koji*), and red pepper powder, and it is often mixed with malt-digested rice, barley, or sweet potatoes.

Kochujang is divided into traditional *kochujang*, which is made with *meju* and mass-produced *kochujang*, which is made with *Aspergillus oryzae*. The natural fermentation of *kochujang* is processed by microorganisms and enzymes produced in the red pepper or *meju*. Natural microorganisms are used in the traditional method of *kochujang* production (1). However, in the case of factory production, a *koji* that is a multiplication of *A. oryzae* is usually used. On the other hand, some factories apply combined use with *Bacillus* (2).

Kochujang quality depends on the combination ratio of ingredients, the processing methods, and the aging conditions, as well as the types of *meju* or *koji* (3). Many studies on the improvement of manufacturing methods and the selection of strains for *meju* preparation has been conducted; these studies have assessed the effects of *meju* shapes and strains on the quality of traditional *kochujang* and soy sauce (4-6), on the *kochujang* preparation by liquid *beni-koji* using *A. oryzae* (7), on the improvement of the quality of traditional *kochujang* by using a mixture of yeasts, including *Saccharomyces rouxii* (8), and on the preparation of low-salt *kochujang* by addition of mustard powder (9).

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Here, all products of *kochujang* have to be managed and standardized for the process of fermentation through the management of microorganisms. Therefore, the control of *meju* fermentation is important for the quality of *kochujang* (10). For the production of traditional *kochujang* with standardized and sanitary qualities, modern manufacturing processes utilize selected *Aspergillus* sp. and *Bacillus* sp., which have strong amylase and protease activities.

In the previous study (11), for the preparation of high quality *kochujang* by a traditional fermentation method, we created 8 types of *meju* (brick-shape, grain-shape) prepared with *A. oryzae* and *A. sojae* alone or in combination with *B. subtilis*. Four types of *meju*, brick- and grain-shaped *meju* fermented with *A. sojae* or *A. oryzae+B. subtilis*, were selected based on the physicochemical properties and enzyme activities during fermentation.

In this study, for investigation on the possibility of using the selected strains, with high protease and amylase activity, instead of traditional method in manufacturing of *kochujang meju*, we estimated the quality characteristics of *kochujang* prepared with brick- and grain-shaped *meju*, which were fermented with *A. sojae* and *A. oryzae+B. subtilis*, during fermentation.

Materials and Methods

Materials Non-glutinous rice 'Ilmi' produced in autumn 2005 in Buan (Jeonbuk, Korea) was used for the *koji* preparation. Soybean 'Taekwang' and glutinous rice 'Dongjinchalbye', also harvested in autumn 2005 in Sunchang (Jeonbuk, Korea), were used for the *meju* preparation. Red pepper powder 'Dabok', domestic sun-dried salts, and malt 'Olbory' for *kochujang* preparation were obtained from Sunchang Moonokrae Food Co. (Sunchang, Jeonbuk, Korea).

Microorganisms *A. oryzae* and *A. sojae* used for the *koji* preparation were obtained from the Hagyeong Fermentation Research Center (Suwon, Gyeonggi, Korea). The *B. subtilis* for the *meju* preparation was obtained from Daesang Foods Co. (Sunchang, Jeonbuk, Korea).

***Koji* preparation** Non-glutinous rice was soaked for 12 hr at 20°C, drained, and steam-heated for 30 min at 121°C. The *koji* was then cooled to approximately 30°C, inoculated with a 3%(v/w) spore suspension of either *A. oryzae* or *A. sojae*, and incubated for 5 days at 25°C (11). The spore suspension was made by homogenizing the spores with 10 mL of 0.1% peptone water containing 0.1% Tween 80 in the plate count agar medium.

***Meju* preparation** Four types of *meju* were prepared according to their physical shapes (brick or grain) and the 3 strains (*A. sojae* and *A. oryzae*+*B. subtilis*) used. Soybean were soaked in water for 9 hr at 20°C, allowed to drain for 1 hr, and steam-heated for 30 min at 121°C. The soybeans were then cooled to about 30°C and ground. The non-glutinous rice was soaked in water for 12 hr at 20°C and treated as described for the soybeans. For *meju* fermented with *A. sojae*, a mixture of steam-heated soybean and non-glutinous rice (6:4) was inoculated with 0.5%(v/w) of non-glutinous rice *koji* prepared with *A. sojae*, mixed, and molded in bamboo trays (18×10×3 cm). The soybean and non-glutinous rice mixture were ground for the preparation of brick-shaped *meju*, but not for grain-shaped *meju*. For *meju* fermented with *B. subtilis*, the mixture of steam-heated soybean and non-glutinous rice (6:4) was inoculated with 0.5%(v/w) *B. subtilis* culture medium together with 0.5%(v/w) non-glutinous rice *koji* prepared with *A. oryzae*, and was then mixed and molded in bamboo trays (18×10×3 cm). *Meju* fermented with *A. sojae* or *A. oryzae*+*B. subtilis* were fermented for 12 days at 28°C and dried for 3 days in a drying room. The *meju* was ground finely to pass a 425 μm sieve (Laboratory test sieve, Endecotts Ltd., London, England) for subsequent analysis (11).

***Kochujang* preparation** The mixing ratio of raw ingredients for preparation of *kochujang* according to the traditional methods used in the Sunchang area is shown in Table 1. Glutinous rice was soaked in water for 24 hr at 20°C, ground, mixed with malt extract, saccharified for 1.5 hr at 60°C and then filtered. This solution was named *shikhye* and used in the *kochujang* preparation. Treatments were divided into 5 groups depending on the physical shapes of *meju* and the strains used. Control samples were prepared with *meju* which was used for the preparation of Sunchang traditional *kochujang*. Six kg of each sample were used to fill a polyethylene bag, put in a polypropylene container, and sealed using a string. *Kochujang* was fermented for 100 days at 25°C in an incubator and samples were analyzed at intervals of 20 days.

Chemical analysis and color *Kochujang* (5 g) mixed with 45 mL distilled water was prepared to measure the pH of the *kochujang* with a pH meter (Orion SA520; Orion Research Inc., Beverly, MA, USA) (12). The titratable acidity was presented as the titration volume in mL of 0.1 N NaOH needed to bring the pH to 8.3 after homogenization, filtering and a 10-fold dilution of the *kochujang* (13). Moisture content was determined by the 105°C drying method (14). Salinity was measured using a salt meter (T-30D; Takemura Electric Works, Ltd., Tokyo, Japan), and color was expressed as L (lightness), a (redness), b (yellowness), and ΔE $[(L_0-L_1)^2+(a_0-a_1)^2+(b_1-b_0)^2]^{1/2}$ according to the Hunter scale, as determined using a colorimeter (Color and color difference, TC-360; Tokyo Denshoku Co., Tokyo, Japan).

Amino-type nitrogen *Kochujang* (5 g) was mixed with 25 mL of distilled water and shaken for 1 hr, and the pH was adjusted to 8.4 using 0.1 N NaOH. Twenty mL of neutral formalin (pH 8.3) was added to the mixture as above, and the pH was again adjusted to 8.4 using 0.1 N NaOH. The final titrated volume was used to calculate the amino-type nitrogen content; distilled water was used as the test blank (15).

Ethanol content Distilled water was added to 10 g of *kochujang* in order to adjust the volume to 25 mL and this mixture was then filtered using filter paper (Whatman No. 2, Whatman Int., Ltd., Maidstone, Kent, England) and Sep-Pak® Plus C_{18} cartridges (Waters Co., Milford, MA, USA). The filtrate was used for ethanol analysis by a high

Table 1. The mixing ratio (%, w/w) of raw ingredients for the *kochujang* preparation

Shape	Strains	Raw materials (%)						
		Red pepper powder	Traditional *meju* powder[1)]	*Meju* powder[2)]	*Meju* powder[3)]	Salt	Glutinous rice	Water[4)]
Control		27.8	8.9	0	0	12.5	23.8	27.0
Brick	A.s[5)]	27.8	0	8.9	0	12.5	23.8	27.0
	A.o+B.s[6)]	27.8	0	0	8.9	12.5	23.8	27.0
Grain	A.s	27.8	0	8.9	0	12.5	23.8	27.0
	A.o+B.s	27.8	0	0	8.9	12.5	23.8	27.0

1)*Meju* powder prepared by the traditional method in the Sunchang region.
2)*Meju* powder fermented with *A. sojae*.
3)*Meju* powder fermented with *A. oryzae*+*B. subtilis*.
4)Mixture of malt extract and water.
5)*A. sojae*.
6)*A. oryzae*+*B. subtilis*.

performance liquid chromatography (HPLC) (NSG-Series; Futecs, Daejeon, Korea) equipped with a Water® high performance carbohydrate column and an reflectance index (RI) detector (Waters 410 detector; Waters Co.). Acetonitrile : water (75:25, v/v) was used as the mobile phase at room temperature. A volume of 15 μL of sample was injected for each run (16).

Sensory evaluation Graduate students (n=14) received an orientation about the purpose of the test and the evaluation method prior to the evaluation. The samples were evaluated using a 9-point hedonic scale ranging from 'like extremely' (scale-9) to 'dislike extremely' (scale-1). Using the above numeric scaling system, the mean scores for each of these samples were obtained (17,18).

Statistical analysis All statistical analyses were performed using SAS (Statistical Analysis System) (19). Mean standard deviations were calculated, and Duncan's multiple range tests were applied. All experiments were performed in triplicate. A probability (p) level of 0.05 was considered to indicate significance.

Results and Discussion

Moisture contents Changes in moisture content during the fermentation of *kochujang* prepared with different *meju* are shown in Fig. 1.

Moisture contents in *kochujang* were slightly increased, by 0.36-1.17% from 40.32-42.03% at the initial stage to 40.52-43.20% after a 100-day fermentation period. Choi *et al.* (20) presumed that the moisture content in *kochujang* during fermentation was increased because macromolecules in *kochujang* were hydrolyzed by the actions of microorganisms and free water eluted to the outside. In addition, they estimated that the moisture contents of treatments were different because of difference in enzyme activities and rheological properties. In our study, the moisture contents were slightly increased compare to those of Lee *et al.* (21) and Park *et al.* (22), who reported that moisture contents in *kochujang* were increased by 5% after fermentation using different methods of preparation.

pH and titratable acidity The pH and titratable acidity of *kochujang* were changed by organic acids produced by various microorganisms during fermentation.

Changes in pH during the fermentation of *kochujang* prepared with various *meju* are shown in Fig. 2. The pH in *kochujang* were slightly decreased, by 0.04-1.14 from pH 4.81-4.92 at the initial stage to pH 4.71-4.82 after a 100-day fermentation period. The pH of *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae* was the lowest, with a pH of 4.71, but little difference was observed between the treatments. The results of this study were consistent with those of Lee *et al.* (23), who reported that the pH of *kochujang* was below pH 5.0 after the initial stage of fermentation.

Changes in titratable acidity during the fermentation of *kochujang* prepared with various *meju* are shown in Fig. 3. Titratable acidities in *kochujang* were slightly increased for up to 60 days of fermentation and decreased or were maintained thereafter. The change in the titratable acidity of *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae* was the highest, going from 16.77 mL at the initial stage to 21.87 mL at 100 days of fermentation, consistent with the lowest pH noted above. The reduction of the pH and increase of titratable acidities in *kochujang* at the initial stage of fermentation are caused by organic acids produced by raw materials and the metabolic action of microorganisms during fermentation. After 80 days of fermentation, however, titratable acidities were decrease or maintenance despite the reduction of the pH; this is caused by the utilization of produced organic acids for formation of volatile compounds, including ester (24,25). Major organic acids in *kochujang* were known as succinic acid, citric acid, and malic acid (16) or citric acid, and lactic acid (24). In addition, oxalic acid, formic acid, and acetic acid

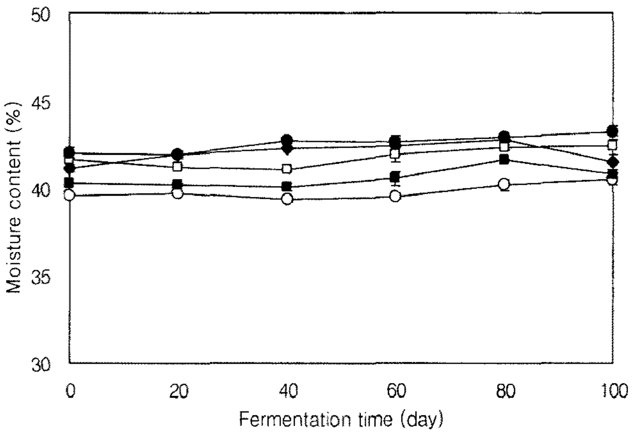

Fig. 1. Moisture content of *kochujang* prepared with different *meju* during fermentation at 25°C. -◆-, Control; -○-, *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae;* -□-, *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae*; -●-, *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis*;, -■-, *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis.*

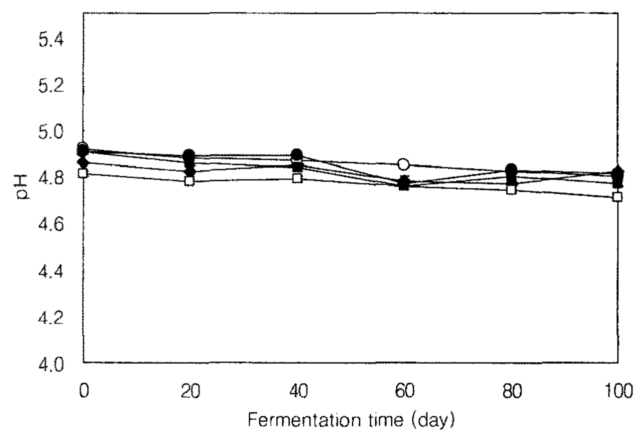

Fig. 2. pH of *kochujang* prepared with different *meju* during fermentation at 25°C. -◆-, Control; -○-, *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae;* -□-, *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae*; -●-, *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis*;. -■-, *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis.*

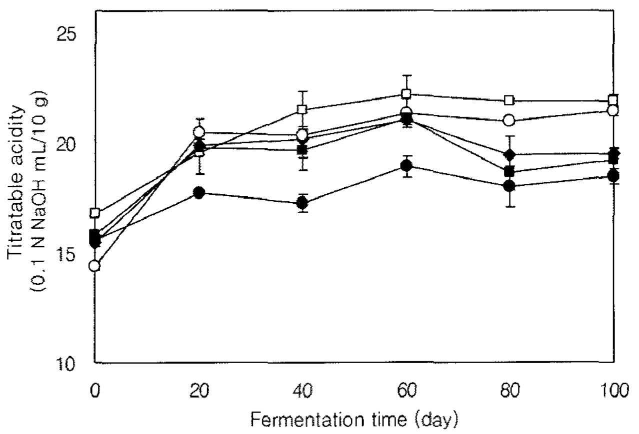

Fig. 3. Titratable acidity of *kochujang* prepared with different *meju* during fermentation at 25°C. -◆-, Control; -○-, *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae;* -□-, *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae*; -●-, *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis;*, -■-, *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis.*

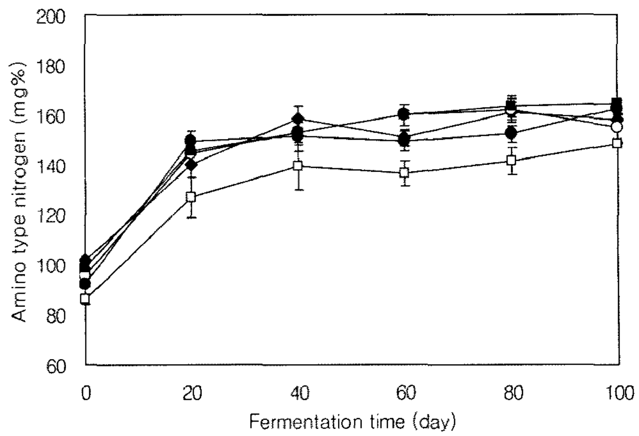

Fig. 5. Amino-type nitrogen content of *kochujang* prepared with different *meju* during fermentation at 25°C. -◆-, Control; -○-, *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae;* -□-, *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae*; -●-, *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis;*, -■-, *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis.*

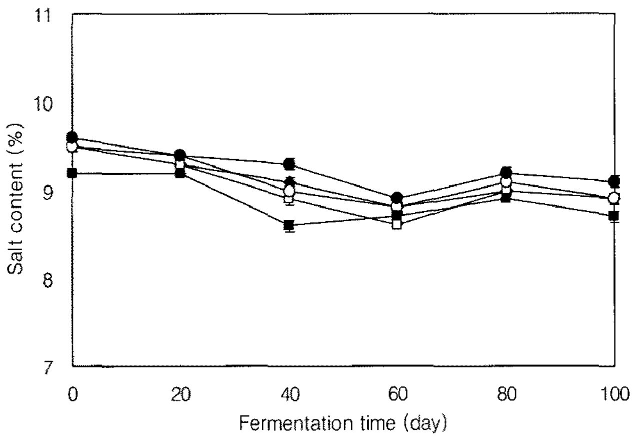

Fig. 4. Salt content of *kochujang* prepared with different *meju* during fermentation at 25°C. -◆-, Control; -○-, *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae;* -□-, *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae*; -●-, *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis;*, -■-, *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis.*

were identified as minor organic acids in *kochujang* (16).

Our results were similar to the results of Lee *et al.* (26), who reported that the pH of traditional *kochujang* decreased for up to 70 days of aging, while the titratable acidity increased for up to 60 days of aging, and decreased thereafter.

Salt contents Changes in salt content during the fermentation of *kochujang* prepared with different shaped *meju* fermented with various strains of microflora are shown in Fig. 4. The salt contents in *kochujang* were slightly decreased, by 0.5-0.6% from 9.2-9.6% at the initial stage to 8.7-9.1% after a 100 day of fermentation period. The salt content of *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis* was the lowest, from 9.2% at the initial stage to 8.7% at 100 days of fermentation. No differences were observed among the other treatments.

Our results were lower than those of Shin (27), who reported that the salt contents of *kochujang* produced in the Gangwon and Geonggi, Chungcheong, Jeonbuk, Jeonnam, and Gyeongsang regions in Korea were 19.65±2.66, 21.01±9.39, 15.20±5.67, 13.35±4.69, and 8.92±5.16%, respectively. Kim (28) reported that the salt contents of *kochujang* of the Sunchang (Jeonbuk, Korea), Boeun (Jeungbuk, Korea), and Sacheon (Gyeongnam, Korea) regions were 9.1, 6.6, and 6.3%, respectively. The salt contents of our *kochujang*, which was prepared according to the Sunchang standard recipe, were similar to that of Kim (28) and higher than that of Kim *et al.* (29).

Amino-type nitrogen contents The amino-type nitrogen content presents the degree of hydrolysis of soy protein by microorganisms, and is related to the content of free amino acid, which affects the tastes of soy products.

Figure 5 shows changes in amino-type nitrogen content during fermentation of *kochujang* prepared with various *meju*. The amino-type nitrogen contents of *kochujang* in all treatments were drastically increased, from 85.96-101.88 mg% at the initial stage to 126.74-149.38 mg% of 20 days of fermentation, and gradually increased thereafter. At 100 days of fermentation, the amino-type nitrogen content of *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis* was the highest (164.20 mg%), while that of *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae* was the lowest (148.42 mg%). In addition, the amino-type nitrogen content of *kochujang* prepared with *meju* fermented with *A. oryzae* and *B. subtilis* was slightly higher (162.40-164.20 mg%) than those of kochujang produced with *A. sojae* alone (148.42-154.66 mg%). Our results were similar to those of Park *et al.* (22), who reported that amino-type nitrogen contents of *kochujang* were gradually increased for up to

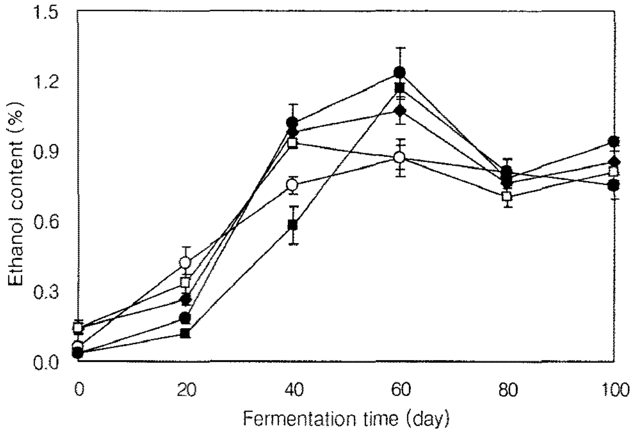

Fig. 6. Ethanol content of *kochujang* prepared with different *meju* during fermentation at 25°C. -◆-, Control; -○-, *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae;* -□-, *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae*; -●-, *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis;*, -■-, *kochujang* prepared with brick-shaped *meju* fermented with *A. oryzae* and *B. subtilis.*

60 days of fermentation. No differences were observed among the treatments.

Oh (16) and Lee *et al.* (30) were reported that major amino acids in *kochujang* were glutamic acid, proline, alanine, aspartic acid, and lysine.

Ethanol contents Changes in ethanol content during the fermentation of *kochujang* prepared with various *meju* are shown in Fig. 6. Ethanol contents of *kochujang* drastically increased for up to 40 days of fermentation, showing the highest contents (0.87-1.23%) at 60 days of fermentation, and decreasing to 0.75-0.94% at 100 days of fermentation. Among the treatments, the ethanol contents of *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae* and *B. subtilis* were the highest (1.23%) at 60 days of fermentation and at 100 days of fermentation (0.94%). Our results were slightly lower than the 1.25% (31) and 1.98% (32) reported for traditional *kochujang* in Jeolla-do regions (Korea), the 1.42% observed at 150 days of aging in traditional glutinous rice *kochujang* (33), and the 1.1% seen at 120 days of aging in Sunchang traditional *kochujang* (28).

The ethanol in *kochujang* was produced by *Zygosaccharomyces rouxii,* a salt-tolerant yeast, after an increase of reducing sugar and a decrease of the pH, and was maintained at a level of 2%. Ethanol provides *kochujang* with flavoring and preservation through esterification (2,34).

Color The color of *kochujang* was an important factor in sensory evaluation by consumers, and may be used as the standard of quality evaluation for *kochujang* (35).

Changes in color during the fermentation of *kochujang* prepared with various *meju* are shown in Table 2. The lightness (L) of *kochujang* did not changed from the initial stage to 100 days of fermentation, starting at 19.76-20.57 and ending at 19.45-21.16, respectively, with no differences among the treatments. The redness (a) gradually increased, from 16.01-16.61 at the initial stage to 18.37-19.08 at 100 days of fermentation. The redness value was the highest (19.08) for *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae.* The yellowness (b) in all treatments was slightly increased for up to 100 days of fermentation. Color difference with control group (ΔE) was gradually increased; from 0.04-0.38 at the initial stage to 3.61-8.80 at 100 days of fermentation, and the difference in *kochujang* prepared with brick-shaped *meju* fermented with *A. sojae* were the highest (8.80).

Table 2. Color difference value of some *kochujang* prepared with different *meju* during fermentation at 25°C

Shape	Strains[1)]	Hunter value[2)]	Fermentation time (day)					
			0	20	40	60	80	100
Brick	A.s	L	20.07	20.07	19.78	19.05	20.88	21.16
		a	16.01	16.47	18.41	18.50	18.62	19.08
		b	11.34	11.46	12.00	11.24	11.65	12.82
		ΔE	0.38	0.02	3.63	4.89	4.70	8.80
	A.o+B.s	L	19.76	19.47	19.39	19.29	19.37	19.45
		a	16.52	16.21	17.84	17.45	17.84	18.37
		b	11.42	11.35	11.71	11.78	11.23	11.57
		ΔE	0.15	0.60	2.12	1.47	2.23	3.61
Grain	A.s	L	20.01	20.09	19.78	19.27	19.58	19.48
		a	16.61	16.77	15.97	15.99	18.22	18.54
		b	11.40	11.34	11.46	11.57	11.89	11.88
		ΔE	0.04	0.08	0.50	1.08	3.06	4.30
	A.o+B.s	L	20.57	20.84	20.22	20.42	20.64	20.88
		a	16.21	17.46	16.68	17.83	18.40	18.59
		b	11.44	11.80	11.89	12.61	12.49	12.68
		ΔE	0.36	1.34	0.12	2.70	4.41	5.80

[1)]*A.s, meju* powder fermented with *A. sojae;* A.o+B.s, *meju* powder fermented with *A. oryzae+B. subtilis.*
[2)]L, lightness; a, redness; b, yellowness; $\Delta E = [(\Delta L)^2 + (\Delta a)^2 + (\Delta b)^2]^{1/2}$.

Table 3. Sensory evaluation of *kochujang* prepared with different *meju* during fermentation at 25°C[1)]

Items	Shape				
	Control	Brick[2)]		Grain	
		A.s	A.o+B.s	A.s	A.o+B.s
Flavor	5.57±1.22[a]	5.07±2.24[a]	5.00±1.75[a]	4.50±2.38[a]	4.50±1.79[a]
Color	4.86±2.28[b]	4.36±1.60[b]	4.50±1.91[b]	6.43±1.87[a]	5.71±1.64[ab]
Sweet taste	4.86±1.99[a]	2.93±1.94[c]	3.07±1.73[bc]	4.71±2.56[ab]	3.64±2.47[abc]
Sour taste	3.43±1.60[a]	3.57±2.14[a]	3.57±2.10[a]	3.71±2.30[a]	3.71±1.98[a]
Salty taste	4.86±1.83[a]	4.86±1.96[a]	5.21±2.49[a]	5.21±2.08[a]	5.00±2.39[a]
Bitter taste	3.29±2.46[a]	3.86±1.56[a]	4.29±2.34[a]	4.00±1.84[a]	4.86±1.96[a]
Pungency	5.71±2.02[a]	5.50±1.29[a]	5.79±1.85[a]	6.07±1.77[a]	4.93±2.24[a]
Overall acceptability	4.86±1.41[b]	5.00±1.57[ab]	4.93±1.69[ab]	6.29±1.44[a]	4.64±2.41[b]

[1)]Means with the same lowercase letters within rows were not significantly different at $p<0.05$.
[2)]*A.s, meju* powder fermented with *A. sojae;* A.o+B.s, *meju* powder fermented with *A. oryzae+B. subtilis.*

Color values in this study were higher than those of other reports, with L, a, and b values of Sunchang traditional *kochujang* being 14.88±1.20, 20.75±1.88, and 9.20±1.11 (36), and 14.49±1.44, 15.45±1.77, and 8.34±1.02 (37). Changes in color during storage varied in separate investigations (7,35). These variations may result from differences in cultivars and drying methods of red pepper, and ratios of red pepper powder and fermentation periods in *kochujang* preparation.

Sensory evaluation *Kochujang* prepared with various *meju* were assessed by sensory evaluated after fermentation at 25°C for 100 day (Table 3).

No significant differences in flavor, pungency, sour, salty, and bitter taste were observed among the treatments. However, in terms of the flavor scores of the control group, the *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae* had slightly higher scores for sour taste, salty taste, and pungency, and the *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae+B. subtilis* had slightly higher scores for sour and bitter tastes, but these differences were not significantly different from those of other treatments. High score for sour taste of *kochujang* prepared with grain-shaped *meju* fermented with *A. oryzae+B. subtilis* were consistent with the results of titratable acidity (Fig. 3). Sweet taste showed the highest score (4.86±1.99) in the control group and this difference was significant. The color and overall acceptability of *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae* shows the highest significant scores, 6.43±1.87 and 6.29±1.44, respectively, among the treatments. Results on overall acceptability of *kochujang* prepared with grain-shaped *meju* fermented with *A. sojae* were consistent with the results of amino-type nitrogen contents, which affect the tastes of soy products (Fig. 5).

In this study, the possibility of using the selected strains, with high protease and amylase activity, instead of traditional method in manufacturing of *kochujang meju* were investigated. Therefore, it was estimated that *kochujang* prepared with *meju* fermented with different strains (*A. sojae, A. oryzae+B. subtilis*) had better quality characteristics than those of the control group made by the traditional method, and could be used to prepare *kochujang* with high quality.

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