Characteristic Flavor Components in the Brew of Cooked Clam (*Meretrix lusoria*) and the Effect of Storage on Flavor Formation

Youko Sekiwa,* Kikue Kubota, and Akio Kobayashi

Laboratory of Food Chemistry, Department of Nutrition and Food Science, Ochanomizu University, 2-1-1 Ohtsuka, Bunkyo-ku, Tokyo 112, Japan

The odorants in the brew of cooked clam (*Meretrix lusoria*) were separated by adsorption into Tenax TA resin. An analysis by GC and GC−MS enabled 49 compounds to be unambiguously or tentatively identified. Among them, 2,5-dimethyl-4-hydroxy-3(2*H*)-furanone (DMHF) (caramel-like), maltol (sweet), 2-acetyl-2-thiazoline (roasted), 2-acetylthiazole (popcorn-like), and 3-methylthiopropanal (boiled potato-like) were determined as the main potent odor compounds by an aroma extract dilution analysis. Since the concentrations of these compounds were increased with the longer cooking time, it was considered that the increase of these compounds was deeply related to the formation of the characteristic odor in the brew of cooked clam. Furthermore, by keeping the body of the clam at 4 °C for periods of 30 min, 2 h, 12 h, and 24 h before cooking, the formation of maltol and DMHF was increased significantly with increasing length of storage time, thus enhancing the flavor of the cooked clam.

Keywords: *Cooked clam; potent odorant; Tenax TA adsorption; DMHF; maltol; storage*

INTRODUCTION

Bivalves, with their distinctive odor and taste, are used as a popular food throughout the world. In particular, the clam (*Meretrix lusoria*) has long been enjoyed in Japan and is often eaten as a soup ingredient, since heating creates a pleasant flavor. Recent progress in food processing and distribution enables not only live clams but also clams with shells removed to be made available on the market. Most studies of the flavor of cooked bivalves have focused on taste-active components such as amino acids. Konosu *et al.* (1990) investigated the taste-active components in a soup of cooked scallop and determined some potent compounds by an omission test. As regards odor-active compounds, Nishibori *et al.* (1972) analyzed the volatile compounds of cooked short-necked clam and corbicula. Kubota *et al.* (1991) compared the composition of volatiles from boiled clam, short-necked clam, and corbicula, which were isolated by simultaneous steam distillation and extraction. The odor compounds of heated short-necked clam formed at different temperatures have been identified by Kawai *et al.* (1990). All of these reports indicate that nitrogen- and/or sulfur-containing compounds play important roles quantitatively and qualitatively in the odor of heated bivalves. However, there are no data on the main compounds that organoleptically contribute to the odor, or any investigation to relate the effect of storing the unshelled body on formation of cooked clam odorants. Enomoto (1994) has reported that the composition of the constituents in scallop could be changed by storage under anaerobic conditions and that the taste-active components increased. With clam, it can also be expected that the nonvolatile constituents would change during storage of the body and influence formation of thermally-generated odorants by cooking. In this study, the potent odor compounds formed in the brew by cooking the clam just after shucking were identified, and changes in these main odor compounds were evaluated for various storage times of the shucked clam before cooking.

The column adsorption method, using porous polymer resin packing, has recently been developed for isolation of volatiles, and it has been reported that this method was very effective for recovering volatile compounds in water (Osajima *et al.*, 1989; Wasino *et al.*, 1989; Sugawara *et al.*, 1990). In our preliminary experiment, an aroma concentrate extracted by this method retained the characteristic odor of cooked clam soup. Furthermore, more compounds were detected than could be obtained by steam distillation under reduced pressure. In addition, the aroma extract dilution analysis (AEDA) has been developed by Ullrich and Grosch (1987) for evaluating potent odor compounds. This method, performed by GC−sniffing, can be used to clarify the contribution of individual compounds to the whole odor.

In the current study, the volatile constituents in the brew of cooked clam extracted by the column adsorption method were identified by GC and GC−MS, and the main significant aroma compounds were determined by an aroma extract dilution analysis (AEDA). To investigate the effect of storage before cooking on the flavor in the brew of cooked clam, the concentrations of the main potent odor compounds from shucked clams stored for different periods were also quantified by selected ion monitoring−mass spectrometry.

MATERIALS AND METHODS

Materials. Live clams (*M. lusoria*) from the Mie coast of Japan were purchased from Tokyo central market and kept for 24 h in water including 1% NaCl. The weight of each clam with the shell was about 70 g, the body comprising almost 20% of the whole weight. The clams were separated from shells by hand-shucking for use as samples, which were kept at 4 °C for 30 min, 2 h, 12 h, or 24 h. In this study, the sample was used without homogenizing because shell fish is usually cooked in that way.

Standard Compounds for Identification. Most of the standard compounds were commercially available. 2,5-Dimethyl-4-hydroxy-3(2*H*)-furanone and 5-methyl-4-hydroxy-3(2*H*)-furanone were gifts from T. Hasegawa Co. Ltd., and 2,5-dimethylpyrazine, 2,6-dimethylpyrazine, 2,3-dimethylpyrazine, and 2-acetyl-3-methylpyrazine (10, 11, 12, and 26 in Table 1) were from Ogawa & Co. Ltd. 2-Acetyl-2-thiazoline was a gift from Takasago Co. Ltd.

Isolation of the Volatile Compounds. The clam bodies (200 g) were boiled for 30 min with 200 mL of purified water

S0021-8561(96)00433-5 CCC: $14.00

Table 1. Volatile Compounds Identified in the Brew of Cooked Clam

peak no.	compound	KI[a]	fraction[b]	peak area % on GC[c]
	alcohols			
4	ethanol	922	N	8.44
7	pentanol	1252	N	1.27
13	2-butoxyethanol	1404	N	0.41
17	2-ethylhexanol	1491	N	0.38
22	octanol	1560	N	t[e]
24	2-(2-methoxyethoxy)ethanol	1589	N	t
35	2-(2-butoxyethoxy)ethanol[d]	1797	N	t
37	benzyl alcohol	1881	A, N	1.02
42	phenol	2004	A, N	0.40
	carbonyl compounds			
1	ethanal[d]		N	
2	2-propenal[d]		N	
19	benzaldehyde	1523	A, N	0.95
5	2-pentanone	966	N	5.74
9	3-hydroxy-2-butanone	1286	A, N, B	1.21
29	acetophenone	1656	A	0.38
	acids			
14	acetic acid	1445	A, N, B	1.53
18	formic acid	1492	A	0.68
20	propanoic acid	1532	A	0.48
23	2,2-dimethylpropanoic acid	1579	A, N	t
25	butanoic acid	1628	A	t
31	2-methylbutanoic acid	1670	A	t
32	pentanoic acid	1740	A	t
34	2-methylpentanoic acid	1770	A, N	t
36	hexanoic acid	1847	A, N	1.15
38	2-ethylhexanoic acid	1954	A, N	2.38
39	heptanoic acid	1956	A, N	0.95
44	octanoic acid	2059	A, N	2.55
46	nonanoic acid	2165	A, N	0.28
48	decanoic acid	2273	A, N	0.45
	N,S-containing compounds			
6	pyridine	1187	N, B	0.72
8	2-methylpyrazine	1269	N, B	1.77
10	2,5-dimethylpyrazine	1339	N, B	0.12
11	2,6-dimethylpyrazine	1345	N, B	0.21
12	2,3-dimethylpyrazine	1360	N, B	0.10
26	2-acetyl-3-methylpyrazine[f]	1627	N, B	1.82[f]
27	2-acetylpyrazine[f]	1627	N, B	0.58[f]
41	2-acetylpyrrole	1973	N, B	t
28	2-acetylthiazole	1650	N, B	4.95
33	2-acetyl-2-thiazoline	1760	B	2.39
15	3-methylthiopropanal	1458	N	0.28
21	3-methylthiopropyl alcohol	1539	N	t
	others			
3	ethyl acetate		A, N, B	19.10
16	2-furancarboxaldehyde	1462	N	0.18
30	2-furanmethanol	1660	N	1.02
40	2-methyl-3-hydroxy-4*H*-pyran-4-one (maltol)	1973	A, N	11.10
43	2,5-dimethyl-4-hydroxy-3(2*H*)-furanone (DMHF)	2035	A	1.57
45	4-hydroxy-5-methyl-3(2*H*)-furanone	2114	A	1.63
47	2,3-dihydro-3,5-dihydroxy-6-methyl-4*H*-pyran-4-one[d]	2264	A	1.01
49	2-methyl-3,5-dihydroxy-4*H*-pyran-4-one[d]	2294	A	t

[a] KI, Kovats index. [b] Fraction: Each compounds was identified in the described fraction. A, N, and B refer to acidic, neutral, and basic fractions, respectively. [c] Calculated on the basis of peak area % on GC excluding solvent peak. [d] Tentatively identified only by the mass spectrum. [e] t: These compounds were only detected after the fractionation. [f] These compounds could not be separated clearly by DB-Wax column but were separated by OV-1 at KI values of 1062 (peak 26) or 988 (peak 27). The peak area % of each compound was calculated from GC using OV-1.

in a glass flask by an electric mantle heater. The obtained liquid was filtered through cotton gauze, centrifuged for 10 min at 3000 rpm to remove the solids, and then passed through a 2 cm i.d. × 25 cm bed-height column packed with preconditioned Tenax TA resin (60−80 mesh, GL Science). After the resin was washed with water (50 mL) to remove nonvolatile water-soluble constituents such as sugars and amino acids, the volatile compounds adsorbed on the resin were eluted with diethyl ether (200 mL). The ethereal extract was dried with anhydrous sodium sulfate and then concentrated at 39 °C. Methyl decanoate (6.0×10^{-3} mg) was added to the ethereal extract as an internal standard before concentration. To investigate the volatile constituents in detail the ethereal extract was fractionated to acidic, neutral, and basic fractions. The acidic and basic compounds in the extract were separated with 5% NaOH solution and 5% HCl solution, respectively. After the pH of each solution was adjusted to 2 or 11 with HCl or NaOH, acidic or basic compounds were extracted with diethyl ether. These extracts and the residual ethereal layer were washed with saline, and, after being dried with anhydrous sodium sulfate, each fraction was concentrated.

Gas Chromatography (GC). A Hewlett-Packard 5890 series II gas chromatograph equipped with FID was used for the GC analysis with DB-Wax (60 m × 0.25 mm i.d., J&W) fused silica capillary column. Nonpolar OV-1 (50 m × 0.25 mm i.d., Gasukuro kogyo Inc.) capillary column was also used to separate 2-acetylpyrazine and 2-acetyl-3-methylpyrazine. The oven temperature was held at 60 °C and raised to 180 °C

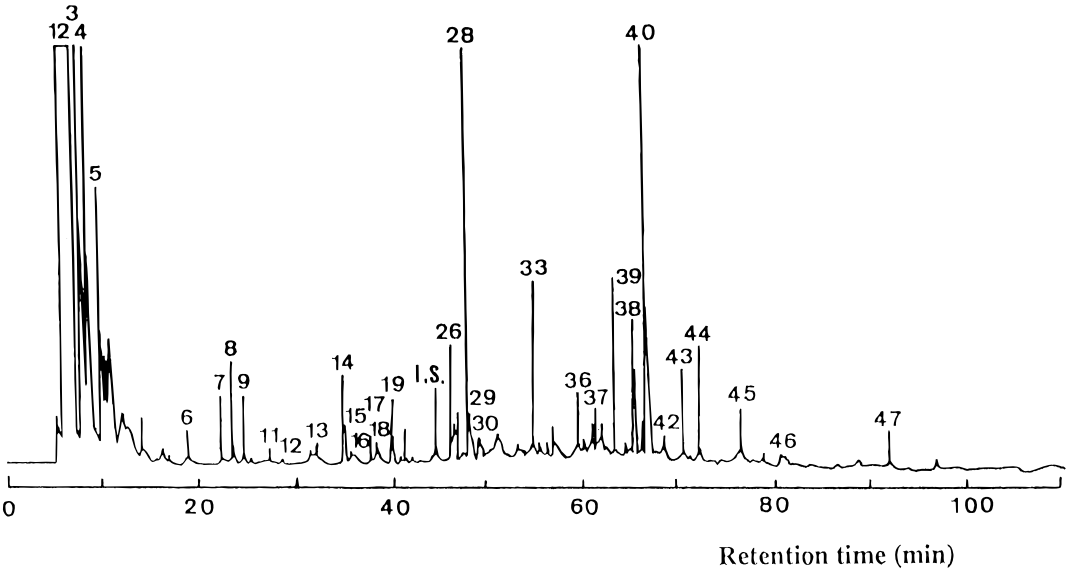

Figure 1. Gas chromatogram of the volatile compounds of cooked clam extracted by the column adsorption method.

at 2 °C/min. The injection and detection port temperatures were both set at 200 °C, and helium at 1.0 mL/min was used as the carrier gas.

Gas Chromatography−Mass Spectrometry (GC−MS). A Hewlett-Packard 5972 series mass selective detector interfaced with a Hewlett-Packard 5890 series II gas chromatograph was used with conditions similar to those used for GC.

Determination of the Potent Odorants. The potent odor compounds of the volatiles from cooked clam were determined with the AEDA method (Ullrich and Grosch, 1987). The GC eluate of the odor concentrate was separated to the FID and to sniffing port immediately in front of the detector port (split ratio = 1:1). After stepwise dilution of the concentrate by adding diethyl ether, GC and GC−sniffing were performed for each diluted sample. The significant odorous peaks in the eluate were determined by evaluating the odor and dilution values (Ullrich and Grosch, 1987).

Quantitative Analysis. The concentrations of maltol, DMHF, 2-acetylthiazole, and 2-acetyl-2-thiazoline in volatile concentrate from stored clam body were analyzed by using the selected ion monitoring (SIM) method. The GC−MS conditions were the same as for the GC−MS analysis described above. The selected ions of each compound was as follows: maltol (*m*/*z* 126, 71, 55, 97); DMHF (*m*/*z* 128, 43, 57, 85); 2-acetylthiazole (*m*/*z* 43, 127, 99, 112); and 2-acetyl-2-thiazoline (*m*/*z* 43, 60, 129, 101). The calibration curve was established with standard solutions containing four kinds of definite amount of each compound. The concentration range of each compound was determined to obtain a good linearity. The experiment was performed three times with 100 g of clam body without cutting, and the changing tendency of the four odorants with the length of storage was investigated.

RESULTS AND DISCUSSION

Yield and Profile of the Aroma Concentrate. The yield of the odor concentrate was 0.37 mg/100 g based on the raw wet weight of the clam bodies. The odor profile showed that the preferred characteristic odor of cooked clam soup involved roasted and sweet notes. A gas chromatogram of the aroma concentrate is shown in Figure 1. Most peaks were identified by comparing their mass spectra and KI values with those of reference or authentic compounds, the results being summarized in Table 1. The concentrations of individual components were shown by peak area % on GC. Some peaks, such as methylbenzenes, detected at early retention times were excluded because they were confirmed to be derived from the resin. Apart from these, 49 compounds were identified as volatile constituents. Among them, almost no carbonyl compounds originating from unsaturated fatty acids were detected in this study. The reason for this observation could be that clam characteristically contains much fewer lipids than other fishes and that the effect of autoxidation or lipoxygenase was very small because the clam used was alive just before heating and, besides, was heated without homogenization. Heterocyclic compounds including pyridine, pyrazines, 2-acetylthiazole, 2-acetyl-2-thiazoline, hydroxypyranones, and hydroxyfuranones, which are commonly thermally generated, were quantitatively detected as main peaks. In particular, 2-acetyl-3-methylpyrazine and 2-acetylpyrazine, each known to have a nutty odor, were detected for the first time in the odor of heated bivalves. Since they could not be separated clearly by DB-Wax column on GC, they were identified using mass chromatography and their concentration ratios were determined by OV-1 column on GC in Table 1. 2-Acetyl-3-methylpyrazine has been identified in the odor of cooked spiny lobster, and it was described as contributing to the nutty/popcorn-like odor of cooked lobster (Cadwallader *et al*., 1995). Since the threshold value for 2-acetyl-3-methylpyrazine is very low (4.0 ppb/water, Fors, 1983), it is also presumed to be important to the cooked clam odor. 2-Methyl-3-hydroxy-4*H*-pyran-4-one (maltol), which is known as a major compound possessing a sweet odor, was detected as the largest peak, its concentration representing 11.1% by GC. Kawai *et al*. (1990) have identified maltol in heated short-necked clam at above 100 °C. Maltol was detected for the first time in this study as a flavor compound of cooked clam. When the acidic fraction was separated, two more hydroxypyranones, 2,3-dihydro-3,5-dihydroxy-6-methyl-4*H*-pyran-4-one and 2-methyl-3,5-dihydroxy-4*H*-pyran-4-one, were detected. Kim and Baltes (1996) reported that maltol and 2-methyl-3,5-dihydroxy-4*H*-pyran-4-one were produced by the decomposition of 2,3-dihydro-3,5-dihydroxy-6-methyl-4*H*-pyran-4-one in an aqueous solution. It is considered that these three hydroxypyranones were formed via the same pathway in the clam. Since the concentrations of 2-methyl-3,5-dihydroxy-4*H*-pyran-4-one and 2,3-dihydro-3,5-dihydroxy-6-methyl-4*H*-pyran-4-one were small and especially because the odor threshold value of

Table 2. Potent Odor Compounds in the Brew of Cooked Clam

odor compound	dilution value (>2)	odor description
2-methylpyrazine	2	nutty, almond-like
2,5-dimethylpyrazine	2	cocoa-like
2,6-dimethylpyrazine	2	nutty
2,3-dimethylpyrazine	2	green
methional	64	boiled potato-like
2-acetyl-3-methylpyrazine	4	nutty, almond-like
2-acetylpyrazine	4	nutty
2-acetylthiazole	16	popcorn-like
unknown a (KI 1730)	4	grassy
2-acetyl-2-thiazoline	256	roasted, popcorn-like
maltol	16	sweet
DMHF	256	sweet, caramel-like
unknown b (KI 2070)	2	boiled beef
unknown c (KI 2282)	8	sweet
unknown d (KI 2289)	8	sour, floral

2,3-dihydro-3,5-dihydroxy-6-methyl-4*H*-pyran-4-one is very high (>200 ppm/orange juice, Fors, 1983), these compounds would be ineffective in the whole odor. Hydroxyfuranones like DMHF and 4-hydroxy-5-methyl-3(2*H*)-furanone were also newly identified as volatile components of cooked clam. We have previously reported that DMHF was the most important odor compound of cooked squid (Kubota *et al*., 1996). Since the odor character of cooked clam resembles that of cooked squid, it seems that DMHF is also one of the most important odor compounds in clam.

Evaluation of the Potent Odor Compounds. To clarify the contribution of each identified compound to the characteristic odor of cooked clam, an aroma extract dilution analysis was carried out. The volatile concentrate was diluted stepwise to 2^n-fold with diethyl ether and then subjected to GC−sniffing. In all, 15 compounds were recognized by sensory evaluation and are shown with their odor profiles in Table 2. Among them, unknown compounds a−d did not appear as GC peaks but were recognized by sniffing. Therefore, these compounds would have had very low threshold values and be undetectable by GC.

Among the identified compounds (Table 2), the main odor-active constituents were 2-acetyl-2-thiazoline, 2-acetylthiazole (popcorn-like), maltol (sweet), DMHF (caramel-like), and 3-methylthiopropanal (methional, boiled potato-like). In particular, DMHF and 2-acetyl-2-thiazoline remained as odor characters after 256-fold dilution. Maltol and 2-acetylthiazole were confirmed up to 16-fold dilution, which is a relatively low level in consideration of their respective quantities in the concentrate. On the other hand, methional was perceived up to 64-fold dilution, in spite of its trace content in the volatile concentrate. The characteristic odor of each of these compounds was generally sweet and roasted and like the whole odor of cooked clam. Therefore, it is concluded that these compounds play important roles as the potent odorants of cooked clam.

Changes in Main Potent Odor Compounds with Heating Time. In order to investigate the formation mechanism of the cooked odor of clam, the quantitative changes of five potent odor compounds, maltol, DMHF, 2-acetylthiazole, 2-acetyl-2-thiazoline, and methional, were examined. The quantities of these compounds at 0, 15, and 30 min after reaching boiling point were calculated as the ratio of the percentage peak area by GC to that of the internal standard and are plotted in Figure 2. As the maximum heating time was only 30 min, similar to a practical cooking time, the yields of each aroma compound did not greatly increase. However, the sweet, roasted, and meaty odors became stronger with increasing cooking time. Maltol, DMHF, 2-acetylthiazole, 2-acetyl-2-thiazoline, and methional increased moderately with increasing heating time, confirming the supposition that they are generated by thermal processes. Among these compounds, the increase in maltol was conspicuous, reaching 4 times the original value within 0−15 min of boiling time. DMHF, which was not found in the concentrate at 0 min, was predominantly formed after 15 min of cooking. As much 2-acetyl-2-thiazoline was generated as internal standard at 0 min, but its final content was only 1.4 times that of the internal standard. On the other hand, 2-acetylthiazole increased from 0.4 times to 2.3 times with 30 min boiling. Mulders (1973) has proposed formation mechanisms for 2-acetyl-2-thiazoline and 2-acetylthiazole from cysteine and α-dicarbonyl compounds and has reported that both of them were formed through the same pathway. Hoffman and Schieberle (1995) have reported that 2-acetyl-2-thiazoline was easily degraded by heating in an aqueous solution. From these reports, it is presumed that 2-acetyl-2-thiazoline was initially produced at a relatively low temperature and then oxidized to 2-acetylthiazole, the final amount of 2-acetylthiazole reaching more than that of 2-acetyl-2-thiazoline. Although the increase of methional was very small, its low odor threshold value already described makes it important for generating the characteristic odor. In conclusion, the formation of the five main potent odorants, maltol, DMHF, 2-acetylthiazole, 2-acetyl-2-thiazoline, and methional, influenced the thermally-generated odor in the brew of cooked clam.

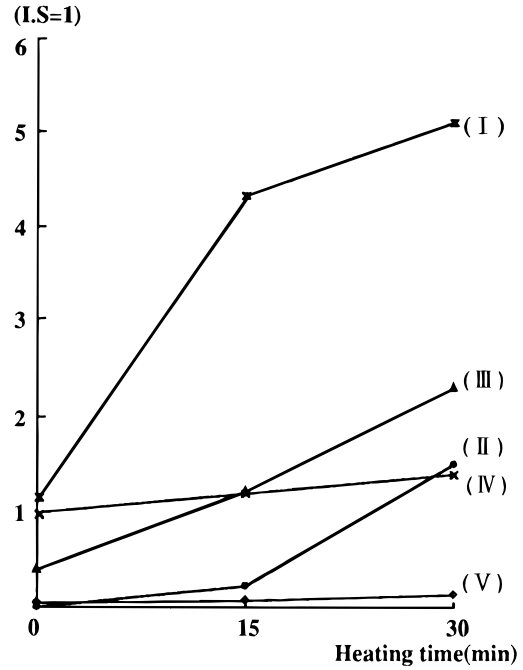

Figure 2. Change of main potent odor compounds with heating time: (I) maltol, (II) DMHF, (III) 2-acetylthiazole, (IV) 2-acetyl-2-thiazoline, and (V) methional.

Effect of Storage of the Clam Body on the Formation of Odorants. Since the cooked flavor in the brew from the clam body kept at 4 °C was organoleptically evaluated to be superior to that obtained just after removing the shell, the concentration of the main odor compounds was analyzed and compared. After the clam bodies had been kept at 4 °C for 30 min, 2 h, 12 h, or 24 h, they were heated for 30 min and the odor compounds were extracted in a manner similar to that

Table 3. Concentrations of Maltol and DMHF in the Brew of Cooked Clam after Storage (4 °C)[a]

odor compound	replication[b]	storage time (h) 0.5	2	12	24
maltol	1	154	160	285	292
	2	131	135	304	321
	3	82	92	163	182
DMHF	1	22	30	48	62
	2	21	26	42	51
	3	12	15	26	35

[a] In ng/g, calculated on the basis of the weight of raw clam. [b] Replications were performed using the samples bought on different days.

described above. The quantities of main potent odor compounds, maltol, DMHF, 2-acetylthiazole, and 2-acetyl-2-thiazoline were measured by SIM analysis. Three replications were performed for their analyses with samples bought on different days. They were calculated on the basis of the wet weight of raw clam. 2-Acetylthiazole and 2-acetyl-2-thiazoline did not show a marked tendency by storage time, that is, they did not increase or smaller amounts were sometimes observed. Though the reason was not clear at present, it was presumed that the low molecular weight degradation products, which would be the precursors of 2-acetyl-2-thiazoline and 2-acetylthiazole, were not produced sufficiently in viscera because the sample was stored at low temperature, 4 °C, and for only 24 h in this study. On the other hand, maltol and DMHF increased with longer storage times, the amount of maltol generated after storage for 24 h being approximately twice that of the 30 min storage. DMHF also increased in proportion to storage time and was about 2.5−3 times greater by storage for 24 h. In Table 3, the results of DMHF and maltol for three different samples are shown. Though a small difference of the amount among samples was observed, almost the same increasing tendency was exhibited. From these results, since each of these compounds had a sweet odor, it was considered that maltol and DMHF would have improved the flavor of cooked clam soup. In particular, since the odor threshold level for DMHF is very low (0.04 ppb/water, Fors, 1983), its increase seems to contribute significantly to the whole flavor of cooked clam. It is well-known that both maltol and DMHF are the major compounds formed from the sugar molecule. As regards to the formation of DMHF, Schieberle (1992) has mentioned that it was produced predominately from fructose-1,6-diphosphate in wheat bread crust. On the other hand, it is known that free sugars, including phosphate sugar, are increased by degradation of glycogen in glycolysis under anaerobic conditions. Therefore, it is expected that the increase in free sugars and their derivatives during storage would influence the formation of maltol and DMHF in clam. The relationship between their formation and free sugars as precursors in the clam will be described in another report.

LITERATURE CITED

Cadwallader, K. R.; Tan, Q.; Chen, F.; Meyers, S. P. Evaluation of the aroma of cooked spiny lobster tail meat by aroma extract dilution analysis. *J. Agric. Food Chem.* **1995**, *43*, 2432−2437.

Enomoto, T. Hotategai no Teimi. *Kagaku to Seibutsu* **1994**, *32*, 765−766.

Fors, S. Sensory properties of volatile maillard reaction products and related compounds. (A Literature Review). In *The Maillard Reaction in Foods and Nutrition*; G. R. Waller, M. S. Feather, Eds.; American Chemical Society: Washington, DC, 1983; pp 185−286.

Hoffman, T.; Schieberle, P. Studies on the formation and stability of the roasted-flavor compound 2-acetyl-2-thiazoline. *J. Agric. Food Chem.* **1995**, *43*, 2946−2950.

Kawai, T.; Ishida, Y.; Kakiuchi, H.; Ikeda, N.; Tsuneya, T. Volatile components of boiled and roasted short-necked clam *Tapes philippinarum*. *Nippon Suisan Gakkaishi* **1990**, *56*, 795−802.

Kim, M.; Baltes, W. On the role of 2,3-dihydro-3,5-dihydroxy-6-methyl-4(*H*)-pyran-4-one in the Maillard reaction. *J. Agric. Food Chem.* **1996**, *44*, 282−289.

Konosu, S.; Watanabe, K.; Koriyama, T.; Shirai, T. Extractive compounds of scallop and identification of its taste-active components by omission test. *Nippon Shokuhin Kogyo Gakkaishi* **1988**, *35*, 252−258.

Kubota, K.; Nakamoto, A.; Moriguchi, M.; Kobayashi, A.; Ishii, H. Formation of Pyrrolidino[1,2-*e*]-4(*H*)-2,4-dimethyl-1,3,5-dithiazine in the volatiles of boiled short-necked clam, clam, and corbicula. *J. Agric. Food Chem.* **1991**, *39*, 1127−1130.

Kubota, K.; Matsukage, Y.; Sekiwa, Y.; Kobayashi, A. Identification of the characteristic volatile flavor compounds formed by cooking squid (*Todarodes pacificus Steenstrup*). *Food Sci. Technol. Int.* **1996**, *2*, 163−166.

Mulders, E. J. Volatile components from the non-enzymatic browning reaction of the cysteine/cystine-ribose systems. *Lebensm. Unters. Forsch.* **1973**, *152*, 193−201.

Nishibori, K.; Kanamitsu, S.; Okamoto, K. Flavor of boiled short-necked clam (*Tapes* (*Amygdala*) *philippinarum*) and corbicula (*Corbicula japonica*). *J. Home Econ.* **1972**, *23*, 27−31.

Osajima, T. Recent advances in food analysis (8). *Nippon Shokuhin Kogyo Gakkaishi* **1989**, *36*, 609−614.

Schieberle, P. Formation of furaneol in heat-processed foods. In *Flavor Precursors*; R. Teranishi *et al.*, Eds.; American Chemical Society: Washington, DC, 1992; pp 164−174.

Sugawara, E.; Ito, T.; Odagiri, S.; Kubota, K.; Kobayashi, A. Comparison of miso aroma components in preparations by different methods. *Nippon Nogeikagaku Kaishi* **1990**, *64*, 171−176.

Ullrich, F.; Grosch, W. Identification of the most intense volatile flavour compounds formed during autoxidation of linoleic acid. *Z. Lebensm. Unters. Forsch.* **1987**, *184*, 277−282.

Washino, Y.; Kubota, K.; Kobayashi, A. Aroma component of dried bonito soup stock. *Nippon Kaseigakkaishi* **1989**, *40*, 265−270.

Received for review June 17, 1996. Accepted December 5, 1996.[⊗]

JF960433E

[⊗] Abstract published in *Advance ACS Abstracts,* February 1, 1997.