CHAPTER V

DISCUSSION AND CONCLUSION

In the present investigation, sodium hydroxide was used to replace potassium hydroxide proposed by Schafer (13) for the digestion of fish samples. The weight ratio between sodium hydroxide and the fish sample was 1:1. The recovery yield obtained from sodium hydroxide digestion was not significantly different from that obtained from potassium hydroxide digestion. Thus, sodium hydroxide was prefered because its cost was lower. Acid hydrolysis could be used as well as alkaline hydrolysis. In the former, the lipids in sample would give emulsion to interfere in the extraction of methylmercury(II) with benzene. The later was prefered although it was time consuming, since the lipids would be converted to the salts. Generally, methylmercury (II) was determined in the form of methylmercury (II) chloride, neither methylmercury (II) bromide nor methylmercury (II) iodide because the chloride form had the most stability. The recommended method for determining the methylmercury (II) was shown in Figure 3.6. The fish sample was digested with sodium hydroxide. After converting methylmercury (II) hydroxide to methylmercury (II) chloride with hydrochloric acid. The latter was extracted into benzene. However, some hydrogen sulfide and sulfur compounds might go into the benzene layer and

cause the interference in the gas chromatographic analysis. Cysteine was used to clean up the benzene layer that methylmercury (II) chloride formed methylmercury (II) cysteine complex ($CH_3HgSCH_2\underset{NH_2}{C}HCOOH$) in aqueous phase. After acidification the aqueous phase with hydrochloric acid, methylmercury (II) chloride was quantitatively determined by gas chromatography with an electron capture detector. The detection limit of the chromatographic system for methylmercury(II) chloride was found to be 52 pg. Standard addition method was applied to the analysis of methylmercury (II) in 16 fish samples since the content of methylmercury (II) in fish was normally lower than the detection limit (0.02 ppm), although it was time consuming. It was therefore not surprising that it took two days for analysing three samples. Nevertheless the method developed in this thesis can be applied to determine the methylmercury (II) in liver, meat and foodstuff.

APPENDIX

LEAST SQUARE METHOD

Least square method is employed to treat the data for calibration curve and standard deviation curve to estimate the peak height for any measured value of the content of methylmercuric chloride. If "y" is the peak height which is expected to be linearly dependent upon the variable x (the content of methylmercuric chloride), so the best linear equation to estimate "y" for any measured value of x is shown as following ;

$$y = \bar{y} + b\,(x - \bar{x}) \qquad \text{.....(1)}$$

where

$$\bar{y} = \sum_{i=1}^{n} y_i/n \qquad \text{.....(2)}$$

$$\bar{x} = \sum_{i=1}^{n} x_i/n \qquad \text{.....(3)}$$

$$b = \frac{\sum_{i=1}^{n} (x_i - \bar{x})\,(y_i - \bar{y})}{\sum_{i=1}^{n} (x_i - \bar{x})^2} \qquad \text{.....(4)}$$

y = the estimate value

Example of estimation

Table 1

ng of methylmercuric chloride (x)	0.13	0.21	0.31	0.52
peak height (y) (cm)	1.50	2.50	5.03	9.20

Table 2

x	y	$x-\bar{x}$	$y-\bar{y}$	$(x-\bar{x})(y-\bar{y})$	$(x-\bar{x})^2$
0.13	1.50	-0.16	-3.06	0.49	0.0256
0.21	2.50	-0.08	-2.06	0.16	0.0064
0.31	5.03	0.02	0.47	0.01	0.0004
0.52	9.20	0.23	4.64	1.07	0.0529
$\bar{x} = 0.29$	$\bar{y} = 4.56$			$\Sigma = 1.73$	$\Sigma = 0.0853$

from Eq (4) $b = 1.73/0.0853 = 20.28$

Table 3 From Eq.(1)

x	estimated "y"
0.13	4.56 + 20.28(-0.16) = 1.32
0.21	4.56 + 20.28(-0.08) = 2.94
0.31	4.56 + 20.28(0.02) = 4.96
0.52	4.56 + 20.28(0.23) = 9.22
