

# Studies on processing and storage of eel, *Anguilla japonica*

## 1. Changes in free amino acids during processing

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뱀장어의 加工 및 貯藏에 關한 研究

第 1 報. 加工중의 유리아미노산의 變化

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뱀장어, *Anguilla japonica*, 를 原料로 하여 焙燒 工程과 天日乾燥중의 유리아미노산 變化를 實驗하였다. 生原料와 加工工程중 다 같이 17種의 유리아미노산이 分離, 同定되었다. 生原料의 유리아미노산중 含量이 많은 것은 taurine, histidine, glycine 그리고 alanine 으로 이들 4種 아미노산이 全 유리아미노산의 66.1% 를 차지 하였으며 arginine, tyrosine, phenylalanine 및 proline 은 痕跡量이었다.

뱀장어 加工중 유리아미노산 組成에는 變化가 없었다. 焙燒試料의 유리아미노산 중에는 proline, glutamic acid, histidine, alanine 및 taurine 이 많았고 lysine, isoleucine, serine, tyrosine 은 含量이 적었으며 arginine 은 痕跡량에 지나지 않았다. 天日乾燥의 경우 유리아미노산중에는 glutamic acid, taurine,

leucine, histidine, valine 그리고 lysine 의 含量이 많았으며 arginine 과 proline 은 痕跡量이었다.

加工중의 유리아미노산의 量的 變化로는 焙燒工程중 約 13%, 天日乾燥중 約 57% 그 含量이 增加하였다. 焙燒 工程중 proline 이 특히 많이 增加하였고 그 다음으로 glutamic acid, aspartic acid, phenylalanine 그리고 leucine 이 많이 增加하였으며 taurine, histidine 및 glycine 은 많이 減少하였다. 天日乾燥중에는 leucine, valine, glutamic acid, lysine, isoleucine 등이 많이 增加하였고 glycine 과 histidine 은 특히 많이 減少하였다.

엑스분 窒素중 유리아미노산 窒素가 차지하는 比率 은 生試料, 焙燒試料, 乾燥試料가 모두 12~24% 정도 였다.

### Introduction

Eel, *Anguilla japonica*, has been esteemed as one of the nourishing and medicinal sea foods since a long time ago.

Several papers with food scientific characteristics on eel could be found: report on the composition of free amino acid by Konosu et al.<sup>1)</sup>, studies on fatty acid composition by Junko and Noguchi<sup>2)</sup> and Ha<sup>3)</sup> reports on histological changes by Song et al.<sup>4)</sup>, and study on taste compounds by Yang<sup>5)</sup>

However, there are very few studies on physicochemical changes during processing.

This study is attempted to establish the basic data on the changes of physicochemical characteristics of eel. The changes of free amino acids in eel which received the process of roasting and sun-drying were determined.

## Materials and methods

### 1. Preparation of samples

Eel, *Anguilla japonica* (average body length 52.5 cm, body weight 385 g), caught in a fish farm of Sung-san Fisheries High School were purchased on July 20th 1981 in alive conditions and carried to the laboratory. Then killed instantly and make fillet after removal of the head, intestines, and backbone.

The sun-dried samples were made by sun-drying of the fillets on an iron sieve for 48 hours. During dehydration the fillets were put in a vinyl film and stored at refrigerator at night and sun-dried at day-time. The roasted samples were made by heating for 3 minutes on an electric heater after the fillets were wrapped in a grill.

### 2. Experimental methods

#### (1) Chemical components

Moisture, crude protein, crude lipid and crude ash were determined by general methods and total sugar by Somogyi's method.<sup>6)</sup>

#### (2) Free amino acids

Approximately 5g of sample weighed accurately was blended with 80ml of 1% picric acid solution in a homogenizer. The mixture was diluted to 100ml and centrifuged at 4,000 rpm for 15 minutes. A certain amount of supernatant was introduced on the Dowex 2×8 resin column (chloric form, 100-200 mesh:  $\phi$  2×3 cm) to eliminate the picric acid and the resultant eluate was diluted to 50ml with distilled water. A 20ml portion of the dilution was taken to be absorbed in Amberlite IR-120 resin column (H<sup>+</sup> form, 100-200 mesh:  $\phi$  1.5×5 cm) and washed with 150ml of distilled water. The amino acids were eluted by washing the column with 120ml of 2N ammonia water and the eluate was evaporated until dryness in a rotary evaporator under reduced pressure. The residual fraction was made into 25ml with citrate buffer solution (pH 2.2), then stored at -30°C after ampouling. Amino acids were quantitatively determined by amino acid autoanalyzer (JLC-6AH, No. 610) according to the method of Spackman et al.<sup>7)</sup>

#### (3) Extractive nitrogen (Ex-N)

A 4 to 5g of sample was blended with 80ml of 1% picric acid solution in a homogenizer for 15 minutes. The resultant mixture was diluted to 100ml and centrifuged at 4,000 rpm for 15 minutes. A 20ml portion of the supernatant was introduced onto the column of Dowex 2×8 (chloric form, 200-400 mesh:  $\phi$  2×3 cm) to eliminate the picric acid. The resultant eluate was diluted with distilled water to 50ml and the extractive nitrogen content was determined by semi-micro Kjeldahl method.

## Results and discussion

The contents of moisture, crude protein, crude lipid, crude ash and total sugar of eel during processing were shown Table 1.

As shown in Table 2, 17 species of amino acids including lysine were isolated from the extracts of raw material. In the extracts of the raw samples, contents of taurine, histidine, glycine and alanine were high and followed by threonine, glutamic acid, lysine, serine and methionine in the order named while the contents of leucine, valine, isoleucine and aspartic acid were low and arginine, proline, tyrosine and phenylalanine were only traces in content. The great portion of free amino acid was composed of taurine, histidine, glycine and alanine marked 22.6%, 21.9%,

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11.3% and 10.3% respectively and their overall content was 66.1% of its total free amino acids.

In general, free amino acid composition of animal muscle is quite different from species to species and some kinds of amino acids occupy the most part of total amino acids<sup>8-11)</sup>. Komata et al.<sup>12)</sup> and Lee<sup>13)</sup> reported such amino acids as glycine, alanine and proline occupied a large portion of total free amino acids in many invertebrate animals. Fujita et al.<sup>14)</sup> reported that taurine, glycine and arginine were abundant in the free amino acid of adductor muscle and Konosu et al.<sup>15)</sup> also reported that taurine, glycine, alanine, glutamic acid and arginine were dominant in the free amino acid of short-necked clam. On the other hand, Konosu and Maeda<sup>16)</sup> reported that the content of taurine, arginine and glycine were dominant in the extracts of abalone.

Table 1. Changes in chemical composition of eel during processing (g/100g)

Sample	Moisture	Crude protein	Crude lipid	Crude ash	Total sugar
Raw	77.6	10.2	10.4	1.7	0.2
Roasted	59.2	19.9	18.2	2.2	0.3
Sun-dried	26.8	41.7	26.6	3.2	0.5

Table 2. Changes in free amino acids of eel during processing (on dry basis)

Amino acids	Raw			Roasted			Sun-dried		
	mg%	% in total a. a.	N-mg%	mg%	% in total a. a.	N-mg%	mg%	% in total a. a.	N-mg%
Lys	45.9	5.7	8.8	27.0	3.0	5.2	121.1	9.5	23.2
His	177.1	21.9	48.0	115.9	12.7	31.4	125.1	9.9	33.9
Arg		trace			trace			trace	
Tau	182.4	22.6	20.4	69.3	7.6	7.8	144.9	11.4	16.2
Asp	8.5	1.1	0.9	44.8	4.9	4.7	15.3	1.2	1.6
Thr	61.9	7.6	7.3	34.4	3.8	4.0	24.1	1.9	2.8
Ser	36.3	4.5	4.8	23.3	2.6	3.1	5.1	0.4	0.7
Glu	52.3	6.5	5.0	121.5	13.3	11.6	158.5	12.5	15.1
Pro		trace		142.3	15.6	17.3		trace	
Gly	91.7	11.3	17.1	40.5	4.5	7.6	17.0	1.3	3.2
Ala	83.2	10.3	13.1	79.1	8.7	12.4	98.3	7.7	15.4
Val	13.9	1.7	1.6	39.3	4.3	4.7	123.1	9.7	14.7
Met	27.7	3.4	2.6	46.0	5.1	4.3	62.9	5.0	5.9
Ile	9.6	1.2	1.0	23.9	2.6	2.6	83.7	6.6	8.9
Leu	18.1	2.2	1.9	50.3	5.5	5.4	142.8	11.2	15.2
Tyr		trace		20.2	2.2	1.6	73.5	5.8	5.7
Phe		trace		33.1	3.6	2.8	74.5	5.9	6.3
Total	808.6	100.0	132.5	910.9	100.0	126.5	1269.9	100.0	168.8

Konosu et al.<sup>17)</sup> reported that glycine and alanine occupied approximately 50% of the total free amino acid in the extracts of boiled crabs and Lee<sup>9)</sup> also reported such amino acids as proline, arginine and taurine was abundant in the extracts of squid marked 27.3%, 21.3%, and 18.6%

respectively and their overall contents were 67.2% of the total free amino acids. Lee<sup>13)</sup> analyzed the extracts of dried Gae-bul and found that glycine and alanine were dominant covered 58.5% and 18.6% of the total free amino acids and described these amino acids as the most predominant sweet taste compounds of dried Gae-bul. Konosu<sup>18)</sup> analyzed the free amino acid of abalone and the results of omission test showed that there were little changes in taste when taurine and arginine which was high quantity were removed while sweet taste and meaty taste were obviously decreased when glycine was removed.

From these results it is known that the amount of free amino acid in some species are high and occupied the great portion of total free amino acid. Taurine, histidine, glycine and alanine is abundant in the extracts of eel so that it is presumed these free amino acids play an important role for the characteristics of taste.

As shown in Table 2, no qualitative changes of free amino acids composition in the extracts of eel during the sun-drying and roasting were detected in this experiment. Lee<sup>9)</sup> and Lee et al.<sup>10)</sup> reported that no qualitative changes of free amino acids composition in the extracts of mackerel, jack mackerel, squid and allaska pollack during drying process were detected. Lee et al.<sup>19)</sup> also reported same result during Gul-bi processing.

In free amino acids in the extracts of the roasted sample, proline, glutamic acid, histidine, alanine and taurine was abundant with the contents of 15.6%, 13.3%, 12.7%, 8.7% and 7.6% respectively and the summed contents of these amino acids occupied 57.9% of the total free amino acid and followed by leucine, methionine, aspartic acid, glycine, valine, threonine and phenylalanine in the order named while lysine, isoleucine, serine and tyrosine were low and arginine was only trace in content. In free amino acids in the sun-dried sample, glutamic acid, taurine, leucine, histidine, valine and lysine were dominant with the contents of 12.5%, 11.4%, 11.2%, 9.9%, 9.7% and 9.5% respectively and their overall content was 64.2% of the total free amino acid and followed by alanine, isoleucine, phenylalanine, tyrosine and methionine in these named order while threonine, glycine, aspartic acid and serine were low in its content and proline was only a trace.

The quantitative changes of free amino acids during processing of eel, the content was increased up to 13% during roasting and 57% sun-drying. Proline increased in particularly large amount during roasting and glutamic acid, aspartic acid, phenylalanine and leucine also increased abundantly, and followed by valine, tyrosine, methionine and leucine in the order named. Taurine, histidine and glycine were decreased severely, and followed by threonine, lysine, serine in the order while arginine showed no changes. During sun-drying leucine, valine, glutamic acid, lysine and isoleucine increased abundantly and followed by phenylalanine, tyrosine, methionine, alanine and aspartic acid in the order named and glycine and histidine decreased in a large amount, and followed by taurine, threonine and serine but proline and arginine showed no changes.

Lee<sup>9)</sup> reported that during sun-drying and hot-air drying, leucine, lysine, arginine, glutamic acid and alanine increased abundantly in mackerel but they kept steady state or showed only concentration by dehydration in jack mackerel and squid so the author presumed that the increase of free amino acids might be due to autolysis during dehydration, and that autolytic enzymes in different fishes might result in different levels of free amino acid content in the final product. Manita et al.<sup>20)</sup> determined the changes in free amino acids of mackerel during the storage of 48 hours at 45°C in anaerobic condition and reported that glycine, alanine, aspartic acid, leucine and glutamic acid were increased in a large quantity and lysine, threonine, isoleucine, phenylalanine and serine also increased in the amount of comparative abundance. Konosu and Hashimoto<sup>8)</sup> analyzed free amino acids variation during processing of Katsuo-bushi and reported the decrease of taurine, lysine and serine and Lee et al.<sup>10)</sup> reported lysine, serine and taurine were decreased while the other free amino acids were increased during dehydration of allaska pollack.

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As shown in Table 3, the total free amino acid nitrogen in the raw sample was 12.1% in the muscle extractive nitrogen. The ratio of total free amino acid nitrogen to the muscle extractive nitrogen was 16.8% in the roasted sample and 24.2% in the sun-dried. Several papers with this ratio could be found : in short-necked clam and clam it was both 23%<sup>15)</sup> and 65.6% in abalone<sup>16)</sup>, approximately 50% in sea mussel,<sup>22)</sup> 51.7% in *Corbicula elatior*,<sup>23)</sup> 43.7% in mackerel,<sup>9)</sup> 48.5% in yellow tail, 18.9% in common carp, 25.1% in crucian carp, 23.7% in Korean snake head and 12.2% in Korean catfish.<sup>5)</sup>

**Table 3. Changes in nitrogenous compounds of eel during processing (mg%, on dry basis)**

Compounds	Raw	Roasted	Sun-dried
Extract-N	1092.8	754.2	696.8
Free amino acids-N	132.5	126.4	168.9
Ammonia-N	47.4	50.6	42.5

In this experiment, the ratio of raw material and the products was in the range of 12-24%. This showed similar tendency to fresh-water fishes, however this was much lower rate compare to shellfishes or dark-muscle fishes.

### Summary

Changes in free amino acids of eel, *Anguilla japonica*, during roasting and sun-drying were examined.

17 species of free amino acids were isolated from the extracts of raw material and its products.

The free amino acid composition of the raw, taurine, histidine, glycine and alanine were dominant and these 4 species of amino acids were composed of 66.1% of the total free amino acid while arginine, tyrosine, phenylalanine and proline were trace in content.

During processing no changes were detected in free amino acids composition. In the roasted, proline, glutamic acid, histidine, alanine and taurine were dominant while lysine, isoleucine, serine and tyrosine were poor and arginine was trace amount. In the sun-dried, glutamic acid, taurine, leucine, histidine, valine and lysine were abundant while arginine and proline were trace in its content.

In the quantitative changes in free amino acids during processing, the roasted sample showed the increase of approximately 13% and the sun-dried showed the increase of 57%. Proline increased in particularly large amount during roasting, and glutamic acid, aspartic acid, phenylalanine and leucine also increased abundantly while taurine, histidine and glycine were decreased severely. During sun-drying leucine, valine, glutamic acid, lysine and isoleucine were increased in a large amount while glycine and histidine were decreased in particularly large amount.

The total free amino acids nitrogen in the extractive nitrogen was in the range of 12-24% in the raw, the roasted and the sun-dried samples.

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