Digestion of Oysters for the Determination of Mercury[1]

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Introduction

Interest is increasing in this Country concerning mercury contamination of fish and shellfish (1). Interest stems from the Japanese "minamata disease" and reports from Sweden about mercury contamination of their shellfish and environment (2-3).

A survey by this Laboratory was initiated to reveal any existing concentrations of mercury in oysters harvested from Mobile Bay and waters of southern states.

Analytical support for this project required selection of an acceptable method for the detection and estimation of mercury concentrations in oysters less than 1 ppm. Atomic absorption spectrophotometry had been used successfully for the analysis of oysters, seawater and other material for various metals (4-6). However, this method appeared inadequate for low concentrations of mercury. At least 15 ppm mercury was required for proper instrument response (7).

Anticipated levels of mercury in oysters were less than 1 ppm. A method, therefore, was needed that would detect and quantitatively measure concentrations of mercury in the microgram range. Dithizone chelation was selected with final quantitation by a Beckman DU Spectrophotometer (8) at a wavelength of 490 nm.

Preparation of the sample for dithizone or any other analytical means was very important (8). Conversion of all mercury into an ionizable state required disintegration of the oyster tissue by some suitable means so that mercury would not be lost due to its volatile nature. The usual means of digestion of oysters or any food involved use of concentrated sulfuric and nitric acids while heating in a carefully constructed apparatus designed to prevent loss of mercury (9).

This type of digestion usually was time-consuming. The reason for mercury loss appeared associated with two parameters; concentrated nitric acid and applied heat. The oxides of nitrogen could carry away mercury vapor. The AOAC clearly anticipated loss of mercury with the improper application of concentrated nitric acid

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(9). Therefore, it appeared feasible to eliminate concentrated nitric acid and to utilize the heat generated from the mixing of concentrated sulfuric acid and water as the sole source of heat. Digestion of the oyster tissue is completed with small increments of solid potassium permanganate.

This paper describes a type of digestion showing the excellent retention of low concentrations of added mercury to oyster tissue (Table 1) (Table 2). The method not only prevents mercury loss but expedites the digestion. The time required was usually about 25 minutes, with a maximum temperature of 126 C. Analytical grade reagents were used in all reactions.

TABLE 1

Recovery of Mercury from Oysters Spiked with Mercuric Nitrate[a]

Micrograms Mercury Added	Average Micrograms Mercury Recovered	Percent Mercury Recovered
5	5.04	100.8
10	10.8	108
15	14.5	96
20	20.0	100
25	26.5	106
50	50.0	100
	Average Percent Recovery	101

[a]Standard Based on Inorganic Mercury

TABLE 2

Recovery of Mercury from Oysters Spiked with Methyl Mercury Chloride[a]

Micrograms Mercury Added	Average Micrograms Mercury Recovered	Percent Mercury Recovered
8.0	8.5	106
12.0	12.0	100.0
18.0	17.5	97.0
24.0	25.0	104.0
	Average Percent Recovery	101

[a]Standard Based on Inorganic Mercury

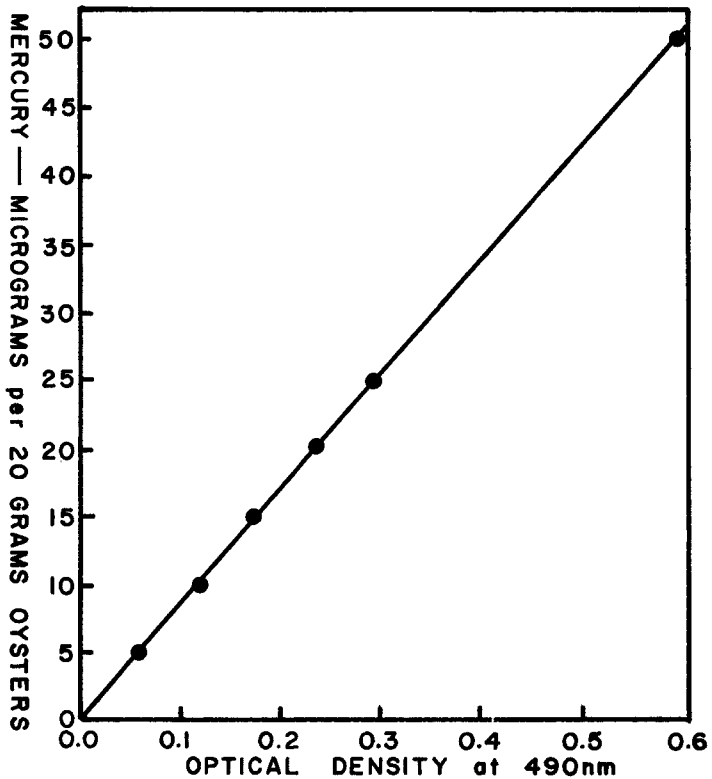

Figure 1. Relationship of added mercuric nitrate concentration and optical density.

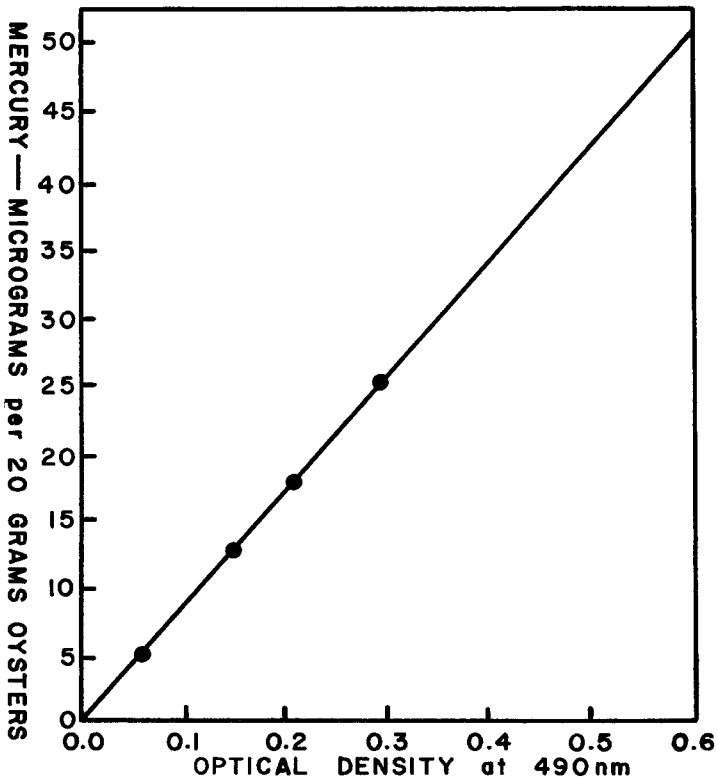

Figure 2. Relationship of added methyl mercury chloride concentration and optical density.

A. Digestion

a. Select about 10 oysters; homogenize the meats in a grinder.

b. Weigh 20 gm of the homogenized oyster tissue in a tall beaker.

c. Introduce 5 drops of diluted Dow Corning Antifoam C Emulsion into the beaker.

d. Transfer 15 ml of water to the homogenate.

e. Carefully add 20 ml of concentrated sulfuric acid to the homogenate.

f. Gently mix the dark acid slurry and add 1/2 gram increments of potassium permanganate crystals to the acid slurry; allow time for the violent reactions to subside. Within 15 minutes the slurry will be decomposed resulting in a whitish solution containing some nondescript charred material.

g. Add more potassium permanganate until a slight purple color persists. Add cool distilled water filling the beaker within 3 inches of the top. Add about 2 drops of hydroxylamine hydrochloride thus resulting in a clear solution.

h. Filter this solution into a 2-liter separatory funnel through a glass funnel containing a small amount of glass wool inserted in the apex.

i. Add water through the funnel so that the final volume is about 1 liter.

B. Analysis. Analysis of this solution is done by the well documented dithizone technique which can be found in many volumes. The method especially useful was taken for this study from Sandell (8). Particular attention is directed to various techniques used for the control of metals other than mercury. A blank is always included in the analysis since traces of mercury can be found in reagents.

Results and Discussion

In order to establish the validity of the newly developed digestion technique, experimental recovery studies were conducted utilizing oyster tissue samples spiked with various concentrations of mercury. One set of oyster samples included inorganic mercury as mercuric nitrate, and another set of oyster samples included organic mercury as methyl mercury chloride. Each concentration

level of the mercury compound was spiked in a 20-gram aliquot of oyster tissue.

In the mercuric nitrate tests, 6 concentrations of mercury were used at levels of 5, 10, 15, 20, 25, and 50 micrograms. Three separate samples were examined for each concentration level, thus providing a total of 18 examinations. The results of the recovery studies are shown in Table 1. The average quantity and average percent of mercury recovered for the three samples is tabulated for each concentration level of added mercury. The overall average recovery of mercury for all samples was 101 percent. The linear relationship between the concentration of inorganic mercury added to the oyster samples and the optical density readings are clearly shown in Figure 1.

In the methyl mercury tests, four concentrations of mercury were used at levels of 8, 12, 18, and 24 micrograms. Three separate samples were examined for each concentration level, providing a total of 12 examinations. The results of the recovery studies are shown in Table 2 in which the average quantity and average percent of mercury recovered are tabulated. The average recovery of mercury for the organic samples was 101 percent. The linear relationship between the concentration of organic mercury added to the oyster samples and the optical density readings are clearly shown in Figure 2.

A method for the measurement of mercury in oysters by dithizone is presented. The dithizone technique is a well-known method for the measurement of many metals, including mercury. The contribution in this paper is concerned with the digestion of oyster tissue in a way preventing loss of mercury. This digestion technique was also applied to lyophilized and dried mercury fortified oyster tissue at 100 C. These drying methods did not cause any loss of mercury.

This digestion technique might be used on other substances easily digested by concentrated sulfuric acid and potassium permanganate.

Future studies are planned with oysters fed known concentrations of mercury. Analysis of these tissues will demonstrate the application of this technique to freshly harvested oysters exposed to mercury concentrations.

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