การผลิตซอสปรุงรสจากผัก
Seasoning Sauce Production from Vegetable Paste

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บทคัดย่อ

น้ำผักเป็นผลิตภัณฑ์อาหารพื้นบ้านของอำเภอแจ้ห่มจังหวัดลำปาง ซึ่งทำจากผักกาดเขียว นิยมบริโภคกันอย่างแพร่หลายในพื้นที่ภาคเหนือตอนบน เนื่องจากมีรสชาติและคุณค่าทางโภชนาการสูง การศึกษานี้มีวัตถุประสงค์เพื่อนำน้ำผักมาทำเป็นซอสปรุงรสเพื่อเพิ่มมูลค่าของผลิตภัณฑ์ในท้องถิ่น โดยการนำน้ำผักมาเจือจางด้วยน้ำอัตราส่วน 60:40 (น้ำผัก:น้ำ) และพัฒนาสูตรโดยใช้การทดลองออกแบบส่วนผสม พบว่าสูตรซอสปรุงรสจากผักที่มีคะแนนการประเมินทางประสาทสัมผัสสูงสุดประกอบด้วย น้ำผักเจือจาง 65 wt% แป้งข้าวโพด 5 wt% น้ำตาลอ้อย 30 wt% เกลือ 4 wt% และพริกไทย 8 wt% โดยสภาวะในการพาสเจอร์ไรส์ที่เหมาะสมคือ 93.3ºC เป็นเวลา 20 นาที ซอสปรุงรสจากผักที่ได้มีองค์ประกอบทางเคมีคือ โปรตีน 3.66% ไขมัน 15.04% เถ้า 13.20% เส้นใย 3.32% ปริมาณคาร์โบไฮเดรต 24.46% และกรดอะมิโนอิสระ 14 ชนิด วิเคราะห์โดย High Performance Liquid Chromatography (HPLC) มีปริมาณกรดทั้งหมดและ pH เท่ากับ 0.74% และ 3.35 ตามลำดับ ปริมาณจุลินทรียทั้งหมด $3.0x10^2$ CFU/g ตรวจไม่พบโคลิฟอร์มแบคทีเรีย ยีสต์ และรา ซึ่งคาที่ได้เป นไปตามคามาตรฐานของซอส (มอก.1317-2538)

คำสำคัญ: ซอสปรุงรสจากผัก น้ำผัก ผักกาดเขียวปลี ผลิตภัณฑ์อาหารพื้นเมือง

ABSTRACT

Vegetable paste is the local food product of Chae Hom District, Lampang Province which was made of mustard green. It is widely used in the Upper Northern region due to its taste and high nutrition. This study aims to use the vegetable paste to be made into seasoning sauce in order to increase the value of the local product. Diluting the vegetable paste with water at ratio 60:40 (vegetable paste: water) and developing the recipe using the mixture design experiment. The results showed that the selected recipe with the highest sensory evaluation scores consisted of 65 wt% diluted vegetable paste liquid, 5 wt% corn flour, 30 wt% cane sugar, 4 wt% salt and 8 wt% pepper. The optimal thermal process condition was at 93.3ºC for 20 minutes. The proximate analysis of sauce showed 3.66 % protein, 15.04 %fat, 13.20 %ash, 3.32 %fiber, 24.46 % carbohydrate content and 14 free amino acids analyzed by High Performance Liquid Chromatography (HPLC) was detected in this developed sauce. Physical and microbiological properties were also determined, acidity and pH were 0.74% and 3.35, respectively. Microorganisms analyzed by Total plate count method were $3.0x10^2$ CFU/g. Coliform bacteria, Yeast and mold were not detected. The result complied with the microbiological criterion standard for sauce established by TISI. 1317-2538.

Keywords: vegetable seasoning sauce, vegetable paste, mustard green, local food product

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INTRODUCTION

As a traditional food of the Chae Hom District, Lampang Province, vegetable paste is popularity widely consumed across Upper-north of Thailand and predominantly in Lampang Province. Vegetable paste has been popular for centuries due to its pleasant flavor and unique taste especially the umami taste. The flavor and taste of vegetable paste is produced from fermentation of leaf mustard (*Brassica juncea* (L.) Czern.) combined with glutinous rice and salt. Vegetable proteins can be utilized by the lactic acid bacteria (LAB) into peptides and amino acids during fermentation. Most of the amino acids and organic acids in the vegetable paste were originated in the raw material. The synergistic effect among free amino acid and sodium salt has improved the umami taste of vegetable paste. For example, Glutamic (GLU), Phenylalanine (PHE), Valine (VAL) and Serine (SER) have potentially been detected in some leaf mustard (*Brassica juncea* (L.) Czern.) [1].

Even though vegetable paste is a well-known cuisine in Up north of Thailand, the new generations are rarely interested in vegetable paste, due to its taste and intense vegetable smell. Therefore, development of vegetable seasoning sauce from vegetable paste will be improving its smell and taste and make more choices in consuming the vegetable paste according to the specific need of the consumer who wants different food. Moreover, the value of local products will increase and be able to extend the commercial opportunities. The purpose of this study was to use the vegetable paste as a raw material for vegetable seasoning sauce production. For this, we performed this study as following experimental scheme; (1) study the physicochemical of vegetable paste, (2) develop the optimal formulation of vegetable seasoning sauce, and (3) effect of time-temperature conditions for thermalon the overall quality of vegetable seasoning sauce.

MATERIALS AND METHODS

1. Analysis of vegetable paste

The vegetable paste used in this study were obtained from local market in Chae Hom District, Lampang Province area between June-September, 2020. Vegetable paste was diluted with water in ratio 40:60 to reduce the total solid content and boiled for 5 minutes to partial sterilization. The diluted vegetable paste liquid was used for the proximate analysis according to the guidelines published by the Association of Official Analytical Chemists [2]. The free amino acid compositions were analyzed according to the method described by [3]. The pH and water activity were measured in triplicate using pH meter (Waterproof IP57, Taiwan) and Aqualab (4TEV series), respectively. Aerobic plate counts and yeast and mold counts were determined by following the procedures of Bacteriological Analytical Manual method [4].

2. Formulation of vegetable seasoning sauce

Development of vegetable seasoning sauce product formulas modified from oyster sauce which is famous and widely used. The diluted vegetable paste liquid prepared with the ratio of water: vegetable paste as 40:60. The optimal percentage of diluted vegetable

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paste, corn flour and sugar were optimized by using Mixture design as follows: diluted vegetable paste liquid, corn flour and sugar were varied in 50-65%, 5-15% and 20-45%, respectively. For other ingredients, used in fixed amounts. All formulas were appeared in Table 1 and the method of vegetable seasoning sauce described in figure 1. Sensory evaluation was completed by simple ranking test with nine-point hedonic scale. The sensory attributes (colour, flavour, taste, after taste and overall liking) were evaluated by 30 untrained panelists [5].

3. Study of quantity of salt and pepper

The quantity of salt and pepper were studied in three different levels, which were 4%, 6% and 8% of the formulas. Sensory evaluation was done for the selection of appropriate salt and pepper quantity with nine-point hedonic scale test [5]. The 30 untrained panelists evaluated the samples for color, flavour, saltiness, spiciness, taste, after taste and overall acceptability.

4. Optimization of thermal conditions of vegetable seasoning sauce

A thermal process for vegetable seasoning sauce using conventional treatment; 180 mL glass bottles containing 170 g of seasoning sauce immersed in water at 199.94°F (93.3°C) for an additional 10, 20, 30 and 40 min, respectively, after the core temperature had reached 199.94°F (93.3°C) for one min. A control vegetable seasoning sauce (without thermal treatment) and five treated vegetable seasoning sauce were cooled and stored at 4°C for 48 hours. Then, the six treated sample were subjected to analysis for Aerobic plate counts, yeast and mold counts [4] and Coliform and *Escherichia coli* [6].

Statistical Analysis

Data from three replications were analyzed by analysis of variance (ANOVA) using SPSS version 17.0 for Windows. Duncan's multiple range tests (DMRT).were used to determine the statistical significance among the means at a significance level of 95%.

Table 1 Formulated vegetable sauce

Ingredient	Formulas								
(gram)	1	2	3	4	5	6	7	8	9
DVPL*	54	50	58	58	65	65	50	65	55
Corn flour	8	15	15	10	15	10	5	5	12
Sugar	38	35	27	32	20	25	45	30	33
Garlic	10	10	10	10	10	10	10	10	10
Sesame oil	15	15	15	15	15	15	15	15	15
Vinegar	15	15	15	15	15	15	15	15	15
Salt	2	2	2	2	2	2	2	2	2
Ground pepper	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5

Note: DVPL, diluted vegetable paste liquid

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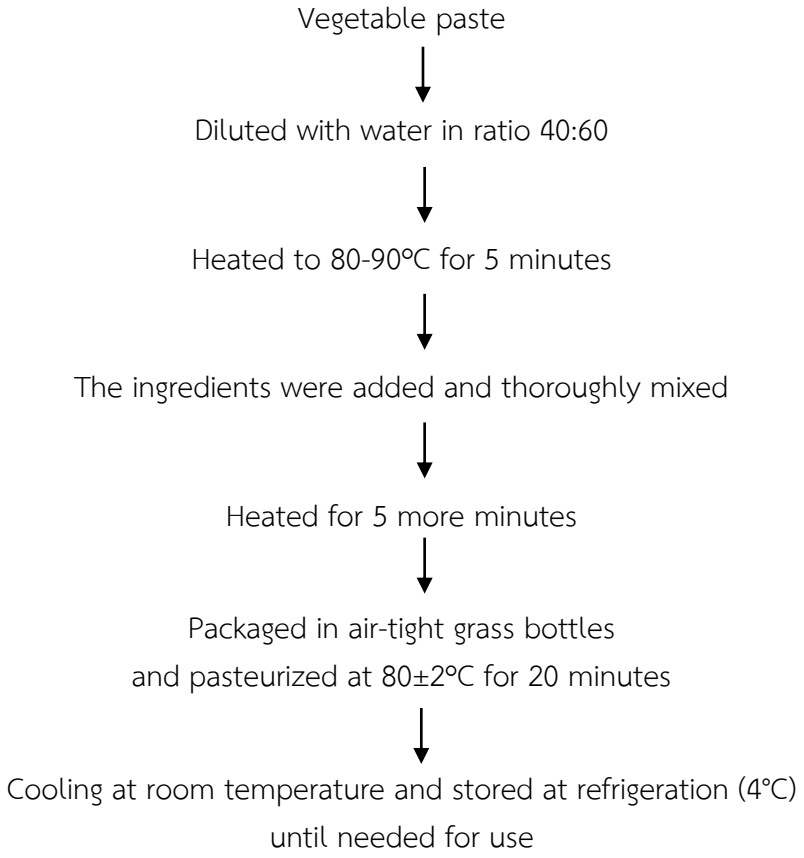

Figure 1 The production of vegetable seasoning sauce

RESULTS AND DISCUSSION

1. Quality of vegetable paste

Vegetable paste of the Chae Hom District, Lampang Province, contained the number of proteins, fat, ash, fiber and carbohydrate for 5.75, 0.64, 32.74, 11.48, and 18.97 g/100g, respectively. The pH values indicated that vegetable paste is an acidic food (pH<3.5). Free amino acids play a crucial role to the development of organisms [7] and enhance the taste of vegetable paste. The 14 free amino acids in vegetable paste were analyzed by HPLC. The contents of free amino acid (mg/g) were shown in Figure 2. Among the 14 amino acids, Asp and Glu are responsible for umami taste [1]; Ser, Pro, Gly and Thr create sweet tastes; Val, Met, Ile, Phe, Lys, Leu, Arg and His create taste bitter; Arg, which accounted for 0 mg/g in sample, has no contribute to the taste. The content of bitter taste amino acids was much higher than sweet and umami-taste amino acids, ranging from 6.29 to 166.20 mg/g. The result indicated that bitter taste amino acids were the dominant amino acids in vegetable paste. The amount of sweet-taste amino acid ranged from 29.75 to 85.86 mg/g. Umami taste amino acid contents ranged from 56.94 to 57.23 mg/g.

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Table 2 Quality of vegetable paste

Components	Vegetable paste
Protein (g/100g)	5.75±0.15
Fat (g/100g)	0.64±0.47
Ash (g/100g)	32.74±0.31
Fiber (g/100g)	11.48±0.11
Moisture (g/100g)	30.42±0.07
Carbohydrate (g/100g)	18.97±0.04
Water activity (aw)	0.73±0.00
pH value	3.65±0.11
Aerobic plate counts (CFU/g)	$1x10^3$
Yeast and mold counts (CFU/g)	$1x10^2$

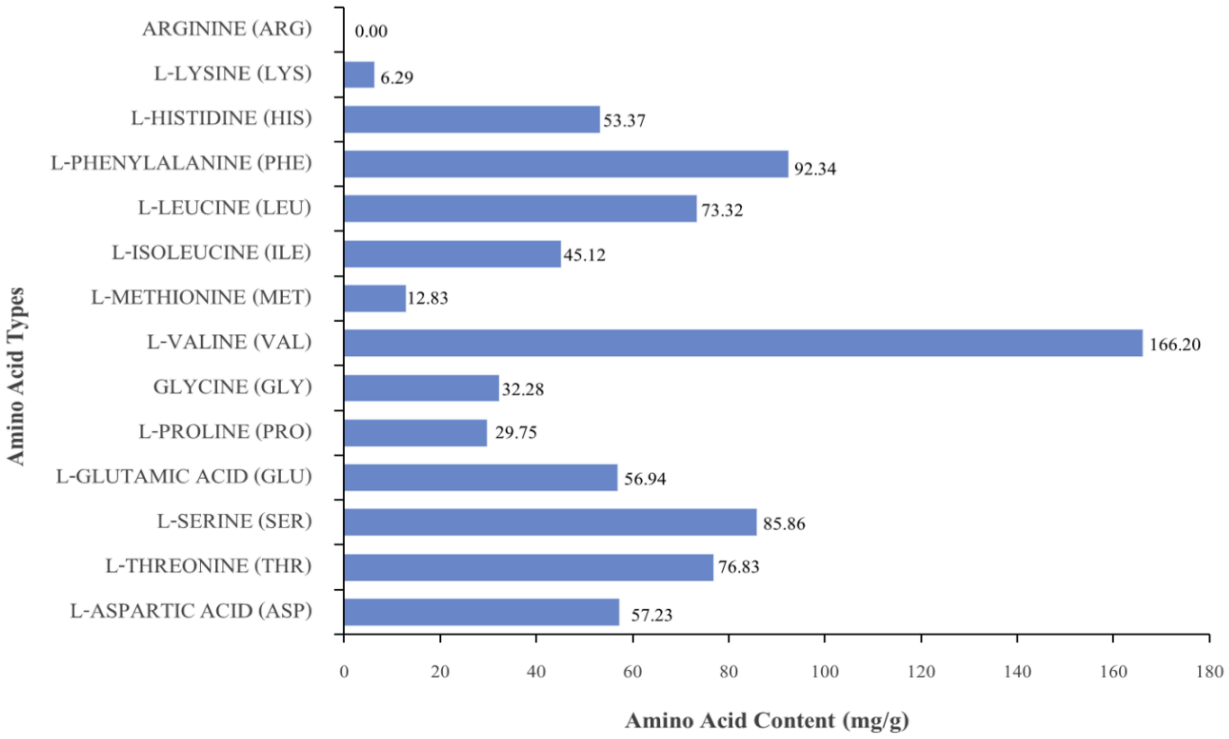

Figure 2 Free amino acid content in vegetable paste

2. Formulation of vegetable seasoning sauce

Using the results from the consumer acceptance, majority of panelist like vegetable seasoning sauce based on hedonic percentage for the specific attributes of flavour, taste, and after taste, which these sensory attributes reflect the consumer acceptance of the product, i.e., overall acceptance. The best formula was chosen to prepare vegetable seasoning sauce consist of 65 g of vegetable paste liquid, 5 g of corn flour and 30 g of sugar. shows highest hedonic score percentage for all attributes. The selected vegetable seasoning sauce formula was used for further analysis

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Table 3 Sensory evaluation scores of vegetable seasoning sauce in different formulas

Formulas	Colour	Flavour ^{ns}	Taste	After taste	Overall acceptability
1	6.40^{ab}±1.24	7.00±1.31	7.13^{b}±1.03	6.87^{a}±0.92	7.07^{b}±0.88
2	6.33^{b}±1.23	6.87±0.99	6.80^{cd}±0.56	6.53^{a}±0.64	6.60^{c}±0.74
3	6.60^{ab}±1.30	6.93±0.96	6.97^{c}±1.13	6.73^{a}±1.03	6.87^{b}±1.19
4	6.53^{ab}±1.25	6.80±0.78	6.40^{f}±0.83	6.53^{a}±0.83	6.53^{c}±0.92
5	6.53^{ab}±1.41	6.60±1.12	6.73^{d}±0.88	6.40^{ab}±0.74	6.73^{b}±0.96
6	6.40^{ab}±1.30	6.73±1.03	6.20^{g}±1.01	6.00^{b}±1.00	6.27^{d}±0.88
7	6.47^{ab}±1.30	6.73±0.70	6.47^{f}±1.13	6.47^{ab}±1.06	6.53^{c}±0.92
8	6.73^{a}±1.39	7.07±1.16	7.80^{a}±1.08	6.53^{a}±0.74	7.39^{a}±1.16
9	6.60^{ab}±1.35	6.87±1.12	6.67^{e}±1.05	6.67^{a}±0.90	6.60^{c}±0.83

Note: ns: non significance

The scores of sensory evaluation was mean ± standard deviation (n=3)

The different lowercase letters between columns represent significant difference ($p<0.05$)

3. Salt and pepper

Improvement of the taste, salt and pepper are varied into 3 level including 4, 6 and 8 g/100g. Table 4 illustrates that the vegetable seasoning sauce adding salt and pepper for 4 and 8 g/100g, respectively, has the highest median score for overall acceptability (7.64). Therefore, the vegetable seasoning sauce containing salt and pepper of 4 and 8 g/100g, respectively, was selected to study about temperature and time in pasteurization.

Table 4 Sensory evaluation scores of vegetable seasoning sauce adding salt and pepper in different portion

Salt (g/100g)	Pepper (g/100g)	Colour ^{ns}	Flavour	Saltiness	Spiciness	Taste	After taste	Overall acceptability
4	4	6.13±1.30	5.20^{b}±1.61	5.87^{ab}±0.99	5.73^{ab}±0.88	5.60^{bc}±1.54	5.60^{ab}±1.40	5.73^{d}±1.33
4	6	5.87±1.24	5.60^{ab}±1.54	5.87^{ab}±1.24	5.67^{ab}±1.39	5.67^{abc}±1.39	5.80^{ab}±1.32	5.93^{c}±1.33
4	8	6.33±1.04	6.47^{ab}±1.18	6.07^{ab}±1.53	6.33^{a}±1.29	6.80^{a}±0.94	6.60^{a}±1.18	7.64^{a}±0.92
6	4	5.93±1.33	5.33^{ab}±1.49	5.87^{ab}±1.06	5.87^{ab}±1.06	5.80^{abc}±0.94	5.86^{ab}±1.72	5.86^{cd}±1.68
6	6	5.87±1.18	5.40^{ab}±1.45	5.40^{ab}±1.12	5.60^{ab}±1.05	5.67^{abc}±1.17	5.73^{ab}±1.03	5.80^{d}±1.01
6	8	6.13±1.12	5.93^{ab}±1.33	5.73^{ab}±1.33	6.20^{a}±1.52	6.13^{ab}±1.40	5.93^{ab}±1.38	7.00^{b}±1.16
8	4	5.93±1.10	5.00^{b}±1.85	4.87^{b}±1.72	5.00^{b}±1.81	4.80^{c}±1.65	4.80^{b}±1.69	5.06^{e}±1.79
8	6	5.93±1.43	5.87^{ab}±1.30	5.67^{ab}±1.39	5.67^{ab}±1.17	5.93^{abc}±1.03	5.86^{ab}±1.06	5.86^{cd}±0.91
8	8	6.53±1.24	5.87^{ab}±1.18	5.60^{ab}±1.54	5.87^{ab}±1.55	6.00^{ab}±1.60	5.53^{ab}±1.50	5.93^{c}±1.62

Note: ns: non significance

The scores of sensory evaluation was mean ± standard deviation (n=3)

The different lowercase letters between columns represent significant difference ($p<0.05$)

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4. Optimization of thermal time of vegetable seasoning sauce

The pH and total plate count in thermally processed (pasteurized) vegetable seasoning sauce decreased significantly ($p<0.05$) compared with the control sample (as shown in Table 5). The data obtained in the study showed that aerobic plate counts of treated samples were decreased in the thermal process. After the 20 min thermal treatment, the aerobic plate counts were reduced 1 log compared with the control sample (from 5.1 $x10^4$ CFU/g to $1.8x10^3$ CFU/g), respectively. The lowest aerobic plate counts were found in the thermal treatment at 93.3°C for 40 min ($2.0x10^2$ CFU/g). However, it was observed that the conventional thermal process at 93.3°C for 20 min was optimal for vegetable seasoning sauce due to aerobic plate counts was less 1 $x10^4$ CFU/g according to the standard for sauce set by TISI. 201-2543 and TISI. 1317-2538. Yeast and mold were not detected.

Table 5 Total plate count, yeast and mold levels of pasteurized vegetable seasoning sauce in different time

Thermaltime (minute)	pH value	Microorganisms	
		Total plate count (CFU/g)	Yeast and mold (CFU/g)
Control	3.67^a±0.00	5.1 $x10^4$	Not detected
10	3.36^b±0.00	$3.5x10^4$	Not detected
20	3.35^b±0.00	$1.8x10^3$	Not detected
30	3.35^b±0.00	$4.5x10^2$	Not detected
40	3.34^b±0.00	$2.0x10^2$	Not detected

Note: The value of pH was mean ± standard deviation (n=3)
The different lowercase letters between columns represent significant difference ($p<0.05$)

5. Physico-chemical properties of the vegetable seasoning sauce

Table 6 shows the physico-chemical properties of the vegetable seasoning sauce. According to Food and Drug Administration Federal Agency in USA has reported that the pH value of sauce should be within 4.4 to 5.4 [8]. The vegetable seasoning sauce in this study had a pH of 3.35 and 0.74% titratable acidity. The pH value and acidity of vegetable seasoning sauce is influenced by vegetable paste. According to the regulations, it is necessary for sauce to maintain a pH of 4.6 or less to preserve the product as production of botulism toxin by *Clostridium botulinum* is inhibited at this pH range [9]. The newly developed vegetable seasoning sauce has 3.66 % protein, 15.04 % fat, 13.20% ash, 3.32% fiber and 24.46% carbohydrate content. Aerobic plate counts were $3.0x10^2$ CFU/g. Coliform and *Escherichia coli*, Yeast and mold counts were not detected. The microorganism were complied with standard for sauce established by TISI. 1317-2538.

Among free amino acids, Aspartate (Asp) Glutamic acid (Glu) and Glycine (Gly) are umami amino acids [1]. Thus, the umami amino acid content we mentioned is the total content of the three amino acid. Figure 3 showed the free amino acid content in vegetable seasoning sauce, the

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total content of umami amino acid could reach 65.68 mg/g, it was lower compared with bitter-taste amino acid (156.51 mg/g). However, The Glu content in vegetable seasoning sauce was significantly higher than others (38.87 mg/g). Glutamic acids containing di- and tri-peptides have been of particular interest because they are related to the unique taste of umami. Besides, Glu is an important umami amino acid in brain tissue biochemical metabolism and participates in the production of a variety of physiologically active substances [10].

Table 6 Quality of vegetable seasoning sauce

Components	Vegetable seasoning sauce
L*	35.49±0.36
a*	2.83±0.06
b*	-1.14±0.19
pH value	3.35±0.01
Total acidity (g/100g)	0.74±0.00
Moisture (g/100g)	40.32±0.58
Fat (g/100g)	15.04±0.14
Protein (g/100g)	3.66±0.33
Ash (g/100g)	13.20±0.74
Fiber (g/100g)	3.32±0.56
Carbohydrate (g/100g)	24.46±0.12
Aerobic plate counts (CFU/g)	$3.0x10^2$
Yeast and mold counts (CFU/g)	Not detected
Coliform and *Escherichia Coli* (MPN/g)	Not detected

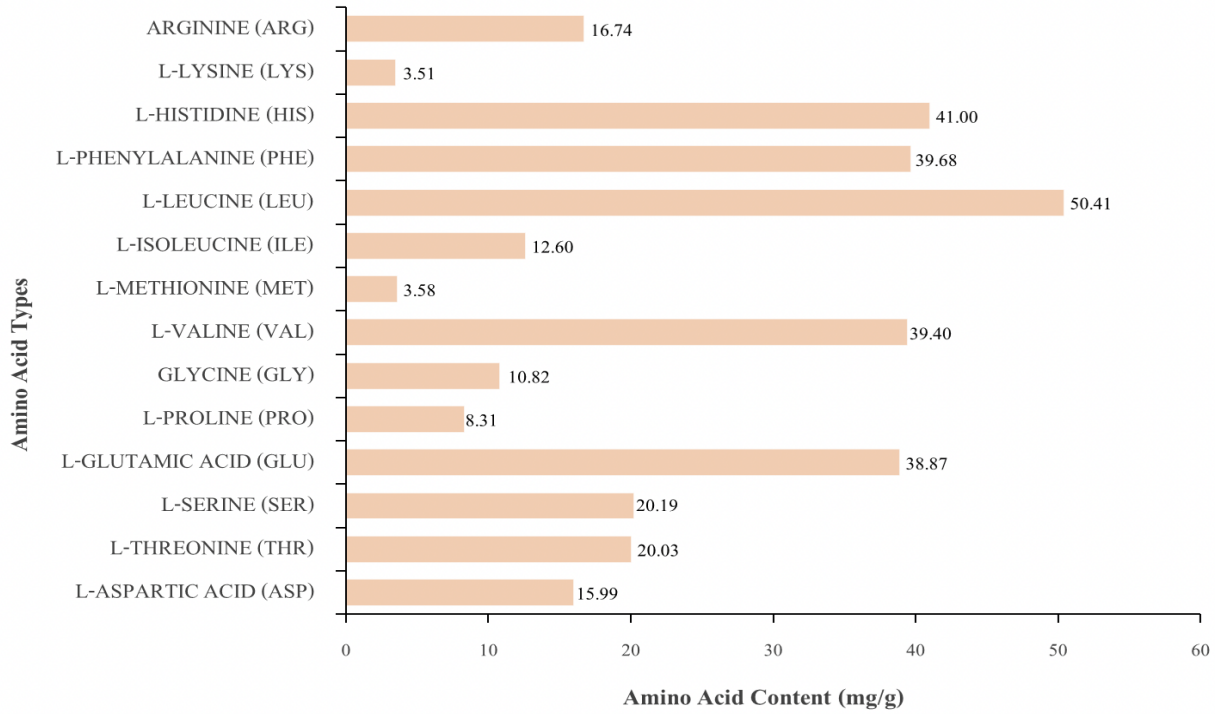

Figure 3 Free amino acid content in vegetable seasoning sauce

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CONCLUSIONS

The vegetable seasoning sauce prepared from vegetable paste was found to be of superior quality in terms of nutritional value and overall acceptability. Vegetable seasoning sauce includes 65 g of vegetable paste liquid, 5 g of corn flour, 30 g of sugar, 10 g of Garlic, 15 mL of sesame oil, 15 mL of vinegar, 4 g of salt and 8 g of ground pepper, in which the consumers have given the highest moderate score. The conventional thermal process at 93.3°C for 20 min was the best in terms of decreased microbiological to comply with standard for sauce established by TISI. 1317-2538. Moreover, vegetable seasoning sauce contained 14 free amino acids play a crucial role to the development of organisms and enhance the taste of sauce. Results of the present study offer a good choice for production of high value vegetable seasoning sauce

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