ELSEVIER

Journal of Chromatography A, 703 (1995) 309–325

JOURNAL OF CHROMATOGRAPHY A

Review

Two-dimensional gas chromatography[1]: principles, instrumentation, methods

Gerhard Schomburg

Department of Chromatography and Electrophoresis, Max-Planck-Institut für Kohlenforschung, 45470 Mülheim-Ruhr, Germany

Abstract

The principles and potentials of two or multidimensional gas chromatographic separations (MDGC) of different types of complex mixtures are outlined with regard to analytical application. Except for preparative-scale and certain process control applications systems of coupled columns should contain at least one capillary column. The separation of selected cuts is to be executed at high efficiency and with change of selectivity in order to achieve the maximum resolution for selected parts of the eluate which are transferred from a preseparation column into a column of high efficiency applying flow switching. The potentials of trapping and cryofocusing of the selected cuts between the columns for the improvement of analytical performance are discussed with special emphasis. The involvement of capillary columns makes high demands on the instrumentation especially with regard to the construction of the coupling piece between the columns. For MDGC application in laboratory analyses valveless and automated, i.e. pre-programmable, operation is an important feature either for the reproducible execution of large series of samples or for the separation of complex isomeric mixtures at the maximum resolution. By automated partial analyses high performance at shorter run times can be achieved. Typical areas of application are hydrocarbon, environmental and food analysis with involvement of the separation of isomers and enantiomers.

Contents

[1] The author does not intend to give a complete review on MDGC. The paper treats the principles and the special instrumentation for MDGC on hand of some applications performed in his own laboratory.

0021-9673/95/$09.50
SSDI 0021-9673(95)00190-5

1. Introduction and terminology

In 1984 Giddings [1] published a fundamental and theoretical paper with the title "Two-Dimensional Separations: Concept and Promise" in which he outlines the large number of two-dimensional (2D) separations which can be realized by combining one-dimensional (1D) displacement processes. He distinguishes between sequential and simultaneous zone "displacement" and states: "Sequential displacements, as those occurring in column chromatography or in two-dimensional thin-layer chromatography, are far more adaptable because optimum conditions can be applied separately to each step. One can carry out the first displacement in one medium under one set of conditions and transfer the linear array of zones to the edge of a 2D system for the second displacement". Concerning the research work on 2D methods in the future Giddings writes: "any 2D technology must stand on the shoulders of 1D building blocks. Whereas the thrust of 1D research is to improve the building blocks, the thrust of of 2D research will be to find powerful and ingenious ways of combining building blocks". In the terminology of Giddings 2D methods such as GC–GC make use of two selective 1D displacements. These are "selective", only, if columns with different stationary phases are combined. Modern gas chromatography in capillary columns is a highly developed building block for 2D methods because of the extraordinary high efficiencies which lead to even higher peak capacities at the separation of multicomponent mixtures with two selective chromatographic building blocks.

According to the classical terminology in column chromatography separations are commonly called two- or multidimensional when separations of all or certain selected groups of sample components are repeated in two or more columns of different polarity, which are coupled in series to the column in which the first separation was performed. The continuous transfer of the eluate or the transfer of selected cuts from the first to further columns is achieved by the carrier gas flow which can also be diverted to exit ("venting") or reversed for backflush by flow switching between the columns. The necessary switching of the carrier gas flow can be effected either by valves or in the valveless mode. The latter mode is to be preferred when separations with the highly efficient but also miniaturized capillary columns are to be performed. The application of capillary columns in MDGC couplings requires special instrumental provisions because of the small carrier gas flows, the small peak volumes and the low sample capacity in general, however.

By the transfer of selected cuts from one column to another column of different polarity and the related selectivity of the separation the resolution of those peak groups which are contained in such cuts is improved. This methodology has very early been recognized as effective also for GC analyses in routine laboratories by Deans [2]. The resolution of the selected cuts can be further increased in columns of high efficiency e.g. in the miniaturized capillary columns, see Schomburg et al. [3,4], Bertsch [5] and Schomburg [6,7]. The selection of cuts which are to be re-separated at different selectivity and additionally also with high efficiency has to be done according to the objective of the intended analytical application. In this way mainly partial analyses of much higher performance for the separation and determination of a single or several components in a certain peak group, can be executed which may be adequate and sufficient for the solution of the analytical problem. In the analytical practice the increase of peak capacity of separations of an entire multicomponent mixture is not as important. The removal of uninteresting sample matrix components from the separation in the first (or pre-) column can be accelerated by higher carrier gas flows and at elevated temperatures or with temperature programs. Such a fast clean-up and reconditioning procedure of the first column can proceed in parallel to the separation of the significant sample components in the second column. The entire MDGC system becomes soon be ready for the next analysis in a series of routine measurements.

The transfer of either the entire eluate or of narrow or wider cuts, taken from the eluate

leaving the first separation, to another separation in the coupled next column is effected by the carrier gas (mobile phase) flow. If the second column is to be operated at lower carrier gas flow only a part of the eluate can be led into the second column. The other part has to be vented between the columns.

According to a stringent theoretical definition, separations with transfer of eluate cuts from a first into a coupled second column should only be called multidimensional, if additional new and significant sets of qualitative (e.g. relative retention) as well as the related better quantitative data are achieved. Basically this can only be the case if either the polarity of the stationary phase or the temperatures for the separation systems in the coupled columns are different. Temperature programs in the columns are, of course, also suited to effect the necessary change of selectivities between the first and the second separation system. One of the major aims of optimization of chromatographic separations is the improvement of resolution of certain components groups in a sample and the related better performance of the intended analytical application.

Optimization of the resolution of a group of sample components selected from the eluate of a first or preseparation can also be achieved if the separation in the second column is performed at higher efficiency or sample capacity i.e. also at higher phase ratio.

In trace analysis the resolution of two components which are present in an extreme ratio of concentrations and which appear as closely neighboured and overlapped peaks in the chromatogram can be improved by the transfer of so-called heart cuts [2]. They are to be taken in a way that the ratio of component concentrations is considerably decreased.

In systems of coupled columns with different stationary phases and lengths (efficiencies) the two columns may, moreover, be operated at different flows of the mobile phase, then separations with mixed selectivities are achieved which depend on the ratio of the residence times of analyte pairs or groups in each column of different polarity, see Deans et al. [8] and Kaiser et al. [9]. Such mixed selectivities are avoided if the sample components which are transferred from the first column are trapped in a cold trap between the columns or in the inlet of second column [3,4] before the second separation is initiated by revolatilisation. In a trap before or in the second column the separated species of certain "cuts" are also cryofocussed; in this mode the selectivity of the second separation is not any more influenced by the selectivity of the first separation. Moreover, cryofocusing allows for a second separation with an efficiency which is not influenced by the band broadening and symmetry distortion which unavoidably occurs in the first separation and in inadequately constructed coupling pieces.

2. Objectives of multidimensional GC separations

(1) Increase of peak capacity especially with the analysis of samples which consist of very many components. High peak capacities can also be achieved in a single column system by temperature programmed column operation.

(2) High resolution for isomers and enantiomers with special (e.g. enantio-) selectivity of the second column. Highly selective columns can successfully be applied only to narrow eluate cuts which contain a limited number of components. Especially enantiomer separations have to be performed in the highly efficient capillary columns in order to combine the optimum enantioselectivity with high efficiency.

(3) Short analysis times by partial analyses of cuts, which were taken from the eluate of a preseparation of very complex mixtures, they are performed in the second separation system under optimized conditions with the objectives of high resolution and also signal-to-noise ratios at detection. In preparative scale MDGC separations, (see Fig. 4), columns with high content of stationary phase and the corresponding higher sample capacity are to be applied; short separation (cycle) times for selected peak groups can only be achieved, when high retention compo-

nents are prevented to reach the main (second) separation which must be longer and correspondingly more efficient than the first column. Matrix components of low chromatographic volatility and the corresponding high retentions can be removed by fore- or backflushing from the short precolumn and not so favorable at elevated temperatures. Temperature programming cannot be applied with advantage in cyclic preparative GC because of the large column diameters and the related slow heat transfer, see Schomburg et al. [10].

(4) Improved determination of trace components eluted closely to the peaks of solvents or major components by "heart cutting", Deans [2].

(5) Avoidance of high column temperatures for the elution and venting of low volatility components, which are not of interest for the analysis, using short precolumns and/or backflush operation.

3. Systems and instrumentation for multidimensional GC

Today analytical GC separations are predominately performed in fused-silica capillary columns at high efficiency and with short analysis times which can be achieved with this miniaturized low sample capacity column type. By the high efficiency achievable especially in thin-film capillary columns and by temperature programming a large number of the components in quite complex samples can be resolved within reasonable analysis time also in a single column system. The peak capacity of analytical separations in capillary columns is already high and can moreover be increased by temperature programming. Adequately chosen temperature profiles during the separation provide good resolution over a wide range of component volatilities.

The analytical application of GC separations is limited to compounds which are volatile enough at temperatures of up to 350°C and to compounds which are stable at such high temperatures. Because of the low content of stationary phase in thin-film capillary columns from fused silica or borosilicate glasses separations of less volatile compounds can be performed (especially with hydrogen as the carrier gas) at considerably lower temperatures than in packed columns. The sensitive ionisation detectors allow for the generation of sufficiently high signal-to-noise ratios even at the low vapour pressures of such low volatility analytes at the very low admissible sample loads of capillary columns. In open-tubular columns temperatures of up to 350°C could be applied to stable compounds such as triglycerides, polyaromatic hydrocarbons, derivatized oligosaccharides, alkylcyclodextrines, etc. Multidimensional separations at such high temperatures have not yet been reported, probably because of the instrumental problems involved with the coupling technique but also because of the lack of more polar stationary phases which are stable at such high temperatures.

For comparison Fig. 1 shows different instrumental systems for chromatographic separations performed in the single- and multichannel or multidimensional mode. A is a single-channel system which is characterized by the three elements: injector, column and detector.

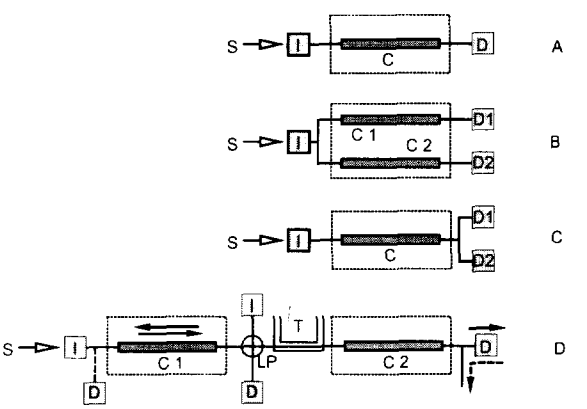

Fig. 1. Possible configurations of coupled GC systems. (A) One-dimensional, one-channel system. (B) Multichannel system with parallel columns. (C) Multichannel system with parallel detectors. (D) Multidimensional GC system. With this system, temperatures can be independently adjusted in columns, detectors, and injectors. S = sample; I = injector; C = column; D = detector; LP = "live" coupling piece; T = trap.

The column may contain stationary phases with different polarity which can be changed for selectivity optimization in series of consecutive measurements to develop applications with adequate resolution. The column is operated in an oven isothermally or under temperature programming. In the latter case absolute and relative chromatographic volatilities (i.e. retentions and selectivities) are changed during the separation. In the multi-channel system B two columns are operated in parallel, which should have different polarity. By a split after injection the same sample is transferred into both columns. Both channels of separation are connected to a detector, which could also be of different specificity to monitor the elution of the separated species. In such systems, in which the separation temperatures of both columns may also be programmed, screening analyses with samples of unknown composition can be performed in the analytical practice. On the basis of retention data and peak areas derived from the chromatograms which were obtained with different polarities of the columns conclusions can be made about the presence of components of different polarity in the sample and about the volatility (carbon number) range in which they appear. Components of different polarity can be recognized from the retention shifts arising between the chromatograms obtained from the two parallel separations. System C is of the single-channel type with regard to the separation but is multichannel with regard to the parallel detection with detectors of different specificity.

System D is typical for multidimensional chromatographic separations and is treated in more depth here with regard to GC separations, only. Two columns of different stationary phase polarity are coupled in series. Preferably each column is operated in a separate oven either isothermally or temperature programmed, because of the strong influence of temperature on absolute and relative retentions in GC. The coupling of narrow bore open tubular columns requires miniaturized dead volume-free devices in which, moreover, the surfaces are inert against adsorption and catalytic conversion of the analytes. In many cases preseparations can also be performed in packed columns with advantage because of their high sample capacity. Larger sample volumes e.g. in trace analysis can be preseparated with advantage in packed columns. The separation of selected cuts in the coupled second column should be performed with the high efficiency of a capillary column in order to achieve the maximum resolution, however. Different systems have been applied in which the injector is either positioned before the first or between the columns, depending on the desired elution direction in the column C_1 during the first separation. Detectors are incorporated between the two columns and at the end of column C_2 to monitor both the first and the second separation. The decision about the windows for the selection of suitable narrow or wide cuts from the eluate of a first separation are to be made in preceding test separations from the signal of an additional detector between the coupled columns.

As most of the GC separations are to be performed at elevated temperatures, the switching of the carrier gas flows for the transfer of cuts and the venting or backflushing of other sample components, also under flow reversal, should be done valvelessly i.e. without movable parts to which sample components could get contact at elevated temperatures. Flow switching by valves can only be applied at not too high separation temperatures i.e. for gaseous or volatile samples.

Coupling pieces for valveless flow switching should also allow for the dead volume-free connection of an additional detector (FID, ECD, etc.) between the columns to monitor the separation process in the first column. Via such a coupling piece (called "live switching piece" by F. Müller et al., see [11,12]), a system of flows between and in the columns can be established which allows for the reversal of the elution direction in column C_1 and the transfer of selected eluate cuts into column C_2. In the cases of backflushing from C_1, or when the sample had been injected between the coupled columns with C_1 operated in the reverse direction of the carrier gas flow, the elution of components can also be monitored by another detector which is located at the inlet of column C_1. Injection of

certain types of samples between two coupled columns is of advantage if the sample is highly diluted and contains solvents or major components with short separation. The less volatile components may be strongly retarded in the early part of the precolumn C_1 whereas the volatile solvent leaves the precolumn with very short retention time. The transfer of these less volatile components from the early section of C_1 by 'fore-flushing' through the entire length of the column C_1 into column C_2 takes much time or has to be performed at increased flow and/or elevated temperature in C_1. If the sample is injected between the columns and the flow in C_1 has the reverse direction to the left side of the system the less volatile components can be backflushed from the early section of C_1 in a short time. Short cycle times for such applications could be achieved in this mode for process control analyses. Between the columns devices can be incorporated for the trapping and/or cryofocusing of the components which are contained in the eluate cut which was selected for transfer into column C_2. By trapping and cryofocusing the resolution of the subsequent separation can be strongly influenced. Trapping before the second separation has the consequence that the so called "mixed polarities" are avoided. The main separation is then performed with the polarity of the column C_2 only. If the components leave C_1, one after the other, with retentions which depend on the polarity of the stationary phase in C_1, and if they are not trapped before entering C_2, they enter this column at different starting times. The total retentions at the end of C_2 therefore depend on the time when they enter C_2 and on their retentions in the subsequent separation in the stationary phase of C_2. In this way completely different relative retentions of the components are achieved without trapping. These have been called by several authors "mixed polarities" and depend on the single polarities of the coupled columns and also on the relative residence times of the sample components in the two columns. With trapping the selectivities of the separation in C_2 depend only on the polarity of the stationary phase and the separation temperature in this column. The trapping and/or cryofocusing can also be performed directly within the inlet of the column C_2 which is cooled by blowing cooled gases (N_2 or Ar) onto a short or longer section of the capillary. Trace components can be accumulated in the trapping section of the column by repetitive preseparations for subsequent detection of the accumulated material at higher signal-to-noise ratio after separation in the second column.

Valveless and pre-programmable automated flow switching has been applied in industrial process control by GC analysis long before its application in laboratory analysis. The application of such techniques to gas chromatographic analyses in the laboratory was firstly described by Deans [2], at the time when he introduced coupled column systems into laboratory analysis with automated valveless equipment. Deans was of the opinion that the usage of capillary columns in such systems would not be necessary in the common industrial applications, because by selectivity optimisation for selected cuts higher efficiencies for the separation of such cuts would not be required. This opinion has worked out to be not quite appropriate today considering the analytical problems involved with isomeric and enantiomeric separation problems with complex mixtures as occurring in environmental and flavor analysis, as will be shown below. The instrumental requirements to the coupling of packed columns are of course not as rigid as to couplings with involvement of capillary columns. Packed columns are operated with larger carrier gas flows which cause less problems with dead volumes in the flow system, and much higher sample loads. Moreover, in the scheme of system D the possibility of bypassing the detector is also incorporated [7]. By venting of the eluate from column C_2 solvents or other major sample components can be prevented from reaching the detector and impairing the performance of detection. This technique of detector bypassing could be useful in trace analyses with ECD or thermionic detectors, as has been demonstrated by Schomburg [7]. The "gate" to the detector is opened only when the components which are relevant for a certain analytical problem are to enter the detector. Commercial equipment for MDGC in capillary columns first became avail-

able by developments of F. Müller and M. Oréans et al. from Siemens (Karlsruhe, Germany) [11,12]. This instrumentation was developed on the basis of systems previously designed and applied by Schomburg et al. [3,4]. Fig. 2 shows the instrumental set-up for MDGC in a system of two coupled capillary columns, taken from [4], which contained most of the principally required features as later used in commercial systems, including a specially designed coupling ("double T") piece, cryofocussing in the inlet of the second column, specially arranged scavenge gas flows for dead volume compensation in the coupling piece, a second detector for the monitoring of the preseparation and the possibility of injection of samples through an injector between the columns.

Fig. 3 shows the flow system of the Siemens instruments for automated capillary MDGC as developed by Müller, Oréans et al. [13]. Other commercially available systems for MDGC were constructed later by the Gerstel [14], SGE [15] and the Chrompack companies [16].

The "live" column switching system in the Siemens instrumentation which is also applied for process control GC contains a coupling device which is based on a platinum capillary of suitable external diameter which is introduced into the ends of the capillary column. The three principal modes "cut transfer", "venting" and "backflush" are initiated by changes of the pressure drop within the connecting capillary and within the columns 1 and 2. In the coupling capillary and in column 1 the flow direction, e.g. the pressure drop, can be reversed. In column 1 this reversal is achieved by venting of the backflush eluate at NV4 between injector 1 and the inlet of column 1. The venting of the eluate from column 1 is effected when the flow direction in the coupling capillary is from B to A, then the eluate flow from column 1 cannot enter column 2. The coupling piece is carefully constructed with regard to the avoidance of dead volumes by scavenge flows and the corresponding impairment of peak width and symmetry. The coupling of columns with application of intermediate cryofocussing is instrumentally less demanding because the band broadening arising at the preseparation, and any additional band broadening in the coupling piece would be suppressed by the focussing, before the main or second separation of the components trapped from the eluate cut is initiated.

In Fig. 4 the scheme of a MDGC system for

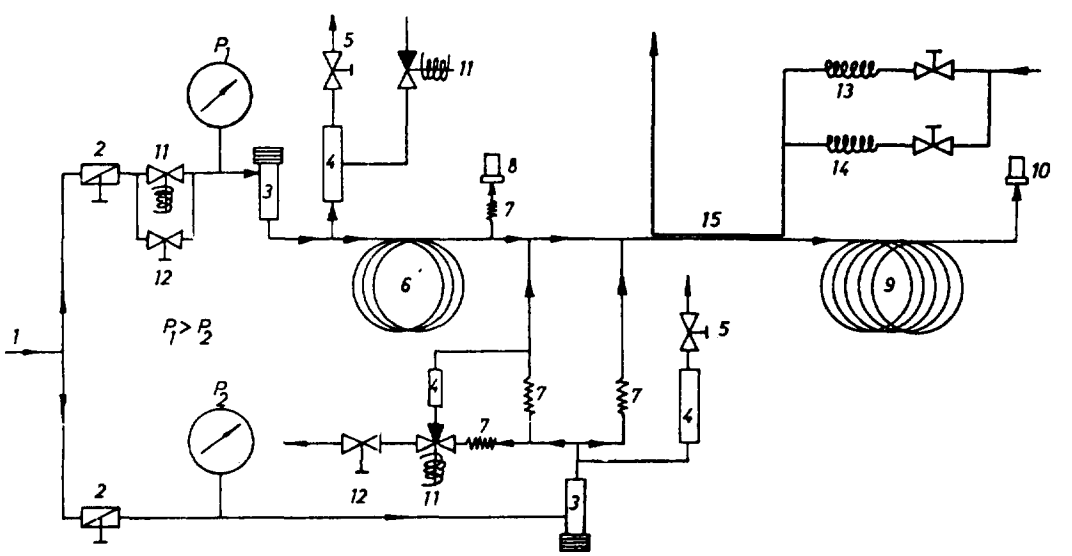

Fig. 2. Automatized gas chromatographic system with two coupled capillary columns and intermediate trapping. System status: Eluate of the pre-column is transferred to trapping. 1 = Carrier gas; 2 = pressure controller; 3 = injector; 4 = filter; 5 = split; 6 = glass capillary pre-column; 7 = throttles; 8 = control FID; 9 = glass capillary main column; 10 = FID; 11 = solenoid valves; 12 = needle valves; 13 = cooled coil; 14 = heated coil; 15 = trap.

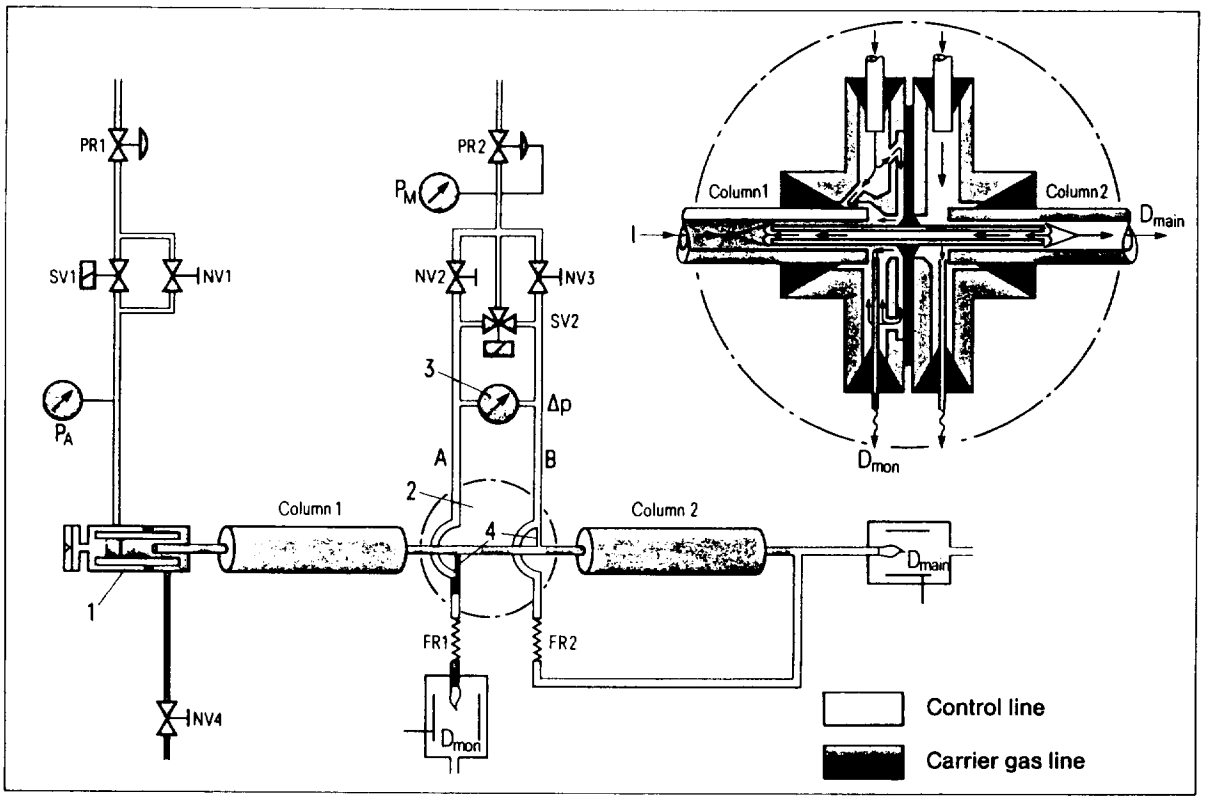

Fig. 3. "Live" column switching system. 1 = Injector with split valve and septum purge; 2 = live-T-piece; 3 = differential pressure manometer; 4 = ring slot. NV = Needle valve; SV = solenoid valve: PR = pressure regulator; FR = flow restrictor; D_{mon} = monitor detector; D_{main} = main detector; P_A = inlet pressure; P_M = mean pressure; Δp = differential pressure; A = control line A; B = control line B.

preparative (large scale) gas chromatography is shown. Such instrumentation was developed by Schomburg et al. [10] in cooperation with the Gerstel company (Mülheim a.d. Ruhr, Germany) on the basis of the preparative GC instrument APG 402 originally designed by the Hupe and Busch company. In this instrument packed columns of a diameter between 10 and 40 mm are applied. In the system shown 14 is the pre column, 18 and 23 the main columns. 13 and 22 are the monitor detectors for pre- and main separation, 26 and 28 cold traps for condensation of the purified materials; 10 and 15 are the traps for the collection of backflushed material. 1–5 and 6 are devices for pneumatic introduction and gentle vaporization of the sample. For further details see [10].

4. Some typical applications of MDGC

In the following a few typical multidimensional GC separations of different types of samples are shown. The features and special modifications of the instrumentation used for the optimal execution of these applications are also given.

4.1. Separation of a coal derived gasoline fraction

The gasoline sample contained polar compounds such as ketones, alcohols and nitriles besides isomeric including aromatic hydrocarbons (see Fig. 5). For the shown separations a coupling of a polar polyethylene glycol and a

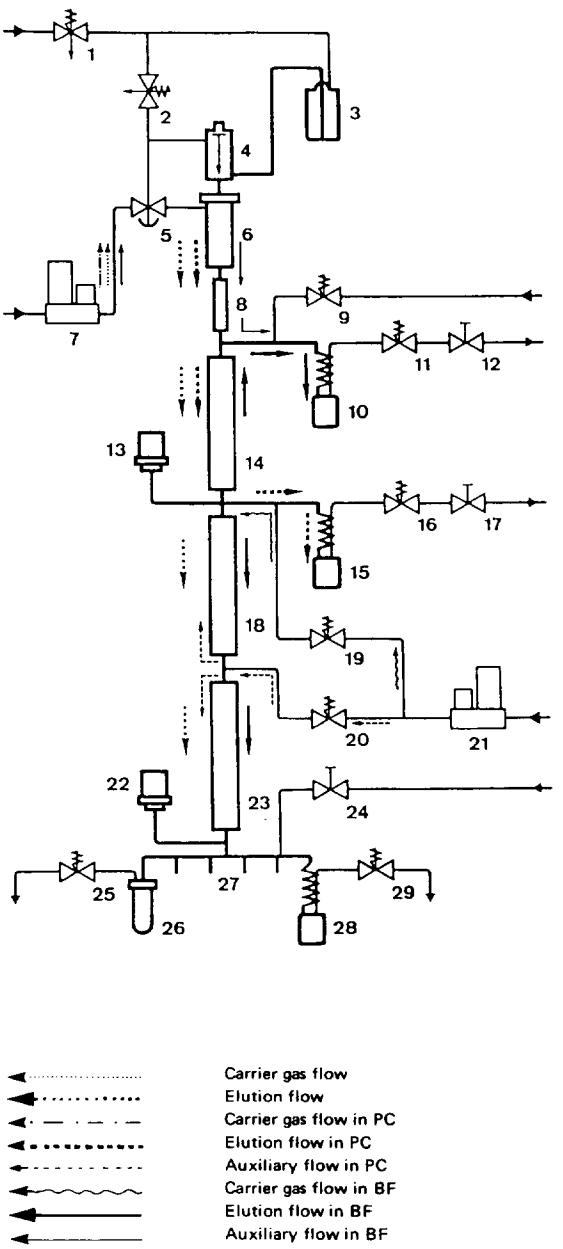

Fig. 4. Chromatographic and pneumatic system of a PSGC instrument for multidimensional operation. PC = Peak cutting mode, Bf = Backflushing mode. 1–5 = Pneumatic system for sample introduction; 6 = vaporization chamber; 7, 21 = "thermal" mass flow controller; 8 = buffer column; 9, 11, 16, 19, 20, 25, 29 = 2-way solenoid valve; 10 = backflush trap; 12, 17, 24 = needle valve; 13 = monitor detector for preseparation; 14 = precolumn; 15 = peak cutting trap; 18, 23 = main column; 22 = detector for main separation.

non-polar methylpolysiloxane OV 1 capillary columns was applied. The polar column was operated isothermally, the non-polar main or second column with temperature programming. The carrier gas was hydrogen. In chromatogram A the polar sample components are shifted far behind the bulk of unresolved volatile hydrocarbon isomers in the carbon number range from C_4 to C_8. The hydrocarbon isomers could be well resolved with a temperature programm which began at subambient temperatures, see separation B in Fig. 5 [6,7]. The pre- or first separation is characterized by long retentions for the polar ketones, nitriles and alcohols in the highly polar column. The resolution of the hydrocarbon group is very poor because of the weak and unselective intermolecular interaction of these non-polar hydrocarbons with the polar stationary phase.

4.2. Multidimensional separation of a PCB mixture (Clophen A 30/A 50), see Fig. 6

The separation of all of the PCB isomers contained in such a mixture, which is a frequent problem of environmental analysis, can only be done in a single capillary column if the selectivity of a polysiloxane phase and the temperature program of operation are specially adjusted for this separation. High efficiencies in narrow-bore capillaries with an internal diameter of about 100 μm facilitate the achievement of the desired resolution of all isomers even more. The overlapping peaks in a single column separation by unresolved isomers can easily be elucidated by GC–MS measurements, provided molecular weights and the mass spectra of the overlapping components are different. It is also possible to resolve overlapped species in MDGC separations with polarity change for selected cuts. Without the approach of transferring relatively narrow cuts into a column of different polarity new cases of overlapping species would arise. The separations were performed in a Siemens instrument which was equipped with ECD detectors for both the first and the second separation, see

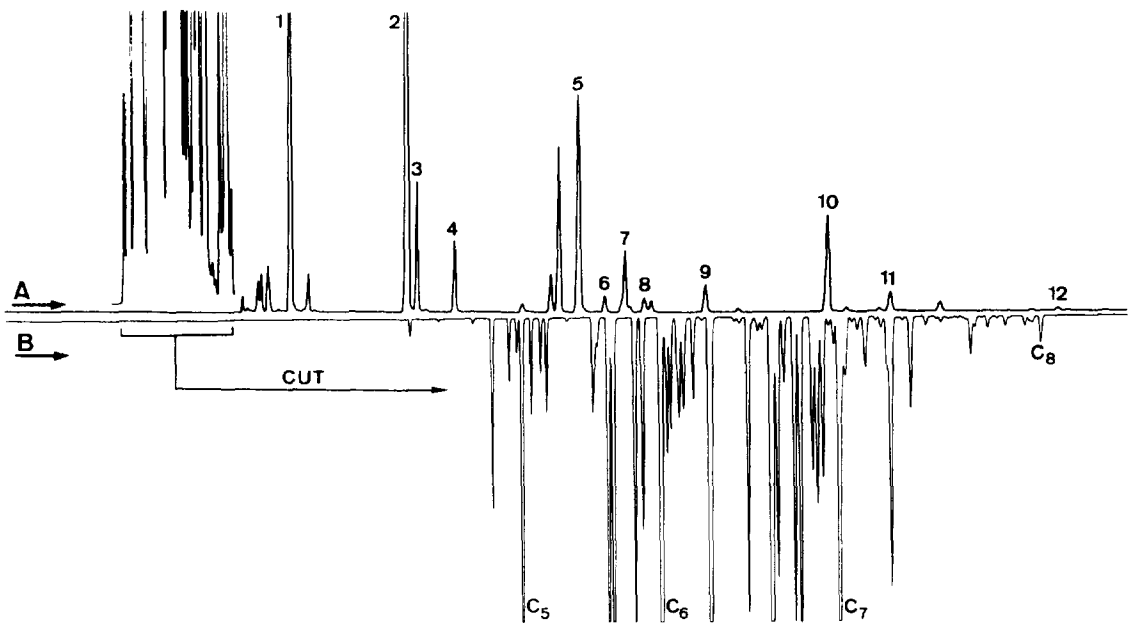

Fig. 5. Analysis of a coal derived gasoline fraction applying multidimensional gas chromatography in capillary columns. Sample: 0.2 μl; 1 = acetone; 2 = 2-butanone; 3 = benzene; 4 = isopropyl methyl ketone; 5 = isopropanol; 6 = ethanol; 7 = toluene; 8 = propionitrile; 9 = acetonitrile; 10 = isobutanol; 11 = 1-propanol; 12 = 1-butanol. Pre-column: (A) 121 m polyethylene glycol CW 400; temperature: 12.6 min isothermal at 50°C, from 50 to 80°C at 3°C/min; carrier gas: (A) 0.13, (B) 0.07 MPa hydrogen; cut: 12.6–17.4 min; main column: (B) 64 m methylpolysiloxane OV 1, thick film; temperature: 17.4 min isothermal at −30°C, from −30 to 50°C at 20°C/min, from 50 to 150°C at 3°C/min; carrier gas: 0.07 MPa hydrogen; analysis time: 50 min.

Schomburg et al. [17]. A Siemens double oven MDGC instrument was applied by Duincker et al. [18] for the identification and determination of single PCB species.

4.3. *Multidimensional separation of the species of PCDD and PCDF in an artificial mixture*

The separations of Fig. 7 illustrate that the required resolution for the determination of the more toxic species in such a complex isomeric mixture can only be achieved if quite narrow cuts are transferred. In this application [17] the first column was the polar cyanopropyl polysiloxane Silar 10 C, whereas the second column contained the non-polar methylpolysiloxane OV 1. If the transfer (cut) times are too large, more of the many overlapping isomers may enter the second column and cause new peak overlappings as a consequence of the polarity change. See peak 6 which overlaps the "target" peak 7 because the cut was too wide (60 s). How narrow the cut has to be depends on the polarity shift of a neighboured non-target isomer (6) which may be different from that of the target isomer (7). In the present practice of dioxin analysis consecutive single column separations with different stationary phase polarities are performed in GC–MS systems. Overlappings of interesting species are either resolved by the polarity change and by GC–MS-SIM detection in cases were the molecular weights of the overlapping species are different. For forensic applications the latter mode which involves the more reliable characterisation of toxic species by MS instead of by chromatographic retentions, only, is to be preferred, although the detection by ECD, as applied in the

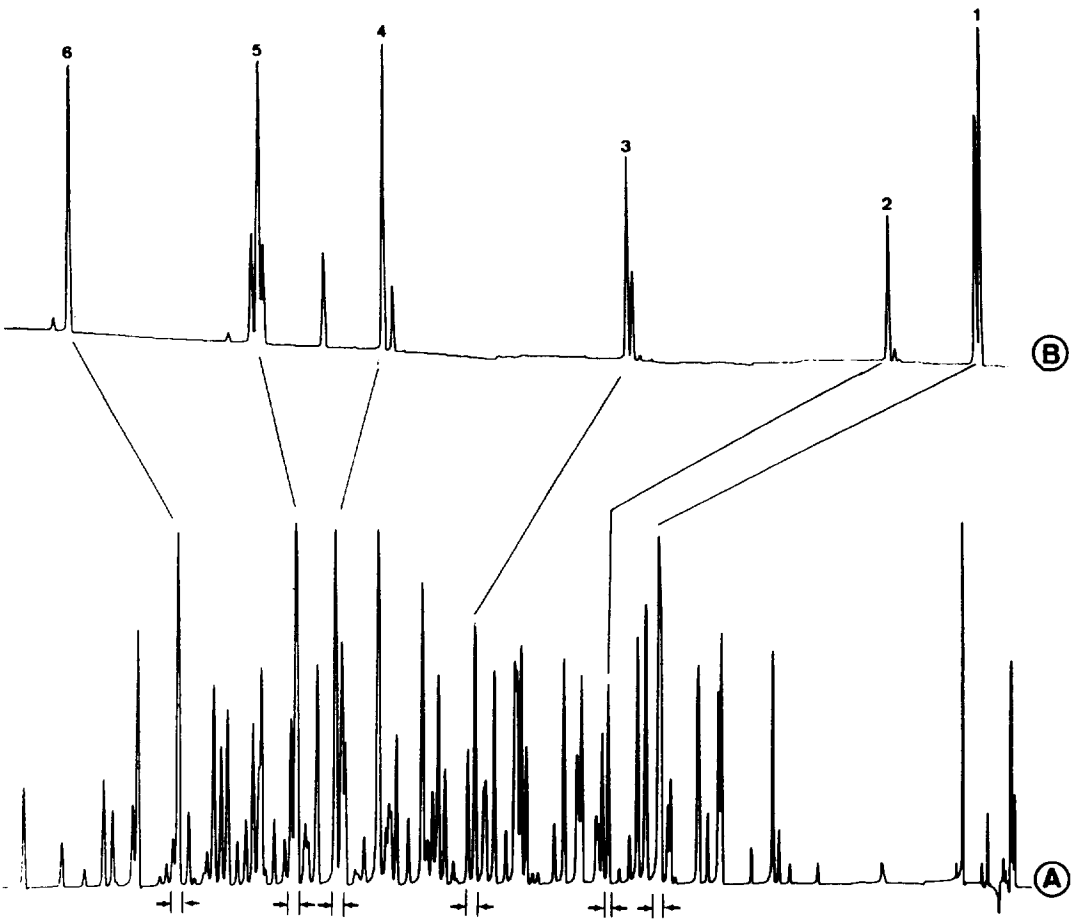

Fig. 6. Multidimensional separation of isomeric PCB mixtures. Determination of the 6 "Ballschmiter" isomers by heart cutting, intermediate trapping and subsequent temperature programmed separation of the 6 cuts in a single run. Sample: 0.2 μl Clophen A 30/A 50 mixture; Split: 1:20; columns: (A) 35 m methylpolysiloxane PS 345.5 on FS, 0.25 mm I.D.; (B): 30 m cyanopropyl polysiloxane Silar 10 C on AR, 0.32 mm I.D.; temperatures: (A) sampling at 60°C, bal. 140°C, then 3°C/min to 280°C; (B) 180°C, 22 min isothermal, from 180 to 260°C, 2°C/min; carrier gas: (A) 0.1 MPa hydrogen, (B) 0.04 MPa hydrogen; detection: ECD for both separations. The cuts are marked in chromatogram A, as obtained from the temperature programmed first separation. Chromatogram B was obtained by temperature programmed separation of the trapped 6 cuts in column B.

MDGC procedure, is quite sensitive for the analysis of traces.

4.4. *Enantiomeric separations with MDGC in capillary columns*

Chiral separation require the application of highly efficient capillary columns which contain chiral stationary phases which are capable of intermediate formation of diastereomers with chiral analytes. Chiral capillary columns with a more or less universal capability of achieving suitable high enantioselectivities are available now. A large variety of chiral compounds including such which are non- or weakly polar, such as saturated and unsaturated hydrocarbons, can now be separated into the enantiomers using derivatized cyclodextrins as chiral selectors which are either dissolved in methylpolysiloxanes or may also be chemically bonded to polysiloxanes backbones. Another analytical problem

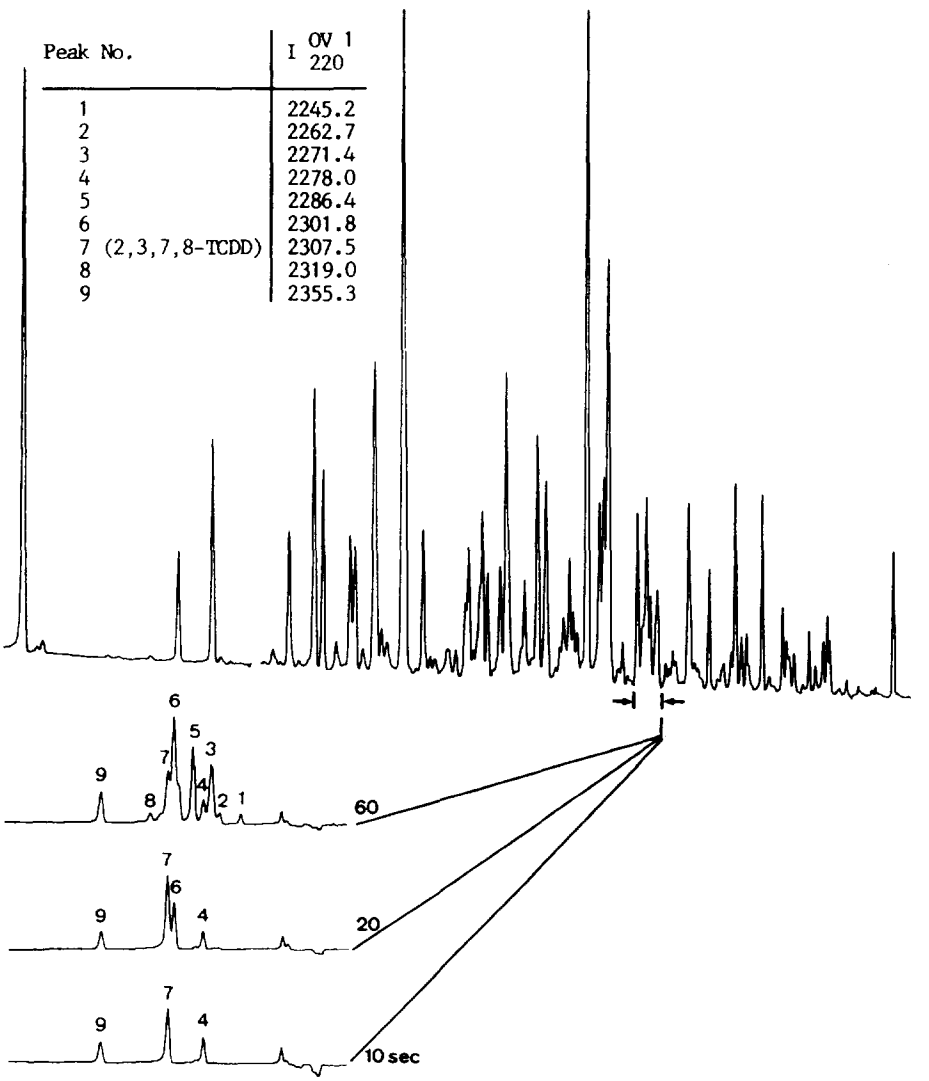

Fig. 7. MDGC separation of an artificial isomeric mixture of PCDD and PCDF. Heart cutting for identification by Kováts index and for determination of special isomers such as 2, 3, 7, 8-TCDD. Influence of cut width on resolution. Sample: 0.6 μl solution of PCDD and PCDF; columns: (A) 30 m × 0.27 mm I.D. cyanopropyl polysiloxane Silar 10 C, alkali glass; coupled to (B) 25 m × 0.32 mm I.D. methylpolysiloxane OV-1, fused silica; temperatures: (A) 60 to 160°C ballistically, 160 to 260°C at 3°C/min; (B) 220°C isothermal; carrier gas: (A) 0.115 MPa hydrogen; (B) 0.050 MPa hydrogen; detectors: ECDs; analysis time: 35 min.

arises when the enantiomer ratio is to be determined of chiral compounds which are contained in complex mixtures of other compounds (as in essential oil analysis) or generally in matrices which are less compatible with GC.

The first chiral separations in MDGC systems were performed by Bayer and co-workers [19] and Schomburg et al. [20]. Bayer et al. applied a Siemens double oven instrument i.e. a system which was based on instrumental arrangements described in previous work by Schomburg.

In Figs. 8 and 9 two different MDGC separations of a mixture containing the isopropylurethane derivatives of four diastereomeric menthols in a matrix of solvents and derivatization reagents. The system of coupled columns

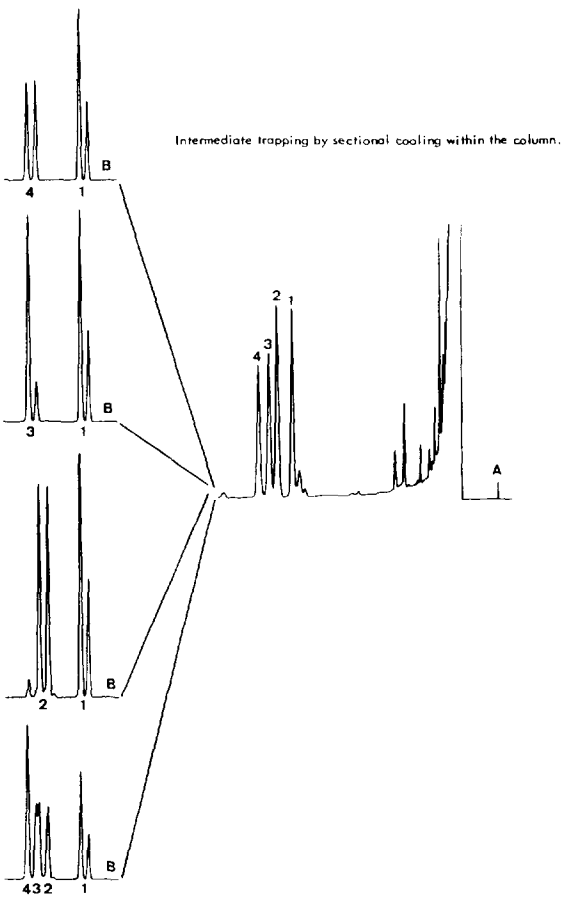

Fig. 8. Enantiomeric separation of the four diastereomeric menthols as isopropylurethane derivatives. Multidimensional separation in a system of coupled columns with a chiral main column. Sample: 0.2 μl IPU-derivatives diluted in methylene chloride. Isopropylurethane derivatives of: 1 = neomenthol, 2 = menthol, 3 = isomenthol, 4 = neoisomenthol; columns: (A) pre-: 16 m OV 1701 on FS, 0.25 mm I.D., (B) main: 50 m XE-60-S-Valine-S-α-phenylethylamide on FS, 0.23 mm I.D. (Chrompack); temperatures: (A) 117°C isothermal, (B) 147°C isothermal (trapped in column at −80°C), injector: 200°C; carrier gas: (A) 0.16 MPa hydrogen, (B) 0.13 MPa hydrogen; split: 1:30.

consisted of a polar cyanomethylethyl polysiloxane XE 60 column and a chiral XE 60-S-valine-S-α-phenylethylamide column as second column. The Siemens SiChromat 2 instrument with live switching coupling piece and cold trap device was used for these separations. In Fig. 8 it is shown how different cuts were transferred from the first into the chiral second column. In the first experiment two cuts containing the diastereomers 1 (neomenthol) and 4 (neoisomenthol) were trapped in the inlet of the chiral column and subsequently, after revolatilisation, separated into their 4 enantiomers. The isomers 2 and 3 were vented between the columns and did not reach the chiral column. In the next two experiments isomers 1 and 3, as well as 1 and 2 were transferred into the chiral column for the separation of the enantiomers. Finally all four menthol diastereomers were trapped and transferred into the chiral column. Only six of the eight enantiomers could be resolved. The complete separation of all eight enantiomers is possible, however, when the transfer of the four diastereomers into the chiral column is done directly and without intermediate trapping. Now the four pairs of enantiomers are separated with a mixed polarity of the column system and with a selectivity which does not lead to overlappings of the enantiomers which are resolved when these pairs enter the chiral column. This mixed polarity can also be adjusted in different ways, i.e. by the length and stationary phase content (film thickness) of the columns, by the temperatures in the two separate column ovens and by the ratio of the residence time of the analytes in the two column. This ratio can be changed by variation of the carrier gas flows in the two columns. The series of MDGC experiments shown illustrate in which way systems of coupled columns with different polarity, which are operated at different temperatures and carrier gas flows, can be applied in analytical gas chromatography.

In recent years MDGC with and without involvement of chiral columns was extensively applied to the analysis of flavors and essential oils by several groups. See for example Schreier and co-workers [21], Mosandl et al. [22] and König et al. [23]. In Fig. 10 a chiral MDGC separation of a component contained in a complex mixture is shown which was taken from the work of König. The stereomers of methyljasmonate in jasmine concrete were separated. Four separations were performed with a column system which contained a chiral column.

The lower chromatogram which was obtained

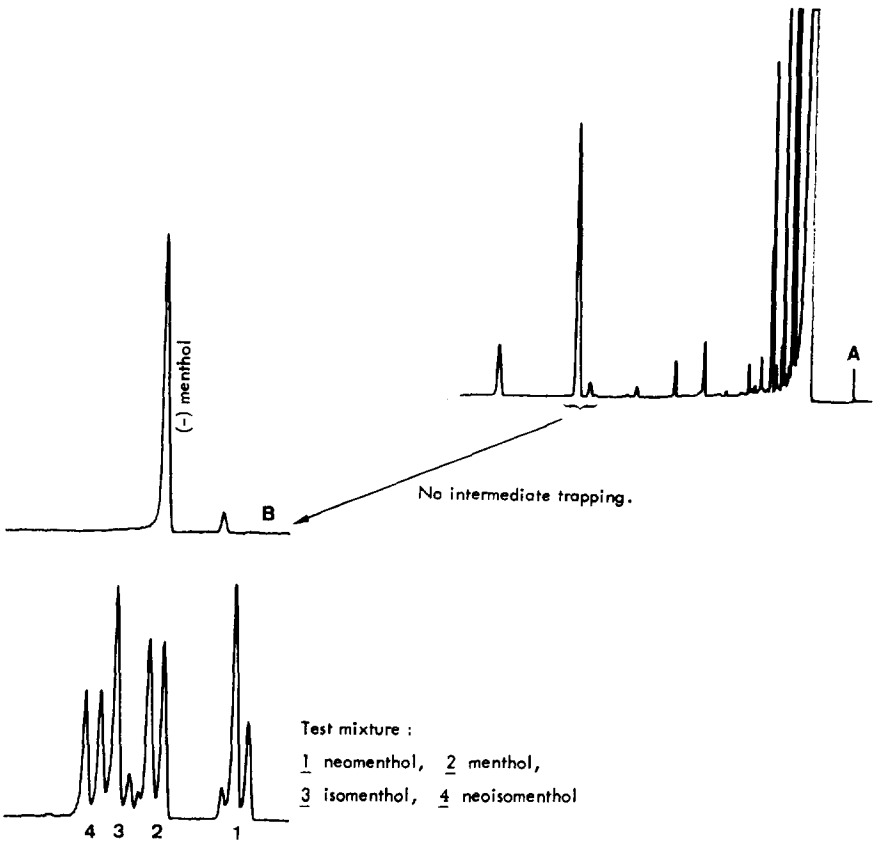

Fig. 9. Enantiomeric separation of the four diastereomeric menthols as isopropylurethane derivatives. Multidimensional separation in a system of coupled columns with a chiral main column. Sample: 0.4 μl of derivatized product, menthols of peppermint oil diluted in methylene chloride; columns: (A) 18 m cyanomethylethyl polysiloxane XE 60, immobilized, on FS, 0.25 mm I.D., (B) 50 m XE-60-S-Valine-S-α-phenylethylamide on FS, 0.2 mm I.D.; temperatures: (A) 110°C isothermal, (B) 147°C, injector: 200°C; carrier gas: (A) 0.16 MPa hydrogen, (B) 0.13 MPa hydrogen, split: 1:30; instrument: Siemens SiChromat 2 with live switching device.

from the precolumn with a standard mixture of the different stereomers of methyl jasmonate showed three peaks, whereas the chromatogram from the second chiral column showed six peaks. Four of these six peaks were the methyl jasmonate stereomers 1*R*, 2*R*; 1*S*, 2*S*; 1*S*, 2*R* and 1*R*, 2*S*. By the separation of this standard the position of the cut for the three further separations could be fixed.

By comparison of the test chromatogram of the standard obtained by the second column with the three separations obtained with the cuts taken from the jasmine concrete sample it can be concluded that jasmine concrete consists of 1*R*, 2*R* stereomer. The two upper chromatograms from column 2 were obtained after addition of the standard to the original jasmine sample in different amounts.

4.5. MDGC of flavors: preparative isolation, sniffing and MS of selected species

An interesting application of a Siemens MDGC system in coupling with different enrichment techniques and also sensoric detection with a sniffing mask behind the first column and GC–MS identification behind the second column was described by Nitz et al. [24].

Nitz et al. also described a multidimensional

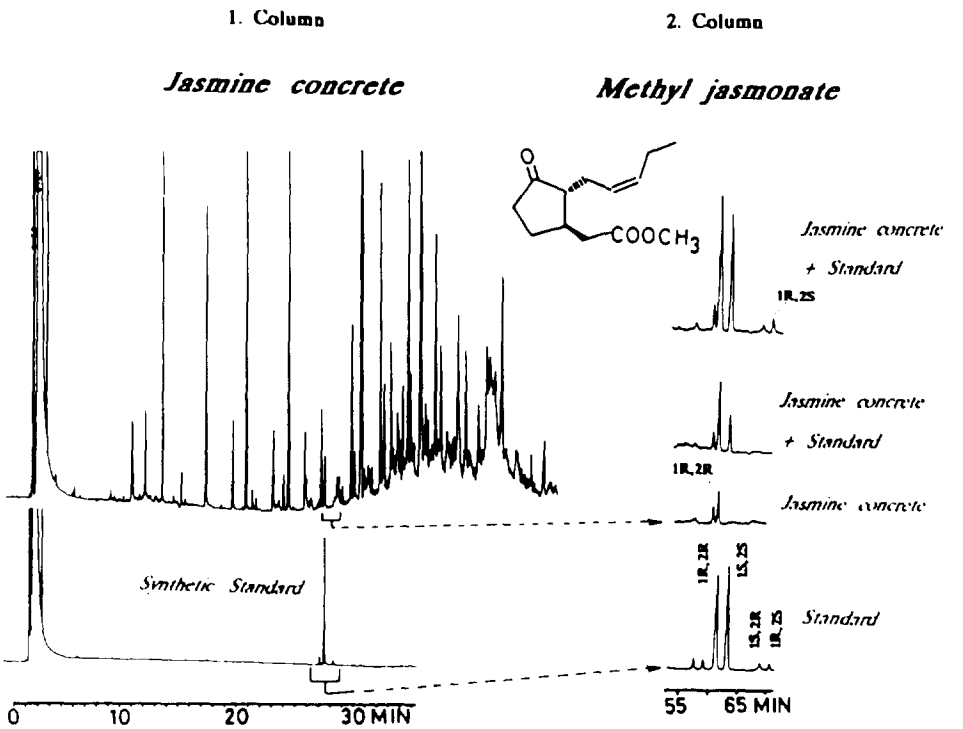

Fig. 10. Identification of the stereoisomers of methyl jasmonate in jasmine concrete by two-dimensional capillary gas chromatography: left, 25 m fused-silica capillary coated with CPSil-5 (Chrompack) programmed from 70 to 250°C at 5°/min; right, 25 m fused-silica capillary column coated with octakis(2-O-methyl-3,6-di-O-pentyl)-γ-cyclodextrin (125°C, isothermal); for further details, see text. Taken from Ref. [23].

GC–IRMS (isotope ratio mass spectrometry) system [25] and a system with automated preparative isolation of MDGC separated flavor components [26].

4.6. Separation of phenols in waste water from a coking plant

Separations of aqueous solutions for GC-compatible trace components by gas chromatography are usually executed by extraction, because real aqueous samples may also contain salts and other non-volatile compounds which may be deposited in the injector. Another problem of trace analyses using capillary columns is the limited sample capacity of this column type; generally large sample volumes have to be handled even after preceding enrichment in order to achieve high enough signal-to-noise ratios for the target trace components at high resolution. Extensive and important work on the direct sampling of large volumes of highly diluted solutions originating from extraction or preceding LC separations onto capillary columns (or into coupled "retention gaps") has been performed by K. Grob jr. et al. [27,28] which has been described in many publications. The MDGC experiment described here (see Fig. 11) was executed in a combination of a packed Tenax column (1 m long) with a capillary column with tri cresyl phosphate as stationary phase. From the Tenax column the highly polar water is eluted with very fast retention; the elution of the water could be monitored by a thermal conductivity detector which is not shown in the system of Fig. 11. The polar but more lipophilic phenols are strongly retarded on the Tenax, so strongly that they are accumulated already in the early section of the column whereas the matrix water leaves the column very fast, i.e. with very short

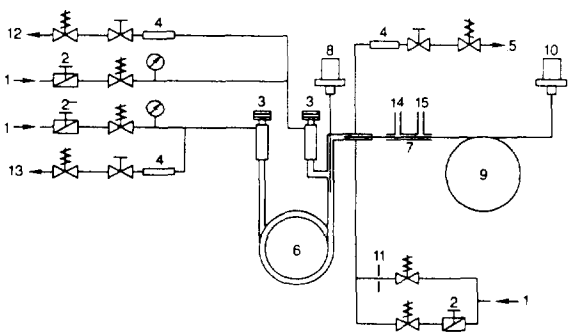

Fig. 11. Multidimensional GC in systems of coupled columns. Isothermally operated system of packed pre-column coupled to capillary column. Focusing of the transferred analytes in the temporarily cooled inlet of the capillary column. 1 = Carrier gas inlets; 2 = pressure regulators; 3 = injectors; 4 = filter columns; 5 = split; 6 = packed column; 7 = cold trap (column inlet); 8 = FID; 9 = capillary column; 10 = FID for detection of the significant analytes after main separation; 11 = blend type flow controllers for control of make-up gas flow into the coupling piece; 12 = carrier gas outlet for packed column when operated from left to right and using the left-hand injector (3); 13 = carrier gas outlet for packed column when operated from right to left using the right-hand injector (3); 14 = inlet for heating gas (nitrogen); 15 = inlet for cooling gas (nitrogen).

retention. The transfer of the lipophilic phenols from this stationary phase in the same direction in which the elution of the water was performed is difficult and would require elevated temperatures. If, however, the injection of the aqueous sample is done between the columns, the elution of the water is now performed in the reverse direction as the transfer of the phenols into the capillary column for the separation of the different, also isomeric, phenols at high resolution. In the Tenax column no selective separation of the phenols can be achieved and the displacement of the accumulated analytes is slow. The profile of the displacement of the phenols from the Tenax column has also been monitored by FID detection and takes place in the form of a very broad peak. Such a peak profile is of course unsuitable for a subsequent efficient separation in a capillary column which is operated isothermally. With temperature programmed operation with initially low column temperature cryofocusing of the transferred not too volatile compounds would take place. By cryofocusing in the initial part of

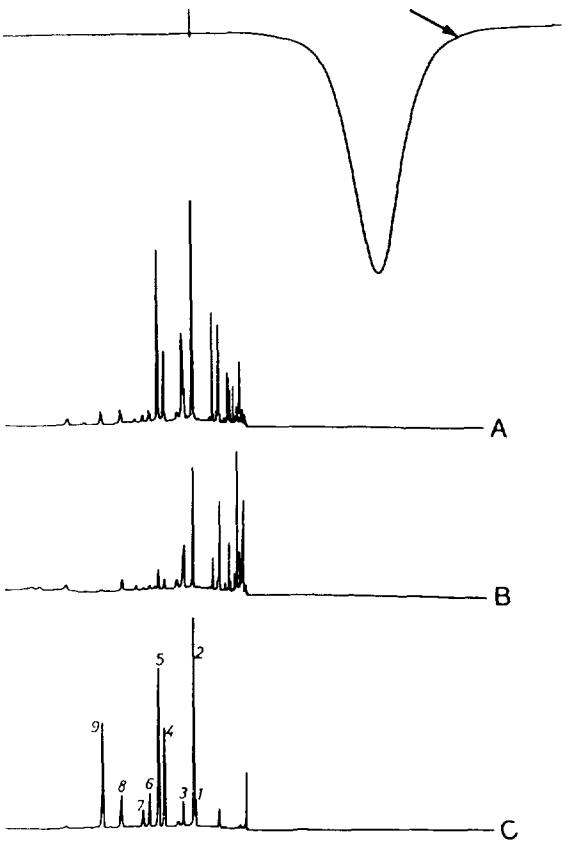

Fig. 12. Separation and determination of low concentrations of phenols in aqueous solutions. Sample: 1 = 2,6-dimethylphenol; 2 = phenol; 3 = *o*-cresol; 4 = *p*-cresol; 5 = *m*-cresol; 6 = 2,4-dimethylphenol; 7 = 2,5-dimethylphenol; 8 = 2,3-dimethylphenol; 9 = 3,5-dimethylphenol. Pre-column: 1 m packed Tenax, 80–100 mesh. Main column: 19 m × 0.25 mm I.D. tricresyl phosphate. Temperatures: 130°C in both columns (single-oven MDGC system). (A) 3 ml (split 1:30), waste water from coking plant before the phenol scrubbing; (B) 3 ml (split 1:30), after scrubbing; (C) 3 ml (split 1:30), test mixture.

the capillary column this problem can easily be overcome as demonstrated by the separations A and B of Fig. 12. At that time these separations were still performed in a single oven instrument at a temperature of 130°C.

5. Conclusion

Based on the results of the author's own experimental and instrumental work in the past,

since the introduction of automated multidimensional separations into laboratory analysis using packed columns (by Deans [2]), typical separations of samples were treated which are difficult to handle in single-column systems even with capillary columns and temperature programming. The application of MDGC with involvement of capillary columns in routine type analysis is, however, still limited, probably because of the higher complexity of the necessary instrumentation. With the well-developed instrumentation of GC–MS also using capillary columns and temperature programmed operation the identification and determination of target compounds in highly complex mixtures can be achieved especially with higher forensic reliability, but only if the target compounds have characteristic spectra for specific detection.

References

[1] J.C. Giddings, *Anal. Chem.*, 56 (1984) 1259 A.
[2] D.R. Deans, *Chromatographia*, 1 (1968) 18.
[3] G. Schomburg and F. Weeke, in S.G. Perry and E.R. Adlard (Editors), *Gas Chromatography* 1972, The Institute of Petroleum, United Kingdom, 1972, p. 285.
[4] G. Schomburg, H. Husmann and F. Weeke, *J. Chromatogr.*, 112 (1975) 205.
[5] W. Bertsch, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 1 (1978) 85, 187, 289.
[6] G. Schomburg, in P. Sandra (Editor), *Sample Introduction in Capillary Gas Chromatography*, Vol. 1, Hüthig, Heidelberg, 1985, p. 235.
[7] G. Schomburg, *LC-GC*, 5 (1987) 304.
[8] D.R. Deans and I. Scott, in A. Zlatkis (Editor), *Advances in Chromatography 1973*, University of Houston, Texas, 1973, p. 77.
[9] R.E. Kaiser and R.I. Rieder, *Labor Praxis*, 12 (1985) 1465.
[10] G. Schomburg, H. Kötter, D. Stoffels and D. Reissig, *Chromatographia*, 19 (1984) 382.
[11] G. Schomburg, F. Weeke, F. Müller and M. Oréans, *Chromatographia*, 16 (1982) 87.
[12] F. Müller, H. Müller and H. Straub, *J. Chromatogr.*, 477 (1989) 25.
[13] Siemens AG, 76187 Karlsruhe, Germany, Information A 19100-E687-A51-V2.
[14] Gerstel-DCS (Dual Column Switching), Internal report of Gerstel Company, Mülheim a.d. Ruhr, Germany.
[15] MDS 2000 Column switching system, Catalog of SGE Scientific Glass Engineering Company, Weiterstadt, Germany.
[16] MUSIC Column switching system, Catalog of Chrompack Company, Frankfurt, Germany.
[17] G. Schomburg, H. Husmann and E. Hübinger, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 8 (1985) 395.
[18] J.C. Duinker, D.E. Schulz and G. Petrick, *Anal. Chem.*, 60 (1988) 478.
[19] W. Chinghai, H. Frank, W. Guangha, Z. Liangmo, E. Bayer and L. Peichang, *J. Chromatogr.*, 262 (1983) 352.
[20] G. Schomburg, H. Husmann, E. Hübinger and W.A. König, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 7 (1984) 404.
[21] A. Bernreuther, N. Christoph and P. Schreier, *J. Chromatogr.*, 481 (1989) 363.
[22] A. Mosandl, U. Hener, U. Hangenauer-Hener and A. Kustermann, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 12 (1989) 532.
[23] W.A. König, B. Gehrke, D. Icheln, P. Evers, J. Dönnecke and W. Wang, *J. High Resolut. Chromatogr.*, 15 (1992) 367.
[24] S. Nitz, H. Kollmannsberger and F. Drawert, *J. Chromatogr.*, 471 (1989) 173.
[25] S. Nitz, F. Drawert and M. Albrecht, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 11 (1988) 322.
[26] S. Nitz, B. Weinreich and F. Drawert, *J. High Resolut. Chromatogr.*, 15 (1992) 387.
[27] K. Grob jr., D. Fröhlich, B. Schilling, H.P. Neukom and P. Nägeli, *J. Chromatogr.*, 295 (1984) 55.
[28] K. Grob jr., H.P. Neukom and R. Etter, *J. Chromatogr.*, 357 (1986) 416.