Simultaneous Separation of Six Kinds of Organic and Inorganic Mercury Compounds by Means of Thin-Layer Chromatography

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Recently, the authors[1] had measured mercury content in the hairs of approximately 850 inhibitants in the coast of Tokuyama Bay by flameless atomic absorption spectrophotometry and the results were already reported. On the other hand, by flameless atomic absorption spectrophotometry, the separation and determination of organic and inorganic mercury compounds can be achieved, but that for individuals of organic mercury compounds by gas chromatography is entirely impossible. Accordingly, if we want to determine each of them by it, they are previously separated by a certain mean.

Up to date, a great number of studies have been made on their separation by paper chromatography[2,3], thin-layer chromatography[4,5,6], ion-exchange paper chromatography[7], column chromatography[8], and so on. However, these means may be said to leave room for further study. Therefore, first of all, we have carried out some studies for the simultaneous separation of six kinds of organic and inorganic mercury compounds by using of thin-layer chromatography.

MATERIALS AND METHODS

1) Materials

Mercuric chloride, methylmercuric chloride, methylmercuric iodide, methylmercuric cysteine, ethylmercuric chloride, and phenylmercuric acetate. Out of these materials, mercuric chloride and methylmercuric cysteine were dissolved in water and the others in chloroform. The solutions each was to the concentration of 5 μg/5 μl.

2) Preparation of plate

Thirty g of silica gel G (Merk, gibs 13%) was placed in a motor, To this was added 60 ml of water and the mixture was rigorously

stirred until uniform. About 5 g of the slurry was poured onto a glass plate (20 × 20 cm) and spread uniformly with a rod of glass equipped with the cellophane tape of 0.25 mm in thickness to one's both ends. After drying in air over a night, the plate was left in an oven at 110°C for 1 hour and kept a box containing sillica gel desiccant.

3) Mobile solvents

(A): Benzene-petroleum benzine (50:10 v/v)
(B): n-Hexane-acetone (85:15 v/v)
(C): Petroleum ether-ethyl ether (70:30 v/v)
(D): Chloroform-ethanal (98:2 v/v)
(E): Benzene-chloroform (50:10 v/v)
(F): Benzene-dioxane-conc. ammonia water (60:20:10 v/v)
(G): Benzene-n-hexane-chloroform (30:20:10 v/v)
(H): Benzene-cyclohexane-petroleum benzine (30:20:10 v/v)
(I): n-Butanol saturated with N-ammonia water
(J): n-Butanol-pyridine-N-ammonia water (35:34:31 v/v)
(K): Cyclohexane-chloroform-pyridine (20:60:5 v/v)

4) Spray reagent

0.005% dithizone chloroform solution

5) Tank for development

A tank is vertical type and 10×21×21 cm in size.

Under these conditions mentioned above, each 5 μl of the sample solutions was spotted onto the adsorbent at a point of 2 cm apart from one end of the plate and developed in the tank which the atmosphere was saturated with the used mobile solvent vapour. The developed distance is 12 cm. After the completion of development, the plate was dried in air and the spots were detected by the spray of 0.005% dithizone chloroform solution.

RESULTS AND DISCUSSION

1) Development with 11 kinds of mobile solvents

The Rf values of alkyl, aryl, and inorganic mercury compounds obtained are shown in Table 1. The order of Rf value of each mercury compounds was almost constant irrespective of the kind of mobile solvents. The Rf value was the largest in methylmercuric iodide, followed by ethylmercuric chloride, methylmercuric chloride, phenylmercuric acetate, mercuric chloride, and methylmercuric cysteine, in order of decreasing Rf value. Methylmercuric cysteine did not nearly ascend and in about all cases, it remained at a start line. The mobile solvents of

Table 1. Rf values of organic and inorganic mercury compounds in thin-layer chromatography
MC: mercuric chloride, MMCY: methylmercuric cysteine, MMC: methylmercuric chloride,
MMI: methylmercuric iodide, PMA: phenylmercuric acetate, EMC: ethylmercuric chloride.

Materials	Mobile solvents										
	A	B	C	D	E	F	G	H	I	J	K
MC	0.09	0.20	0.67	0.03	0.13	0.01	0.08	0.05	0	0.02	0.66
MMCY	0	0.03	0	0.01	0.01	0.01	0	0.01	0.22	0.34	0
MMC	0.33	0.50	0.63	0.87	0.48	0.15	0.43	0.21	0.05	0.06	0.73
MMI	0.65	0.68	0.75	0.82	0.76	0.52	0.87	0.60	0.79	0.44	0.90
PMA	0.28	0.50	0.80	0.83	0.47	0.11	0.34	0.17	0.11	0.32	0.73
EMC	0.55	0.60	0.72	0.86	0.69	0.30	0.70	0.40	0.33	0.40	0.87

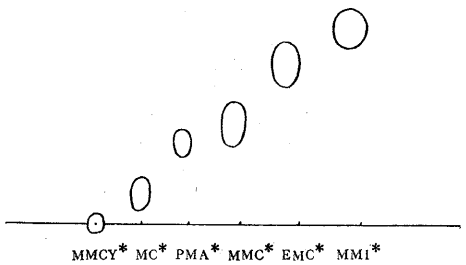

Fig. 1. Thin-layer chromatogram with benzene-petroleum benzine solvent (50 : 10v/v)
* : See Tab. 1

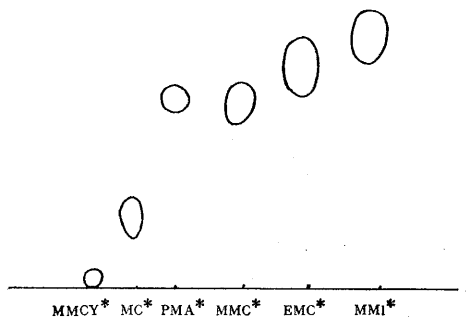

Fig. 2. Thin-layer chromatogram with n-hexane-acetone solvent (85 : 15 v/v)
* : See Tab . 1

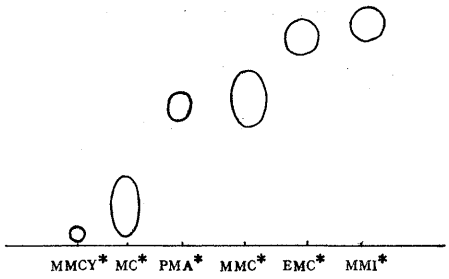

Fig. 3. Thin-layer chromatogram with benzene-chloroform solvent (50 : 10 v/v)
* : See Tab. 1

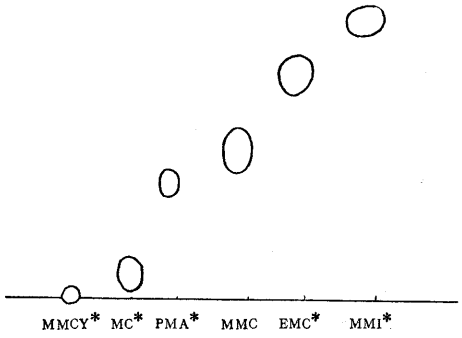

Fig. 4. Thin-layer chromatogram with benzen-n-hexane-chloroform solvent (30 : 20 : 10 v/v)
* : See Tab. 1

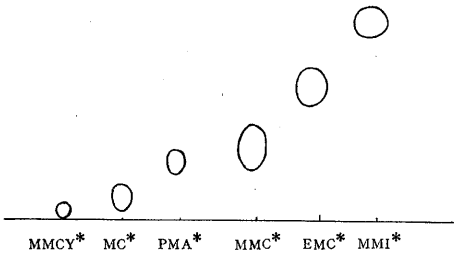

Fig. 5. Thin-layer chromatogram with benzene-cyclohexane-petroleum benzine solvent (30 : 20 : 10 v/v)
* : See Tab. 1

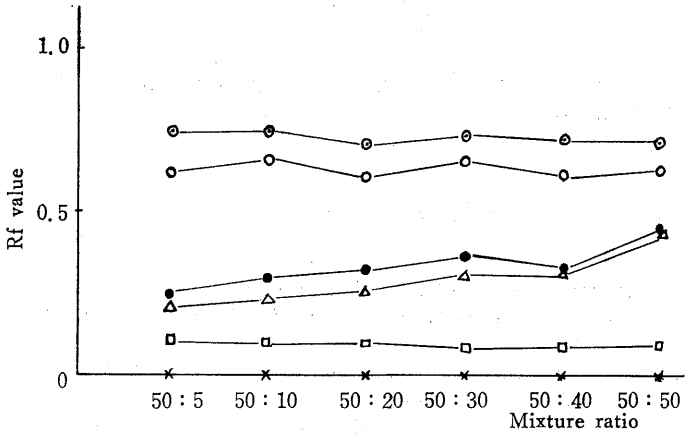

Fig. 6. Relationship between Rf values of mercury compounds and the mixture ratio of benzen-chloroform solvent

⦿ : methymercuric iodide ○ : ethylmercuric chloride
● : methylmercuric chloride △ : phenylmercuric chloride
□ : mercuric chloride × : methylmercuric cysteine

I, J, and K which are rich in the polarity could not be satisfied in their separation. In addition, the magnification and tailing of a spot from time to time appeared. On the contrary, the non-polar mobile solvents of A, B, E, G, and H were more excellent than others in their separation and furthermore, thus undesirable phenomena of a spot could not be observed at all (Figs. 1, 2, 3, 4, and 5).

Previously, Nakazawa et al.[5] have reported that aryl and phenylmercuric compounds which are different only in their union radicals could not be separated through thin-layer chromatography. However, in our experiment, the separation of methylmercuric chloride and methylmercuric iodide that have the same alkyl radical was completely achieved.

2) Development with benzene-chloroform solvent at various mixture ratios

The mixture ratio of benzene-chloroform solvent variously changed and a comparative study was made on their separation. As shown in Fig. 6, the variation of mixture ratio had not so much a significant effect on the Rf values. Namely, the Rf values of phenylmercuric acetate and methylmercuric chloride varied in some extent, while those of others did not nearly changed. As the result, the satisfactory separation of mercuric compounds on the basis of the change of mixture ratio could not be expected.

SUMMARY

This report deals with the simultaneous separation of six kinds of organic and inorganic mercury compounds by means of thin-layer chromatography.

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