THE ANALYSIS OF CHLOROPESTICIDES AND PCB IN WATER
A STATISTICAL EVALUATION OF FOUR ENRICHMENT METHODS

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ABSTRACT

Chloropesticides and PCB in the ng/l-concentration range are best extracted from water by liquid/liquid extraction. Two methods showed higher recoveries and lower method standard deviations than liquid/solid extraction with commercial C-18 phase cartridges or extractive steam distillation.

INTRODUCTION

The widespread use of chloropesticides and polychlorinated biphenyls has lead to an ubiquitous occurrence of these compounds in the environment. Even after a ban on the use of some chloropesticides and of PCB's in open systems, the global deposition of PCB's in Germany is around 100 mg/ha per year due to global dispersion [1].

For the analysis of low polar organic micropollutants from water, numerous methods are described. But for the extraction step prior to the analysis, only three strategies are known:

- liquid/liquid extraction
- solid/liquid extraction
- steam distillation

The aim of our investigations was to compare these three established procedures (liquid/liquid extraction by German DIN [2], solid/liquid extraction with C-18 cartridges [3] and extractive steam distillation [4]) to each other and - in addition - to the improved liquid/liquid extraction according to Brodesser and Schöler [5] in the concentration range of 1.5 to 50 ng/l. This is the essential range for control-
ling chloropesticides and PCB's in drinking water. The limit value required by the German drinking water regulation is $0.1 \mu g/l$ [6].

18 chloropesticides and 6 PCB-congeneres were investigated (table 1). Analyses were performed with GC-ECD. Slope, rest standard deviation, method standard deviation of linear calibrations [7], limits of detection [8] and recoveries were determined applying statistical means.

| 1.2.4-Trichlorobenzene ('Tr') | Dieldrin ('Di') |
| 1.2.3.5-Tetrachlorobenzene ('Te') | $\alpha$-Endosulfan ('aE') |
| Pentachlorobenzene ('Pe') | $\beta$-Endosulfan ('bE') |
| Hexachlorobenzene ('He') | pp'-DDE ('DE') |
| Lindane ('Li') | Methoxychlor ('Me') |
| Quintocen ('Qu') | pp'-DDD ('DD') |
| Heptachlor ('He') | pp'-DDT ('pD') |
| Heptachlorepoxide ('Ep') | op'-DDT ('oD') |
| Aldrin ('Al') | PCB: # 28, # 52, # 101, # 138, # 153 and # 180 |
| Endrin ('En') | |

Table 1: Chloropesticides and PCB's used in the investigations

**EXPERIMENTAL**

For each method (which are specified below), 5 parallel experiments were carried out on 5 concentration levels between 1.5 and $50 \, ng/l$ - along with 5 blind value determinations.

All chemicals used were of analytical quality. The purity of the solvents was checked by evaporating amounts in the order of magnitude as they were used in the extraction procedures down to $0.5 \, ml$ (with $1 \, ml \, i$-octane as keeper) and investigating them with GC. All solvents were suitable except for pentane, which was rectified to obtain a tolerable blank.

The water used to prepare the synthetic samples had been bidistilled and furtheron extracted to eliminate organic residues. In the case of the solid phase enrichment, the water was extracted with C-18 phase cartridges corresponding to the way the samples were treated. For the DIN-method, the water was extracted several times with pentane, for the other two methods with hexane and - after separation of the solvent - heated to boil.

1 L portions of the precleaned water were fortified with a solution of the investigated compounds in $500 \, \mu l$ acetone and extracted immediately.

**Methods of enrichment**

**Light phase rotation perforator:**

125 ml n-pentane are added to the 1 l water sample in the rotation perforator (figure 1a). The solvent in the solvent vessel is heated to evaporate, enters the con-
denser, is liquified and flows into the rotating distributor. From there it is dispersed into the water as minute droplets, which percolate the water phase. Thus the organic phase is continuously exchanged. After 1 h of extraction, the solvent is transferred to a calibrated flask (a 250 ml pointed flask, to which a tip with a calibrated volume of 3 ml is attached) and 2 ml i-octane are added as a keeper. The solvent is evaporated by help of a rotary evaporator through a 10 cm Vigreux-column first on a heated water bath (55 °C), then with vacuum of a water jet pump in air bath, allowing the extract temperature to decrease below 0 °C.

Figure 1: a) Light Phase Rotation Perforator  b) Microseparator  
c) Solid Phase Extraction Cartridge  d) Extractive Steam Distillation App.
**DIN-method:**

Placed in a 1 l Erlenmeyer flask, the water sample is intensively stirred by a magnetic stirrer with 10 ml n-pentane for 10 min. After standing for another 10 min, the organic phase is separated by help of a microseparator (figure 1b) and after addition of 1 ml i-octane reduced to 0.5 ml as described above.

**Solid phase extraction:**

The commercially available cartridges (C-18 phase, 500 mg, Analytichem, figure 1c) are first conditioned with 5 ml hexane, methanol and purified water respectively. The water sample (containing 1 % methanol) is then extracted by help of a slight vacuum with a flow rate of 7-17 ml/min. After drying with nitrogen, the solid phase is treated with 3 x 1 ml hexane. 1 ml i-octane is added to the eluate and the volume reduced to 0.5 ml as described above.

**Extractive steam distillation** (figure 1d):

The water sample is refluxed in a round flask for 2 hours (heating output 450 watts). The condensed steam flows into an extraction device, where it percolates a 15 ml layer of cooled hexane before continuously returning into the round flask. After transfer to a calibrated flask and addition of 1 ml i-octane, the hexane phase is reduced to 0.5 ml as described above.

**Gas chromatography**

- **gas chromatograph:** Vega 6130a, Carlo Erba
- **injection:** split/splitless according to Grob, Carlo Erba; temperature 270 °C; "hot needle", volume "1 µl"; 1 min splitless, then split ratio 1:40
- **detection:** 63Ni ECD, Carlo Erba; temperature 300 °C
- **retention gap:** FS-phenyl-sil desact. 2.5 m x 0.32 mm; C&S
- **column:** CP-Sil-8 CB, 50 m x 0.32 mm, 0.26 µm df, Chrompack
- **carrier:** nitrogen, 99.999 %; flow rate 3.0 ml/min
- **make-up:** argon/methane 95:5, flow rate 40 ml/min
- **temperature program:** 60 °C, 1 min; 60 °C - 180 °C, 25 °C/min; 180 °C - 232 °C, 4 °C/min, 11 min; 232 °C - 280 °C, 48 °C/min, 10 min
- **data aquisition:** integrator HP 3390 A, Hewlett Packard

**Data Evaluation**

To eliminate the influence of detector instabilities and changes in the working conditions of the whole gas chromatographic system, we choose to record a standard chromatogram of comparable concentration directly preceding each sample chromatogram. The standard values were used differently for the determination of either recoveries and characteristic method values.
Recoveries were determined by relating the area values of the corresponding standard and sample chromatograms. Each experiment was carried out five times under identical conditions. So five values were obtained for each compound, for each concentration and for each method, from which average values and standard deviations were determined.

As characteristic values for the procedures, slope (sensitivity), rest standard deviation and method standard deviation of linear regressions and limits of detection were determined. The regression function was determined by the method of the least squares. The calibration value for each concentration resulted from averaging the measured values of five experiments. To minimize systematical errors, these five values were standardized beforehand: One of the five samples was deliberately chosen as reference. The reference standard and the standards of the other four samples were related to form correction factors by which the sample values were multiplied. The means over these standardized values were calculated and used for the linear regression.

**RESULTS**

**Characteristic values**

A linear regression was justified for the enrichment with the rotation perforator, the DIN-method and the extractive steam distillation. In the case of the solid phase extraction, we observed a strong decrease of sensitivity at water sample concentrations of 25 and 50 ng/l. The cause for this behaviour we see in an altogether insufficient extraction capacity of the used 500 mg cartridges accompanied with a selective adsorption of compounds with higher affinity to the active sites. For sake of comparibility, a linear regression was also applied in this particular case.

No results are given in the case of the solid phase extraction for the compounds \( \beta \)-endosulfan and methoxychlor. These compounds could not be reliably enriched (see also section “recoveries”.)

**Sensitivity \( A_1 \)**

In figure 2 the slopes of the linear regression functions \( A_1 \) in the equation \( Y = A_1 \cdot X + A_0 \) are compared. The different detector response of the compounds is reflected in comparably low values for 1,2,4-trichlorobenzene and 1,2,3,5-tetrachlorobenzene or relatively high values for aldrin e. g. Regarding the respective methods, both liquid/liquid extractions performed equally well, their means being about 50 % higher than the average of the solid phase extraction. The steam distillation procedure showed relatively low sensitivities especially for heptachlor, \( \beta \)-endosulfan and methoxychlor.

**Rest standard deviation \( S_y \) (figure 3)**

The rest standard deviation describes the deviation of the measurements from the regression function. It was determined as
\[ S_y = \left( \sum \left( \frac{(Y_i - \bar{Y}_i)^2}{N - 2} \right) \right)^{1/2} \]

with \( Y_i \) = measurement of the calibration at the concentration \( i \)
\( \bar{Y}_i \) = function value of the linear regression at the concentration \( i \)
\( N \) = number of measurements

As can be seen in the diagram, similar results were obtained for the two liquid/liquid extraction methods and the extractive steam distillation. Still, the lowest values were generally found for the steam distillation. Very high rest standard deviations on the other hand resulted for the solid phase extraction. As we stated above, a saturation effect was observed with this method for most compounds.

**Method standard deviation** \( S_{x0} \) (figure 4)

The method standard deviation is a measure for the quality of an analytical procedure, relating the slope and standard deviation of the regression function:

\[ S_{x0} = \frac{S_y}{A_1} \]

The mean over all compounds reveals a slight advantage of the extraction with rotation perforator \((S_{x0} = 2.9 \text{ ng/l})\) over the DIN-method \((S_{x0} = 3.4 \text{ ng/l})\) and the extractive steam distillation procedure \((S_{x0} = 3.8 \text{ ng/l})\). Very high values were found for the solid phase extraction, due to the high rest standard deviations.

**Limit of detection** \( NG \) (figure 5)

The limits of detection were determined by the formula:

\[ NG = 2 \cdot t_{f,95} \cdot \sigma_{C} / S \]

with \( \sigma_{C} = \left( \frac{(m - 1) \cdot \sigma_a^2 + (n - 1) \cdot \sigma_b^2}{m + n - 2} \right)^{1/2} \) (weighted standard deviation)
\( \sigma_a \) = standard deviation of the measurements at the lowest investigated concentration level
\( \sigma_b \) = standard deviation of the blind values
\( m \) = number of measurements (= 5)
\( n \) = number of blind value measurements (= 5)
\( t_{f,95} \) = t-factor for twosided questioning and 95 % statistical safety \((f = m + n - 2)\)
\( S \) = slope of the linear calibration

The limits of detection lay generally between 0.5 and 5 ng/l. High values above 10 ng/l had their origin in high blank values, which on the other hand cannot be totally attributed to the respective extraction procedures. In some cases, blanks were due to contaminations of the gas chromatographic system. Leaving the values above 10 ng/l aside, we receive quite comparable mean values for all methods of about 2.5 ng/l.
Figure 2: Sensitivity $A_1$

Figure 3: Rest Standard Deviation $S_y$
concentration (ng/l)

Figure 4: Method Standard Deviation $S_{x_0}$

Figure 5: Limit of Detection NG
Recoveries

The results for the 10 ng/l concentration level of the water samples are shown in figures 6 - 9. In figure 9 for the solid phase procedure also for 50 ng/l.

Recoveries ranged between 80 and 90% for the liquid/liquid methods. At 50 ng/l, the DIN-method was found to be more efficient, while near the detection limits, extraction by rotation perforator led to higher recoveries. For both procedures, recoveries were generally better than 70%. Standard deviations were typically about 10%.

Unacceptably low recoveries were on the other hand found for the extractive steam distillation, in contradiction to other investigations [4, 9]. Only in exceptional cases, values above 70% were obtained. A greatly prolonged extraction time of 4 hours or more would probably have allowed higher recoveries, but was not chosen for practical reasons. \( \beta \)-endosulfan and heptachlor showed recoveries even below 20%. Either these compounds hydrolyse or they are insufficiently steam volatile.

The solid phase extraction gave recoveries between 80 and 100% at 4 and 10 ng/l water sample concentration, although with standard deviations between 15 and 30%. At 25 and 50 ng/l, recoveries declined, so that only for lindane and quintocen, values above 70% were received at 50 ng/l. Methoxychlor, \( \beta \)-endosulfan, and to a minor degree also endrin, dieldrin, and \( \alpha \)-endosulfan showed an unreliable enrichment behaviour. Concerning methoxychlor for example, in less than half of the experiments and independent of the concentration, the compound could be recovered at all, while in the other experiments, recovery was mostly high. No recoveries are therefore given for methoxychlor and \( \beta \)-endosulfan.

Figure 6: Rotation perforator: Recoveries at 10 ng/l
Figure 7: DIN-method: Recoveries at 10 ng/l

Figure 8: Extractive Steam Distillation: Recoveries at 10 ng/l
Figure 9: Solid Phase Extraction: Recoveries at 10 and 50 ng/l
DISCUSSION

The best enrichment results were obtained with the liquid/liquid extraction methods. A slightly better performance of the extraction with rotation perforator stands beside practical advantages of the DIN-method, which requires only simple, inexpensive equipment. The high enrichment potential of perforation extraction for medium or highly polar organic micropollutants from water (10, 11) is not necessarily required for the enrichment of low polar organic residues. Comparably high amounts of highly purified solvent are needed for the perforator method. A practical problem with this method turned up when we analyzed mineral water: as dissolved carbon dioxide escaped, calciumcarbonate precipitated, which blocked the rotating distributor. Sometimes severe phase separation problems may occur with both methods on analyzing real samples.

Extractive steam distillation and solid phase extraction both revealed high deficiencies in their respective enrichment performance. Steam distillation is an extraction method especially useful for the investigation of highly polluted waters or sediments, as it comprises a clean-up step: lipids, waxes and related natural substances will not be codistilled [4]. We assume that the low enrichment potential of the steam distillation is due to an insufficient exchange between the condensed steam and the organic solvent, which would be largely increased by a forced mixing of the two phases.

Concerning the solid phase extraction, the use of higher capacity - for example 1 or 2 g - cartridges should reduce the observed effects of insufficient enrichment capacity. As a practical limitation of this method, we observed a severe clogging tendency of the solid phase when extracting real samples containing iron hydroxide or considerable amounts of microorganisms.

LITERATURE


