

ANNEX

**DETERMINATION OF DICHLOROMETHANE AND 1,1,1-TRICHLOROETHANE
IDENTIFICATION AND DETERMINATION OF ORGANOMERCURY
COMPOUNDS****SCOPE AND FIELD OF APPLICATION****A. IDENTIFICATION****1. PRINCIPLE**

The organomercury compounds are complexed with 1,5-diphenyl-3-thiocarbazonone. After extraction of the dithizonate with carbon tetrachloride, silica gel, thin-layer chromatography is carried out. The spots of the dithizonates appear as an orange colour.

2. REAGENTS

All the reagents should be of analytical purity.

- 2.1. Sulphuric acid, 25 % (v/v).
- 2.2. 1,5-diphenyl-3-thiocarbazonone (dithizonone): 0,8 mg in 100 ml carbon tetrachloride (2.4).
- 2.3. Nitrogen.
- 2.4. Carbon tetrachloride.
- 2.5. Development solvent: hexane /acetone, 90:10 (v/v).
- 2.6. Standard solution, 0,001 % in water of:
sodium 2-(ethylmercuriothio)benzoate,
ethylmercury chloride or methylmercury chloride,
phenylmercury nitrate or phenylmercury acetate,
mercury dichloride or mercury di(acetate).
- 2.7. Ready prepared silica gel plates (e.g. Merck 5721 or equivalent).
- 2.8. Sodium chloride.

3. APPARATUS

- 3.1. Normal laboratory equipment.
- 3.2. Normal TLC apparatus.
- 3.3. **Phase-separating filter.**

4. PROCEDURE**4.1. Extraction**

- 4.1.1. Dilute 1 g of sample in a centrifuge tube by titration with 20 ml of distilled water. Obtain the maximum dispersion and warm to 60 °C in a water bath. Add 4 g of sodium chloride (2.8). Shake. Allow to cool.

Status: EU Directives are being published on this site to aid cross referencing from UK legislation. After IP completion day (31 December 2020 11pm) no further amendments will be applied to this version.

- 4.1.2. Centrifuge for at least 20 minutes at 4 500 rev/min in order to separate the greater part of the solid from the liquid. Filter into a separating funnel and add 0,25 ml of sulphuric acid solution (2.1).
- 4.1.3. Extract several times with 2 or 3 ml of dithizone solution (2.2) until the last organic phase remains green.
- 4.1.4. Filter each organic phase sequentially through a phase-separating filter (3.3).
- 4.1.5. Evaporate to dryness in a stream of nitrogen (2.3).
- 4.1.6. Dissolve with 0,5 ml of carbon tetrachloride (2.4). Apply this solution immediately as indicated in 4.2.1.
- 4.2. ***Separatum and identification***
- 4.2.1. Apply immediately 50 µl of the carbon tetrachloride solution obtained in 4.1.6 on to a silica gel plate (2.7). Treat simultaneously 10 ml of standard solution (2.6) as in 4.1 and apply 50 µl of the solution obtained in 4.1.6 on the same plate.
- 4.2.2. Place the plate in the solvent (2.5) and allow the latter to rise 150 mm. The organomercury compounds appear as coloured spots whose colour is stable, provided the plate is covered by a glass plate immediately the solvent evaporates.

For example, the following Rf values are obtained:

	Rf	Colour
Thiomersal	0,33	Orange
Ethylmercury chloride	0,29	Orange
Methylmercury chloride	0,29	Orange
Phenylmercury salts	0,21	Orange
Mercury (II) salts	0,10	Orange
Mercury di(acetate)	0,10	Orange
1,5-diphenyl-3-thiocarbazon	0,09	Pink