Studies on the Volatile Fatty Acids and Volatile Bases in “Shiokara”-II. Some Changes in the Process of the Ripening of Ika-Shiokara

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In the previous paper¹⁾, the authors investigated the acidic and basic fractions of Shiokara from commercial sources and detected a number of these substances. Acetic acid, propionic acid and iso-butyric acid were found as the main components in the acidic fraction, and ammonia, iso-butylamine and an unknown substance in the basic fraction.

The present paper deals with the appearance and the variation of these odoriferous components in the process of ripening of Ika-Shiokara.

Experimentals

Processing of Ika-Shiokara. Ten kilograms of cuttle-fish-meat was used for the processing as Shiokara. Two kilograms of common salt and 500 g of the hepato-pancreas of the cuttle-fish was added to this meat which were mixed thoroughly, and stored at 20°C for 40 days. Throughout the whole term the Shiokara was stirred thoroughly once a day to enable it to ripen properly. After 10, 20, 30 and 40 days, volatile acidic and basic components were examined with gas-chromatography. The procedure of the processing mentioned above is nearly equal to that of commercial processing²⁾.

Determination of volatile fatty acids (VFA) and volatile basic nitrogen (VBN). Thirty grams of Shiokara was subjected to steam distillation, and passed through a definite volume of 0.1 N-sodium hydroxide solution, and 0.1 N-hydrochloric acid solution in order to determine VFA and VBN. VFA was calculated as acetic acid.

Gas-chromatographic analysis of volatile substances. Five hundred grams of Shiokara was used for the analysis of the volatile components. For collecting volatile components and fractionation, the same technique as adopted in the previous experiment¹⁾ was followed. Gas-chromatography was carried out under the same conditions as those of the previous report¹⁾, too. The identification of peaks was carried out in order to compare them with those of authentic samples.

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Abbreviation used: VFA, volatile fatty acids; VBN, volatile basic nitrogen.

Results and Discussion

Table 1. Variations of volatile fatty acids and volatile bases during the ripening process of Ika-Shiokara.

Storage time (days)	Volatile fatty acids* (mg%)	Volatile basic nitrogen (mg%)
0	18.9	0.7
10	31.5	4.0
20	34.5	6.7
30	37.8	10.8
40	56.0	9.4

* Volatile fatty acids were calculated as acetic acid.

The results of experiment are shown in Tables 1-4 and Figs. 1-3. In this study, the amounts of VFA increased continually during the ripening process, though VBN decreased slightly at the 40-th day. Through the whole period of ripening the content of VFA surpassed that of VBN. The results of gas-chromatography of acidic fraction are shown in Tables 2, 3 and Fig. 2. In case of newly made Shiokara 3 peaks

Table 2. Retention time of volatile components detected with gas-chromatography during the ripening process of Ika-Shiokara.

Acidic fraction			Basic fraction		
Peak	tR	Corresponding fatty acids	Peak	tR	Corresponding bases
1	1.00	Acetic acid	1	1.00	Trimethylamine
2	1.25	Propionic acid	2	1.28	Ammonia
3	1.79	iso-Butyric acid	3	1.75	Dimethylamine
4	0.81	Formic acid	4	4.30	iso-Butylamine
5	2.17	n-Butyric acid			
6	2.33	n-Valeric acid			
7	3.35	n-Caproic acid			

Conditions on volatile fatty acids:

Column; diethylene glycol succinate+H₃PO₄

Temperature; 130°C

Carrier gas and its flow rate; He and 40 ml/min.

Conditions on volatile bases:

Column; triethanolamine

Temperature; 90°C

Carrier gas and its flow rate; He and 45 ml/min.

Table 3. Change in the ratio of acidic components during the ripening process of Ika-Shiokara.

Storage time (days)	Ratio of volatile fatty acids (%)						
	Formic	Acetic	Propionic	iso-Butyric	n-Butyric	n-Valeric	n-Caproic
0	—	73	20	7	—	—	—
10	—	73	19	8	—	—	—
20	3	56	21	14	6	—	—
30	2	53	19	23	3	—	—
40	1	50	20	24	2	2	1

Table 4. Change in the ratio of basic components during the ripening process of Ika-Shiokara.

Storage time (days)	Ratio of volatile bases (%)			
	Trimethylamine	Ammonia	Dimethylamine	iso-Butylamine
0	—	18	—	82
10	—	11	—	89
20	3	15	1	81
30	3	17	—	80
40	3	17	—	80

were detected in the gas-chromatogram and they were identical in tR with acetic acid, propionic acid and iso-butyric acid.

After 10 days, the same 3 peaks were detected, though their ratios were different. After 20 days, considerable changes were observed in the components as well as in the ratios. Two new peaks were seen, which were assumed to be formic acid and n-butyric acid, respectively. In this sample the ratio of acetic acid decreased and that of iso-butyric acid increased remarkably. After 30 days, no new component was to be detected, though a considerable changes were seen in the ratios of each component. After 40 days, 7 peaks were detected, though no noticeable changes were found in the main components. The two new peaks were ascertained to be n-valeric acid and n-caproic acid.

The results of the gas-chromatography of the basic fraction are shown in Tables 2, 4 and Fig. 3.

Two peaks were detected in newly-made Shiokara, and they were identical in tR with that of ammonia and iso-butylamine. After 10 days, no change could be found in the components, though their ratios were different slightly. After 20 days, 4 peaks were detected, which were identical with trimethylamine, ammonia, dimethylamine and iso-butylamine. After 30 days, no marked change could be found in the components, though peak 3 (dimethylamine) has disappeared at the 30-th day.

In our study, it seemed that the ripening was nearly completed within 40 days, and only 3 peaks were observed in the gas-chromatogram as basic components, while 10 peaks were detected in Shiokara from commercial sources¹⁾. This discrepancy in the samples may be attri-

Fig. 1. Changes in amount of volatile fatty acids and volatile bases in the process of the ripening of Ika-Shiokara.

- : Volatile fatty acids (calculated as acetic acid).
- : Volatile basic nitrogen.

Fig. 2. Gas-chromatogram of acidic components during the ripening process of Ika-Shiokara.

A: Newly made, B: After 10 days, C: After 20 days,
D: After 30 days, E: After 40 days.

Fig. 3. Gas-chromatogram of basic components during the ripening process of Ika-Shiokara.

A: Newly made, B: After 10 days, C: After 20 days,
D: After 30 days, E: After 40 days.

buted to the relative freshness of the raw material or to some differences in processing conditions.

In this experiment, no notable change could be found in the ratio and the sorts of components in the basic fraction, but the amount of bases increased markedly during the ripening. On the other hand, in acidic fraction, some remarkable changes were perceived in the sorts of acids as well as in their ratios. Total amount of acid in-

creased gradually in the process of ripening. The main components were found to be equal in both; the newly-made and the ripened product. However, the small quantities of other components increased gradually with the lapse of time.

After 10 days, the quantities of volatile acids and bases increased considerably, however, the Shiokara was not palatable fully at the time as reported by the workers³⁾⁻⁵⁾. But, after 40 days, the Shiokara had a good taste involving favorable flavor. So, in view of the organoleptical examination, Shiokara was conceivable to ripen within 40 days.

From these results, it may be assumed that the ripening of Ika-Shiokara commences at about the 20-th day after its start and is finished within 40 days.

Summary

The variation of acidic and basic components during the ripening process of Ika-Shiokara were investigated. The results obtained are as follows:

1. Acetic acid, propionic acid, iso-butyric acid, ammonia and iso-butylamine were observed to be present in newly-made Ika-Shiokara. Other components such as formic acid, n-butyric acid, n-valeric acid, n-caproic acid, trimethylamine and dimethylamine were produced during the later ripening period (Tables 3, 4, Figs. 2, 3).

2. The variation of the components belonging to the acidic fraction were more marked than those of the basic one.

3. It may be assumed that the ripening of Ika-Shiokara begins at about the 20-th day after the commencement of processing and is finished within about 40 days.

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