Flavor Enhancer From Catfish (*Clarias batrachus*) Bekasam Powder and Angiotensin-I-Converting Enzyme (ACE) Inhibitory Activity in Various Dishes

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Abstract. Flavor enhancer is characterized by high glutamic acid content and it can be obtained from fermented food such as Bekasam. Fermented food had inhibitory effect on Angiotensin-I-Converting Enzyme (ACE) activity which is advantageous for hypertension. However, such activity was not known to sustain in food system. The aim of this research was to study addition of flavour enhancer from Catfish Bekasam Powder (CBP) in various food systems and to determine the ACE inhibitory (ACEI) activity in the food system. Four food system consisted of carrot, champignon, and chicken meat dishes were boiled in water and added with CBP or MSG. Each food system was added with graded level of CBP (0%; 0.5%; 0.8%; 1.1%; and 1.4%) and for control monosodium glutamate (MSG) was used. ACEI activity in each food system and organoleptic test using multiple comparison differentiation on 15 semi-trained panellists were determined. The results showed that there were fluctuation of ACEI activity in the carrot, champignon, and chicken meat dishes (p=0.017; 0.043; and 0.032). The MSG containing dishes showed the lowest ACEI activity. Addition of graded level of CBP on carrot, champignon, and chicken meat dishes were directly proportional to glutamic acid content but inversely proportional to ACEI activity (p<0.05). The addition of commercial MSG on all dishes increased glutamic acid content but reduced ACE-inhibitory activity significantly (p<0.05). Comparing CBP to MSG addition in champignon dish revealed that increasing level of CBP increased the flavour preference of the panellists. On the contrary the higher the addition CBP in noodle and chicken meat dishes the worse were the flavour score (p<0.05). It can be concluded that the addition of CBP as flavour enhancer on various dishes can deliver better flavour and ACE-inhibitory activity than the addition of commercial MSG.
Key words: umami, taste, natural, fish, diet

1. Introduction
Hypertension is still recently becomes a major problem in the world. According to WHO, hypertension is the number one cause of death in the world. Data 2010 in the United States shows that 28.6% of adults aged 18 years and over are suffering hypertension [1]. Based on Riskesdas (Basic Health Research) in Indonesia of 2013, 25.8% of adults aged 18 and over are suffering hypertension [2]. Hypertension should be prevented and treated both by medication and dietary modification. Low salt diet program is one form of dietary modification for patients with hypertension. Adding flavor enhancer to the dish may become one effort to reduce the use of salt but still can provide taste sensation to the diet program of people with hypertension. The flavor enhancer enhances the attractiveness at low flavor dishes [3]. Monosodium Glutamate (MSG) is the most favorite flavor enhancer and common used as a flavoring which is a combination of sodium salt and glutamic acid [4]. MSG strengthen salt sensation taste of the dish. The addition of 0.38% MSG at 0.4% to the salt may provide the same saltiness with 0.8% NaCl [5].

Bekasam (soaking salt fish) is a product of preserved freshwater fish using high salinity spontaneous fermentation. Research shows that the bekasam has an inhibitory activity of Angiotensin-I-Converting Enzyme (ACE) which causes hypertension. Fermentation process of milkfish bekasam on 6th day shows 51.77% ACE inhibition [6]. Catfish (Clarias batrachus) is one freshwater fish that can be used as a flavor enhancer. Catfish becomes favorite because of its savory taste, reasonable price, and high nutrient (protein, essential fatty acids, lysine, leucine, and phosphorus) [7,8]. Bekasam has specific flavor that can increase appetite, but due to its uninteresting color and shape so that people is not interested. Therefore, it is necessary to reform the food processing technology of the products to be more interesting and safe to consume for people who suffers hypertension. This study aims to analyze ACE inhibitory activity and organoleptic properties of catfish bekasam powder (CBP) flavor enhancer of several food products.

2. Materials and Methods

2.1. Preparation of Catfish Bekasam Powder (CBP)
The local catfish (Clarias batrachus) weighing + 200-225 g/head was bought from the local market in Semarang. Its viscera and head were discarded then the flesh was split into two parts. Afterward, they were washed and added 10% salt (w/w) and let them for 24 hours in a sealed container (37°C). Next procedure was adding samu (roasted sticky rice) by the ratio of fish: sticky rice 1: 1 then ripened for 6 days. Bekasam that was ready-made then steamed for 40 minutes and drained. The lees was dried with a drying cabinet (45°C for 6 hours) then finely ground. CBP were stored at room temperature inside closed containers which treated with silica gel prior to analysis.

2.2. Sample Preparation of Organoleptic Test
The sample consisted of 4 types of dishes, namely noodles (carbohydrate source), carrot (vegetable source), Champignon (vegetable protein source), and chicken meat (source of animal protein). Raw ingredients were boiled until cooked for 15 minutes then drained. About 50 grams of dishes were weighed and then boiled again in 250 mL boiling water and added salt and complementary spices (garlic powder, onion, bombay onion, white pepper, ginger, galangal and coriander) as the main flavors which each added as much 0.25% of weight of dish. Decoction of dishes was then added by CBP to the treatment group and added MSG for the standard sample. Boiling continued for + 2 minutes.

2.3. Properties Tests of Organoleptic Flavor
The organoleptic test used multiple comparisons. This test assessed the presence or absence and enormity of the difference intensity of the sample organoleptic properties if compared with the standard sample [9]. The organoleptic properties which studied were the flavors. The flavor are the complexity of combination of sensations of aroma, taste and involving the somatosensoric senses [10]. The assessment was conducted by 15 semi-trained panelists. Six samples were presented together in a tray which consist of one standard sample containing 0.38% MSG, one sample of processed dishes without CBP, and four processed dishes samples added with CBP (dose 0.5 %; 0.8%; 1.1%; and 1.4%). Each sample was consecutively coded using 3 (three) digit numbers, namely 189; 492; 278; 760; 251; And the standard samples are coded R. Organoleptic properties used Likert scale [11]. The enormity and intensity differences of flavor were scored by ranging from 1-5 (1 = much better than R and 5 = is much worst than R for the difference assessment of flavor, while 1 = there is no difference and 5 = very much for the difference intensity assessment).

2.4. Sample Extraction
The samples were firstly blended and finely diluted with ratio of 1:3 (volume between sample and deionized water). Samples were then homogenized for ±10 minutes and centrifuged at 20.000 x g for 10 minutes. The supernatant at the top of the tube was piped and filtered. Fresh extracts sample were directly used for analysis.

2.5. Determination of Glutamic Acid Level
Glutamic acid levels were analyzed by spectrophotometric method at 565 nm wavelength. 20 μL sample extracts were mixed with 60 μL buffers, 1 μL enzyme, 5 μL (Nicotinamide adenine dinucleotide) NAD, and 14 μL [3-(4,5-Dimethylthiazol-2-yl)-2,5-Diphenyltetrazolium Bromide] MTT and homogenized. The solution was then incubated for 30 minutes at room temperature. The analysis was conducted for three times repetition. Standard curves used standard glutamate (containing 100 mM glutamate / mL) [12].

2.6. Determination of Angiotensin-I-Converting Enzyme (ACE) Inhibition
ACE inhibitory activity was conducted by Cheung & Chusman (1971) modified spectrophotometric method. 65 μL sample extracts were mixed with 75 μL Hip-His-Leu (HHL) substrate 5 mM in HEPES buffer (pH 8.3 containing 1 M NaCl) and 25 μL of ACE (3.3 mU) enzyme. The solution was incubated at 37°C for 1 hour and then added 125 μL HCl 1 M to stop the reaction. Hypuric acid was extracted and added by 750 μL of ethyl acetate and then solution was homogenized. 200 μL of the solution was piped and evaporated (waterbath 65°C for 30 minutes) until ethyl acetate dried. The formed residual were dissolved by 1 mL of demineralization water. Absorbance was measured on 228 nm wavelength. The analysis was conducted 3 times repetition. Inhibitory activity by the formula: [13]

\[
\% \text{ ACE Inhibition} = \frac{(B / A)}{(B-C)} \times 100\
\]

Information:
A = absorbance with enzymes and ACE enzyme inhibitory compounds
B = absorbance without enzymes and without ACE enzyme inhibitory compounds
C = absorbance without enzymes but with enzyme inhibitors

2.7. Data Analysis
Data were analyzed using SPSS 16.0 for Windows. Data of glutamic acid levels and ACE inhibitory activity were analyzes by variance test using the One-Way Anova by 95% reliable level. It was continued by Duncan Multiple Range Test (DMRT) test. Further analysis by comparing the addition of CBP with standard value (addition 0.38% MSG of processed) to each dishes group using One-Sample T-Test [14]. Properties of organoleptic data were analyzed by Kruskal-Wallis test and continued by Wilcoxon Test (Wilcoxon Sign rank test) with 95% reliable level. The analysis was continued by comparing the treatment of addition CBP doses with standard values to each foodstuff
group using the Wilcoxon test (Wilcoxon sign rank test) [15]. The analysis was performed only for the addition of CBP doses (0%, 0.5%, 0.8%, 1.1%, and 1.4%) and conducted to each foodstuff group.

3. Result and Discussion

3.1. Glutamic Acid Content of Various Dishes

The results showed that CBP contains fewer glutamic acids (6.22 ± 0.008) than fresh bekasam (11.88 ± 0.025). Reduction of glutamic acid levels as a result of processing and drying of fresh bekasam for a long time. Glutamic acid is formed as a result of microorganism’s proteolytic activity during fermentation which breaks the protein complex molecules into smaller components of peptides and amino acids. Miroorganism’s proteolytic growth during fermentation was affected by salt concentration of substrate. The increased of salt concentration on substrate, microorganism’s proteolitic growth would be declined [16,17].

Table 1. Glutamic Acid Content of Various Dishes

| Treatment            | Glutamic Acid Level (mg/g) | Difference (∆) | P-Value |
|----------------------|-----------------------------|----------------|---------|
| **Noodle**           |                             |                |         |
| Dose 0%              | 3.65 ± 0.011a               | 0.28 ± 0.007   |         |
| Dose 0.5%            | 3.93 ± 0.004b               | 0.13 ± 0.012   | 0.0001*(#) |
| Dose 0.8%            | 4.06 ± 0.009c               | 0.10 ± 0.014   |         |
| Dose 1.1%            | 4.16 ± 0.006d               | 0.07 ± 0.002   |         |
| Dose 1.4%            | 4.23 ± 0.004e               |                |         |
| Standard (0.38% MSG) | 4.43 ± 0.003                | 0.0001*(^)     |         |
| **Carrot**           |                             |                |         |
| Dose 0%              | 2.71± 0.003a                | 0.14 ± 0.006   |         |
| Dose 0.5%            | 2.85 ± 0.007b               | 0.11 ± 0.008   | 0.0001*(#) |
| Dose 0.8%            | 2.96 ± 0.006c               | 0.09 ± 0.100   |         |
| Dose 1.1%            | 3.05 ± 0.005d               | 0.13 ± 0.003   |         |
| Dose 1.4%            | 3.18 ± 0.006e               |                |         |
| Standard (0.38% MSG) | 3.32 ± 0.003                | 0.0001*(^)     |         |
| **Champignion**      |                             |                |         |
| Dose 0%              | 5.03 ± 0.005a               | 0.60 ± 0.003   |         |
| Dose 0.5%            | 5.63 ± 0.003b               | 0.59 ± 0.003   | 0.0001*(#) |
| Dose 0.8%            | 6.21 ± 0.005c               | 0.23 ± 0.008   |         |
| Dose 1.1%            | 6.44 ± 0.004d               | 0.33 ± 0.030   |         |
| Dose 1.4%            | 6.77 ± 0.032e               |                |         |
| Standard (0.38% MSG) | 7.32 ± 0.006                | 0.0001*(^)     |         |
| **Chicken Meat**     |                             |                |         |
| Dose 0%              | 9.12 ± 0.009a               | 0.61 ± 0.012   |         |
| Dose 0.5%            | 9.73 ± 0.006b               | 0.67 ± 0.017   | 0.0001*(#) |
| Dose 0.8%            | 10.40 ± 0.024c              | 0.51 ± 0.020   |         |
| Dose 1.1%            | 10.91 ± 0.004d              | 0.45 ± 0.019   |         |
| Dose 1.4%            | 11.35 ± 0.015e              |                |         |
| Standard (0.38% MSG) | 11.84 ± 0.007               | 0.0001*(^)     |         |

* Significant difference average at level of 0.05
(#) One-Way Anova (comparison among treatment)
Table 2. ACE Inhibitory Activity of Various Dishes

| Treatment | ACE Inhibitory Activity (%) | P-Value |
|-----------|-----------------------------|---------|
| Noodle    |                             |         |
| Dose 0%   | 10.42 ± 0.46                |         |
| Dose 0.5% | 9.92 ± 0.51                 | 0.287 (#) |
| Dose 0.8% | 9.87 ± 0.64                 |         |
| Dose 1.1% | 10.02 ± 0.42                |         |
| Dose 1.4% | 9.42 ± 0.50                 |         |
| Standard (0.38% MSG) | 9.56 ± 0.56 | 0.017(*) |
| Carrot    |                             |         |
| Dose 0%   | 10.49 ± 0.72^b              |         |
| Dose 0.5% | 10.49 ± 0.62^b              |         |
| Dose 0.8% | 8.80 ± 0.52^a               | 0.017(#) |
| Dose 1.1% | 9.83 ± 0.58^ab              |         |
| Dose 1.4% | 9.09 ± 0.54^ab              |         |
| Standard (0.38% MSG) | 6.41 ± 0.17 | 0.0001(*) |
| Champignon |                             |         |
| Dose 0%   | 9.02 ± 0.63^ab              |         |
| Dose 0.5% | 8.59 ± 0.63^a               | 0.043(#) |
| Dose 0.8% | 9.83 ± 0.58^bc              |         |
| Dose 1.1% | 10.27 ± 0.64^c              |         |
| Dose 1.4% | 9.93 ± 0.67^bc              |         |
| Standard (0.38% MSG) | 5.93 ± 0.77 | 0.0001(*) |
| Chicken Meat |                             |         |
| Dose 0%   | 7.73 ± 0.41^b               |         |
| Dose 0.5% | 7.86 ± 0.47^b               | 0.032(#) |
| Dose 0.8% | 6.83 ± 0.50^a               |         |
| Dose 1.1% | 6.76 ± 0.38^a               |         |
| Dose 1.4% | 7.73 ± 0.41^b               |         |
| Standard (0.38% MSG) | 6.33 ± 0.41 | 0.0001(*) |

* Significant difference average at level of 0.05
(#) One-Way Anova (comparison among treatment)
(*) One Sample T-Test (comparison between treatment and standard)
Letter notation applies to the average of each foodstuff group
The process of drying the catfish *bekasam* was assumed may alter the molecular structure of L-glutamic acid into α-glutamic acid crystals. The α-glutamic acid crystal would not undergo transformation into β-glutamic acid form as long as it was still in powder form. When the CBP was added to boiling liquid during the processing of the food, the α-glutamic acid crystals dissolve in the boiling liquid and if all the crystalline α-glutamic acid was dissolved it would induce form change to β-glutamic acid. A solution with a high glutamic acid concentration at 45°C would produce β-glutamic acid crystals, whereas a solution with a low glutamic acid concentration would only produce α-glutamic acid crystals in both high and low temperature conditions [18].

As or the initial levels of glutamic acid in foodstuff is known to contribute to glutamic acid levels in the final product. The higher the initial level of glutamic acid in the food stuff, will result in higher glutamic acid level. It is also known that the processed food added 0.38% MSG always shows the highest levels of glutamic acid (Table 1). This is because of the high level of glutamic acid contained in MSG, equal to 733.29 mg/g so as to produce high glutamate levels also on the dishes added MSG [19].

Table 3. The Assessment of Panelists Against Comparative Level of Dish Flavor Against Standard Sample

| Treatment       | Flavor Comparison against R (n=17) | Median Score | P-Value |
|-----------------|-----------------------------------|--------------|---------|
|                 | Better | Equal | Worse | n | % | N | % | N | % | % | n |
| Noodle          |        |       |       |    |   |    |   |    |   |   |    |   |
| Dose 0%         | 1      | 5,9   | 9     | 52,9 | 7 | 41,2 | 4 |
| Dose 0.5%       | 2      | 11,8  | 1     | 5,9  | 14 | 82,4 | 3 |
| Dose 0.8%       | 1      | 0     | 2     | 11,8 | 15 | 88,2 | 3 |
| Dose 1.1%       | 2      | 11,8  | 2     | 11,8 | 13 | 76,5 | 3 |
| Dose 1.4%       | 2      | 11,8  | 3     | 17,6 | 12 | 70,6 | 3 |
| Standard (0.38% MSG) | 0.0001*(*') |
| Carrot          |        |       |       |    |   |    |   |    |   |   |    |   |
| Dose 0%         | 4      | 23,5  | 2     | 11,8 | 11 | 64,7 | 3 |
| Dose 0.5%       | 4      | 23,5  | 4     | 23,5 | 9  | 52,9 | 4 |
| Dose 0.8%       | 4      | 23,5  | 4     | 23,5 | 9  | 52,9 | 4 |
| Dose 1.1%       | 4      | 23,5  | 4     | 23,5 | 9  | 52,9 | 4 |
| Dose 1.4%       | 1      | 5,9   | 0     | 0    | 16 | 94,1 | 3 |
| Standard (0.38% MSG) | 0.0001*(*') |
| Champignon      |        |       |       |    |   |    |   |    |   |   |    |   |
| Dose 0%         | 2      | 11,8  | 2     | 11,8 | 13 | 76,5 | 2 |
| Dose 0.5%       | 2      | 11,8  | 2     | 11,8 | 13 | 76,5 | 2 |
| Dose 0.8%       | 2      | 11,8  | 4     | 23,5 | 11 | 64,7 | 3 |
| Dose 1.1%       | 4      | 23,5  | 3     | 17,6 | 10 | 58,8 | 2 |
| Dose 1.4%       | 3      | 17,6  | 1     | 5,9  | 13 | 76,5 | 3 |
| Standard (0.38% MSG) | 0.0001*(*') |
| Chicken Meat    |        |       |       |    |   |    |   |    |   |   |    |   |
| Dose 0%         | 6      | 35,3  | 4     | 23,5 | 7 | 41,2 | 4 |
| Dose 0.5%       | 2      | 11,8  | 2     | 11,8 | 13 | 76,5 | 2 |
| Dose 0.8%       | 2      | 11,8  | 4     | 23,5 | 11 | 64,7 | 3 |
| Dose 1.1%       | 4      | 23,5  | 3     | 17,6 | 10 | 58,8 | 2 |
| Dose 1.4%       | 3      | 17,6  | 1     | 5,9  | 13 | 76,5 | 3 |
| Standard (0.38% MSG) | 0.0001*(*') |

Notation differences show significant average differences.
3.2. Angiotensin-I-Converting Enzyme (ACE) Inhibitory Activity
The results of the analysis indicated that the CBP had ACE inhibitory activity of 34.025%. This is in accordance with previous study which stated that milkfish bekasam had ACE inhibitory activity of 35.67 + 6.78% on 5th day of fermentation and 51.77 + 8.80% on 6th day of fermentation [6].

Table 4. The Assessment of Panelists of Intensity of The Different Dishes Flavor

| Treatment   | Flavor Comparison against R (n=17) | Median Score* | P-Value |
|-------------|----------------------------------|---------------|---------|
|             | Better  | Equal | Worse |               |               |
|             | n  | %   | N   | %   | N   | %   |               |               |
| Dose 0.5%   | 4  | 23,5 | 5  | 29,4 | 8  | 47,1 | 2             |               |
| Dose 0.8%   | 2  | 11,8 | 3  | 17,6 | 12 | 70,6 | 3             |               |
| Dose 1.1%   | 4  | 23,5 | 0  | 0    | 13 | 76,5 | 3             |               |
| Dose 1.4%   | 1  | 5,9  | 2  | 11,8 | 14 | 82,4 | 3             |               |
| Standard (0.38% MSG) | 0.0001*(^) |

* Significant difference average at level o 0.05
(#{Kruskal-Wallis Test} (median comparison among treatments)
(*{Wilcoxon Signed Rank Test} (comparison between median of treatment and standard)
*) Median Score : 1=so much better; 2=better; 3=just as good; 4=worse; 5=very worse

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The results of the analysis indicated that the CBP had ACE inhibitory activity of 34.025%. This is in accordance with previous study which stated that milkfish bekasam had ACE inhibitory activity of 35.67 + 6.78% on 5th day of fermentation and 51.77 + 8.80% on 6th day of fermentation [6].

Table 4. The Assessment of Panelists of Intensity of The Different Dishes Flavor

| Treatment   | Difference of Flavor Intensity to R (n=17) | Median Score* | P-Value |
|-------------|---------------------------------------------|---------------|---------|
|             | Almost Nothing  | Moderat/ Medium | Many |               |               |
|             | N  | %   | N  | %   | N  | %   | n  | %   |               |               |
| Noodle      | Dose 0%       | 5  | 33,3 | 6  | 40,0 | 4  | 26,7 | 3  |               |               |
|             | Dose 0.5%     | 9  | 60,0 | 1  | 6,7  | 5  | 33,3 | 2  |               |               |
|             | Dose 0.8%     | 9  | 60,0 | 1  | 33,3 | 5  | 6,7  | 2  |               |               |
|             | Dose 1.1%     | 6  | 40,0 | 4  | 26,7 | 5  | 33,3 | 3  |               |               |
|             | Dose 1.4%     | 5  | 33,3 | 6  | 40,0 | 4  | 26,7 | 3  |               |               |
|             | Standard (0.38% MSG) | 0.0001*(^) |
| Carrot      | Dose 0%       | 7  | 46,7 | 5  | 33,3 | 3  | 20,0 | 3  |               |               |
|             | Dose 0.5%     | 9  | 60,0 | 2  | 13,3 | 4  | 26,7 | 2  |               |               |
|             | Dose 0.8%     | 6  | 40,0 | 5  | 33,3 | 4  | 26,7 | 3  |               |               |
|             | Dose 1.1%     | 7  | 46,7 | 5  | 33,3 | 3  | 20,0 | 3  |               |               |
|             | Dose 1.4%     | 4  | 26,7 | 6  | 40,0 | 5  | 33,3 | 3  |               |               |
|             | Standard (0.38% MSG) | 0.0001*(^) |
| Champignon  | Dose 0%       | 13 | 86,7 | 2  | 13,3 | 0  | 0    | 2  |               |               |
|             | Dose 0.5%     | 9  | 60,0 | 3  | 20,0 | 3  | 20,0 | 2  |               |               |
|             | Dose 0.8%     | 3  | 20,0 | 8  | 53,3 | 4  | 26,7 | 3  |               |               |
|             | Dose 1.1%     | 2  | 13,3 | 3  | 20,0 | 10 | 66,7 | 4  |               |               |
|             | Dose 1.4%     | 4  | 26,7 | 4  | 33,3 | 6  | 40,0 | 3  |               |               |
|             | Standard (0.38% MSG) | 0.0001*(^) |
| Chicken Meat| Dose 0%       | 9  | 60,0 | 0  | 0    | 6  | 40,0 | 2  |               |               |
|             | Dose 0.5%     | 8  | 53,3 | 2  | 13,3 | 5  | 33,3 | 2  |               |               |
|             | Dose 0.8%     | 7  | 46,7 | 4  | 26,7 | 4  | 26,7 | 3  |               |               |
|             | Dose 1.1%     | 8  | 53,3 | 6  | 40,0 | 1  | 6,7  | 2  |               |               |
|             | Dose 1.4%     | 6  | 40,0 | 6  | 40,0 | 3  | 20,0 | 3  |               |               |
|             | Standard (0.38% MSG) | 0.0001*(^) |
The results also stated that ACE inhibitory activity decreased to 12.799% after being processed into CBP. This was associated with the process of making CBP in a long time and high temperature. High temperatures would cause enzyme inactivation. Most enzymes exhibited optimum activity at 30-40°C and started denatured at 45°C [20]. Besides catfish bekasam and CBP, ACE inhibitory activity was also studied on various dishes and compared to MSG (Table 2).

Differences of ACE inhibitory activity that fluctuate at the carrots, champignon, and chicken meat dishes were associated with the difference content of specific ACE inhibitory peptides in each foodstuff. Peptides which had optimum ACE inhibitory activity, at the chain base had to have a series of amino carboxyl acids which was composed of three amino acids (Phe-Ala-Pro). The side chains of these three amino acid sequences residue were assumed to interact specifically with the active part of the ACE enzyme (S1, S'1 and S'2). The active part of the enzyme was a carboxyl group attached to a Zn mineral. The presence of a series of three Phe-Ala-Pro amino acid carboxyls in the foodstuff component could increase the ACE inhibition potential by 1000-fold [21].

The low ACE inhibitory activity on dish which was added 0.38% MSG (standard) associated with the active side of the ACE enzyme crystalline structure. Glutamate is the constituent molecule of one of the active sides of the crystal structure of the ACE enzyme, so that MSG is able to provide large amounts of glutamate donors for the activation of the ACE enzyme. The active Zn side of the ACE enzyme structure binds to two molecules of histidine residue and one glutamate molecule [22,23]. The higher the glutamic acid content in a dish, the more ACE enzyme will be activated. Therefore, ACE inhibitory activity will get lower along with increasing amount of activated ACE enzyme.

### 3.3. Flavor Organoleptic Properties of Various Dishes

The result of panelist assessment showed that the higher the dose of addition of CBP on the noodle and chicken meat dishes, the worse flavor was produced. The higher addition of glutamic acid in a dish, would not make the flavor and taste better, but worse. The addition of excessive dose of glutamic acid to dishes, would not make the taste of dish tastier but would worsen the flavor. The dosage of glutamic acid addition which was capable to improve the taste and flavor umami of dish was only 0.1-0.8% by weight of total ingredients [24].

The addition of CBP on dishes might produce flavor that tended to equal even better than standard (Table 3). The results of the varying panelist assessments were attributed to panelist perceptions that vary on the sensory nature of the dish flavor. This variation was due to the differences of panelist flavor thresholds, because each individual had different threshold sensitivity to a particular flavor. When the food was digested, the taste component of the foodstuff contacted with the receptors on taste cell through the taste bud hole so that it caused a taste sensation. While the nervous system in the oral cavity would act as a signal receiver and could identify the taste sensation of different chemical components. The sensitivity of the tongue as a sense of taste in recognizing a flavor depend on the threshold, when the tongue just able to taste of a flavor with the minimum concentration at the flavor component [25]. Sweet and salty flavors also had varying optimum concentrations of each-individual cases. Some individuals might like the sensation of the flavor but the other hand some individuals might also disliked it, depending on the sensitivity belong to each individual [24].

The threshold values and perceptions of a food's taste were often subjective and different for each individual. Moreover, the results of a panelist assessment of the sensory properties of dishes flavor
might also be influenced by the presence of different flavor components of a foodstuff. This resulted in an interaction and rose the different perceptions of taste. Such interactions could be either mutually enhancing or eliminating flavors, flavor modifications, multiplying and flavor adjustments [25].

The result showed that the higher the dosage addition of CBP on Champignon dishes, the greater the intensity of the flavor difference. When it was compared with the standard, the intensity of the Champignon dishes flavor added by the CBP has a moderate / medium difference. It also meant that the Champignon dishes added with CBP have a better flavor than the standard (Table 4). Dishes which were added by CBP had savory flavor that was not much different than the standard. The savory flavor on dishes which were added by CBP, was produced from free glutamic acid contained in dishes, CBP and complementary spices. Savory taste or better known as umami arose because of the natural glutamic acid content in dishes and spices. Although CBP had a mixture of sour, salty, and slightly bitter taste, but when it was combined with herbs and ingredients, the sour and bitter tastes would disappear, and the sweet and salty taste would become sharper. Free glutamic acid not only increases the sweetness and salt perception but also minimizes sour and bitter taste. The addition of glutamic acids in the dishes could also increase the food's palatability through the umami flavor, although in fact umami was not palatable. Glutamic acid had no effect on aroma but can increase overall flavor intensity [26,27].

Panelist perceptions of food flavors may also be influenced by the presence of glutamic acid components in dishes. Basically the food already contains glutamic acid in certain levels and if the dish was added by glutamic acid, certainly it would change the basic flavor of the foodstuff. The higher the addition of glutamic acid, the dish taste would not be better but worse [25].

This study had limitations such as catfish samples used were obtained from the market so it was not known how if the catfish from cultivation (feed type, feeding method, and environment). Moreover, this study had not been supported by research data and physicochemical powder properties standard. This study also had not been able to describe in detail about the flavor because it only used multiple comparison test.

4. Conclusion
Carrots, Champignon, and chicken meat dishes showed varying ACE inhibitory activity, while MSG added dishes showed the lowest ACE inhibitory activity. On the one hand, the addition of CBP on the processed of carrots, champignon, and chicken meat was directly proportional to glutamic acid levels but inversely proportional to ACE inhibitory activity. On the other hand, the addition of MSG to all dishes could increase glutamic acid levels but could significantly reduce ACE inhibitory activity. Based on the results of organoleptic tests, the Champignon dish which was added by CBP resulted in better flavor preference compared to Champignon dish which was added by MSG. Conversely, the higher the dose of addition of CBP to the processed wet noodles and chicken meat, resulting worse flavor.

The researcher suggests that the addition of CBP as much as 0.5% has been able to provide better cooking flavor and show ACE inhibition activity which is higher than MSG. The researcher also suggests the need of further research on in-vivo ACE inhibitory activity, identification of appropriate use of processing technology to optimize the catfish bekasam typical flavor as a flavor enhancer of food, identification of ACE-inhibiting peptides and the descriptive of organoleptic properties testing.

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