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Determination of methylmercury in fish using focused microwave digestion following by Cu^{2+} addition, sodium tetrapropylborate derivatization, *n*-heptane extraction, and gas chromatography–mass spectrometry

Syr-Song Chen [a,b], Shin-Shou Chou [a], Deng-Fwu Hwang [b,*]

[a] *Department of Health, National Laboratories of Foods and Drugs, Taipei, Taiwan*
[b] *Department of Food Science, National Taiwan Ocean University, 2 Pei-Ning Road, Keelung 202, Taiwan*

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Abstract

The analytical procedure for analysis of methylmercury in fish was developed. It involves microwave-assisted digestion with alkaline solution (tetramethylammonium hydroxide), addition of Cu^{2+}, aqueous-phase derivatization of methylmercury with sodium tetrapropylborate, and subsequent extraction with *n*-heptane. The methylmercury derivative was desorbed in the splitless injection port of a gas chromatograph and subsequently analyzed by electron impact mass spectrometry. Optimum conditions allowed sample throughout to be controlled by the instrumental analysis time (near 7 min per sample) but not by the sample preparation step. At the power of 15–30, 45, and 60–75 W, sample preparation time is only 3.5, 2.5, and 1.5 min, respectively. The proposed method was finally validated by the analysis of three biological certified reference materials, BCR CRM 464 tuna fish, NRC DORM-2 dogfish muscle, and NRC DOLT-2 dogfish liver. The detection limit of the overall procedure was found to be 40 ng/g of biological tissue for methylmercury. The recovery of methylmercury was 91.2–95.3% for tuna, 89.3–94.7% for marlin, and 91.7–94.8% for shark, respectively. The detected and certified values of methylmercury of three biological certified reference materials were as follows: 5.34 ± 0.30 μg/g (mean ± S.D.) and 5.50 ± 0.17 μg/g for CRM 464 tuna fish, 4.34 ± 0.24 and 4.47 ± 0.32 μg/g for NRC DORM-2 dogfish muscle, and 0.652 ± 0.053 and 0.693 ± 0.055 μg/g for NRC DOLT-2 dogfish liver, respectively. It indicated that the method was well available to quantify the methylmercury in fish.

Keywords: Fish; Derivatization, GC; Food analysis; Organomercury compounds

1. Introduction

Mercury pollution has become a global problem because of the occurrence from natural and anthropogenic sources and food chain processes. Mercury is a well-known toxic element, especially in the form of methylmercury ($MeHg^+$), which is considerably more toxic than inorganic mercury (Hg^{2+}) [1]. In the environment, $MeHg^+$ is formed by biotic and abiotic methylation of Hg^{2+} and it accumulates in the tissue of fish and other biota [2,3]. Mercury as $MeHg^+$ usually represents more than 85% of total mercury present in fish [4,5]. Mercury poisonings are mainly caused by consumption of contaminated fish through $MeHg^+$ accumulation in the food chain, such as in the case of Minamata disease [6]. As a result, the US Food and Drug Administration (FDA) has set an action level of 1 μg/g (wet mass) for concentration of $MeHg^+$ in fish. Fish containing concentrations of $MeHg^+$ above this level are considered to be hazardous for human consumption and cannot be sold in interstate commerce. Canada and several US states have developed consumption advisories of 0.5 μg/g for $MeHg^+$ in fish [7]. In Taiwan, the guideline level of $MeHg^+$ is set at 2.0 μg/g for migratory fish and 0.5 μg/g for other fish [8]. As public awareness regarding the toxicity and the environmental impact of mercury contamination increases, the demand for suitable analytical methodology to be used on a routine basis in control laboratories needs to be developed.

* Corresponding author. Tel.: +886-2-2462-2192x5103; fax: +886-2-2462-6602.
E-mail address: dfhwang@mail.ntou.edu.tw (D.-F. Hwang).

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The majority of procedures applied in analytical laboratories are based on the classical Westöö procedure [9] that is specific to $MeHg^+$, and on the Magos procedure [10] that implies an operational definition of the inorganic and organic mercury. Despite continuous improvements during the last three decades, the procedures based on these principles have remained time consuming, tedious, and often unreliable, as comprehensively discussed by Emteborg et al. [11]. Nevertheless, they are the basis of the AOAC Official Methods for $MeHg^+$ in fish and shellfish [12].

An alternative popular approach is the species-selective analysis for $MeHg^+$ and Hg^{2+} based on hydride generation [13–15] (or ethylation [17–19]) followed by purge-and-trap thermal desorption cold vapor atomic absorption spectrometry (CVAAS) [13,17], cold vapor atomic fluorescence spectrometer (CVAFS) [18], Fourier transform infrared (FT-IR) and Raman microspectroscopy [14], microwave-induced plasma atomic emission detection (MIP-AES) [15,16] or furnace atomization plasma emission spectrometry (FAPES) [19]. The drawbacks of these procedures include the need for a pretreatment step to liberate mercury from a biological matrix, the need for custom-built instrumentation that is not commercially available and the numerous problems related to the dismutation reactions, formation of artifacts and uncontrolled losses due to the interaction of analytes with the chromatographic stationary phase [17].

The third category of methods imply the use of standard capillary gas chromatographic equipment, either with a split/splitless injector [20] (in the case Hg^+ and $MeHg^+$ are extracted from a sample solution as diethyldithiocarbamate complexes into toluene followed by their butylation with a Grignard reagent to ensure their thermal stability and sufficient volatility), or with a capillary purge-and-trap injector [13,15]. In the former case, the sample preparation procedure is again tedious whereas in the latter case the high cost of the purge-and-trap injector with the temperature programmed gas chromatography (GC) oven and the limited number of commercially available species- or element-selective detectors which accept sample introduction by capillary GC prevent the methods from being applied in routine foodstuff control laboratories.

Pereiro et al. [21] developed a simple and rapid procedure for the simultaneous determination of $MeHg^+$ and Hg^+ in fish reference to combine the advantages of the above-discussed approaches and eliminate their drawbacks. The procedure involved isothermal multicapillary gas chromatography with atomic emission detection after microwave-assisted solubilization and solvent extraction. When gas chromatography–mass spectrometry (GC–MS) was used to substitute atomic emission detection, the addition of Cu^{2+} in the digestion solution would elevate the recoveries of methylmercury level. Hence, GC–MS after microwave-assisted digestion, propylation and solvent extraction to determine methylmercury in fish reference was described in this study.

2. Experimental

2.1. Reagents

All chemicals used were of analytical-reagent grade unless stated otherwise. Sodium tetra-*n*-propylborate ($NaBPr_4$, 98% purity) was purchased from GALAB (Geesthacht, Germany). Tetramethylammonium hydroxide (TMAH, 25% in water) was purchased from Fluka (USA). Acetic acid (suprepure grade), sodium acetate, copper acetate, potassium hydroxide, *n*-hexane, *n*-heptane, isooctane, and tetrahydrofuran (THF) were purchased from E. Merck (Darmstadt, Germany).

The derivatization solution was prepared by dissolving 1 g of sodium tetrapropylborate in 100 ml of 2% potassium hydroxide solution. The solution was stored in a refrigerator and protected from light. Buffer solution was prepared by dissolving 1 M sodium acetate in water and adjusting the pH to 4.0 with concentrated acetic acid. Copper solution was prepared by dissolving 40 mM copper acetate in water. Milli-Q quality water (Millipore) was used throughout.

2.2. Calibration, biological reference materials and recovery test samples

Methylmercury(II) chloride standard solution ($MeHg^+$, 1000 μg/ml as Hg) was purchased from Alfa Aesar (USA). Working calibration solution of methylmercury was prepared by appropriate dilution of methylmercury standard solution with water and stored for a maximum of 1 week.

One certified reference materials CRM 464 tuna fish (5.50 ± 0.17 μg/g $MeHg^+$) obtained from the Community Bureau of Reference (CBR), and two certified reference materials, DORM-2 dogfish muscle (4.47 ± 0.32 μg/g $MeHg^+$) and DOLT-2 dogfish liver (0.693 ± 0.055 μg/g $MeHg^+$), obtained from the National Research Council of Canada (NRCC), were used to validate the proposed method. The other laboratory tuna fish sample containing 1.98 ± 0.15 μg/g of $MeHg^+$ detected by Westöö method [9].

For recovery test of methylmercury in fish, a sample of 0.5 g of fish muscle (tuna, marlin, and shark) was placed in a microwave vessel and spiked 1.0 ml of methylmercury standard solution (250, 500, and 1000 ng/ml). The sample was stored in a refrigerator overnight before analysis.

2.3. Devices and instrument

Focused microwave digester Microdigest 3.6 (2.45 GHz, maximum power 300 W) was the product of Porlabo (France). Gas chromatograph–ion trap mass spectrometer Saturn 2200 was the product of Varian (USA). Capillary gas chromatograph columns CP-SIL 1 CB (100% dimethylpolysiloxane), and CP-SIL 8CB (5% diphenyl–95% dimethylpolysiloxane), and CP-SIL 24 CB (50% diphenyl–50% dimethylpolysiloxane) lowbleed/MS (30 m ×

Table 1
Parameters of the analytical system

Microwave digester	Porlabo Microdigest 3.6
Irradiation power	45 W
Irradiation time	2.5 min
GC–ion trap	Varian Saturn 2200
Column	CP-SIL 8 CB lowbleed/MS (5% phenyl–95% dimethylpolysiloxane; 30 m × 0.25 mm i.d., d_f 0.25 μm)
Injection technique	Splitless
Injection volume	1 μl
Injection temperature	260 °C
Temperature program	60 °C (1 min), 20 °C/min, 280 °C (10 min)
Carrier gas (flow rate)	He (1 ml/min)
Xferline temperature	280 °C
Trap temperature	170 °C
Quantitative ion	217
Reference spectra (*m*/*z*)	214–215

0.25 mm i.d. with a 0.25 μm film) were the product of Chrompack (The Netherlands).

All volumetric bottles and other glassware were Pyrex brand. Before use, they were washed with detergent and water, soaked with 50% (v/v) nitric acid overnight, rinsed with water, and dried.

2.4. Procedure

For a sample of 0.5 g tissue, 5 ml of TMAH solution were placed in a microwave vessel and used a reflux condenser to prevent evaporation losses, than exposed to the microwave field at 45 W for 2.5 min. After microwave digestion, samples were neutralized by acetic acid and transferred into a 40 ml Pyrex vial with a Teflon cap and diluted with 20 ml water and 1 ml of 40 mM copper solution. The pH was adjusted to 4.0 using 5 ml of 1 M acetate buffer. One milliliter of the 1% sodium tetrapropylborate/potassium hydroxide solution was added by syringe, mixed and stayed 10 min at ambient temperature. Then, 2 ml of *n*-heptane were added and shaken for 10 min. An aliquot of the supernatant was analyzed by GC–MS. Instrument parameters used in this study are listed in Table 1.

3. Results and discussion

3.1. Optimization of microwave-assisted digestion

Using the conditions in Section 2.4 and changing the microwave digestion conditions, 5 ml of TMAH was spiked with methylmercury (1 μg as Hg) and exposed to 15–30, 45, or 60–75 W irradiating power for various heating times. After irradiation, the sample solution was diluted with 20 ml water and methylmercury content was determined as described earlier. Fig. 1A shows the percentage of methylmercury content after different irradiating power and times. A highest methylmercury content was achieved after 3.5 min heating of irradiation at 15–30 W, 2.5 min of irradiation at 45 W, or 1.5 min of irradiation at 60–70 W. Only 72% and 61% of the initial signal was obtained after 3.5 min heating of irradiation at 60 and 75 W. And 95 and 90% of the initial signal was obtained after 3.0 and 3.5 min heating of irradiation at 45 W. The analytical signal obtained was strongly dependent on the heating time and irradiating power. The low content of methylmercury might be caused by long heating times or high irradiating power. This is similar to the report of Tseng et al. [13].

The methylmercury extraction recovery was investigated using one biological certified reference material, CRM 464 tuna fish and a laboratory tuna fish sample. Fig. 1B and C show the percentage of methylmercury content after different irradiating power and heating times. The methylmercury content of CRM 464 tuna fish and laboratory tuna fish sample increased with heating times setting irradiation at 15 and 30 W, and decreased with heating times setting irradiation at 60 and 75 W. The highest methylmercury content was obtained after 2.5 min of irradiation at 45 W. Results showed high irradiation power provided high energy and reduced heating time to extract, but methylmercury might decompose due to high irradiating power. The condition of 45 W irradiation power and 2.5 min heating time could have the optimal extraction efficiency without decomposing methylmercury.

3.2. Optimization of the derivatization parameters

Using the same conditions of Section 2.4, the effect of cupric ion on the derivatization yield of methylmercury was examined by adding 1 ml of 0, 10, 20, 40, 60, and 80 mM copper acetate solution. In agreement with Olson et al. [23], we found the cupric ion could compete with methylmercury already combined with the sulfuryl amino acid in fish protein and resulted in release of methylmercury. Therefore, more than 90% methylmercury content was released from fish sample by adding 1 ml of 40 mM copper acetate solution.

Furthermore, three derivatization agents including water, 2% KOH solution and tetrahydrofuran were tested for effect of solvent on methylmercury detection of CRM 464 tuna fish, laboratory tuna fish sample, and methylmercury standard solution. Fig. 2 shows no significant difference in methylmercury detection peak for mercury standard solution. Water and 2% KOH solution did not affect the propylation of methylmercury in CRM 464 tuna fish, laboratory tuna fish sample, and methylmercury standard solution. THF did not affect the propylation of methylmercury in standard solution, but reduced the propylation of methylmercury in CRM 464 tuna fish and laboratory tuna fish sample. The reason seemed to be due to the reaction of THF with fish protein to retard the release of methylmercury in fish protein. The true mechanism needs further study.

The influence of pH on the derivatization of mercury species was examined by using 5 ml of pH 3.0, 3.5, 4.0, 4.5, 5.0, and 5.5 buffer solutions in CRM 464 tuna fish,

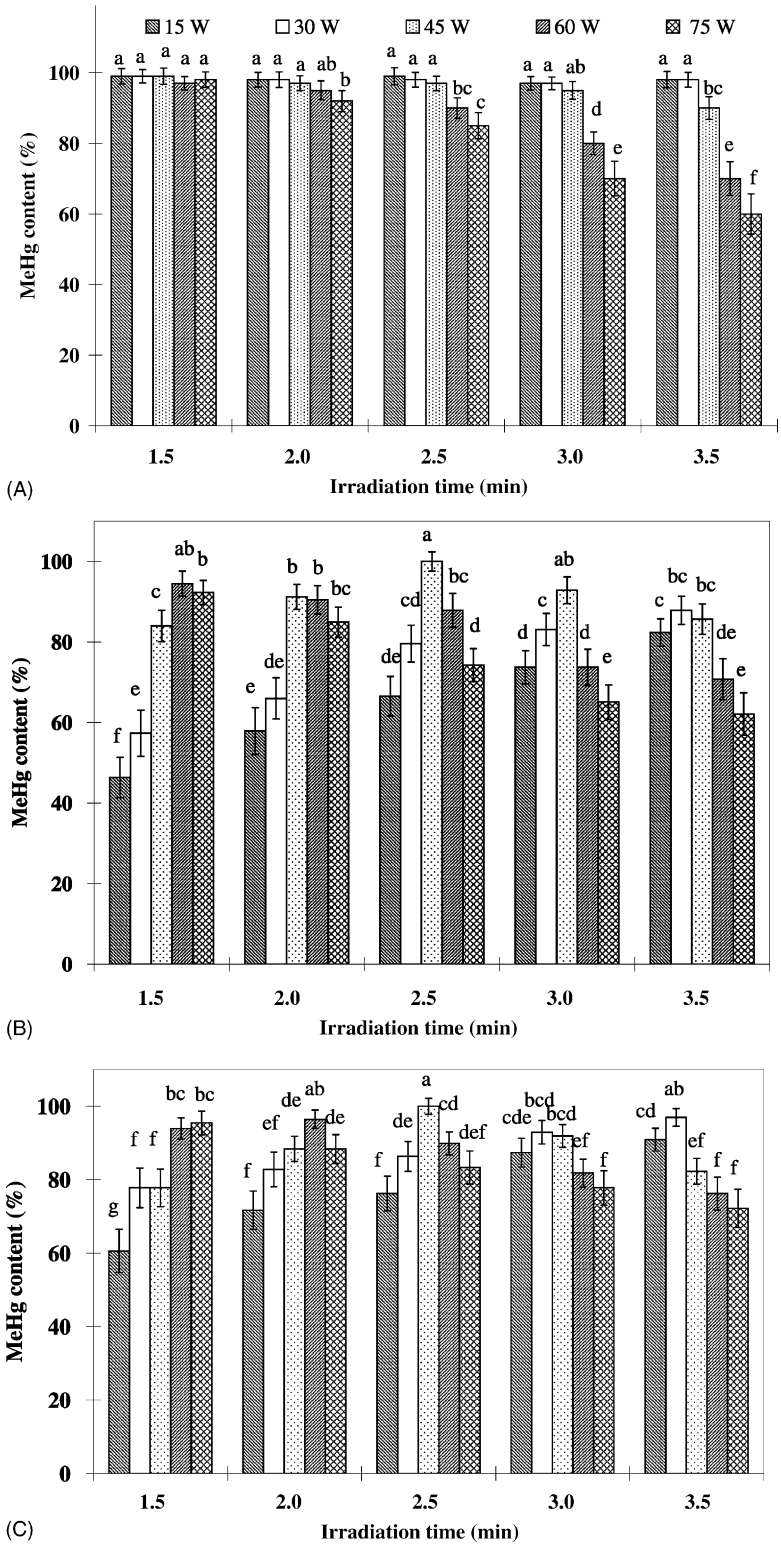

Fig. 1. Percentage of methylmercury (1 μg as Hg) spiked in 25% TMAH solution (A), methylmercury in CRM 464 tuna fish (B), and laboratory tuna fish sample when exposed to microwave irradiation (C). (a–g) Values in the figure with different superscripts are significantly different at $P < 0.05$.

laboratory tuna fish sample, and methylmercury standard solution. The highest derivatization yield for all samples was obtained at pH 4.0. This agrees with the report of Smaele et al. [22].

The influence of acetate concentration on the derivatization of methylmercury was checked by using 5 ml of 0.01, 0.05, 0.1, 0.5, 1.0, and 2.0 M acetate buffer solutions in CRM 464 tuna fish, laboratory tuna fish sample, and

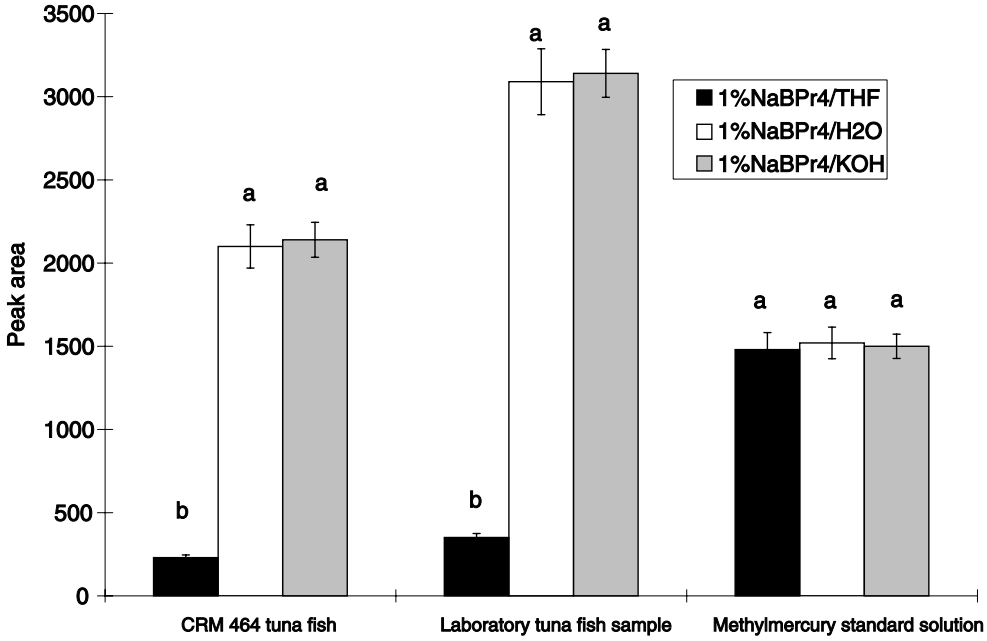

Fig. 2. Effects of derivation agents including water, 2% KOH, and THF on methylmercury detection of CRM 464 tuna fish, laboratory tuna fish sample, and methylmercury standard solution. (a and b) See Fig. 1.

methylmercury standard solution. It was found that the highest methylmercury content was obtained for all samples when acetate concentration was 0.5–2.0 M. Though the derivatization yield of methylmercury decreased significantly when acetate concentration was less than 0.1 M in methylmercury standard solution, it did not vary between 0.01 and 2 M acetate buffer in CRM 464 tuna fish and laboratory tuna fish sample. The pH of CRM 464 tuna fish and laboratory tuna fish sample added different concentrations of acetate buffer was about 4.07. The pH of methylmercury standard solution was about 4.05 when added 0.50, 1.0, and 2.0 M acetate buffer, but 11.2, 10.7, and 11.2 when added 0.01, 0.05, and 0.10 M acetate buffer, respectively. The following procedure was addition 1% $NaBPr_4$/KOH. Because the buffer capacity was not enough when acetate concentration was less than 0.1 M for standard solution. CRM 464 tuna fish and laboratory tuna fish sample pretreated by TMAH and neutralized by acetic acid. The buffer capacity was increased and independent for the acetate concentration.

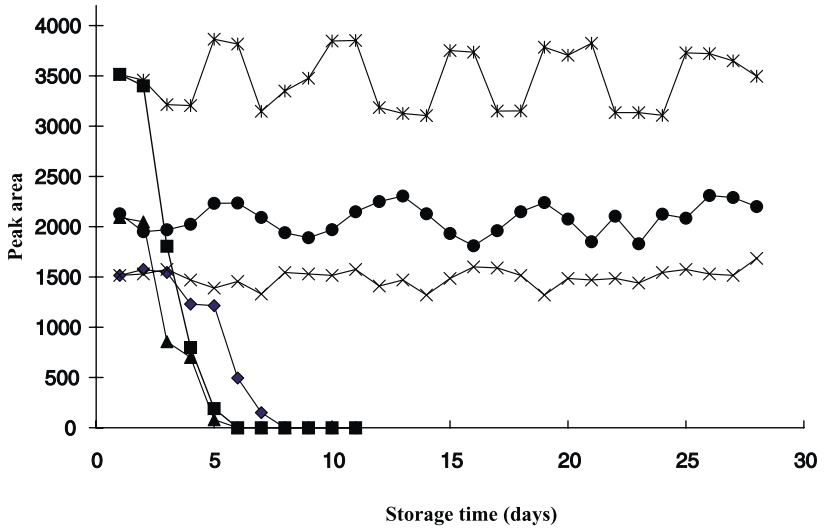

Fig. 3. The inter-day derivation ability of 1% $NaBPr_4$/KOH of CRM 464 tuna fish (●), laboratory tuna fish sample (❊), and methylmercury standard solution (×), and 1% $NaBPr_4/H_2O$ of CRM 464 tuna fish (▲), laboratory tuna fish sample (■), and methylmercury standard solution (◆).

Using the same conditions of Section 2.4 and changing the concentration of derivatization agent, we found 1 ml of a 1% $NaBPr_4$/KOH solution was sufficient in derivatization of methylmercury. The optimal reaction time on the derivatization yield of methylmercury was also found to be 10 min. There results were similar to those of Smaele et al. [22].

The inter-day derivatization abilities of 1% $NaBPr_4$/KOH and 1% $NaBPr_4/H_2O$ solution were examined by using a frozen solution after thawing everyday. Fig. 3 shows the methylmercury derivative was stable within 28 days in 1% $NaBPr_4$/KOH solution, but dropped quickly in 1% $NaBPr_4/H_2O$ solution.

The intra-day derivatization ability of $NaBPr_4$/KOH solution was examined by using freshly prepared solution to stand for 1–10 h. It is found that the intra-day stability was good and relative standard deviation was 0.80%. Hence, 1% $NaBPr_4$/KOH solution was the best derivatization agent for methylmercury.

Using 2 ml of *n*-hexane, *n*-heptane and isooctane examined the influence of extract solvent on the derivatization yield of methylmercury. The chromatogram peak of methylmercury was clearly obtained by using *n*-heptane and isooctane. The peak presented tailing with *n*-hexane. The extract of *n*-heptane has a good peak area count and S/N ratio of methylmercury.

3.3. Optimization of the GC–MS parameters

Later, the optimal temperature condition of using CP-SIL 8CB column in GC–MS was studied. Setting column at 60 °C for 5 min, the temperature of column was increased with 5, 10, 15, and 20 °C/min. It was found that methylmercury derivative was well obtained in performing at 20 °C/min. Under this condition, methylmercury derivative was eluted at 6.10 min and the peak was narrow and symmetric (Fig. 4B).

To avoid the decomposition of methylmercury derivative, the optimal temperature of the injection port in GC was investigated. It was found that the peak area of methylmercury increased from 220 to 230 °C, and kept stable during 230–260 °C. The decomposition of methylmercury derivative was not found even at 260 °C. Hence, the

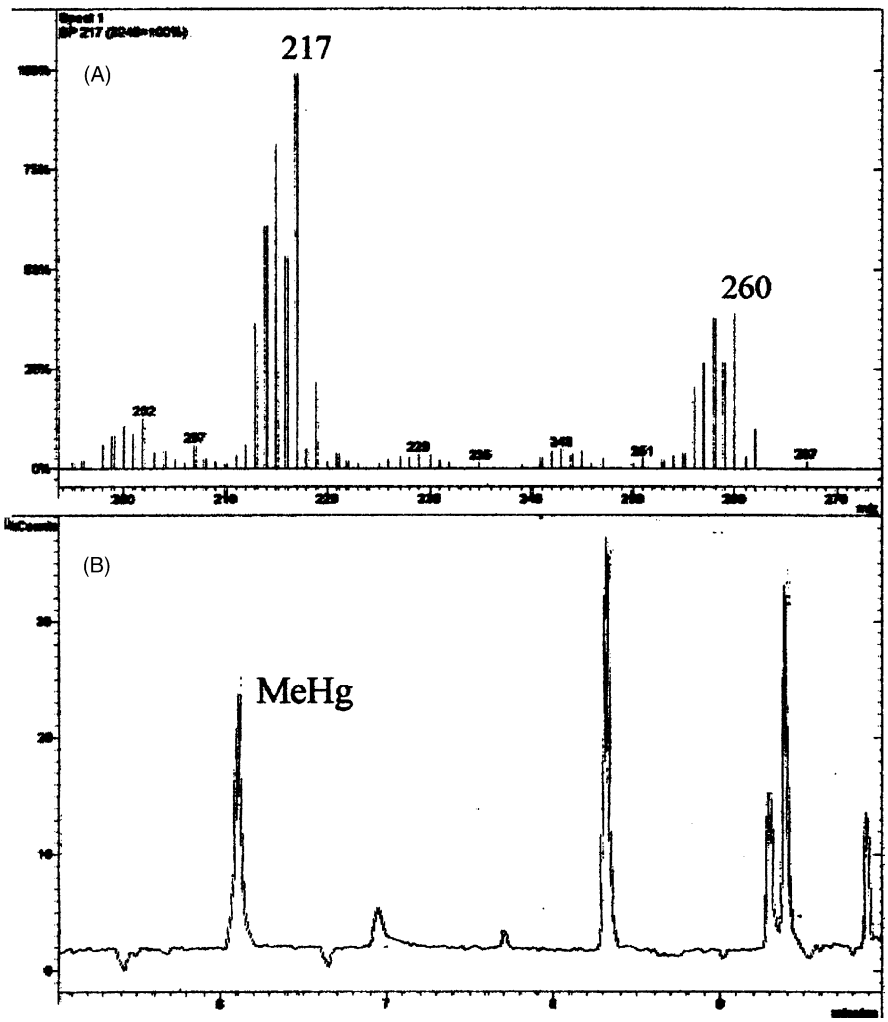

Fig. 4. Electroimpact mass spectra of methylmercury derivative obtained at 70 eV (A), and gas chromatogram (B).

optimal temperature of the injection port in GC was set at 260 °C.

Mass spectra of methylmercury derivative are shown in Fig. 4A. The reference ions for quantification were *m*/*z* 214–215 for methylmercury derivative. Among these reference ions, the peak *m*/*z* 217 was used as quantitative ion to monitor the retention time (6.10 min) of methylmercury in the GC chromatogram. Meanwhile, the CP-SIL 8 CB column was used in this study because other two columns (CP-SIL 1 CB and CP-SIL 24 CB) was found to induce the tailing problem.

The calibration curve was linear with slope 1.7891 and intercept −3.3057 within 10–1000 ng/ml of methylmercury. The regression coefficient was 0.9966. The detection limit of methylmercury derivative in GC–MS was 10 pg as Hg, which was calculated from three times of the baseline noise peak. Hence, the detection limit of method for fish sample was 0.04 μg/g.

3.4. Validation of the determination method

The recoveries of methylmercury in tuna, marlin, and shark muscle spiked with 0.5, 1.0, and 2.0 μg/g (as Hg) were tested and were 91.2 ± 4.6, 91.9 ± 3.6, and 95.3 ± 3.5% for tuna, 89.3 ± 5.2, 91.5 ± 4.7, and 94.7 ± 4.1% for marlin, and 91.7 ± 4.9, 93.4 ± 4.1, and 94.8 ± 3.8% for shark, respectively.

The intra-day and inter-day relative standard deviations (R.S.D.) were calculated during 1 week from data of seven replicates. The R.S.D. values for retention time of methylmercury were 0.2% intra-day and 0.5% inter-day, respectively. The R.S.D. values of peak area of methylmercury were 5.8% intra-day and 7.6% inter-day, respectively. These low R.S.D. values indicate the method has a very good reproducibility.

The developed method was validated by determination of three reference materials: CRM 464 tuna fish, DORM-2 dogfish muscle, and DOLT-2 dogfish liver. The tested methylmercury concentration was 5.34 ± 0.30, 4.34 ± 0.24, and 0.652 ± 0.055 mg/kg for CRM 464 tuna fish, DORM-2 dogfish muscle, and DOLT-2 dogfish liver, respectively. The obtained levels also agree with the certified values 5.50 ± 0.17, 4.47 ± 0.32, and 0.693 ± 0.055 μg/g for CRM 464 tuna fish, DORM-2 dogfish muscle, and DOLT-2 dogfish liver, respectively. It indicates that the developed method was well available to detect methylmercury in fish.

4. Conclusions

A procedure for detecting methylmercury in fish has been developed. Microwave-assisted digestion with TMAH solution, cupric ion addition, pH adjusting, derivatization with 1% $NaBPr_4$/KOH solution, *n*-heptane extraction, and GC–MS analysis were stepwise performed. Optimum condition for microwave-assisted digestion of methylmercury from fish with TMAH was found to be 3.5 min at 15–30 W, 2.5 min at 45 W, or 1.5 min at 60–70 W. Adding 20 mM cupric ion could perfectly release methylmercury. Optimum condition for propylation with 1% $NaBPr_4$/KOH solution was found to be adjusting pH to 4.0. The detection limit of the overall procedure was found to be 0.04 μg/g. The recoveries were 91.2–95.3% for tuna, 89.3–94.7% for marlin, and 91.7–94.8% for shark, respectively. The combination of microwave-assisted digestion, aqueous-phase derivatization and GC–MS analysis has resulted in a rapid, safety and accurate method for determining methylmercury in fish.

References

[1] M. Hempel, Y.K. Chau, B.J. Dutka, R. Mclnnis, K.K. Kwan, D. Liu, Analyst 120 (1995) 751.
[2] P.J. Craig, in: P.J. Craig (Ed.), Organometallic Compounds in the Environment Principles and Reactions, Longman, Essex, 1986, pp. 65–101.
[3] J.W. Moore, S. Ramamoorthy, in J.W. Moore, S. Ramamoorthy (Eds.), Heavy Metals in Natural Waters, Applied Monitoring and Impact Assessment, Springer, New York, 1984, pp. 125–160.
[4] N.S. Bloom, Can. J. Fish. Aquat. Sci. 49 (1992) 1010.
[5] K. May, M. Stoppler, K. Reisinger, Toxicol. Environ. Chem. 13 (1987) 153.
[6] A. Kudo, S. Miyahara, Water Sci. Technol. 23 (1991) 283.
[7] US Environmental Protection Agency (EPA), Water Quality Criterion for the Protection of Human Health: Methylmercury, EPA-823-R-01-001, 2001.
[8] Department of Health, Executive Yuan, Hygienic Standard of Fish and Shellfish, Ordinance no. 8143635, 1992 (in Chinese).
[9] G. Westöö, Acta Chem. Scand. 20 (1966) 2131.
[10] L. Magos, Analyst 96 (1971) 847.
[11] H. Emteborg, N. Hadgu, D.C. Baxter, J. Anal. At. Spectrom. 9 (1994) 297.
[12] Official Methods of Analysis, Association of Official Analytical Chemists, 1995, Chapter 9, pp. 22, 24, 26.
[13] C.M. Tseng, A.D. de Diego, F.M. Martin, D. Amouroux, O.F.X. Donard, J. Anal. At. Spectrom. 12 (1997) 743.
[14] M. Filippelli, F. Baldi, F.E. Brinckman, J.A.C. Broekaert, Environ. Sci. Technol. 26 (1992) 1457.
[15] D. Gerbersmann, M. Heisterkamp, F.C. Adams, J.A.C. Broekaert, Anal. Chim. Acta 350 (1997) 273.
[16] C. Dietz, Y. Madrid, C. Camara, J. Anal. At. Spectrom. 16 (2001) 1397.
[17] R. Fisher, S. Rapsomanikis, M.O. Andrea, Anal. Chem. 65 (1993) 763.
[18] L. Liang, M. Horvat, E. Cernichiari, B. Gelein, S. Balongh, Talanta 43 (1996) 1883.
[19] M.S. Jimenez, R.E. Sturgeon, J. Anal. At. Spectrom. 12 (1997) 597.
[20] E. Bulska, D.C. Baxter, W. Frech, Anal. Chim. Acta 249 (1991) 545.
[21] I.R. Pereiro, A. Wasik, R. Lobinski, J. Anal. At. Spectrom. 13 (1998) 743.
[22] T.D. Smaele, L. Moens, R. Dams, P. Sandra, J.V. der Eycken, J. Vandyck, J. Chromatogr. A 793 (1998) 99.
[23] M.L. Olson, L.B. Cleckner, J.P. Hurley, D.P. Kraabbenhoft, T.W. Heelan, J. Anal. Chem. 358 (1997) 392.