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DEVELOPMENT OF A METHOD FOR THE EXTRACTION AND DETERMINATION OF NON-POLAR, DISSOLVED ORGANIC SUBSTANCES IN SEA WATER

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SUMMARY

A method for extracting sea water by sorption on Amberlite XAD is described. The apparatus, sampling technique, working-up procedure and determination by gas chromatography are considered. Recovery tests with DDT, DDE, lindane, aldrin, 2-chlorobiphenyl, phenanthrene, pristane and *n*-hexadecane demonstrated the applicability of the method to non-polar substances. The recoveries were about 80%. Gas chromatograms of two samples are shown.

INTRODUCTION

The pollution of sea water by hydrocarbons, pesticide residues, polychlorinated biphenyls (PCBs) and many other organic compounds has stimulated the development of analytical methods for determining dissolved organic compounds in recent years. Previously, emphasis was placed on substances of so-called natural origin, such as amino acids, fatty acids and carbohydrates; now more widely applicable methods are required for determining a large number of different types of substances, even if they are present in only trace amounts. If it is taken into consideration that the sum of all dissolved organic substances in sea water is in the parts per million range, then the following conditions are required for a method for their determination:

(1) The concentration factor should be sufficiently great that even individual substances at the parts per 10^9 level could be detected with specific detectors after separating the pool.

(2) The working-up procedure should not change the identity or content of the compounds.

(3) Contaminants in the chemicals used in the methods, *e.g.*, solvents, should not be concentrated during the adsorption analysis.

(4) Inorganic salts, the contents of which are 10^4 times greater than those of the organic substances, should be excluded from the concentration steps.

Several methods have been developed for determining qualitatively and quantitatively dissolved organic compounds. A method that does not involve concentration has been proposed for determining benzene in water with the aid of laser Raman spectroscopy[1]. As the Raman spectrum of water is not very significant, pollutants with a characteristic Raman spectrum could be detected in water without a working-up procedure (*e.g.*, benzene at the 50 ppm level[1]). In most instances a direct determination without separation and concentration is not possible. Two principal approaches to the problem of separation have been employed:

(1) sorption on a solid adsorbent (sometimes coated);
(2) solvent extraction.

Batchwise solvent extraction is usually restricted to relatively small amounts of water masses. Although this method of extraction was methodically improved, (Kawahara *et al.*[2] constructed a semi-automatic device to shorten the extraction time; Schafer *et al.*[3] extracted 3.5 l of sea water by rapid stirring with 10 ml of *n*-hexane), continuous solvent extraction is the superior method. Werner and Waldichuk[4] modified a Scheibel apparatus[5] with interchangeable solvent circulation. Kahn and Wayman[6] constructed a three-stage liquid–liquid extractor, in which non-polar substances were extracted with an efficiency of up to 97% by a rapidly circulating flow of light petroleum (500–1100 ml/h). Goldberg and co-workers[7,8] constructed a similar apparatus for solvents lighter than water (five stages) and heavier than water (four stages). With a flow-rate of water of 7–8 l/h they were able to recover up to 80% of non-polar substances with five different solvents with a concentration factor of about 10^5. They pointed out that the efficiency of extraction depended on the difference between the dipole moments of the solution and the extraction solvent. A further improved apparatus was constructed by Ahnoff and Josefsson[9], which avoided the need to renew the solvent by distillation. It was suitable for fieldwork because of its good handling properties. They restricted the apparatus to one extraction vessel, specially constructed so that in a mixing chamber with a flow-rate of water of 2–5 l/h effective magnetic stirring resulted in efficient mixing of the water and the solvent. Separation of emulsions was also achieved, resulting in low losses of the solvent in the waste water. The recovery of pesticides was nearly 90%.

The limiting factor for all extraction methods is the distribution coefficient of the substances between the water and the solvent. Particularly for slightly polar substances, the factor for water–non-polar solvent is not very satisfactory. The time for exchanging is often too short; the disadvantage could be overcome only by several extractions in series. Adsorption by a solid adsorbent in a column does not suffer from these drawbacks.

Activated charcoal is very suitable for adsorption[10,11], but it is difficult to elute the adsorbed substances completely and, furthermore, there is the possibility that chemical changes will occur[12].

Aue and co-workers[13,14] successfully used silicones chemically bonded to Chromosorb G to adsorb chlorinated hydrocarbons from fresh water. Ito[15] used alkylchlorosilanes chemically bonded to Celite, with less success, for the adsorption of pesticides. For the extraction of PCBs, Gesser *et al.*[16] used porous polyurethane foam, while Uthe *et al.*[17] used a polyurethane foam coated with selective adsorbents and obtained better results. For the extraction of sea water, Ahling and Jensen[18] used Carbowax 4000 together with *n*-undecane on Chromosorb DMCS, the particulate material being removed first by precipitation with aluminium sulphate. The recovery of pesticides and PCBs was 80–100%, depending on the experimental conditions used. Riley and Taylor[19] were the first to use cross-linked polymers such as styrene–divinyl benzene copolymers of Amberlite XAD to extract [^{14}C]DDT from sea water. Burnham *et al.*[20] applied resins of different types (XAD-2 and XAD-7) to extract phenols, amines, ketones and sulphonates from fresh water with success, and Harvey and coworkers[21,22] for the extraction of DDE, DDT and dieldrin from sea water. We have improved the separation method of the previous workers and have found it to perform satisfactorily for the analysis of non-polar substances in sea water.

EXPERIMENTAL

Apparatus

The apparatus (Fig. 1) consists of a Simplidos double-piston pump (Bran and Luebbe, Hamburg, G.F.R.), which is connected with a PTFE tube by a quick-closing clamp. The piston stroke is continuously adjustable, so that the flow-rate can be regulated from 3 to 70 l/h. After passing through a pulsation damper connected to an excess pressure valve, the water is filtered. On the filter device is a pressure gauge, which enables one to measure the pressure on the filter up to 2 atm.

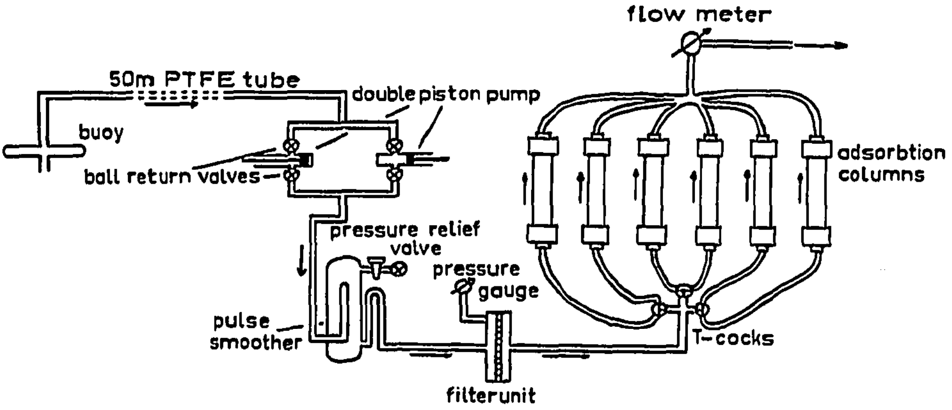

Fig. 1. Schematic diagram of extraction apparatus.

After filtration, the water flow is divided and directed in parallel into six columns, each of which can be disconnected by means of a stop-cock. One adsorption column (Fig. 2) consists of a cylindrical glass body, 9 cm in length and 2.3 cm I.D., which has two screw-threads at each end and a sintered glass plate inside at one end. The tubes for influx and outflow are connected by fittings to a PTFE body, which distributes the water flow equally over a PTFE gauze to the column section. The PTFE body is tightened to the glass cylinder by a nut. The water outflows are combined and the extracted sea water is measured and afterwards discarded.

Sampling procedure and work-up

The sea water is pumped directly on board ship through the previously described apparatus. In order to avoid contamination by materials from the ship, a

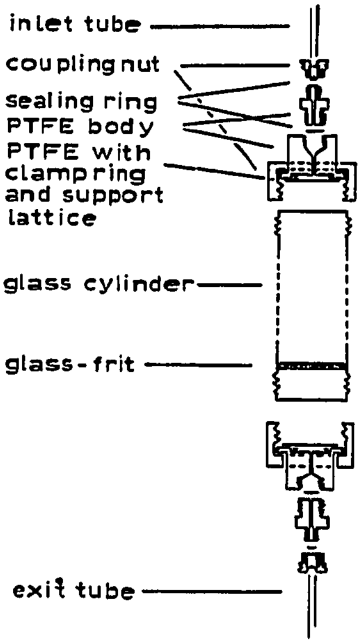

Fig. 2. Adsorption column.

small buoy with the suction inlet is situated about 50 m away from the ship and connected to the apparatus by a PTFE tube. It is possible to attach vertical PTFE tubes of different lengths to the buoy, so that one can sample water from different depths.

The adsorption material in the six columns is Amberlite XAD-2 and each column is filled with 65 ml of aqueous slurry, so that the total content is 390 ml. The flow-rate is about 1.4 bed volumes/min, equivalent to 2.0 ml/min·cm^3. After sampling, the apparatus and the columns are washed with 2 l of distilled water. The screw connections are removed, the columns are washed outside with methanol and directly extracted for 8 h in a Soxhlet apparatus with 150 ml of methanol plus 100 ml of distilled water per column (during this time, it is possible to process another sample with a second set of columns). After extraction, each column is washed free from methanol with 50 ml of distilled water in the Soxhlet apparatus so that they are ready for the next sampling procedure. The combined methanol–water extracts are re-extracted with three 25-ml volumes of *n*-hexane in a separating funnel and the combined extracts are dried with anhydrous sodium sulphate and either made up to 100 ml (high content of substances of interest) or evaporated under reduced pressure to 0.5 ml using a water-bath (low content of substances of interest). In the same manner, the filters from one sample are extracted and the extracts are worked up and an aliquot is injected into the gas chromatograph. A schematic diagram of the procedure is shown in Fig. 3.

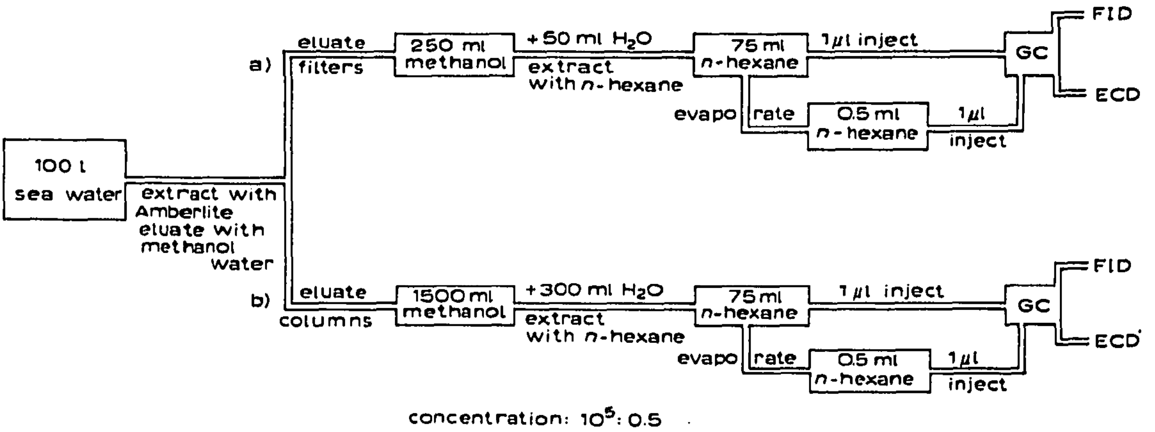

Fig. 3. Schematic diagram of the analytical procedure.

Instrumentation

The gas chromatograph is a Varian 2740 with a flame ionization detector (FID) and an election capture detector (ECD). The column (glass, I.D. 2 mm, length 2 m) contains 2.5% QF-1 and 2.5% DC 11 on Chromosorb W HP, 100–120 mesh. The carrier gas is nitrogen at a flow-rate of 30 ml/min. The temperatures used are injection, 270°; detector, 225°; temperature programme, 100–180° at 8°/min and 180–200° at 6°/min.

The effluent is split into two equal portions and measured simultaneously on the FID and ECD, so that it is possible to determine hydrocarbons and chlorinated hydrocarbons with one injection. The peaks were identified by comparing the R_F values with standards in the recovery tests; with unknown substances, it is possible to use a coupled gas chromatography–mass spectrometry system. The quantitative determination was carried out with the FID by comparing peak areas with known amounts, and with the ECD by using an automatic integrator with a calibration curve (integrator units per milligram of sample weight). It is recommended that the calibration be repeated often, as the sensitivity of the ECD may change with time.

Materials

The construction of the apparatus allows the sea water to come into contact only with PTFE, stainless steel and glass. All glassware is carefully cleaned with Detex®, rinsed with water, methanol and *n*-hexane and heated in an oven for at least 12 h at 200°.

The methanol solvent is distilled on a 1500-mm distillation column before use and each batch is tested by gas chromatography. The *n*-hexane solvent is boiled for 3 h with sodium and then distilled as for methanol.

Distilled water is prepared by boiling ion-exchanged water with $K_2S_2O_8$ for 4 h and then distilling it.

Commercial Amberlite XAD-2 is suspended several times in water and decanted to remove the finer particles and then extracted with methanol in a Soxhlet apparatus for at least 24 h.

Schleicher & Schüll No. 6 glass-fibre filters heated at 450° for 24 h were used.

TABLE I

RECOVERIES (%) FROM SEA WATER SPIKED WITH 100 μg/kg OF SUBSTANCES

Substance	*Not pre-extracted sea water**			*Pre-extracted sea water***		
	*Dissolved****	*Filter*§	*Total*	*Dissolved****	*Filter*§	*Total*
Phenanthrene	61.8	–	61.8	24.4	11.7	36.1
Pristane	—	62.5	62.5	34.6	20.3	54.9
n-Hexadecane	—	58.8	58.8	35.8	12.4	48.2

* Natural sea water, sampled in 25-l glass bottles and spiked in the laboratory.

** Water pumped once through the apparatus on board the ship, sampled in 25-l glass bottles and spiked in the laboratory.

*** Extracted from the Amberlite.

§ Extracted from the filter.

TABLE II

RECOVERIES (%) FROM SEA WATER SPIKED WITH 10 μg/kg OF SUBSTANCES

Substance	*Pre-extracted sea water**		
	*Dissolved***	*Filter****	*Total*
Aldrin	58.5	34.0	92.5
Lindane	39.8	—	39.8
DDE	49.2	51.8	101.0
DDT	47.6	40.8	88.4
PCB-2	95.2	—	95.2
Phenanthrene	42.5	59.5	102.0
Pristane	64.9	32.4	97.3
n-Hexadecane	43.2	40.3	86.4

* Water pumped once through the apparatus on board the ship, sampled in 25-l glass bottles and spiked in the laboratory.

** Extracted from the Amberlite.

*** Extracted from the filter.

TABLE III

RECOVERIES (%) FROM SEA WATER SPIKED WITH APPROXIMATELY 1 μg/kg OF SUBSTANCES

Substance	*Sea water not pre-extracted*			*Sea water pre-extracted*		
	*Dissolved**	*Filter***	*Total*	*Dissolved**	*Filter***	*Total*
Aldrin	23.7	61.1	84.8	38.6	44.7	83.3
Lindane	23.7	—	23.7	5.3	—	5.3
DDE	21.5	67.0	88.5	56.4	27.5	83.9
DDT	15.2	80.3	95.5	71.4	30.9	102.3
PCB-2	86.3	—	86.3	93.7	—	93.7
Phenanthrene	57.0	—	57.0	25.1	9.3	34.4
Pristane	—	86.0	86.0	17.5	30.9	48.4
n-Hexadecane	—	69.0	69.0	15.0	28.7	43.7

* Extracted from the Amberlite.

** Extracted from the filter.

Recovery tests

In order to determine the specificity and accuracy for non-polar substances, we conducted recovery tests. A known amount of test substance was dissolved in 10 ml of acetone and the solution was added to 100 l of sea water, the mixture being shaken for a short time. Immediately after shaking, the water was pumped over the columns. The sea water was either "pre-extracted", *i.e.*, it had already been filtered and extracted by the apparatus, and was therefore free from most particles and dissolved substances, or it was "not pre-extracted", *i.e.*, it was natural sea water from the Baltic.

The following substances were tested: DDT, DDE, Aldrin and lindane (pesticides); 2-chlorobiphenyl (PCB-2); and *n*-hexadecane, pristane and phenanthrene (hydrocarbons). Their concentrations ranged from 10 μg/kg (maximum) to 0.01 μg/kg for the chlorinated hydrocarbons and from 100 μg/kg (maximum) to 0.1 μg/kg for the hydrocarbons.

RESULTS AND DISCUSSION

The construction of the apparatus permitted large amounts of sea water to be extracted on board a ship. Contamination is minimized by avoiding contact of the sea water with plastic material, except PTFE. It is not necessary to store and preserve water samples. The concentration ratio is 1:10^5 for a water sample of 100 l; with larger water samples, it is possible to increase the concentration ratio. With two sets of columns, continuous sampling and working up at the same time is possible.

If one column is damaged, the sampling need not be interrupted, for this column could be disconnected, the remaining five columns being capable of extracting the sea water at the same flow-rate. The adsorption capacity is such that 1000 l of sea water with a content of about 1 mg/kg of dissolved organic material can be extracted without exhaustion of the columns. The flow-rate is limited first by the pressure arising from the filter which should not exceed 1 atm so as to prevent extensive damage to living cells, as this damage would increase the content of dissolved organic material. Secondly, the water should be exposed to the adsorbent for a sufficient time to allow the partition to occur. For our purpose, a flow-rate of 1–2 bed volumes/min was sufficient to extract hydrocarbons and chlorinated hydrocarbons dissolved in sea water at levels of 0.01–100 μg/kg.

As shown by the recovery tests, Amberlite XAD-2 is suitable for separating hydrocarbons and chlorinated hydrocarbons from inorganic salts even at a level of 1 μg/kg. With the exception of lindane, the recovery of all substances was about 80%. At the flow-rate used, Amberlite XAD-2 also adsorbs polar substances; for this reason the elution in the Soxhlet apparatus was carried out with a polar solvent (methanol) so that the columns were fully regenerated for the next sampling. In the next step, the non-polar substances were extracted into the *n*-hexane layer, whereas the polar substances remained in the methanol–water layer.

From the results shown in Tables I–IV, two conclusions can be drawn. Firstly, sea water, already filtered and extracted by the apparatus (pre-extracted sea water) did not give such good recoveries as natural sea water; secondly, the amount of "dissolved fractions" (extracted from the Amberlite) is greater than that retained by the filter in the recovery tests with pre-extracted sea water. The latter result was particularly evident with the hydrocarbons. The reverse was found in the recovery tests with

TABLE IV

RECOVERIES (%) FROM SEA WATER SPIKED WITH APPROXIMATELY 10 ng/kg OF SUBSTANCES

Substance	*Sea water not pre-extracted*			*Sea water pre-extracted*		
	*Dissolved**	*Filter***	*Total*	*Dissolved**	*Filter***	*Total*
Aldrin	34.5	55.2	89.7	50.2	29.5	79.7
Lindane	36.4	—	36.4	29.3	—	29.3
DDE	22.3	71.4	93.7	53.4	29.5	82.9
DDT	22.6	68.6	91.2	48.5	38.2	86.7
PCB-2	87.5	—	87.5	84.2	—	84.2

* Extracted from the Amberlite.
** Extracted from the filter.

TABLE V

SOLUBILITIES OF THE SUBSTANCES IN WATER AT 25°

Substance	*Solubility (mg/kg)*
Aldrin[25]	<0.05
Lindane[25]	7.3
DDE	0.25
DDT[25]	0.0012
Clophen A-30	0.3–1.5
Phenanthrene[26]	1.6
Pristane[27]	<0.01
n-Hexadecane[27]	<0.01

natural sea water. From these results, one could conclude that the substances used for spiking were adsorbed on small particles as soon as they came into the water and that they are not truly dissolved. This conclusion could explain the higher recoveries with natural sea water, for the retention of particles is likely to be more efficient than the sorption of material from solution. In the pre-extracted sea water, the amount of "dissolved fraction" is greater, of course, because most of the particles are removed. One must bear in mind that glass-fibre filters do not have a definite pore size but effect filtration by a process of random contact; also, they have large surface area for sorption, so that it is very difficult to decide if a filter operates as collector for small particles or as an adsorber for dissolved substances. Our results suggest that both processes occur, but that the former is more efficient if particles are present. Our results also show that substances with a higher solubility in water (Table V), i.e., phenanthrene, lindane and PCBs, are mostly recovered in the dissolved portion, whereas the more insoluble substances, which obviously are adsorbed more rapidly on particles, are mostly recovered in the filter portion. It is not possible to extrapolate simply from laboratory experiment to natural conditions, for it is well known that particles or other substances (hydrotrophy) will change the solubility of substances normally insoluble in pure water. Borneff and Knerr[23] equilibrated 1,2,5,6-dibenzanthracene for 2 years with distilled water and found nothing at the level of 0.01 μg/l, whereas a substance at this level is soluble in natural waters. Andelman and Suess[24] confirmed that the solubility of polynuclear aromatics in water could be increased by

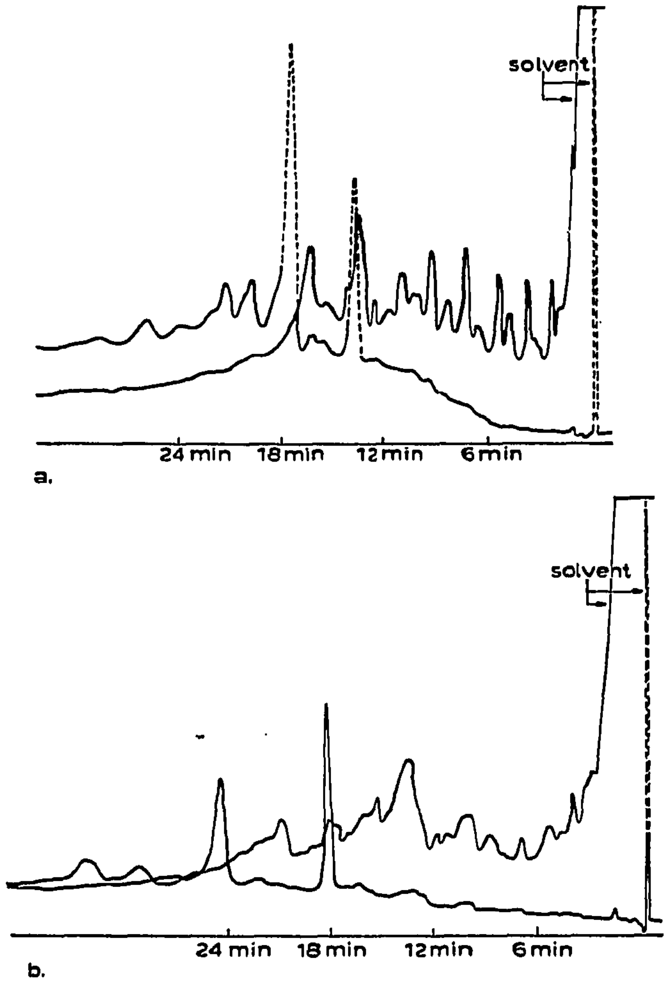

Fig. 4. Gas chromatograms of sea water sample from the Kieler Innenförde. (a) Extract filtered; (b) extract dissolved. Solid line, FID; broken line, ECD.

adding organic acids such as lactic or butyric acid. However, if one looks for the dissolved non-polar substances in sea water, it is necessary to look for the small particles, as most of the non-polar substances will be adsorbed on them.

The gas chromatograms of two samples (Figs. 4 and 5) demonstrate the applicability of the apparatus and the method. The first sample was obtained from the inner part of the Kieler Innenförde, which is a polluted area, and the other from Schönberger Strand, which is an area not directly exposed to sewage or other pollution sources. They were taken from surface water (50 cm depth). Although the identification of single peaks has not been attempted, the filter extract from the Kieler Innenförde sample has the typical pattern of gas chromatograms of a mixture of alkanes, whereas in the other sample no substances sensitive to the FID could be detected, but some compounds sensitive to the ECD with R_F values identical with those of pesticides were detected.

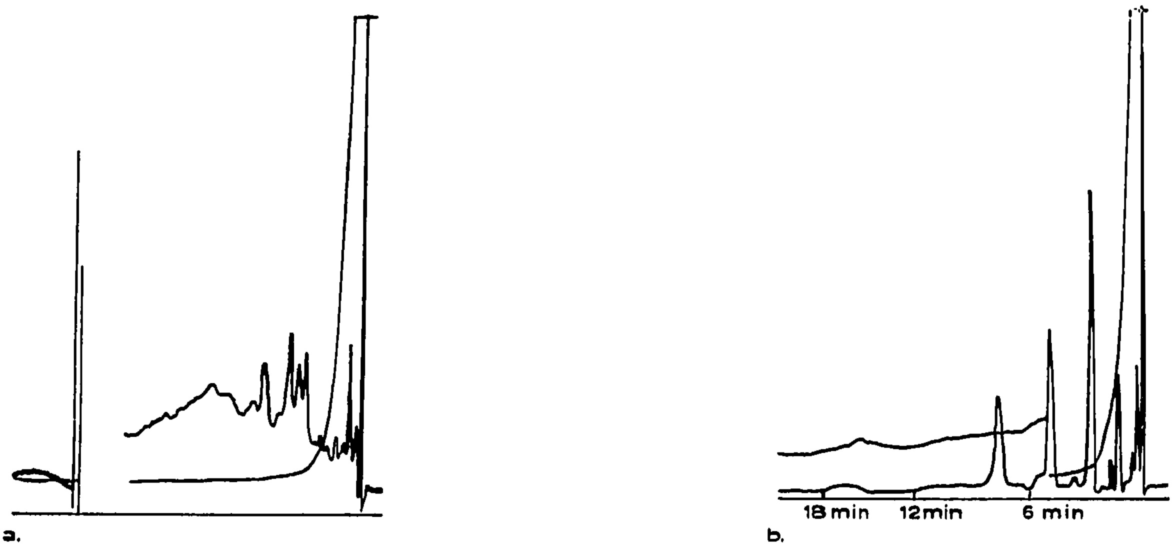

Fig. 5. Gas chromatograms of sea water sample from Schönberger Strand. (a) Extract filtered; (b) extract dissolved. Solid line, FID; broken line, ECD.

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