Thesis for the Degree of Master of Science

Shelf-life of Bottled Sea-squirt Meat (*Halocynthia roretzi*) Products Packed in Vegetable Oil

By

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Department of Food and Life Science

The Graduate School

Pukyong National University

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우렁쉥이 (멍게) 기름 담금 병조림 제조와 저장성

Advisor: Professor Hong-Soo Ryu

By

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Table of Contents

Lists of Tables

Lists of Figures

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Abstract

The sea squirt (*Halocynthiaroretzi*), is a famous cultured mollusk consumed as a raw fresh meat or fermented products in Korea and Japan with a unique flavor and texture. Fresh sea-squirt meat needs a modified processing and preservation process due to its short shelf-life from its high moisture content and proteolytic enzyme activity. In this study, bottled sea squirt meat prepared in vegetable oil (BSMO) to enhance the consumer acceptability and ɣ-ray (Co60, 10KGy/h) irradiation was tried to extend shelf-life without heating process. Response surface methodology was used in determining the optimal-mixing ratio of BSMO using 5% dehydrated fresh meat. Texture analysis and nutritional evaluation were also performed on control and BSMO. The VBN (volatile basic nitrogen) content and viable cell count were determined in order to find out the shelf-life of irradiated BSMO products during chilled storage at 4℃ for 60 days. The optimal mixing formulation was 80g meat in 60mL of mixed vegetable oil (30mL of olive oil and 30mL of sesame oil) by the trained 10 of twenties panelists. The highest rated formulation, by trained 9 of panelists over thirties, was 80g meat in 60mL mixed vegetable oil (42mL of olive oil and 18mL of sesame oil). Moisture, ash, and protein contents in BSMO were not changed significantly ($p<0.01$) compared with control. It did have a higher lipid content due to added vegetable oil on the surface of BSMO ($p<0.01$) from 0.84±0.23 to 2.13±0.61. The added vegetable oil raised the hardness, springiness, cohesiveness, gumminess, chewiness and resilience of BSMO. BSMO products were available during 50 days of storage at 4℃ based on VBN content (BSMO 1: 27.92±0.96 mg/100g, BSMO 2: 24.84±1.95 mg/100g) and viable cell count (BSMO 1: 4.60±0.80 log CFU/mg, BSMO 2: 3.65±0.20 log CFU/mg) when compared with standard levels of VBN (25.00 mg/100g) and viable cell count (5 log CFU/mg) respectively. Results showed that irradiated BSMO products could contribute in expanding the processed seafood market and raising the familiarity of seafood to the younger generations.

Ⅰ. Introduction

As income levels have been increased, the processed food consumption trends have been also changed from a higher temperature sterilized or preservatives added products to fresh or semi prepared food using packaging and storaging techniques. Fish market is no exception to this trend and the market volume of prepared or freshly processed sea food products have been growing and its production has increased. As the edible invertebrates and mollusks such as sea squirt, its meat structure is so tender that are easily damaged mechanically and attacked by microbial organisms enough to drop those freshness fast and spoils easily compared to other sea food meat. Therefore, it is very difficult to manufacture them for fresh processed food. However, those animal sea food resource play important role as excellent sources of nutrients including protein, taste compounds, bio-active substance, vitamins and minerals (Fisheries Economic Institute, 2008)

According to phylogenetic systematics, sea squirt having unique characteristics between vertebrates and invertebrates belongs to protochordate and urochorda. As representative Korean cultured sea food, the importance of sea squirt has been so emphasized as developed ocean agriculture technology and expanded farming ground. A lot of researches and studies on sea squirt have been conducted for the chemical ingredients and taste components of sea squirt (Lee et al, 1993), and precusor and production mechanism of scent compounds (Fujimoto K et al, 1982). However, a higher moisture content in sea squirt meat (80% above) can result its freshness quickly and brown meat discoloration by strong enzyme activity. Due to those componants characteristics, it is hard to be manufactured as processed food. In addition, most of its harvesting is done from late spring and summer, and thus it need an advanced research and studies on new techniques for food process and preservation.

Moisture content and water activity have a great role in food freshness and microorganism growth. If moisture content of sea squirt meat could be reduced, those must have lots of advantages in decreasing freshness and potentials in extending shelf-life. In this study, we used 5% dehydrated sea squirt meat by expressing technique, and added sesame oil and olive oil in order to have a higher consumer's acceptability and anaerobic condition for bottled products.

The smell of Korean style sesame oil, which is roasted and hot pressured, is aromatic and shows severe rancidity term (Nam and Chung, 2008) But sesamol of solvent extracted sesame oil is effective in inhibiting oxidization of linolenic acid (Lee and Choe, 2008). It is also reported that sesame oil inhibits automatic oxidization by reducing radical chain reaction of oxygenated radicals (Yoshida and Takagi, 1999). It is known that olive oil is effective in reducing cholesterol level and against heart disease, arteriosclerosis and stomach disorder (Choi, 2006).

Thus, it could be predicted that mixed sesame oil and olive oil with sea squirt meat would be effective against rancidity to improve storage stability of sea squirt meat and reduce instinct smell of such meat.

It is basically important to find out the best optimizing and standardizing mixture of such oils and sea squirt meat based on the consumer's acceptability for the bottled products. For the first step of such product, accurate statistical analysis model should be applied to test planning method for proper mixture (Naes et al, 1999; Saguy et al, 1984; Han, 2003) and Response Surface Methodology (RSM) may be used for this step for optimization. Response surface methodology which is actually used for the first step for mixture food optimization referred to an statistical analysis method about the response-surface resulted from response variables when independent variable (X) interacts with and influences on dependent variable (Y) (Giovanni, 1983; Mudahar et al, 1989). Variables causing response is independent variables or factors and variables responding is defendant variables showing certain responses (Lee et al, 2000; Seo et al, 1983). The outcomes from this response surface methodology are: firstly, it can be assumed meaningful effects and functional relation between independent variables and dependant variables so that it could predict the changes according to the variation of those variables; and, secondly, the level of independent variables could enable the maximum desirable independent variables (Cornell, 1990; Ellkaer et al, 1996). Therefore response surface methodology can be usefully applied for food mixture optimization for bottled food production. In particular, in case of various ingredient mixture, great information can be gained with the minimum test trials through proper mixture test planning. Many of researches and studies were tried on the optimal mixing ratio using this response surface methodology are the reports about steamed white rice cake (Hong et al, 1999; Kim et al, 1999; Lee et al, 1999), pine nut porridge (Jang et al, 2003), steamed bun with red bean paste filling (Oh et al, 2002), and red pepper jam (Park et al, 2000). Those reports were to gain data for optimization of amount of ingredient and cooking condition. However, research and studies on new sea food design are not sufficient.

Sea food processing is made up with such various ingredients that the researches on the mixing ratio optimization of test planning must be conducted before processing. Also, it is very important that they should focus on the consumer's tastes (Fishken, 1983).

Thus, in this study, central composite design of mixture test planning was tried to find the best ingredient mixing ratio focusing on consumer's tastes to gain basic data for sea squirt meat products. To develop proper model for each generation (Prinyawiwatkul et al, 1983) consumer's taste and evaluations were carried on various perception about values, behavior pattern and time value of each generation (Park et al, 2002). In case of sea squirt meat, most of younger generation do not like it because of its instinct fishy smell and thus it is urgent for us to develop products suitable for new generation.

Response surface methodology of mixture test planning was tried to find the best ingredient mixture rate based on consumer's acceptability. In order to gain the basic data for improving nutrition and storage stability of oil based bottle product of sea squirt meat, it is divided testers group as group 1 and group 2, and each generation and conducted consumer taste survey and texture analysis. Through the nutritional evaluation and storage stability test of bottled products of sea squirt meat made by optimal mixing ratio, it was checked whether response surface methodology could be applied to sea food mixing ratio optimization.

Ⅱ. Materials and Method

2.1 Sample preparation

Live sea squirts (155±11g of weight in average) samples were purchased and exuviated in Namcheon Seafood Market, Busan city. Alter delivering sea squirt meat in cooler (4℃) within 5 min from market, meat samples were dehydrated by pressing with kichen towel immediately, and prepared vacuumized pack (70g and 80g). Vacuum packed samples were frozen at -20±5℃ and those frozen samples were used further experiments. Sterilized bottles and irradiated frozen sea squirt meat samples without viscera were used in preparing the bottled products to extend its shelf-life. Irradiation was performed ɣ-ray(Co60) 10KGy/h of absorbed dose by Advanced Radiation Technology Institute in Jeongeup, Korea. Sterilization was performed at 121℃ for 15 minutes using Autoclave (HB-506-4, Han Baek Scientific Co, Korea)

2.2 Experiment plan for respose surface methodology (RSM)

Response surface methodology (RSM) was used to study the effects of three independent variables (Table 1). The three factors are expressed using X_1, X_2 and X_3 respectively. Effects of Independent variables: [X_1]

ratio of olive oil and sesame oil, [X_2] amount of sea squirt meat, [X_3] amount of added total oil (Ryu et al. 2004).

2.3 Bottled sea squirt meat in oil (BSMO) preparation

Various amount of sea quirt meat, complying with the ratios determined by the central composite design showed in Table 1 and 2, were mixed with olive oil and sesame oil according to the ratios in proportionate to the total amount.

2.4 Sensory evaluation

Sensory evaluation was carried out to optimize the best bottled sea squirt meat. The sensory panelist were divided into two types for group 1 (10 twenties) and group 2 (9 over thirties). Evaluation was 9 grade method (1 point is very bad, 5 point is normal, 9 point very good), with hedonic scale of questionnaire test on color, flavor, taste, texture, overall acceptability. Fresh drinking water was provided when they evaluate the samples, they had to rinse the their mouth using water prior to begin other samples evaluation. Because same surrounding was difficult about every evaluation, balanced evaluation was performed by the block for two days.

Table 1. Independent variable and their levels for central composite design.

Independent variable	Symbol	Coded variable levels				
		-2	-1	0	1	2
Oilve vs Sesame	X_1	4:6	5:5	7:3	8:2	6:4
Sea squirt(g)	X_2	60	70	80	90	100
Oil(ml)	X_3	40	50	60	70	80

X_1 : Ratio of oilve oil and Sesame oil X_2 : Amount of sea squirt meat
X_3 : Added total oil

Table 2. Central composite design arrangement and variable levels.

Exp. no	Variable levels		
	X_1	X_2	X_3
1	−1	−1	−1
2	0	−1	−1
3	1	−1	−1
4	0	−1	0
5	1	−1	0
6	−1	0	−1
7	−1	0	−1
8	0	0	−1
9	1	0	−1
10	0	0	0
11	1	0	0
12	0	0	1
13	−1	1	−1
14	0	1	−1
15	1	1	−1
16	0	1	0
17	1	1	0
18	0	1	1
19	−2	0	0
20	2	0	0
21	0	−2	0
22	−1	2	0
23	−1	0	−2
24	0	0	2

X_1 : Ratio of oilve oil and Sesame oil X_2 : Amount of sea squirt meat
X_3 : Added total oil

2.5 Nutritional evaluation

After the sensory evaluation, the best bottled sea squirt meat products, raw sea squirt meats and dehydrated meats was used in every experiment.

2.5.1 Proximate composition

The proximate composition were experimented with raw and freeze dried samples by AOAC (1990) procedure. Moisture was determined by oven-drying (J-DS4 JISICO) at 105℃ until constant weight. Crude ash was determined by dry ashing method in muffle furnace (KUKJE SCIEN HY-8000S series). Crude fat was determined using the Soxhlet solvent extractor (VELP SCIENTIFICA SER 148). Crude protein was determined by the semi-micro kjeldahl method using Kjeldahl apparatus (Gerhardt Vapodest 30). The carbohydrate contents were calculated by the difference of proximate composition, those were calculated except moisture, crude ash, crude fat, crude protein.

2.5.2 Water activity measurement

Water activity measurements were taken for all the samples, using the water activity-measuring equipment (BT-RSI-7557 012, Switzerland).

2.5.3 Texture analysis

Texture profile analysis (TPA) was performed using texture analyser (TA-XT2i, Stable Microsystem, England) according to the method of Kim and Lee (1999). Before the testing hardness, fracturability, adhesiveness, springiness, cohesiveness, gumminess, chewiness and resilience, sea squirt meats were cleaned with paper towel and cut into 2.0 × 2.0cm sections. The sample were pressed twice at 50%, using 3.0cm cylinder probe (P/3) at a test speed 1.0mm/s (Table4).

2.5.4 Volatile basic nitrogen (VBN) measurement

Volatile basic nitrogen (VBN) was measured using Conway micro diffusion method (KFDA, 2002). 2g samples was added to 16 ml of distilled water and 2 ml of 20% trichloro acetic acid, stirred for 2 minutes using a homogenizer and then filterated. The remaining 1 ml of sample was put into the outer chamber of a Conway dish 1 ml of 0.01N H_3BO_3 into the inner chamber and 1ml of saturated K_2CO_3 was quickly poured into the outter chamber. Then sample were mixed K_2CO_3 in the outter chamber and left at 37℃ for 80 minutes. After the reaction, 0.02N HCl was dripped into the dish to take a measurement. To check saving time for sample , VBN was measured for 50 days.

VBN(mg%) = 0.14 × [(Sample titration amount - Blank titration amount)] × Factor × 100 / Sample(g)

2.5.5 Viable cell count measurement

The method is to mix the clinical material with standard agar medium so that it is coagulated in order to culture bacteria and then aggregate the number of bacteria colony count to calculate the viable cell count: prepare more than two sterilized petri-plates; and then pour 1ml of test solution and 1ml of 10 times diluted solution into each of those plates; spray 15ml of sterilized standard agar medium, whose temperature has been maintained in 45℃; spin and tilt those plates from side to side while being careful not to make those medium attached to the plate cover; then, mix well the clinical material with standard agar medium until it is coagulated; then, make those plates upside down and culture bacteria in the 35~37℃ incubator for 24~48 hours; and, lastly, aggregate the number of bacteria colony count generated (KFDA, 2002).

2.5.6 *In vitro* digestibility

The *In vitro* digestibility values of all the samples were determined by the Satterlee (1979) method with modification by the AOAC procedure (AOAC, 1982), the procedure used four enzymes method. Oduro et al (2011) tried the three enzyme method, determined the correlation coefficient between two assays (R^2=0.9955). The α-chymotrypsin (Sigma

41 units/mg solid, trypsin (Sigma 17,600 BAEE units/mg solid), protease (Streptomyces griceus, Sigma 46 units/mg solid) were used in the three enzymes method. The reference protein used was ANRC casein and digestibility was calculated as follows:

%Digestibility (three enzymes) = 234.84 - 22.56x where x is the pH of sample at 20 minutes

%Digestibility (four enzymes) = 1.03x (three enzymes digestibility) - 0.34

2.6 Statistical analysis

The sensory evaluation results was performed using Minitab by Respose Surface Methodology, other data were was performed by followed Duncan's multiple range test using analysis of variance (one way ANOVA), expressed as mean±S.D differences among treatments. The significance of results was at 5%, used was SPSS (version 18.0)

Table 3. Operation condition of texture analyzer for sea squirt meat.

Mode	Measure force in compression
Option	TPA
sample size (cm)	2.0×2.0×2.5
Pre-test speed (mm/s)	1.5
test speed (mm/s)	1.0
post-test speed (mm/s)	1.0
distance (%)	50
time (sec)	3.0
trigger type (g)	Auto-10g
probe	3.0cm

Ⅲ. Result and Discussion

1. Sensory evaluation

1.1. Sensory evaluation by group 1 panel (10 of twenties)

Sensory evaluation performed in this study was 9 grade method (1 point is very bad, 5 point is normal, 9 point is very good), with hedonic scale of questionnaire test on color, flavor, taste, texture, overall acceptability by 10 of twenties (group 1).

According to the central composite design, bottled sea squirt meat were prepared using quadratic canonical polynomial model. The average scores of sensory evaluation on those products were shown in Table 4.

1.1.1 Color

Figure 1 showed surface plot and contour plot of color of bottled sea squirt meat by group 1 panel (10 of twenties). The response surface regression equation was as follows :

$$\hat{y}_1 = 6.16924 + 0.03546x_1 + 0.13016x_2 - 0.31094x_3 - 0.00898x_1^2 - 0.11744x_2^2 - 0.46050x_3^2 + 0.02704x_1 x_2 + 0.11034x_1 x_3 - 0.03492x_2 x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis, R^2 (coefficient of determination) was 0.1105. And as the result of the variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The clear difference in color between olive oil and sesame oil prior to mixing with sea squirt meat resulted a significant color difference between bottled sea squirt meat products. From results of contour plot and surface plot shown in Figure 1, the panel score of color was increased with increasing the ratio of olive oil. The higher panel score of color were checked when 80 or 90g of sea squirts meat and 50 or 60ml of vegetable oil used samples could have a higher panel score of color compared with other volume of samples.

1.1.2 Flavor

Figure 2 showed surface plot and contour plot of flavor of bottled sea squirt meat by group 1 panel (10 of twenties). The response surface regression equation as follows :

$$\hat{y}_2 = 6.86638 - 0.03615x_1 + 0.00681x_2 - 0.26052x_3 - 0.32265x_1^2 - 0.34068x_2^2 - 0.65342x_3^2 + 0.17480x_1x_2 + 0.52086x_1x_3 - 0.19659x_2x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

It was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil, and R^2 (coefficient of determination) was 0.2301. Roasted and pressing processes adopted in Korean style sesame oil processing result a strong flavor and those flavor could change the original flavor of materials mixed with it. Because of the reason mentioned above, the original sea smell of bottled sea squirt products was disappeared as the added amount of sesame oil was increased. As the result of sensory evaluation, the score of the treatment No. 13 appeared to be the highest value(7.2), and the treatment No. 24 appeared to be lowest value(3.5). From results of contour plot and surface plot shown in Figure 2, the panel score of flavor was increased with increasing the ratio of sesame oil. The higher panel score of flavor were checked when 80 or 90g of sea squirts meat and 50 or 60ml of vegetable oil used samples could have a higher panel score of flavor compared with other volume of samples.

1.1.3 Taste

Figure 3 showed surface plot and contour plot of taste of bottled sea squirt meat by group 1 panel (10 of twenties). The response surface regression equation was as follow :

$$\hat{y}_3 = 6.53235 + 0.08497x_1 - 0.15056x_2 - 0.23632x_3 - 0.19462x_1^2 - 0.40013x_2^2$$

$- 0.40599x_3^2 + 0.04034x_1 x_2 + 0.36069x_1 x_3 - 0.28778x_2 x_3$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on taste R^2 (coefficient of determination) was 0.1105. And as the result of the variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. Usually Korean style sesame oil was familiar with all of Korean generations including twenties. Therefore, Korean style sesame oil could change the taste of the mixture with it positively. Because of the reason mentioned above, the score of the treatment No. 13 appeared to be the highest value(6.8) and the treatment No. 22 and 24 appeared to be the lowest value(4.0). From results of contour plot and surface plot shown in Figure 3, 7:3 or 5:5 (olive oil : sesame oil) samples, and 80g of sea squirts meat and 60ml of vegetable oil used samples could have a higher panel score of taste compared with other samples.

1.1.4 Texture

Figure 4 showed surface plot and contour plot of texture of bottled sea squirt meat by group 1 panel (10 of twenties). The response surface regression equation was as follows :

$$\hat{y}_4 = 6.87258 - 0.06284x_1 - 0.06703x_2 - 0.16299x_3 - 0.09934x_1^2 - 0.14193x_2^2 - 0.36359x_3^2 + 0.26027x_1 x_2 + 0.48076x_1 x_3 - 0.03351x_2 x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on texture, R^2 (coefficient of determination) was 0.0625 and as the result of variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. Sensory scores of texture is closely related with taste and flavor, and the higher score of those parameter showed sample could have a higher scores of texture, respectively. Because of the reason mentioned above, the score of the treatment No. 23 appeared to be the highest value(7.2) and the treatment No. 24 appeared to be the lowest value(4.4). From results of contour plot and surface plot shown in Figure 4, 4:6 or 5:5 (olive oil : sesame oil) samples, and 60 or 70g of sea squirts meat and 50 or 60ml of vegetable oil used samples could have a higher panel score of texture compared with other samples.

1.1.5 Overall acceptability

Figure 5 showed surface plot and contour plot of overall acceptability of bottled sea squirt meat by group 1 panel (10 of twenties). The response surface regression equation was as follows :

$$\hat{y}_5 = 6.49143 + 0.00332x_1 - 0.14693x_2 - 0.18071x_3 - 0.13760x_1^2 - 0.38005x_2^2 - 0.41915x_3^2 + 0.13668x_1 x_2 + 0.49492x_1 x_3 - 0.27347x_2 x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on overall acceptability, R^2 (coefficient of determination) was 0.1103 and as the result of variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 13 appeared to be the highest value(6.6) and the treatment No. 24 appeared to be the lowest value(4.0). From results of contour plot and surface plot shown in Figure 5, the panel score of overall acceptability would be increased with increasing volume of olive oil. The higher panel score of overall acceptability were checked when 70 or 80g of sea squirts meat and 60ml of vegetable oil used samples could have a higher panel score of overall acceptability compared with other volume of samples.

As the result of variance analysis on the color, flavor, texture, taste and overall acceptability, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 13 appeared to be the highest value(6.68) and the treatment No. 24 appeared to be the lowest value(3.8). From results of optimization curve shown in Figure 6, 5:5 (olive oil : sesame oil) samples, and 80g of sea squirts meat and 60ml of vegetable oil used samples could have a higher panel score of color, flavor, taste, texture, overall acceptability compared with other samples.

1.2 Sensory evaluation by group 2 panel (9 of more thirties)

Sensory evaluation performed in this study was 9 grade method (1 point is very bad, 5 point is normal, 9 point is very good), with hedonic scale of questionnaire test on color, flavor, taste, texture, overall acceptability by 9 of more thirties (group 2).

According to the central composite design, bottled sea squirt meat were prepared using quadratic canonical polynomial model. The average scores of sensory evaluation on those products were shown in Table 5.

1.2.1 Color

Figure 7 showed surface plot and contour plot of color of bottled sea squirt meat by group 2 panel (9 of more thirties). The response surface regression equation was as follows :

$$\hat{y}_1 = 8.70619 + 0.03116x_1 + 0.17044x_2 - 0.32860x_3 - 0.99486x_1^2 - 0.65409x_2^2 - 0.80908x_3^2 - 0.07537x_1 x_2 - 0.19041x_1 x_3 - 0.09533x_2 x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on color, R^2 (coefficient of determination) was 0.4118. And as the result of the variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. Like the result of group 1 panel (10 of twenties), the clear difference in color between olive oil and sesame oil prior to mixing with sea squirt meat resulted a significant color difference between bottled sea squirt meat products. The score of the treatment No. 11 and 12 appeared to be the highest value(7.9) and the treatment No. 19 appeared to be the lowest value(4.1). From results of contour plot and surface plot shown in Figure 7, 5:5 or 7:3 (olive oil : sesame oil) samples, and 70, 80 and 90g of sea squirts meat, and 50, 60 and 70ml of vegetable oil used samples could have a higher panel score of color compared with other samples.

1.2.2 Flavor

Figure 8 showed surface plot and contour plot of flavor of bottled sea squirt meat by group 2 panel (9 of more thirties). The response surface regression equation was as follow :

$$\hat{y}_2 = 6.85385 - 0.00137x_1 + 0.05721x_2 - 0.35602x_3 - 0.37006x_1^2 - 0.09867x_2^2$$

$- 0.35597x_3^2 - 0.15935x_1 x_2 - 0.07771x_1 x_3 + 0.18138x_2 x_3$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on flavor, R^2 (coefficient of determination) was 0.0657. And as the result of the variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 2 appeared to be the highest value(7.1) and the treatment No. 24 appeared to be the lowest value(4.7). From results of contour plot and surface plot shown in Figure 8, 5:5, 7:3 and 8:2 (olive oil : sesame oil) were showed the highest ratio of vegetable oil and 60, 70 and 80ml of vegetable oil used samples could have a higher panel score of flavor compared with other samples. But the weight sea squirt meat did not result a significant much difference between samples.

1.2.3 Taste

Figure 9 showed surface plot and contour plot of taste of bottled sea squirt meat by group 2 panel (9 of more thirties). The response surface regression equation was as follows :

$$\hat{y}_3 = 6.80113 - 0.06053x_1 + 0.07903x_2 - 0.43141x_3 - 0.30525x_1^2 - 0.10346x_2^2 - 0.14984x_3^2 - 0.20013x_1 x_2 - 0.14474x_1 x_3 + 0.15063x_2 x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on taste, R^2 (coefficient of determination) was 0.0364. And as the result of the variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 2 appeared to be the highest value(7.6) and the treatment No. 19 appeared to be the lowest value(5.3). From results of contour plot and surface plot shown in Figure 9, 7:3 and 8:2 (olive oil : sesame oil) samples, and 70 or 80g of sea squirts meat used samples could have a higher panel score of taste compared with other samples. But the volume of vegetable oil did not result a significant much difference between samples.

1.2.4 Texture

Figure 10 showed surface plot and contour plot of texture of bottled sea squirt meat by group 2 panel (9 of more thirties). The response surface regression equation was as follows :

$$\hat{y}_4 = 8.12042 - 0.05427x_1 + 0.20185x_2 - 0.43421x_3 - 0.62969x_1^2 - 0.28105x_2^2 - 0.31877x_3^2 - 0.06436x_1x_2 - 0.36023x_1x_3 - 0.02408x_2x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on texture, R^2 (coefficient of determination) was 0.2033 and as the result of variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 15 appeared to be the highest value(8.6) and the treatment No. 19 appeared to be the lowest value(5.1). From results of contour plot and surface plot shown in Figure 10, 7:3 (olive oil : sesame oil) samples, and 80 or 90g of sea squirts meat and 50 or 60ml of vegetable oil used samples could give a higher panel score of texture compared with other samples.

1.2.5 Overall acceptability

Figure 11 showed surface plot and contour plot of overall acceptability of bottled sea squirt meat by group 2 panel (9 of more thirties). The response surface regression equation was as follows :

$$\hat{y}_5 = 7.32344 - 0.12374x0 + 0.18147x_2 - 0.33903x_3 - 0.38120x_1^2 - 0.17553x_2^2 - 0.21920x_3^{\ 2} - 0.01316x_1\ x_2 - 0.28103x_1\ x_3 + 0.06296x_2\ x_3$$

where y : sensory evaluation score,

x_1 : ratio of olive oil and sesame oil,

x_2 : amount of sea squirt meat, x_3 : added total oil

As the result of regression analysis on overall acceptability, R^2 (coefficient of determination) was 0.0745. And as the result of the variance analysis, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 15 appeared to be the highest value(7.7) and the treatment No. 19 appeared to be the lowest value(5.4). From results of contour plot and surface plot shown in Figure 11, 7:3 (olive oil : sesame oil), and 40 or 50ml of vegetable oil used samples could have a higher panel score of overall acceptability compared with other samples. But the amount of sea squirt meat did not give much difference between samples.

As the result of variance analysis on the color, flavor, texture, taste and overall acceptability, it was appeared a significant difference ($p<0.01$) between the ratio of olive oil to sesame oil, and the weight of sea squirt meat and the total volume of vegetable oil. The score of the treatment No. 15 appeared to be the highest value(7.58) and the treatment No. 19 appeared to be the lowest value(5.0). From results of optimization curve shown in Figure 12, 7:3 (olive oil : sesame oil), and 80g of sea squirts meat and 60ml of vegetable oil used samples could have a higher panel score of color, flavor, taste, texture, overall acceptability compared with other samples.

Table 4. Central composite design response by group 1 panel (10 of twenties).

Exp.	Variable levels			Response				
no	X_1	X_2	X_3	Y_1	Y_2	Y_3	Y_4	Y_5
1	−1	−1	−1	5.6	6.5	5.9	7.0	6.4
2	0	−1	−1	5.5	5.7	5.8	6.2	5.4
3	1	−1	−1	5.7	5.0	5.3	5.8	5.1
4	0	−1	0	5.8	6.7	6.7	6.9	6.5
5	1	−1	0	6.0	5.6	6.1	5.8	5.9
6	−1	0	−1	5.6	5.6	6.0	6.7	6.1
7	−1	0	−1	5.7	6.6	5.9	6.8	6.0
8	0	0	−1	6.1	6.2	6.1	6.2	6.0
9	1	0	−1	5.9	5.9	6.3	6.7	6.0
10	0	0	0	6.3	6.3	6.2	6.7	6.2
11	1	0	0	5.9	6.8	6.0	6.7	6.2
12	0	0	1	6.0	6.6	6.4	6.9	6.2
13	−1	1	−1	5.5	7.2	6.8	6.9	7.0
14	0	1	−1	5.9	6.0	5.8	6.1	5.5
15	1	1	−1	6.1	5.8	5.7	6.0	5.7
16	0	1	0	6.4	6.5	5.8	7.0	6.1
17	1	1	0	6.2	6.0	6.0	6.5	5.7
18	0	1	1	5.5	5.8	5.6	6.7	5.8
19	−2	0	0	6.2	5.5	5.6	6.6	5.9
20	2	0	0	6.2	5.5	5.9	6.4	5.9
21	0	−2	0	5.4	5.7	5.2	6.7	5.3
22	−1	2	0	6.0	4.6	4.0	5.4	3.9
23	−1	0	−2	5.8	5.4	6.0	7.2	6.2
24	0	0	2	3.1	3.5	4.0	4.4	4.0

X_1 : Ratio of oilve oil and Sesame oil X_2 : Amount of sea squirt meat
X_3 : Added total oil Y_1 : Average score of color Y_2 : Average score of flavor Y_3 : Average score of taste Y_4 : Average score of texture
Y_5 : Average score of overall acceptability

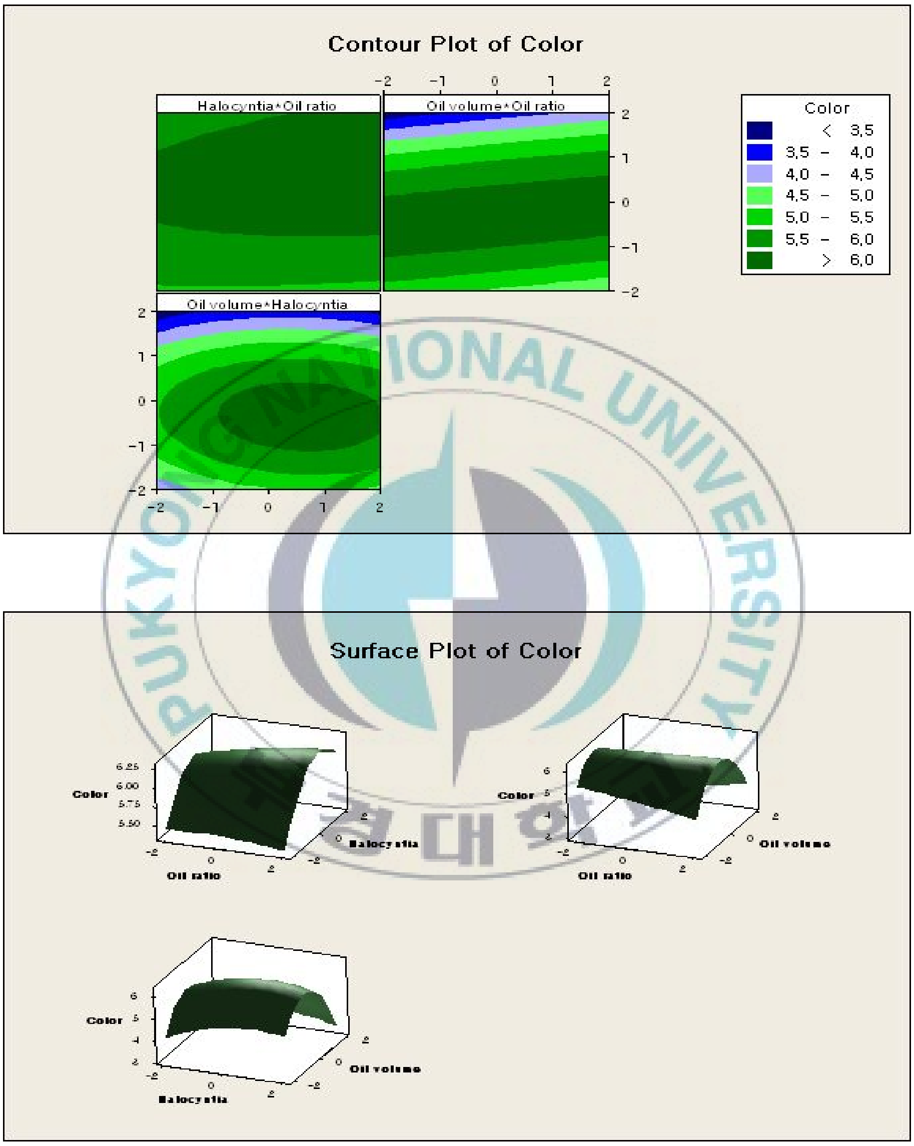


Fig. 1. Surface plot and contour plot of color of bottled sea squirt meat by group 1 panel (10 of twenties).

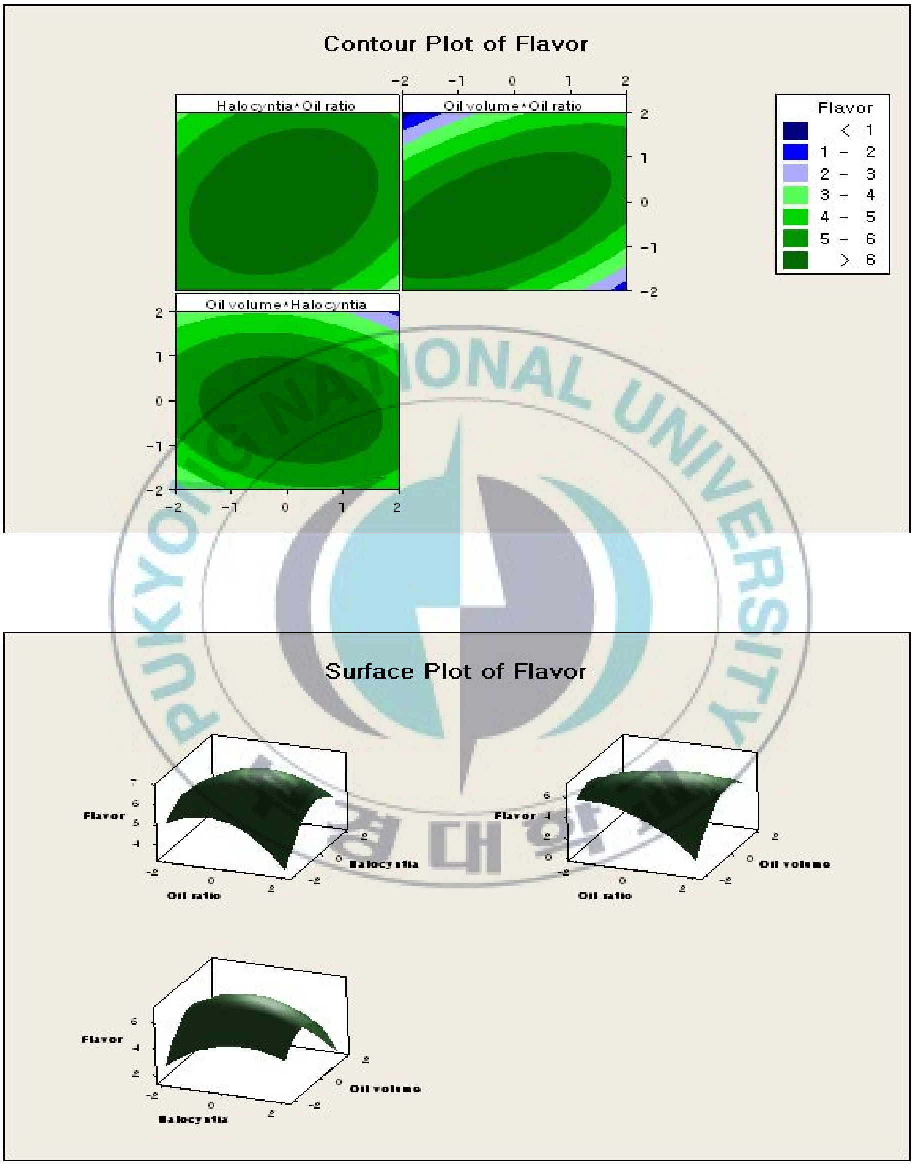


Fig. 2. Surface plot and contour plot of flavor of bottled sea squirt meat by group 1 panel (10 of twenties).

Fig. 3. Surface plot and contour plot of taste of bottled sea squirt meat by group 1 panel (10 of twenties).

Fig. 4. Surface plot and contour plot of texture of bottled sea squirt meat by group 1 panel (10 of twenties).

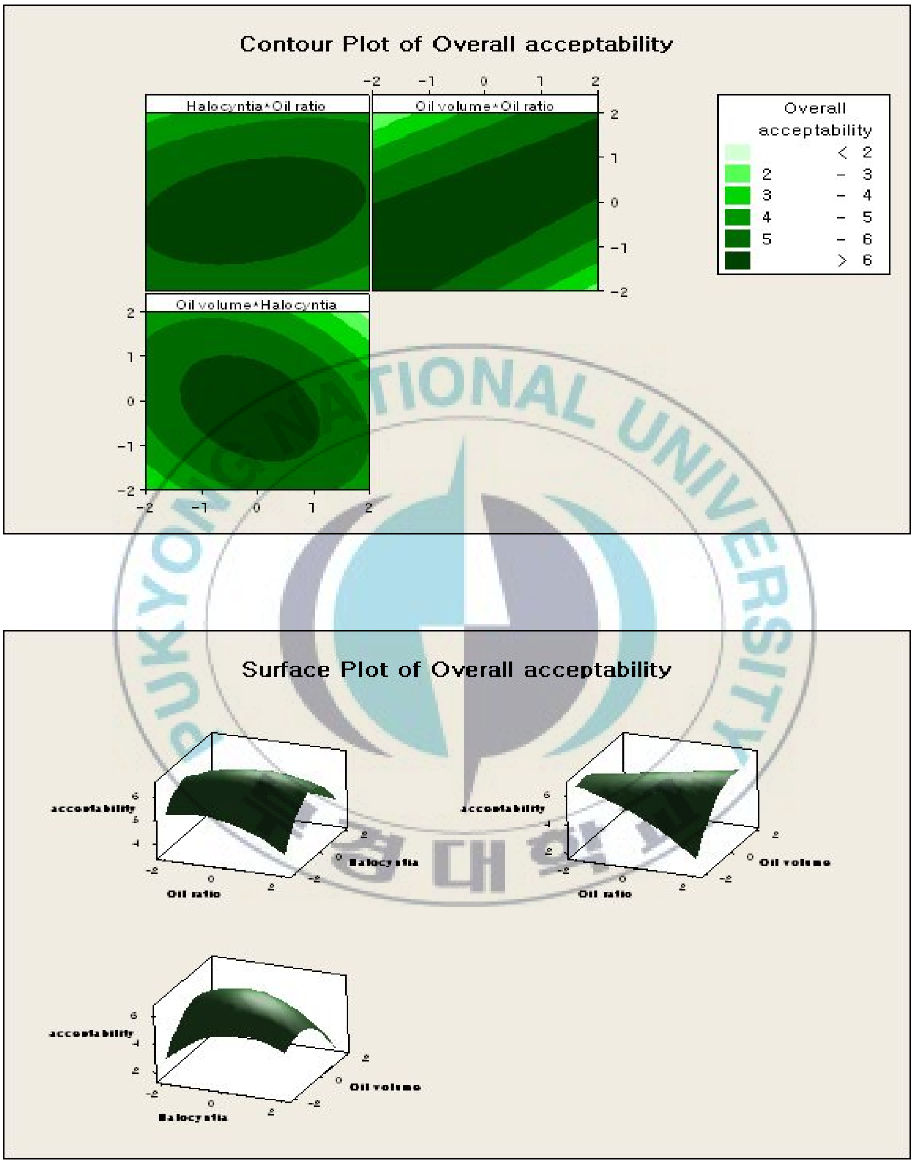


Fig. 5. Surface plot and contour plot of overall acceptability of bottled sea squirt meat by group 1 panel (10 of twenties).

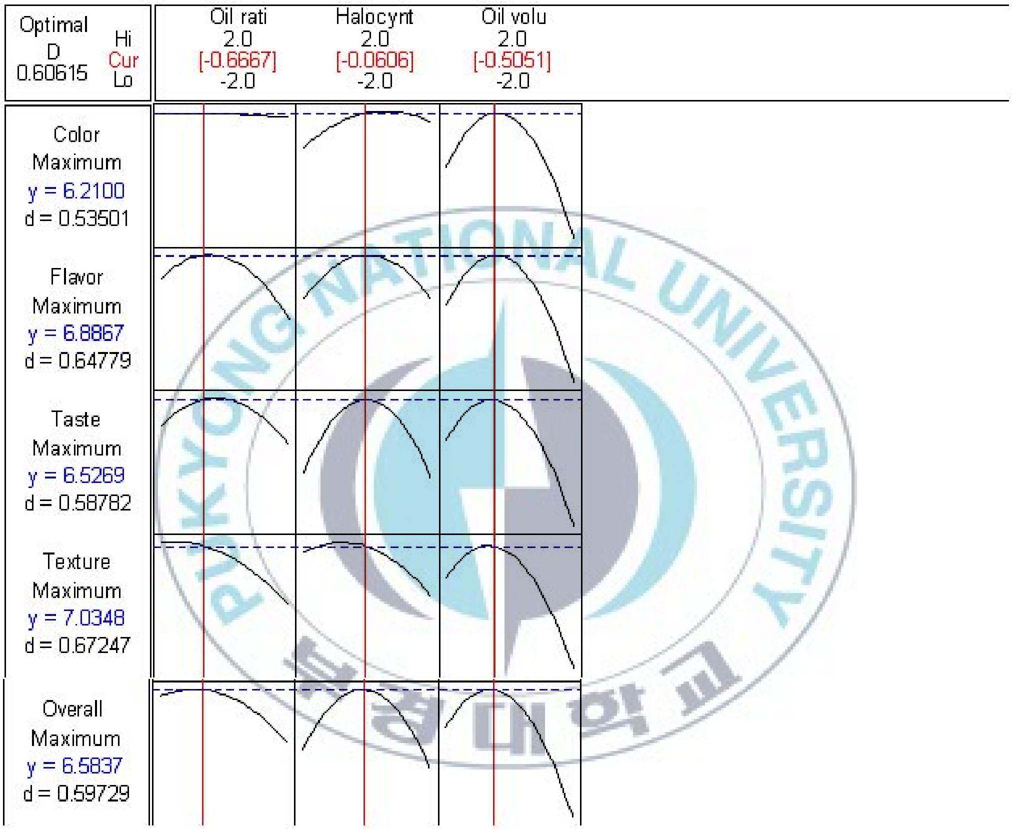


Fig. 6. Optimization curve for the sensory attributes of bottled sea squirt meat by group 1 panel (10 of twenties).

Table 5. Central composite design response by group 2 panel (9 of more thirties).

Exp. no	Variable levels X_1	X_2	X_3	Response Y_1	Y_2	Y_3	Y_4	Y_5
1	−1	−1	−1	7.1	7.0	7.1	8.0	7.6
2	0	−1	−1	7.7	7.1	7.6	7.9	6.9
3	1	−1	−1	7.2	6.8	6.9	7.8	7.2
4	0	−1	0	7.8	6.7	6.2	6.7	7.0
5	1	−1	0	7.2	6.9	6.3	7.1	6.1
6	−1	0	−1	7.7	6.1	6.4	8.0	7.0
7	−1	0	−1	7.7	6.7	7.1	7.8	7.4
8	0	0	−1	7.4	6.6	6.9	7.4	7.1
9	1	0	−1	6.8	6.2	6.7	7.7	7.0
10	0	0	0	7.7	6.3	6.8	8.2	7.2
11	1	0	0	7.9	6.4	6.9	7.6	6.8
12	0	0	1	7.9	6.2	5.9	7.4	7.2
13	−1	1	−1	7.8	6.6	6.8	7.7	6.6
14	0	1	−1	7.6	6.9	6.9	8.1	7.3
15	1	1	−1	8.0	6.7	6.9	8.6	7.7
16	0	1	0	7.6	6.6	6.6	7.8	7.4
17	1	1	0	6.8	5.8	5.6	6.3	5.9
18	0	1	1	7.6	6.6	6.3	7.7	6.7
19	−2	0	0	4.1	5.1	5.3	5.1	5.4
20	2	0	0	4.7	5.4	5.7	5.7	6.0
21	0	−2	0	4.8	5.8	5.8	5.9	5.7
22	−1	2	0	5.4	6.6	7.0	7.1	7.2
23	−1	0	−2	3.8	5.1	6.0	5.8	6.0
24	0	0	2	4.6	4.7	5.4	5.7	5.6

X_1 : Oilve oil and Sesame oil　X_2 : Amount of sea squirt
X_3 : Added total oil
Y_1 : Average score of color　Y_2 : Average score of flavor
Y_3 : Average score of taste　Y_4 : Average score of texture
Y_5 : Average score of overall acceptability

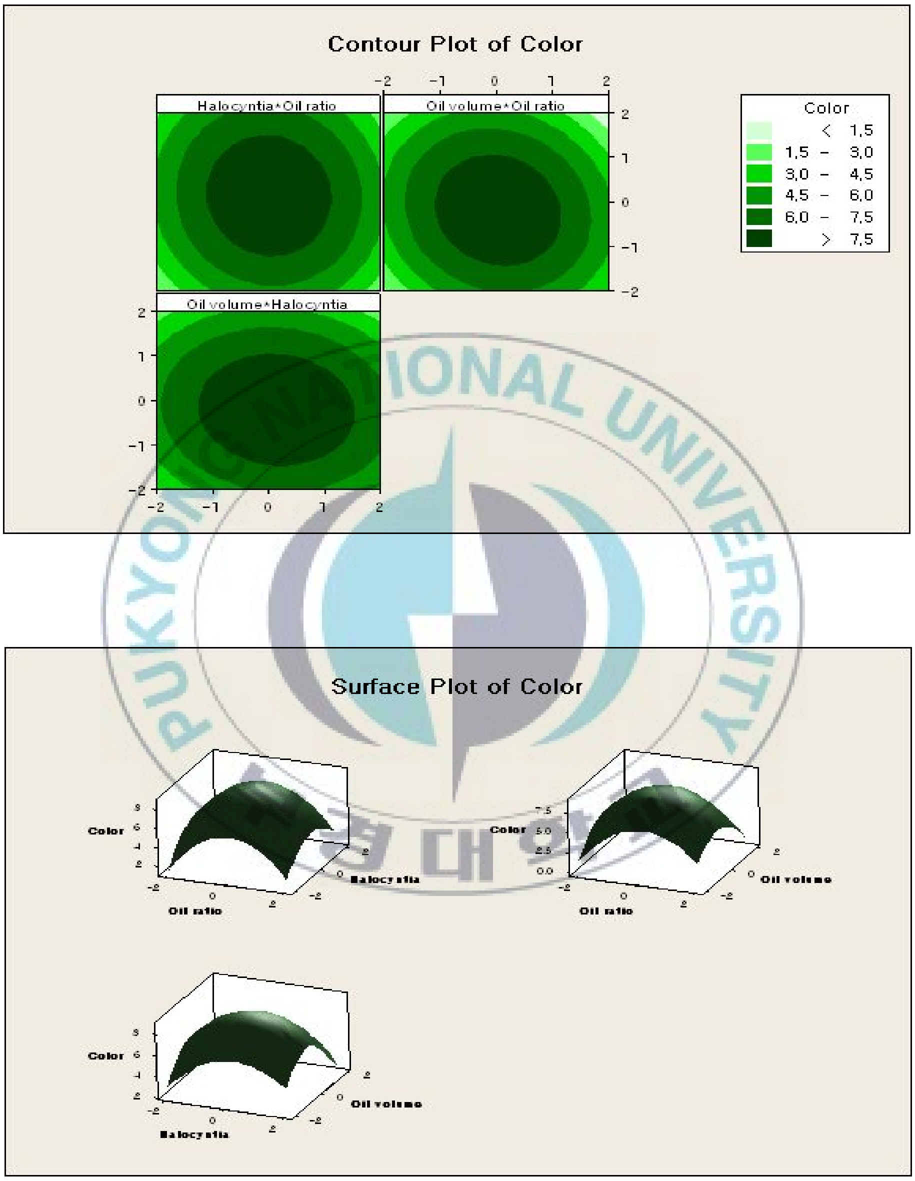


Fig. 7. Surface plot and contour plot of color of bottled sea squirt meat by group 2 panel (9 of over thirties).

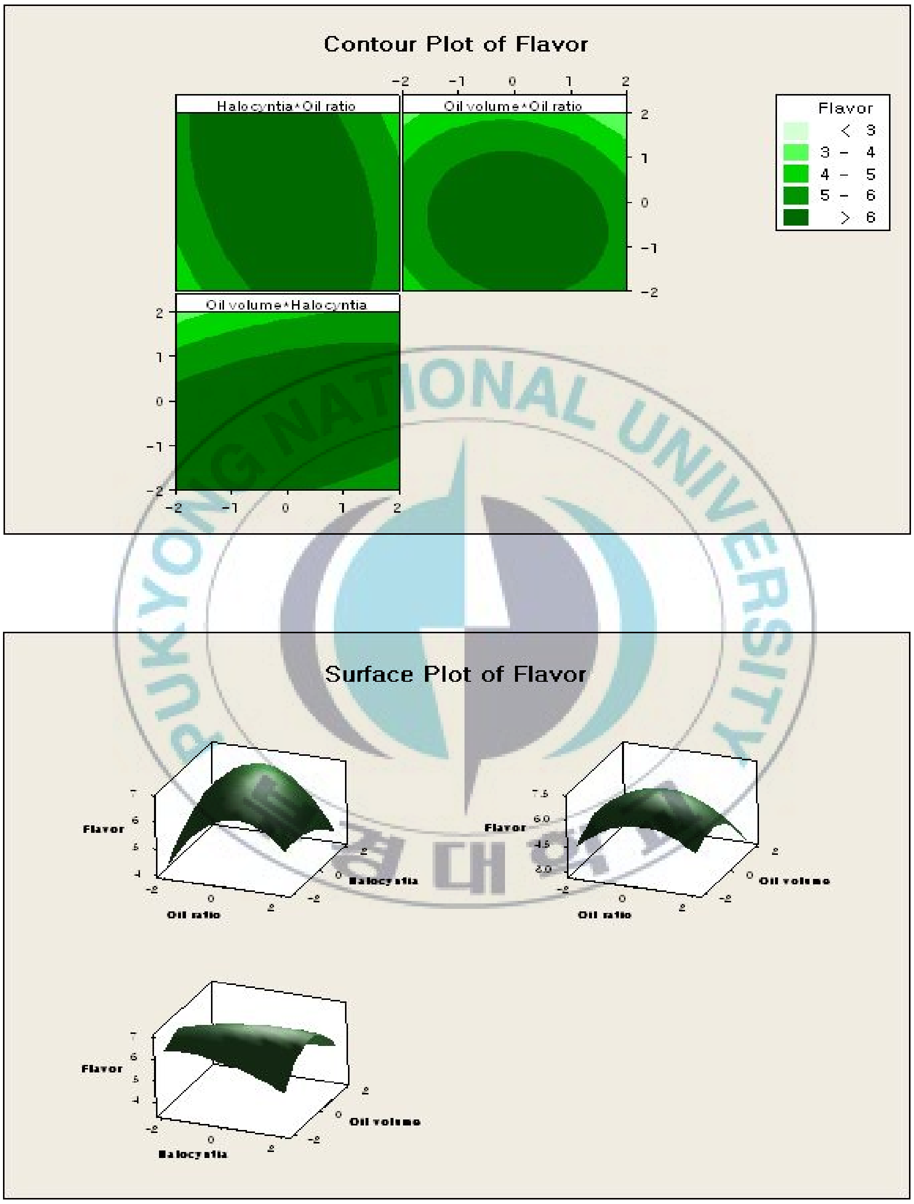


Fig. 8. Surface plot and contour plot of flavor of bottled sea squirt meat by group 2 panel (9 of over thirties).

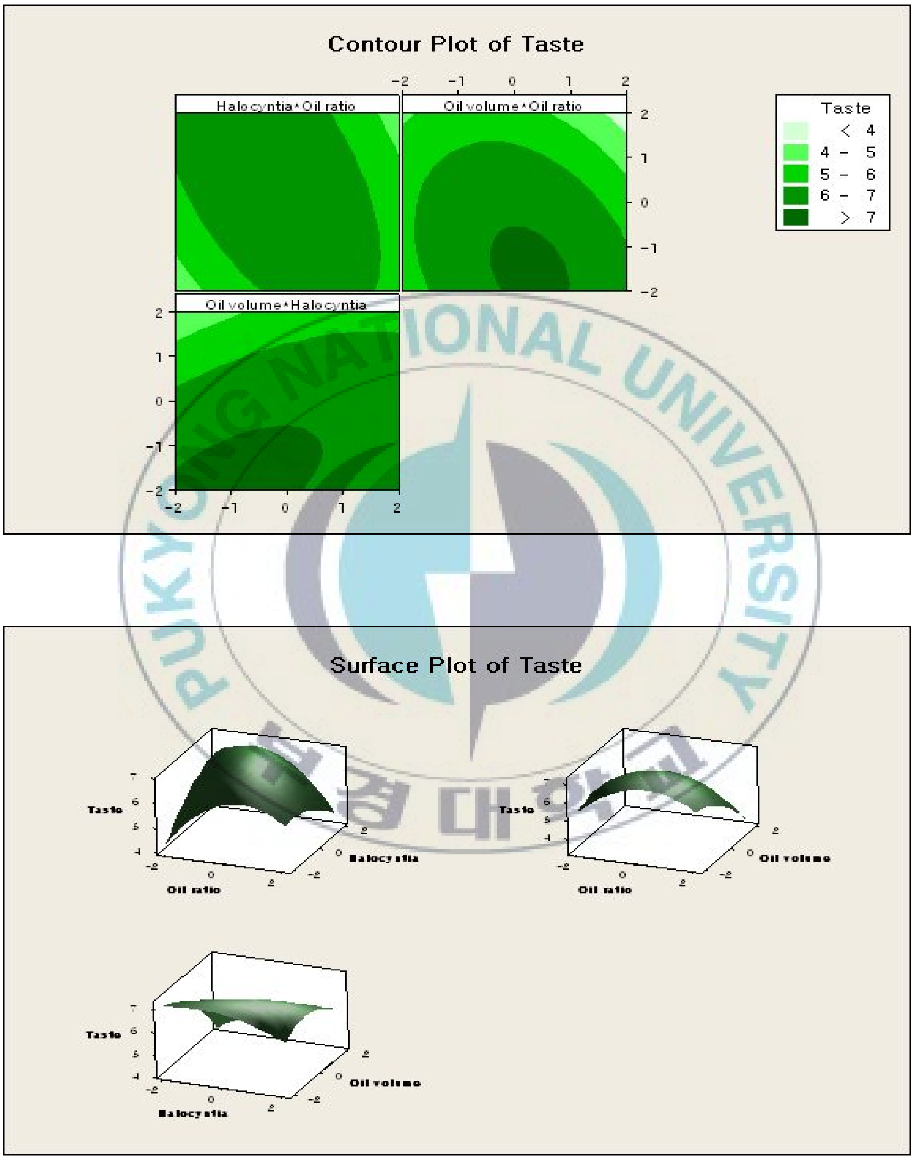


Fig. 9. Surface plot and contour plot of taste of bottled sea squirt meat by group 2 panel (9 of over thirties).

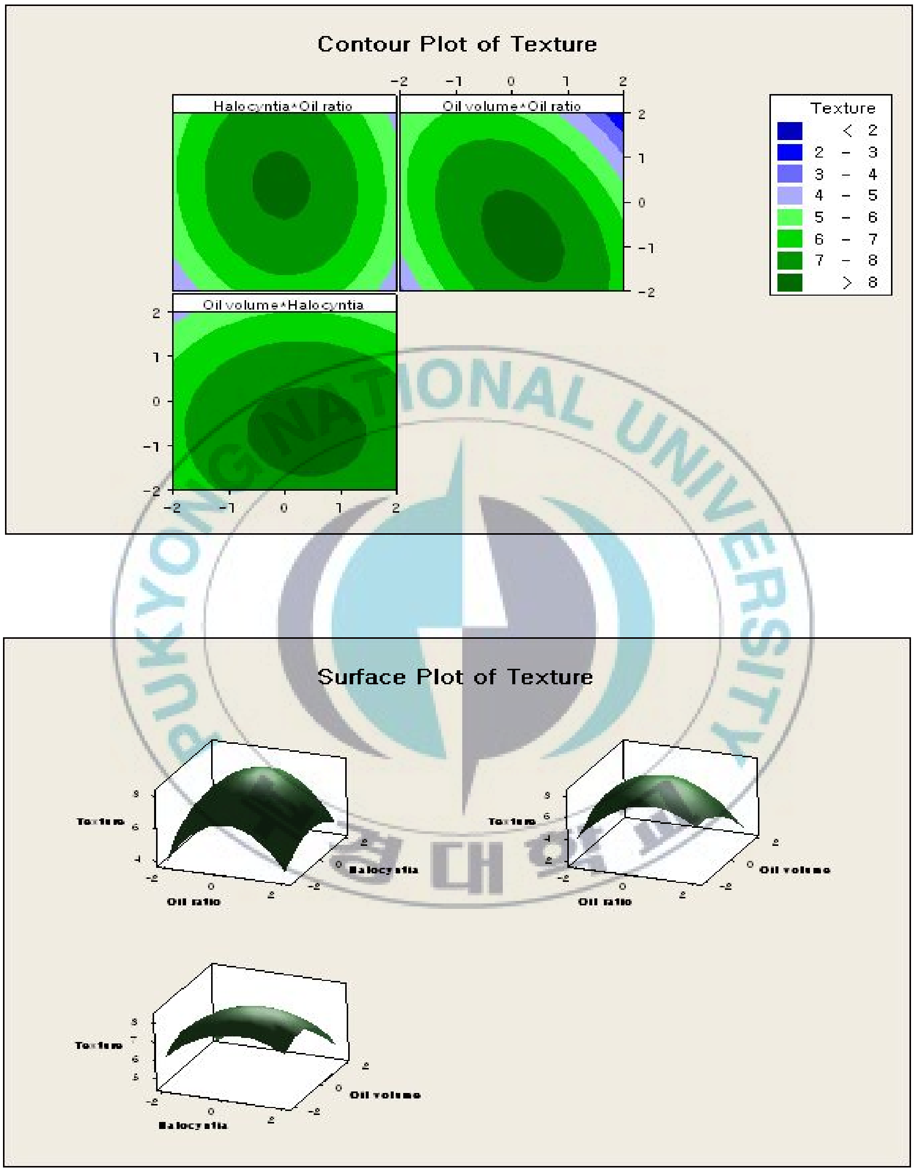
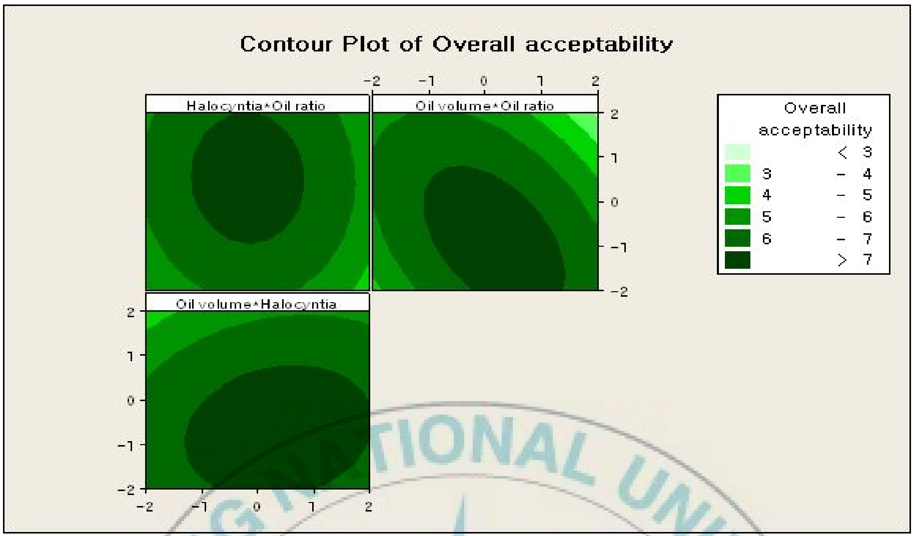


Fig. 10. Surface plot and contour plot of texture of bottled sea squirt meat by group 2 panel (9 of over thirties).



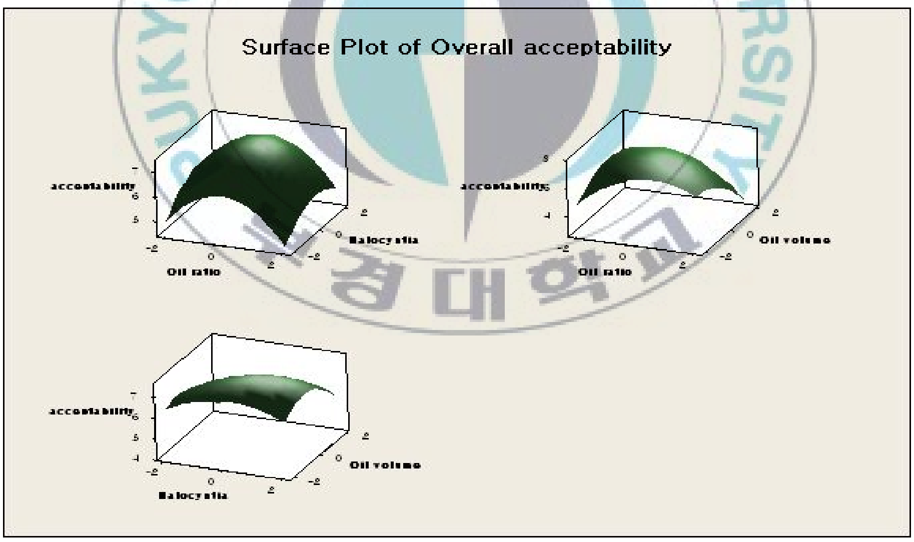


Fig. 11. Surface plot and contour plot of overall acceptability of bottled sea squirt meat by group 2 panel (9 of over thirties).

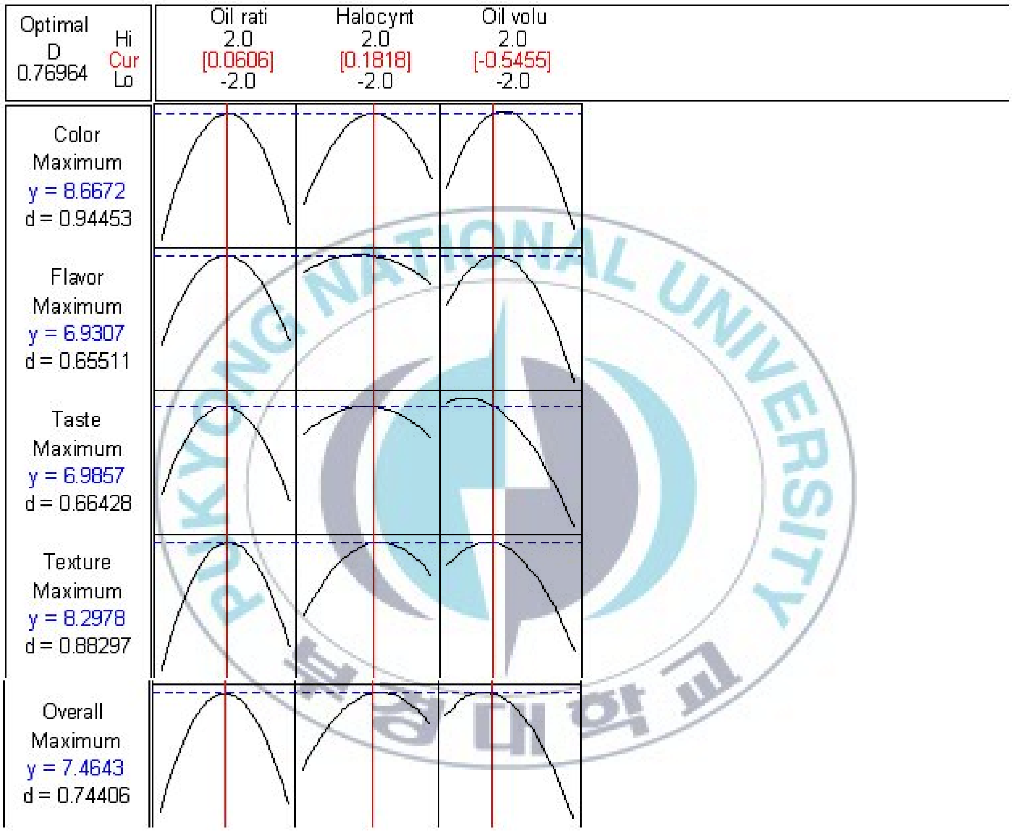


Fig. 12. Optimization curve for the sensory attributes of bottled sea squirt meat by group 2 panel (9 of over thirties).

2. Nutritional evaluation of bottle sea squirt meat products

Nutritional evaluations were carried on the bottled sea squirt meat products prepared following the optimal recipes obtained from Figure 6 and 12 (Table 6)

2.1. Proximate composition

The proximate composition of fresh sea squirt meat, dehydrated sea squirt meat, bottled sea squirt meat in oil (BSMO 1 and BSMO 2) are presented in Table 7. The results of proximate composition of raw sea squirt meat was similar to that reported by National Fisheries Research and Development Institute (NFRDI). A little difference was caused by the characteristics of marine products which can have a little difference easily by the various environmental conditions such as the time of the harvest, season and the place of harvest of specimen. The moisture content of raw sea quirt meat was 88.73%, and that of the dehydrated sea squirt meat was 82.94%. BSMO 1 and BSMO 2 prepared with optimal recipe were about 75%. It could be known that the moisture content was decreased by dehydration through pressing and added vegetable oil compared to raw sample, and those led a higher content of other components. Dipping dehydrated sea squirt meat in vegetable oil appeared to be a higher lipid content than raw sea squirt meat and dehydrated sea squirt meat. The lipid content of the BSMO 1 was 1.73% and BSMO 2 was 2.13%. It was thought that vegetable oil permeated sea squirt meat and this increased lipid content of sea squirt meat itself. The protein content of raw sea squirt meat is 6.04%, and BSMO 1 and BSMO 2 had 10.04% and 10.23% of protein respectively. The ash content was 1.37% in raw sea squirt meat and 2.00% for BSMO 1 and 2.13% for BSMO 2 respectively. Moisture content is the most important factor on the shelf-life of all of food product, especially in seafood products. It was thought that some lower moisture content in dehydrated sea squirt meat used in bottled products could contribute in prolonging the shelf-life of BSMO by freshness related with enzyme activity.

2.2. Water activity (AW)

The water activity (AW) results of samples showed no significant differences as shown in the Table 7, ranging from 0.96 in the raw samples to 0.95 in samples. Usually AW 0.9~1.0 is classfied as water-rich foods and considered to be range to bacterial growth as well as other chemical reactions. This thought that all the sea squirt meat samples had a weakness in storage stability. Thus they had to be preserved to prevent spoilage (Pigott and Tucker, 1990). The predominant component of seafoods is water, and those located as a free water which can easily interacted with other ingredient and enzyme activity and growth of microorganisms. Therefore, the free water content should be lowered or changed to the other form like as bound water. Because those bound water is dependent on water activity, the lower water activity could be guaranted storage stability.

2.3 Texture properties

Table 8 shows texture properties of fresh sea squirt meat, and bottled products (BSMO 1, BSMO 2). The bottled sea squirt meat higher values in hardness, springiness, cohesiveness, gumminess, chewiness and resilience than those of fresh sea squirt meat. Because of olive oil and sesame oil permeated into sea squirt meat the physical demerits such as elasticity and chewiness were inferior to fresh meat.

2.4. Volitile basic nitrogen (VBN) measurement

Determining the volatile basic nitrogen (VBN) content is the tool of investigating the freshness of preserved sample. The experiment was progressed to check the changes in the freshness and storage stability of the bottled sea squirt meat products during 50 days at 4℃. The experiment was carried in 10 days' interval and those results shown in Table 9 and Fig 13. It could be seen that VBN contents of both BSMO 1 and BSMO 2 were increased as the period of storage prolonged. The numerical value of VBN of BSMO 1 rapidly increased until 40 days of storage, but those of BSMO 2 rapidly increased until 30 day storage and then reached maximum value after 40 days longer storage. In the evaluation of freshness of the food by volatile basic nitrogen content, though there are differences in the variety, for the marine product 25mg/100g (Cho et al, 1985) or more is seen as the early decomposition phase. And because in both BSMO 1 and BSMO 2, the volatile basic nitrogen content did not exceed 25mg/100g for 40 days, it was thought that the freshness would be stable but in 50 days' time the BSMO 1 approached closely to 25mg/100g and it is thought that through the volatile basic nitrogen contents the food is in comparatively safe level for 50 days.

2.5. Viable cell count measurement

As the method of inspection on the count of microorganism of BSMO 1 and BSMO 2, the result of viable cell count is shown in Table 10 and Fig 14. It could be seen in both the BSMO 1 and BSMO 2, as the preservation period passes, the viable cell count increased with the longer storaging time until 50 days. In BSMO 1, before 10 days storage, the bacterial count was checked but in case of BSMO 2, same viable cell count was noted at 20 days storage. From the results above, the higher ratio of olive oil was more effective in retarding growth than the sesame oil. The standard of viable cell count of marine product was suggested to be less than 5 log CFU/mg and (KFDA 2002). BSMO 1 and BSMO 2 are thought to be safe in terms of microbiology as the result of measurement of general bacteria was less than 5 log CFU/mg.

2.6. *In vitro* digestibility

Protein is a major component of sea squirt meat products except moisture. It showed 50% above in protein content on the bases of dried weight. In this study, the change of *in vitro* protein digestibility of BSMO 1 and BSMO 2 during 30 days storage is shown in Figure 15. It could be seen that digestibility ($p<0.05$) of BSMO 1 and BSMO 2 were gradually decreased through the storage period. It was thought that proteins in BSMO products interacted with lipid gradually during storage due to rancid products of lipid. In the early period of storage, there was not a notable difference in protein digestibility between both samples but a notable difference was showed after 20 days longer period. The digestibility of BSMO 1 is higher than that of BSMO 2 by about 4%, it is thought that the ratio of sesame oil can have influence on digestibility.

Table 6. Optimizing recipes of bottled sea squirt meat products by response surface methodolgy

(Mean±SD)

	Oil ratio (sesami oil vs olive oil)	Sea squirt meat weight (*Halocynthia roretzi*)	Added total oil volume
BSMO 1	5:5	80g	60ml
BSMO 2	7:3	80g	60ml

BSMO 1 : Bottled sea squirt meat product by group 1 panel (10 of twenties)
BSMO 2 : Bottled sea squirt meat product by group 2 panel (9 of more thirties)

Table 7. Proximate composition of sea squirt meat, dehydrated sea squirt meat, BSMO 1 and BSMO 2

%(dry basis)

	Sea squirt meat *(Halocynthia roretzi)* *	Dehydrated Sea squirt meat *(Halocynthia roretzi)* *	BSMO 1*	BSMO 2*
AW (%)	96.16±0.28	95.5±0.25	95.4±0.2	95.47±0.32
Moisture (%)	88.73±1.62[c]	82.94±1.38[b]	75.73±0.31[a]	75.07±0.57[a]
Ash (%)	1.37±0.16[a] (12.16)	2.84±0.16[c] (16.65)	2.00±0.57[b] (8.24)	2.13±0.23[b] (8.54)
Protein (%)	6.04±0.74[a] (53.59)	9.67±0.58[b] (56.68)	10.04±0.50 [b] (41.37)	10.23±0.74[b] (41.03)
Lipid (%)	0.84±0.23[ab] (7.45)	0.54±0.25[a] (3.16)	1.73±0.83[bc] (7.13)	2.13±0.61[c] (8.54)
Carbohydrate (%)	3.02 (26.79)	4.01 (23.5)	10.5 (43.26)	10.44 (41.87)

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

[a-c]Different letters in column of each sample category show significant differences ($p<0.05$)

*Mean±SD of three determinations

Table 8. Texture properties of sea squirt meat, BSMO 1 and BSMO 2

	Hardness	Fractu ability	Adhesi veness	Springi ness	Cohesi veness	Gummi ness	Chewi ness	Resi lience
Sea squirt meat	1019.419 ±74.017	4.363 ±0.409	−31.198 ±5.557	0.719 ±0.054	0.557 ±0.036	565.283 ±21.265	406.468 ±32.269	0.23 ±0.022
BSMO 1	1135.415 ±84.724	4.213 ±0.564	−8.525 ±1.501	0.691 ±0.011	0.589 ±0.004	668.208 ±44.932	461.578 ±31.971	0.261 ±0.005
BSMO 2	1550.369 ±103.536	4.23 ±0.168	−19.906 ±2.369	0.762 ±0.022	0.619 ±0.089	951.42 ±93.908	722.974 ±50.997	0.279 ±0.029

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

*Mean±SD of three determinations

Table 9. Change in VBN of BSMO 1 and BSMO 2.

Storage days	BSMO 1 (mg/100g)	BSMO 2 (mg/100g)
0 days	7.00 ± 0.50	7.70 ± 1.00
10 days	8.50 ± 0.60	8.70 ± 0.50
20 days	10.13 ± 0.80	10.84 ± 1.44
30 days	13.48 ± 0.49	16.12 ± 0.01
40 days	13.97 ± 0.36	24.44 ± 0.01
50 days	27.92 ± 0.96	24.84 ± 1.95

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

*Mean±SD of three determinations

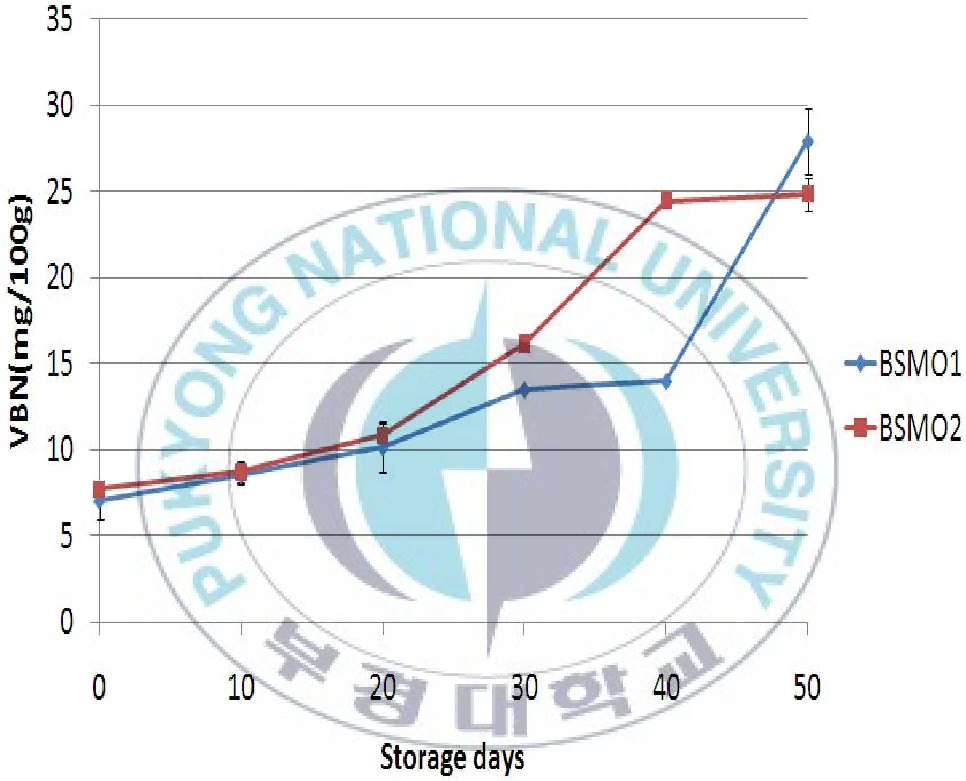


Fig. 13. Change in VBN of BSMO 1 and BSMO 2.

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

Table 10. Change in viable cell count of BSMO 1 and BSMO 2.

Storage days	BSMO 1 (log CFU/mg)	BSMO 2 (log CFU/mg)
0 days	0	0
10 days	2.65 ± 0.30	0
20 days	3.66 ± 0.40	2.72 ± 0.20
30 days	3.66 ± 0.20	3.20 ± 0.20
40 days	4.09 ± 0.40	3.20 ± 0.40
50 days	4.60 ± 0.80	3.65 ± 0.20

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

*Mean±SD of three determinations

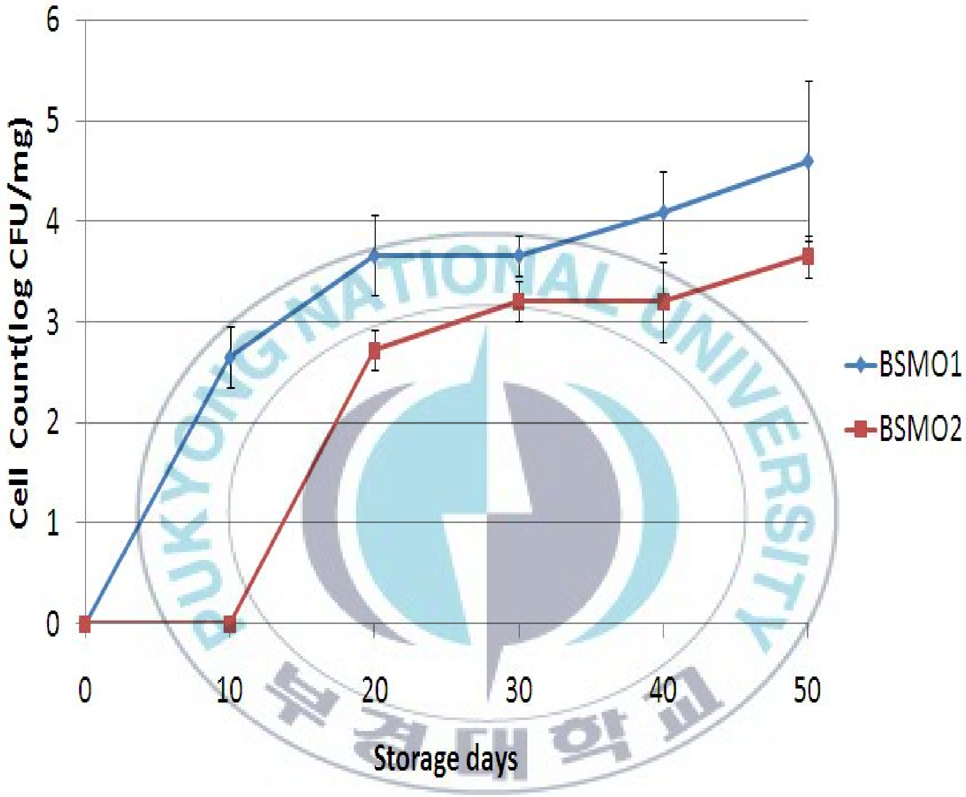


Fig. 14. Change in viable cell count of BSMO 1 and BSMO 2.

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

Table 11. Change in *in vitro* digestibility of BSMO 1 and BSMO 2.

Storage days	BSMO 1 (%)	BSMO 2 (%)
0 days	78.42 ± 0.32	79.11 ± 0.64
10 days	77.71 ± 0.34	76.80 ± 0.32
20 days	76.23 ± 0.13	74.90 ± 0.37
30 days	73.93 ± 0.13	69.76 ± 0.18

BSMO 1 : Bottled sea squirt meat product by group 1 sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 sensory evaluation

*Mean±SD of three determinations

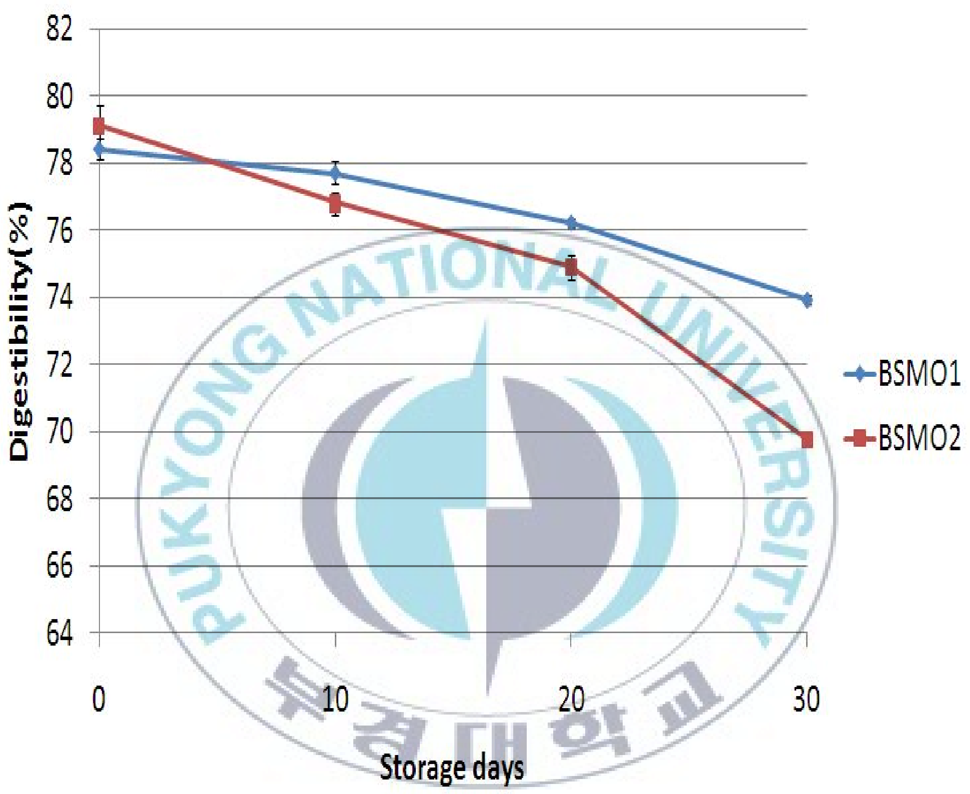


Fig. 15. Change in *in vitro* digestibility of BSMO 1 and BSMO 2.

BSMO 1 : Bottled sea squirt meat product by group 1 (10 of twenties) sensory evaluation

BSMO 2 : Bottled sea squirt meat product by group 2 (9 of more thirties) sensory evaluation

Ⅳ. Conclusion

This study was performed to optimize the recipe of bottled sea squirt meat in vegetable (BSMO) of ratio of vegetable oil (olive oil and sesame oil). Nutritional evaluation and the test of storage stability were also tried on BSMO prepared with optimal recipe.

1. As the result of sensory evaluation on the color, flavor, texture, taste and overall acceptability by group 1 panel (10 of twenties) and group 2 panel (9 more thirties), the optimal ratio of bottled sea squirt meat was find out.

- Bottled sea squirt meat by group 1 panel (10 of twenties)
  the ratio (X_1) of olive oil to sesame oil : 5:5,
  the amount (X_2) of sea squirt meat : 80g,
  the total volume (X_3) of vegetable oil: 60 ml,
- Bottled sea squirt meat by group 2 panell (9 of more thirties)
  the ratio (X_1) of olive oil to sesame oil : 7:3,
  the amount of sea squirt meat (X_2) : 80g,
  the total volume of vegetable oil (X_3) : 60ml

2. Due to added vegetable oil of BSMO, lipid content, hardness, springiness, cohesiveness, gumminess, chewiness, resilience were higher

than those of fresh sea squirt meat. Because of olive oil and sesame oil permeated into sea squirt meat, the physical demerits such as elasticity and chewiness were inferior to fresh sea squirt meat.

3. It was showed that freshness and storage stability were maintained to 50 days by dehydrating process and irradiation of sea quirt meat. Protein digestibility was gradually decreased during 30 days storage.

In spite of the fact that sea squirt meat was enjoyed as representative cultured fishery product, due to the moisture content of more 80% and enzyme activity, It was difficult that manufacturing characteristics of sea squirt meat.

Through this study, it was expected that shlef-life of sea squirt meat and safety of sea squirt meat were extended by bottled sea squirt meat product, and would be enhanced its potentials in terms of the value of fish productions for its food process. In addition, it is thought that it can be the opportunity for the advance of consumer awareness of the functional quality of sea squirt meat and opening of the possibility of planned consumption. And it could be one foundation which hang between the food cultures of generations and could be shared the viewpoints about the food one another.

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