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FURTHER DEVELOPMENT OF DIRECT AQUEOUS INJECTION WITH ELECTRON-CAPTURE DETECTION IN GAS CHROMATOGRAPHY

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SUMMARY

Direct aqueous injection with electron-capture detection (DAI–ECD) is of increasing interest for the determination of halocarbons in water in the 0.01–10 ppb range. It is particularly useful for very volatile halocarbons that may be obscured by a solvent or by solvent byproducts in the course of determination with solvent extraction or with closed-loop stripping. Owing to its simplicity and its straightforward basis of quantification, it also compares favourably with direct headspace analysis. More conclusive information is presented concerning technical details of the method, particularly concerning the column. The chromatographic conditions and the detection have been further refined, resulting in substantially lowered detection limits. The use of DAI–ECD for heavier halocarbons is described.

INTRODUCTION

Our first report on direct aqueous injection with electron-capture detection (DAI–ECD)[1] described the principles and the fundamental technique, including column, sampling and the analytical conditions. As far as we know, the paper by Schulz[2] is the only one on the same subject to have appeared in the meantime. Our first paper was written when little routine experience had been gained, and it is the purpose of this paper to discuss our experience with the method in application laboratories, to complete the description of the fundamentals and to report basic and practical improvements. Our recent work has produced a number of refinements, but no basic modification of the method has proved necessary.

PRACTICAL INTEREST IN DAI–ECD

Interest in DAI–ECD depends on its merits compared with those of other methods. Widely used alternative methods are pentane extraction, closed loop stripping analysis (CLSA) and headspace analysis, all combined with ECD.

The comparison is strongly influenced by the priority given to typical characteristics of a method. DAI–ECD is by far the most rapid (simple injection of water samples), the simplest (no extraction or concentration procedures) and the quanti-

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tatively most reliable method (no recovery problems, no artifacts). On the other hand, the sensitivity of DAI–ECD is limited to the absolute amount of halocarbons present in a 2 μl water sample. In contrast, pentane microextraction[3] offers a 200–500-fold concentration. Recently[4], its concentration power has been increased to about 10,000, by increasing the volume of pentane solution injected to 100–250 μl. It is well known[3] that CLSA concentrates water contaminants by a factor of the order of 10^6.

There is a narrow, but important, application for which DAI–ECD cannot be replaced by pentane extraction or by CLSA. It covers very volatile halocarbons ranging from some frigenes, dichloroethylene and methylene chloride up to chloroform. The last two substances in particular have caused problems in many laboratories. They are readily soluble in water, which renders their extraction from water critical. In addition, the light halocarbons are eluted close to solvents such as pentane and carbon disulphide, which creates problems with the influence of the solvents or their byproducts. Further, during splitless or on-column injection, the same solvents may exert partial solvent trapping effects[5], leading to distorted peaks unsuitable for integration. DAI–ECD does not suffer from these difficulties and is, therefore, suitable for these important water contaminants. The only alternative method is headspace analysis[6], which is, however, more critical in terms of quantification.

When considering even more volatile halocarbons, such as the light frigenes, and vinyl chloride, further problems arise. On the one hand there is no sufficiently sensitive and selective detection method for these substances, and on the other, they are so easily evaporated from water that handling of the water samples becomes critical. Consequently, they are best determined by a headspace technique combined with concentration on an adsorbent, followed by thermal desorption.

In summary, DAI–ECD is selected primarily for the analysis of light halocarbons (methylene chloride to chloroform range), for which an equivalent alternative is not available. In addition, it is also selected for heavier halocarbons (for which there are alternative methods) when the sensitivity is sufficient, and when the emphasis is on quantitative reliability or on simplicity and rapidity.

COLUMNS

As pointed out previously[1], a suitable column for DAI–ECD has to fulfill two primary requirements. It must elute water rapidly and completely, and it must elute water long before light halocarbons such as methylene chloride.

Rapid elution of water is achieved from an apolar column. Complete elution of water is typical of an inert column. Surprisingly, however, different apolar, inert columns, which, according to general testing, are found to be of high quality, may elute substances such as methylene chloride and chloroform with peak areas differing by a factor of 2–3. We attribute the variations to slight differences in inertness. A slight residual activity will cause trace amounts of water to be eluted with delay and this water then lowers the ECD response.

The second requirement is unusual. It means that a more volatile substance (methylene chloride) should be eluted as late as possible after a less volatile one (water). The thickest possible coating is the only means of solving the apparent contradiction. It provides the necessary strong retention of the apolar halocarbons and the high loading capacity, avoiding excessive broadening of the water peak. A factor

limiting film thickness is the loss of separation efficiency. The peak broadening due to excessively thick films means a proportionally reduced sensitivity. As a compromise, 5.0 μm films (in 0.28–0.33 mm I.D. columns) have proved to be optimal. This finding was tentatively presented earlier[1], and has now been confirmed. Owing to its particular ease of coating and immobilization, PS-255 (Petrarch Systems, Bristol, PA, U.S.A.; Fluka, Buchs, Switzerland) has become the standard stationary phase for such columns.

Column length is another matter of compromise. Long columns give better resolution of methylene chloride from water, but with a simultaneous loss of sensitivity. For general use, a coated length of 25–30 m has been established as an optimum. Longer columns may be desirable when the emphasis is on the most volatile halocarbons. Shorter columns are preferred for achieving maximum sensitivity.

Durability is an essential column characteristic under the conditions of direct water injection. Column performance may suffer from two independent influences. The first is typically related to on-column injection. Non-volatile byproducts, primarily from hard water, are deposited in the column inlet. Fortunately, even heavy (easily visible) inorganic deposits do less harm than originally expected, and the bulk of these deposits is accumulated in the first 20–30 mm inlet section. A relatively small, invisible portion of the deposit is distributed over the entire flooded length of the column inlet (about 50 cm under the recommended conditions). A second influence affects the entire column length, and has probably to be interpreted as hydrolysis of the stationary phase. Unfortunately, both influences produce identical damage symptoms. Both an inorganic deposit and a slightly increased activity due to hydrolysis retard the complete elution of water, which is manifested by a broadened water peak and by a generally lowered ECD sensitivity. Thus, the sources of poorer column performance can hardly be distinguished from a regular chromatogram, *i.e.*, without column manipulation.

COLUMN REGENERATION

The first symptom of damage is always broadening of the water peak, resulting in poorer resolution of the most volatile halocarbons from water. Depending primarily on water hardness, the damage will become visible after 50–100 2 μl on-column injections. Shortening the column inlet by 5 cm will greatly improve but not completely restore the column performance. A second inlet shortening after an additional 50–100 runs will provide a less efficient improvement. It will be a matter of judgement whether the regeneration is acceptable. Possibly replacing the inlet section is then a better method.

We recommend mounting a fresh column with a 1–1.5 m long empty persilanized inlet section of 0.32 mm I.D. fused silica, connected to the column by a butt connector (both materials available from Carlo Erba, Milan, Italy). The pre-column serves to retain both the concentrated salt deposit close to the injection point and the deposit distributed over a longer section. As soon as shortening the inlet no longer provides sufficient improvement, the pre-column is replaced. This means that the entire length possibly loaded with inorganic salts is discarded (see Fig. 1 as a practical example). If the replacement is made after 100–200 runs, the original column performance may be nearly restored. If (as in Fig. 1) the replacement is made later,

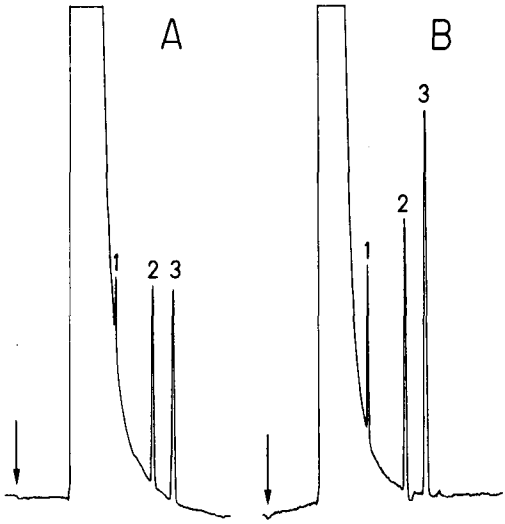

Fig. 1. Regeneration by manipulating the column inlet. The column (28 m × 0.29 mm I.D., 4.2 μm SE-54) had been run with about 300 drinking water samples and 40 samples of extremely hard mineral water. The inlet section had been shortened by a few centimetres five times and re-straightened twice. After this history, the column produced chromatogram A (for substances and conditions, see Fig. 2). Shortening the inlet no longer improved the performance. Methylene chloride (No. 1) was co-eluted with the last portion of water. Chromatogram B was obtained under identical conditions after breaking away 70 cm of the inlet, and connecting 1 m of empty, persilanized fused-silica tubing. Note the improved resolution, and the almost 4-fold increased detection of methylene chloride. Despite the considerable improvement, the performance was not restored (compare with Fig. 2). If a better performance in the area of methylene chloride is required, the column would have to be re-silylated.

hydrolysis of the phase may have proceeded to an extent that precludes a satisfactory solution just by manipulating the column inlet. The original column quality can then be restored by re-silylation.

As re-silylation is sufficiently effective only in the liquid phase[7], some remarks on column washing should be made. Whereas washing a column with an immobilized coating of standard thickness (0.1–0.5 μm) is easy and uncritical, washing a very thick coating for the first time may result in irreversible plugging. The reason is that even the few percent of still extractable phase is sufficient to increase the viscosity of the washing solvent and result in permanent plugging. The problem is overcome by first using a solvent that becomes saturated before a critical viscosity is reached. For apolar silicones, acetone is such a relatively poor solvent. As a first wash, 20–50 ml of acetone are slowly forced through the column overnight. In the morning, the washing is completed with methylene chloride.

After washing, re-silylation is carried out as described previously[7]. We have found that the hydrolysis due to water injection hardly affects the support surface. This allows the re-silylation temperature of 300°C (ref. 7) to be lowered to 200°C.

Figs. 2–5 were produced by a column that had been re-silylated after about 250 runs, followed by another 80 runs. In other words, these traces were intentionally prepared under average routine conditions. Much better results are obtained with a fresh column.

After 100–200 injections, a layer of very fine, needle-shaped crystals will build up in the column outlet section. The crystals are first formed where the tubing is heated from the column to the detector temperature, and then expand over a longer distance. Apparently this deposit does no harm. So far we do not know the origin or nature of the crystals.

SAMPLING AND CHROMATOGRAPHIC CONDITIONS

The importance of on-column injection for avoiding delayed water elution caused by diffusion and dilution in the injector cavity has been discussed[1]. No additional information is required on this aspect.

We also mentioned[1] that on-column injection produced sharper peaks than splitless injection, without being able to explain the difference. The phenomenon is now fully understood[5]. The halocarbons of interest have a sufficiently low solubility in water to be classified as "non-trapped" in the condensed solvent (water). This means that their vapours flow freely over the condensed water covering the wall of the column inlet. Thus, no solvent trapping occurs. In the course of splitless injection the vapours are heavily diluted with carrier in the injector before they enter the column. This leads to a very broad initial band (band broadening with time). As there is no reconcentration effect, the eluted peaks are severely broadened and distorted also. In contrast, on-column injection produces ideally sharp peaks. The reason is that the halocarbons are evaporated from the water *inside* the column, where virtually no dilution with carrier gas occurs. Thus the initial bands are far shorter than with splitless injection.

DAI with on-column injection is not just a general method for water sampling. Slightly more polar, *i.e.*, more water soluble, substances are partially trapped by condensed water[5], and are eluted with distorted peak shapes. Ideal peaks are again obtained with strongly polar substances that are fully trapped in the water layer, thus experiencing a regular solvent trapping effect.

The previously recommended analytical conditions[1] can be roughly confirmed and some optimal values can now be defined more precisely. The optimal column temperature for isothermal work with 5.0 μm columns is restricted to 103–104°C. At lower temperatures the elution of water is delayed, resulting in poorer resolution of very light halocarbons. Higher column temperatures may cause peak splitting. We do not understand the phenomenon. The splitting becomes more severe with increasing molecular weight. It has nothing to do with band broadening in space, and cannot be influenced by secondary cooling.

The carrier flow conditions are more variable, and have to be optimized according to the primary objective of the analysis. One extreme situation is characterized by a pronounced emphasis on the most volatile halocarbons. In this instance, the carrier flow has to be optimized for maximum separation efficiency. For 5 μm columns this requires nitrogen as the carrier gas with an average linear flow-rate of 0.1–0.2 m/sec. However, this very slow analysis results in reduced sensitivity. Thus, if resolution from water is not the crucial point, the carrier gas flow-rate is commonly increased beyond the optimum for separation efficiency. Usual values are 0.2–0.4 m/sec for nitrogen and 0.3–0.6 m/sec for hydrogen. Theoretically, temperature programming is very efficient in combining good resolution with high sensitivity. In

practice, at least when additional attenuation is not permitted, temperature programming is excessively time consuming owing to the long waiting periods for re-equilibrating the system.

The standard sample size is 2 μl. With more concentrated samples the sample size may be reduced in order to protect the column.

DETECTION

The basic features of an electron-capture detector contributing to suitability for DAI–ECD are related to the importance of allowing a large amount of water vapour to pass through the detector within a short time, without any residue.

Geometrically, the detector cell should approach as closely as possible a cylindrical shape. We are aware that other requirements may necessitate a more sophisticated cell shape, so a useful compromise should be sought. A further basic feature is heat resistance. As shown by Verga[8], the most straightforward way of counteracting broadening of the water peak is a detector temperature of 300–400°C. Obviously, this excludes tritium as a source of radiation, whereas nickel-63 is well suited. Fig. 2 shows a substantial improvement of the water elution with a temperature increase from 300 to 340°C. Further heating to 380°C gave little further improvement. Accordingly, our standard detector temperature was fixed at 350°C.

As expected, the elution of water is also influenced by the flow-rate of the make-up gas. According to Fig. 3, the elution improves continuously as the flow-rate increases from 30 to 70 ml/min. A further increase in flow-rate no longer influences the water peak, but reduces the halocarbon signals. Between 30 and 70 ml/min, two

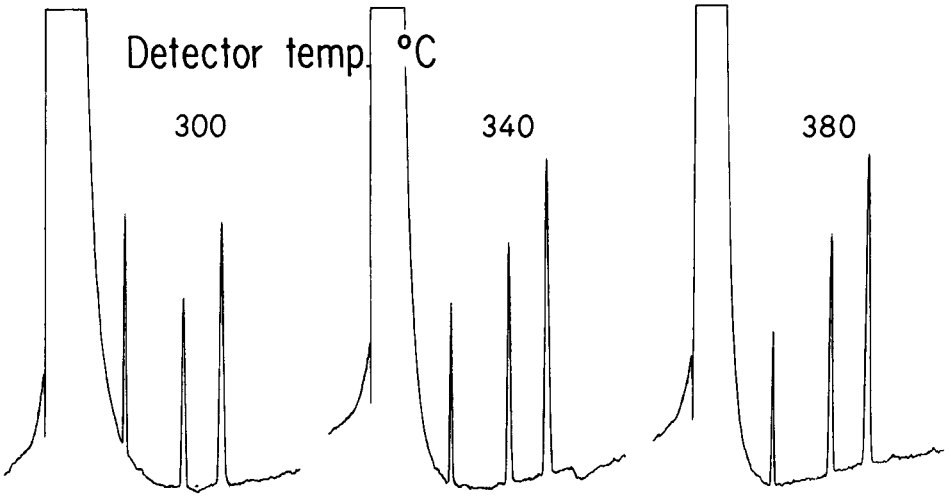

Fig. 2. Influence of ECD temperature. Column: 26 m × 0.32 mm I.D., 5.0 μm PS-255. Column temperature: 104°C (isothermal). Carrier gas: nitrogen at 0.25 m/sec. Injection: 2 μl on-column, with secondary cooling. Gas chromatograph: Carlo Erba Model 4160. Detector: Carlo Erba Model 400 electron-capture detector; temperature as indicated; make-up gas, nitrogen at 50 ml/min; 50 V, 1 μsec, constant current 1.0 nA, 1.15 kHz, attenuation ×4. Electrometer and recorder: 1 mV. Substances: 1 = methylene chloride; 2 = 1,1,1-trichloroethane; 3 = chloroform. Note the improved resolution of methylene chloride from the water peak with increased detector temperature.

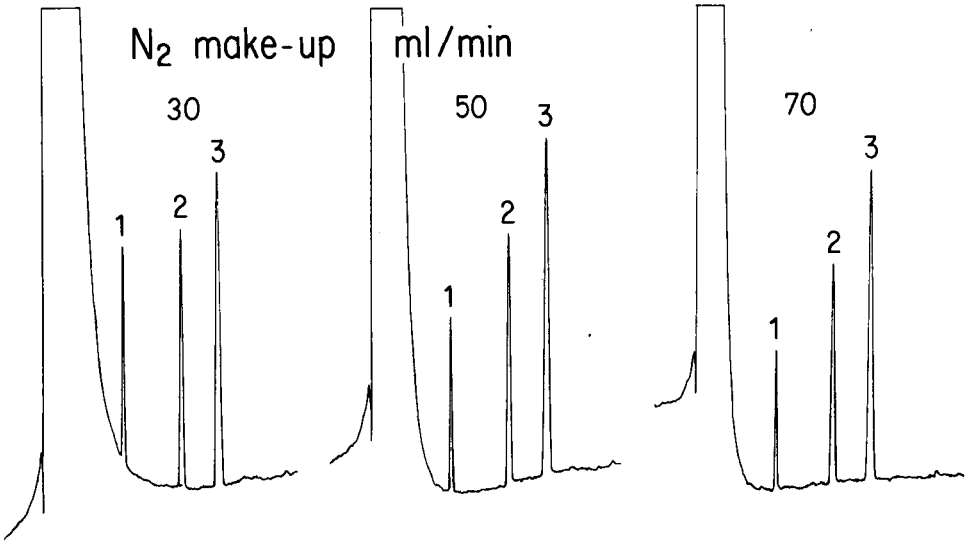

Fig. 3. Influence of make-up gas flow-rate. For column, conditions, equipment and substances, see Fig. 2. Note the decreasing width of the water peak with increasing make-up gas flow-rate.

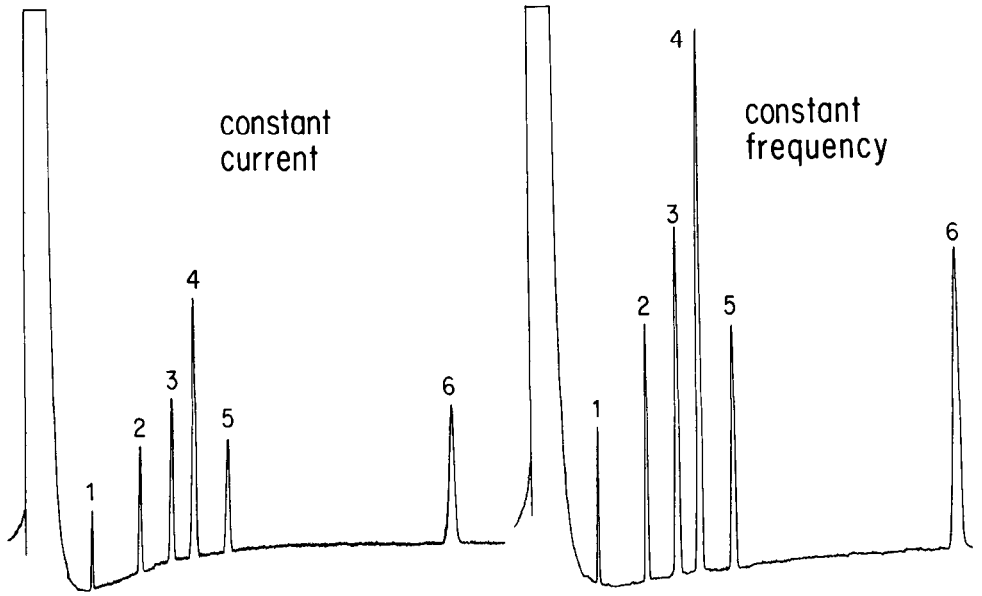

Fig. 4 Influence of ECD operation mode. For column, conditions and equipment see Fig. 2. Constant frequency, 1.20 kHz, 0.95 nA. Substances and concentrations: (1) 10 μg/l methylene chloride; 1.0 μg/l each of (2) 1,1,1-trichloroethane, (3) chloroform, (4) tetrachloromethane, (5) trichloroethylene and (6) tetrachloroethylene. With identical noise levels (slightly higher in the figure with constant current), constant frequency produces 2.5-fold larger peaks.

competing effects nearly compensate each other. With a higher make-up flow-rate the detector sensitivity is increased owing to more efficient water elution, but simultaneously the response is decreased owing to the decreased vapour concentration of the halocarbons.

The detector may be operated with a constant current or constant frequency. Generally, constant current is the preferred mode owing to the greater linearity with various concentrations of halocarbons. However, as Fig. 4 shows, constant frequency is superior in terms of extreme sensitivity. When recorded with identical noise levels, the constant frequency mode produces 2.5–3 times larger peaks than the constant current mode.

As a combined effect of improvements concerning the column, the chromatographic conditions and particularly the detection, the detection limits compared with those reported earlier[1] are now greatly lowered. The new detection limits are as follows: methylene chloride, 0.6; trichloroethane, 0.03; chloroform, 0.02; tetrachloromethane, 0.015; trichloroethylene, 0.03; and tetrachloroethylene, 0.025 μg/l. The detection limits for the last two substances are valid under analytical conditions favouring the determination of the most volatile halocarbons. Under conditions optimized for the heavier halocarbons, the detection limits are lowered by a factor of 2–3.

RELATIVE ECD RESPONSE TO DIFFERENT HALOCARBONS

When the ECD response changes as a consequence of variations in parameters, the changes may not be the same for different halocarbons, particularly for halocarbons with different halogen contents. This well known fact has been studied by numerous workers (*e.g.*, ref. 9). Hence here we discuss only briefly some aspects of the problem in the context of DAI–ECD.

Influence of detector temperature

The detector temperature is known to be a major parameter influencing relative response. As shown by Fig. 2, heating the electron-capture detector greatly reduces its response to methylene chloride compared with trichloro compounds. The fact may seem surprising at first, as heating the detector enhances water elution and would, therefore, be expected to increase the detector response. The reduced response is due to specific temperature-dependent behaviour of the detector.

Influence of make-up gas flow-rate

A similar change is caused by an increased make-up gas flow-rate. The improved water elution due to the increased flow-rate would again be expected to increase the response, particularly for early eluted substances. As shown by Fig. 3, the opposite is true. Fig. 5 shows response changes for some frequently detected water contaminants.

A further parameter influencing the relative detector response is the ground current of the electron-capture detector[8].

Fig. 1 may serve as a practical example illustrating the complex problem of relative detector response. After replacing the inlet section (chromatogram B), a far greater response to chloroform (No. 3) compared with trichloroethane (No. 2) is

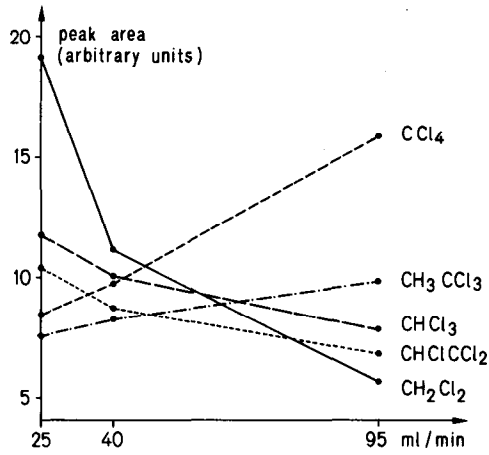

Fig. 5. Influence of make-up gas flow-rate on ECD response. For column and conditions, see Fig. 2. Values determined with 2 μl injections of a 10 μg/l solution for methylene chloride and 1 μg/l solutions for all other substances. The only simple rule to be deduced is that with more halogen atoms per halocarbon molecule, increasing the make-up gas flow-rate tends to increase the ECD response. More detailed correlations are complex. (Courtesy G. Verga, Carlo Erba, Milan).

observed. It is tempting to attribute the effect to more efficient water elution, but we have little evidence for this interpretation.

As a summary, the factors that enhance the detector response to methylene chloride, a compound attracting increasing interest, and for which DAI-ECD is a particularly suitable method of determination, may be listed as follows: low make-up gas flow-rate, low detector temperature and low detector ground current.

QUANTITATIVE ANALYSIS

Quantification is preferably carried out by running standard solutions, from which calibration graphs for all substances of interest are constructed. With samples with widely varying concentrations, it is important to check for a possible non-linear detector response in the range covered.

Internal standards are normally not used, for two reasons. First, the quantitative reproducibility of a 2 μl on-column injection (preferably with a 5 μl syringe) under the described conditions clearly permits work with an external standard. Second, the very variable detector response to individual substances renders internal standards virtually impractical.

The possible influence of hard water[1] on quantification should be emphasized. Hard water may substantially retard water elution, thus reducing the ECD sensitivity, particularly for the most volatile substances. In such instances we recommend carrying out the calibration with standard solutions prepared with water of hardness similar to that of the sample.

DAI-ECD FOR HEAVIER HALOCARBONS

So far we have limited the discussion to volatile halocarbons for the simple

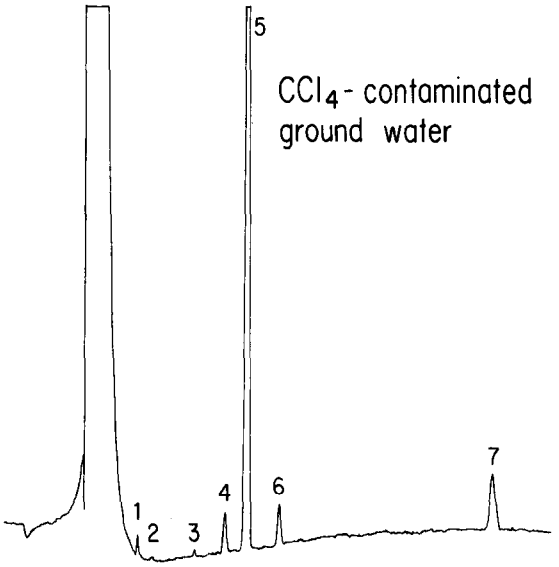

Fig. 6. An arbitrarily selected routine example: analysis of a CCl_4-contaminated ground water. Note that this is the only hard water sample presented; all other chromatograms originated from standard solutions prepared with deionized water. Column, conditions and procedure as described in Fig. 2. Substances and concentrations: 1 = 1,1-dichloroethylene (0.7 μg/l); 2 = methylene chloride (0.2 μg/l); 3 = 1,1,1-trichloroethane (0.02 μg/l); 4 = chloroform (0.12 μg/l); 5 = tetrachloromethane (2.8 μg/l); 6 = trichloroethylene (0.16 μg/l); 7 = tetrachloroethylene (0.18 μg/l).

reason that DAI-ECD is particularly useful in this range. Heavier halocarbons may equally well, or in trace analysis even better, be assessed by solvent extraction or by CLSA. However, owing to its rapidity and reliability, DAI-ECD can also be used for heavier substances.

As discussed already, temperature programming, at least with relatively high sensitivity, is not recommended. Thus, isothermal work at a suitably increased temperature seems to be the solution. However, this would cause problems with the conditions of on-column injection.

We have found that the DAI-ECD technique is best adapted to heavier halocarbons by reducing the film thickness of the column to allow the heavier halocarbons to be ideally eluted at 104°C and the analysis is then carried out exactly as with volatile halocarbons.

A typical example is the analysis of the dichlorobenzenes, particularly 1,4-dichlorobenzene, which are frequently found in water used for sanitary purposes. The ideal column for these substances is coated with a 0.8 μm film. Isothermal analysis at 104°C is then carried out as described for volatile halocarbons.

Fig. 6 shows the analysis of a real sample arbitrarily chosen from the routine work done at EAWAG.

REFERENCES

1 K. Grob and A. Habich, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 6 (1983) 11
2 J. Schulz, *Application Note*, Hewlett-Packard, Waldbronn, 1983.
3 K. Grob, K. Grob, Jr. and G. Grob, *J. Chromatogr.*, 106 (1975) 299.
4 E. Fogelquist and M. Larsson, *J. Chromatogr.*, 279 (1983) 297.
5 K. Grob, Jr., *J. Chromatogr.*, 279 (1983) 225.
6 B. Kolb, M. Auer and P. Pospisil, *J. Chromatogr.*, 279 (1983) 341.
7 K. Grob and G. Grob, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 5 (1982) 349.
8 G. Verga, Carlo Erba, Milan, personal communication.
9 E. D. Pellizzari, *J. Chromatogr.*, 98 (1974) 323.