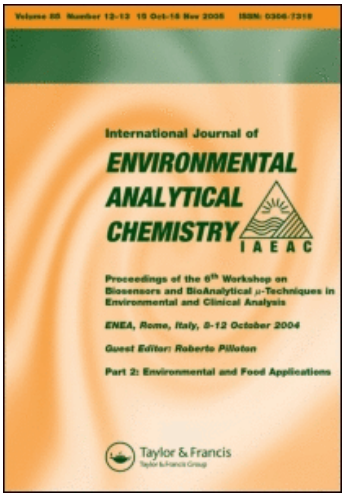

International Journal of Environmental Analytical Chemistry

Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713640455

Application of Multidimensional Gas Chromatography to Analysis for Nitrated Polycyclic Aromatic Hydrocarbons in Airborne Particulate Matter

Stanley L. Kopczynski[a]
[a] Environmental Monitoring Systems Laboratory, U.S. Environmental Protection Agency, Research Triangle Park, North Carolina, U.S.A.

To cite this Article Kopczynski, Stanley L.(1987) 'Application of Multidimensional Gas Chromatography to Analysis for Nitrated Polycyclic Aromatic Hydrocarbons in Airborne Particulate Matter', International Journal of Environmental Analytical Chemistry, 30: 1, 1 — 13
To link to this Article: DOI: 10.1080/03067318708075451
URL: http://dx.doi.org/10.1080/03067318708075451

Intern. J. Environ. Anal. Chem., 1987, Vol. 30, pp. 1–13
Photocopying permitted by license only

Printed in Great Britain

Application of Multidimensional Gas Chromatography to Analysis for Nitrated Polycyclic Aromatic Hydrocarbons in Airborne Particulate Matter

STANLEY L. KOPCZYNSKI

U.S. Environmental Protection Agency, Environmental Monitoring Systems Laboratory, Research Triangle Park, North Carolina 27711, U.S.A.

(*Received July 28, 1986; in final form October 21, 1986*)

Multidimensional gas chromatography (MDGC) in combination with simplified sample handling and detection procedures was evaluated for the determination of nitrated polycyclic aromatic hydrocarbons (nitro-PAHs) in particulate matter. Extraction was carried out by sonication. The extract was cleaned up using a short Florisil column, filtered, concentrated, and injected by means of a capillary on-column injector. Heart-cuts taken from the front column were focused on the back column, which was equipped with parallel detectors (flame ionization detector (FID) and nitrogen selective detector (NSD)). Unambiguous identification of chromatographic peaks from the combined information on front and back column retention times and the NSD/FID peak response ratios was demonstrated in the case of 1-nitropyrene. The limit of detection for 1-nitropyrene at $2 \times$ noise was calculated to be 36 picograms under the NSD/FID splitter ratio employed in this study.

The recovery of 1-nitropyrene in a National Bureau of Standards diesel particulate Standard Reference Material (SRM 1650) was 95%. In heart-cuts taken from the front column several unknown compounds were found with peak intensities comparable to that of 1-nitropyrene. A 1-nitropyrene equivalent concentration $\geqq 1\ \mu g/g$ was found for 122 peaks. Altogether, more than 200 peaks were observed with the NSD.

KEY WORDS: Nitrated polycyclic aromatic hydrocarbons, multidimensional gas chromatography, diesel particulates, nitrogen selective detector.

INTRODUCTION

Organic solvent extracts of ambient aerosols and diesel exhaust particles exhibit direct-acting mutagenicity, which has been attri-

buted, at least in part, to the presence of nitro-PAHs.[1–4] Some members of this class of compounds have been shown to be potent direct-acting mutagens by the Ames Salmonella Bioassay.[2,3,4] M. C. Paputa-Peck *et al.* have found at least 100 nitro-PAHs in diesel particulate extracts, most of which are of undetermined mutagenicity and concentration.[5]

A great variety of procedures has been employed in the analysis of air and diesel exhaust particles for nitro-PAHs. The most commonly used procedures involve soxhlet extraction of particulate samples.[1,6–12] Ultrasonic extraction has also been employed in a few studies.[5,13,14,15] Because the composition of air and diesel particles is extremely complex, fractionation of the extract is generally required prior to analysis. Although high pressure liquid chromatography (HPLC) has been the method of choice in most studies,[1,5–15] column chromatography,[2,16,17] thin layer chromatography,[3,4] liquid–liquid partitioning,[18] and multi-column HPLC[19] also have been used. Very few analyses have been conducted with unfractionated extracts.[20] Most detection systems have been characterized by the use of a detector combined with a capillary gas chromatograph. Gas chromatography/mass spectrometry has been the most widely used detection system,[1,2,6–11,14,16,17,19] but the combination also includes gas chromatography/flame ionization detector[8,17] gas chromatography/nitrogen phosphorous detector or gas chromatography/nitrogen selective detector,[1,5,11,14,15,16,17] gas chromatography/electron capture detector,[12,17] and gas chromatography/thermal energy analyzer.[13,16] Fluorescence,[4] ultraviolet-visible absorption,[3,19] and mass/spectrometry/mass spectrometry systems[10,18,20] have also been employed.

Gas chromatographic analyses of environmental samples commonly employ lengthy clean-up procedures to isolate the nitro-PAHs and other classes of compounds. While a comprehensive analysis can be achieved with such procedures, short, simple procedures providing crude fractions may suffice for screening samples or for analyses of limited scope. Moreover, enhanced chromatographic separations and precise retention times are needed to provide increased confidence in identifying specific compounds in complex mixtures. MDGC offers a means for obtaining enhanced GC separations in complex mixtures as well as additional information content from chromatographic data.[21–25]

EXPERIMENTAL APPARATUS

The MDGC used in this study was a SiCHROMAT 2 (Siemens AG) equipped with an on-column capillary injector (Varian, Model 1095). It was operated with two tandem columns of different polarities. The columns were contained in separate ovens and operated with independent temperature programs. The chromatograph employs a Deans-type switching system to obtain heart-cuts from the front column and transfer them to the back column for analysis. The heart-cuts are made automatically at pre-selected times by appropriate programming of the MDGC. The SiCHROMAT 2 switching system has been described previously.[26] An SPB-35 fused silica capillary column, 15 m × 0.32 mm i.d. × 0.25 μm film thickness (Supelco, Inc.) was the column of choice for the front column. A length of deactivated fused silica capillary tubing (1.5 m × 0.32 mm i.d.) (J&W Scientific, Inc.) served as a guard column to protect the front column from any involatile compounds present in sample injections. The back (analytical) column was a DB-5, 20 m × 0.32 mm i.d. × 0.25 μm film thickness (J&W Scientific, Inc.). The front column effluents were monitored with a FID while the back column effluents were split to a FID and NSD by means of a variable effluent splitter (Siemens AG).

Chromatograms from the front column FID were obtained with a strip chart recorder (Soltec Corp., Model 1320). Computing integrators (Perkin-Elmer Corp., Model LCI-100) were employed for both the FID and NSD of the back column. Analyte retention times on the back column were reported by the integrators while retention times on the front column were read on-the-fly from the chromatograph display panel.

Particulate samples were extracted with a Sonifier Cell Disruptor (Heat Systems-Ultrasonics, Inc., Model W185) using a 1/2 in. diameter ultrasonic probe operated at 40–50 watts.

MATERIALS

Calibration and test solutions of nitro-PAHs consisted of mixtures prepared by Northrop Services, National Bureau of Standards SRM 1587, and solutions of individual nitro-PAHs in toluene procured

from Analabs. Solvents were Burdick and Jackson chromatography grade. National Bureau of Standards diesel particulate sample SRM 1650 served as a test particulate sample. A short open Florisil column (Floridin Co., PR grade, 60/100 mesh) was used for sample clean-up. All procedures were carried out under lights filtered with Kodagraph Yellow sheeting (Eastman Kodak Co.) to reduce the risk of photodegradation of nitro-PAHs.

PROCEDURES

Extraction

A particulate sample (0.1 g) and an internal standard (4.4 μg 3-nitrofluoranthene) were added to a beaker containing 30 ml of a toluene:methylene chloride mixture (3:1) and extracted ultrasonically for 60 min. The beaker was fitted with a custom-made Teflon cap to minimize aerosol and splashing losses during agitation. The extract solution was centrifuged, decanted, and filtered through a 0.5 μm Teflon membrane filter with the aid of a vacuum. The filtrate was blown down to a volume of 1 ml with charcoal-filtered helium.

Clean-up

The concentrated filtrate was fractionated on a Florisil column (10 cm × 6 mm i.d.) using a light vacuum to obtain a solvent flow of approximately 1 ml/min. A sample clean-up step was found to be necessary because the injection of raw filtered particulate extracts caused rapid deterioration in the performance of the front column, despite the use of a guard column. Ten mL of toluene was used to elute the first fraction (toluene eluate I); another 10 mL of toluene was used to elute the second fraction (toluene eluate II); 10 mL of methylene chloride eluted the third fraction (methylene chloride eluate). The eluates were filtered with Teflon membrane filters and blown down to a volume of approximately 300 μl. The exact volumes of the final concentrated solutions were determined using a graduated syringe. The concentrated eluates were stored in the dark in glass vials at room temperature.

Chromatography

The multidimensional gas chromatograph system was operated with helium as the carrier gas at a flow rate of 6 mL/min at room temperature. The needle valve controlling the pressure differential across the back column effluent splitter was adjusted to provide an appropriate split flow between the two detectors. The NSD was operated with the current at 800 mA to achieve maximum sensitivity.

Prior to the analysis of an unknown, a standard sample was run to program the MDGC computer for the heart-cutting and the back column oven temperature appropriate for the analyte of interest. The typical chromatographic procedure consisted of injecting 2 μl of sample on-column with the injector and column ovens at 100°C. After 1/2 min the injector temperature program was begun. One min after injection the GC program was begun with the front column temperature rising under program control to 280°C and the effluent directed to the monitor detector. One half minute before the predetermined elution time of the analyte peak, the effluent was automatically directed to the back column for a period of 1 minute. The effluent was then switched back to the monitor detector. One minute later the temperature of the back column began to rise under program control to the final column temperature (280°C). After 60 min, the GC was re-set automatically to the starting conditions. The GC was ready for the next sample as soon as the injector had cooled down (ca. 15 min).

Computations

Nitro-PAHs in this study were identified by the NSD retention times and quantitated by peak height measurements. The acceptable limits for agreement of the unknown and reference sample retention times was set at $\pm$0.009 min.

The calibration curves for 1-nitropyrene and the other quantitated nitro-PAHs were found to be linear with a zero intercept (e.g., Figure 1). Accordingly, daily 1-point calibrations were used for the nitro-PAHs.

RESULTS AND DISCUSSION

Tests with synthetic mixtures showed retention times on the back

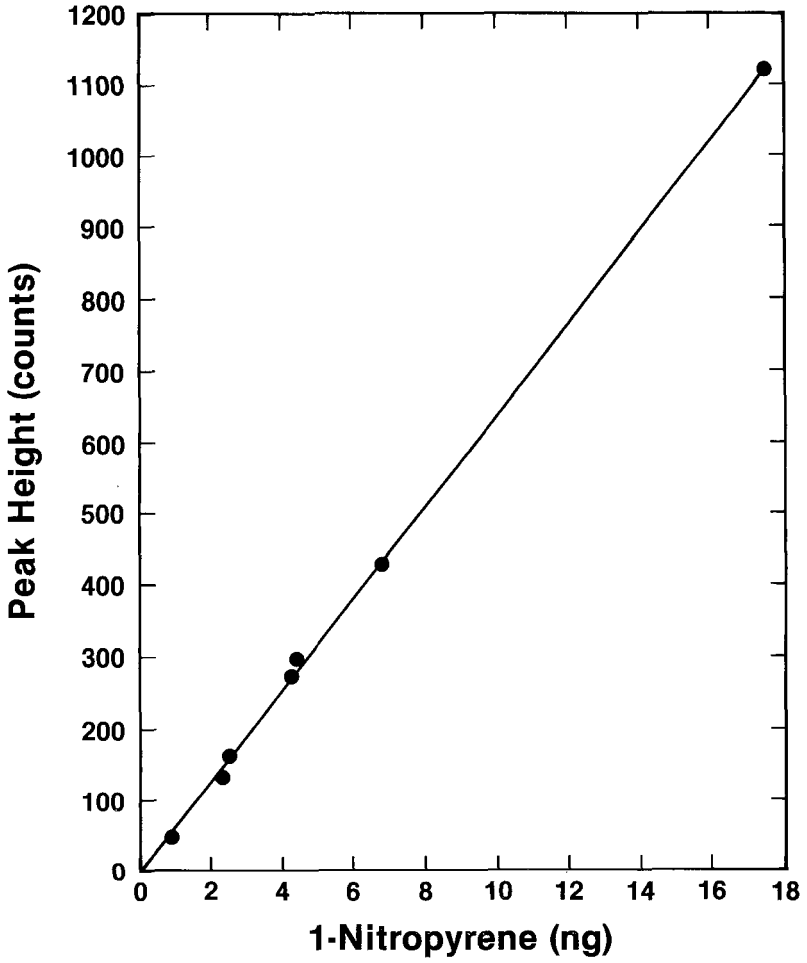

Figure 1 NSD calibration, MDGC back column.

column to be highly reproducible (Table I). The superior reproducibility on the back column is attributed to the heart-cutting and focusing operations which reduce the effects of injection variables.

Although the reproducibility of the area response was found to be better than that of the peak height with synthetic mixtures of 1-nitropyrene (Table II), the area response was found to be much less

Table I Reproducibility of chromatographic retention time

Compound	Time (min)[a]		
	Front column, FID	Back Column, FID	Back column, NSD
9-Nitroanthracene	11.791 ± 0.007	19.745 ± 0.005	
1-Nitropyrene	16.620 ± 0.009	26.966 ± 0.004	26.937 ± 0.005
6-Nitrochrysene	21.602 ± 0.017		
Pyrene	11.554 ± 0.005	19.569 ± 0.003	
Chrysene	14.411 ± 0.005	24.299 ± 0.002	

[a]Mean of 4–8 samples ± SD.

Table II 1-Nitropyrene reproducibility on back column using the LCI-100 *integrator*[a]

	FID		NSD		Ratio, NSD/FID	
	Area (counts)	Peak height (counts)	Area (counts)	Peak height (counts)	Area	Peak height
Mean	1872×10^3	1146	7100×10^3	4622	3.793	4.031
SD	70.1×10^3	69	235×10^3	332	0.017	0.086
RSD (%)	3.75	6.02	3.31	7.18	0.456	2.13

[a]Values obtained from 4 analyses of a standard solution.

accurate in the complex chromatograms obtained with diesel extract samples. Accordingly, quantitation in this study was based on peak height measurements. The limit of detection for 1-nitropyrene at 2× noise was calculated to be 36 picograms under the NSD/FID splitter ratio employed in this study.

Of the three eluates from the toluene:methylene chloride (T:MC) extract, the second toluene eluate (II) produced the greatest total response in the NSD chromatograms of the back column. The first toluene eluate (I) produced, by far, the greatest response in the FID chromatogram of the front column (Figure 2). The front column chromatogram of the methylene chloride eluate (not shown) was similar to that of toluene eluate II, except that no peaks were observed with retention times greater than 14 min. The overwhelming FID response from toluene eluate I indicates that it contains most

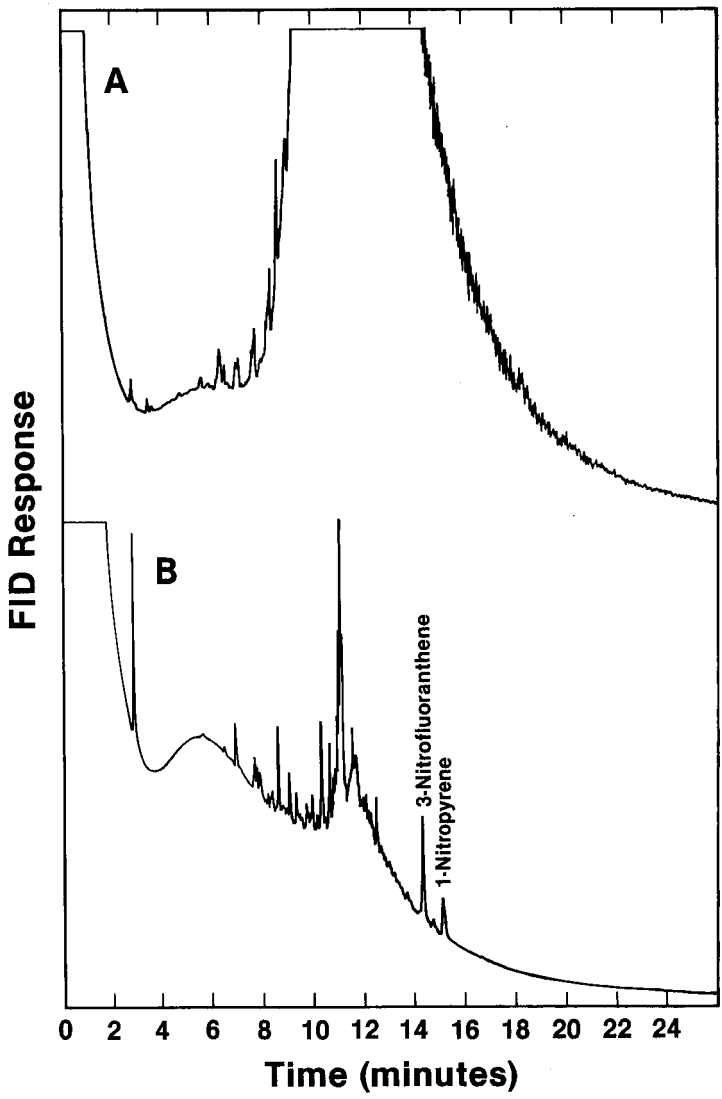

Figure 2 Front column chromatograms of the T:MC extract of SRM 1650: (A) toluene eluate I; (B) toluene eluate II.

of the hydrocarbons present in the extract. An initial elution with *n*-hexane would reduce the amount of hydrocarbons co-eluting with the nitro-PAHs. However, it is not likely that an *n*-hexane elution will simplify the FID chromatograms of the back column enough to make the NSD/FID peak height ratios generally useful for peak identification. In the methylene chloride eluate, for example, only one-half of the NSD peaks could be paired with an FID peak having a corresponding retention time. Shorter cut intervals would help to simplify the back column chromatogram, but at the cost of measuring fewer analytes with each sample injection.

Good results were obtained with the NSD/FID response ratios for 1-nitro-pyrene, a strong, well-resolved peak in a simple chromatogram (Figure 3). In that sample both the NSD and FID retention times agreed with the reference sample to within 0.004 min. The difference in the NSD/FID peak height response ratios for 1-nitropyrene in the eluate and reference samples was 9%, and the difference in the NSD/FID area response ratio was 6%. For those compounds, such as 1-nitropyrene, which are not obscured in the front column chromatogram, the front column retention time provides another piece of information for identification.

Humps of incompletely resolved peaks were present in the NSD chromatograms as well as the FID chromatograms of cuts from toluene eluate I and toluene eluate II (Figure 4). Peaks in cuts taken after 1-nitropyrene were weak and scant. The 9.9–11.9 min cut interval produced approximately 50% of the total NSD response obtained from all cuts of the 3 eluates and contained at least 79 different nitrogenated compounds. Altogether, at least 218 nitrogenated compounds eluting prior to 1-nitropyrene were present in the cuts of the 3 eluates. At least 65 additional compounds were observed eluting after 1-nitropyrene. Only a few nitro-PAHs were identified in this study (Table III). Three compounds detected in SRM 1650 produced an NSD response greater than 1-nitropyrene. 122 of the observed nitrogenated compounds were present at a 1-nitropyrene equivalent concentration $\geqq 1\ \mu g/g$.

Efficiency of the T:MC extraction was checked by re-extracting the SRM 1650 residue with 30 ml methylene chloride. Only 2% of the 1-nitropyrene found in SRM 1650 was contained in the methylene chloride extract.

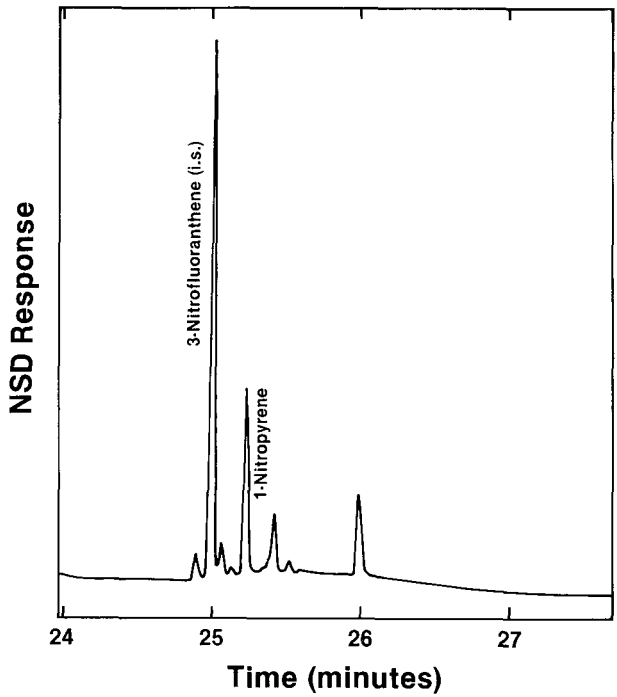

Figure 3 NSD chromatogram of toluene eluate II of SRM 1650 cut at 13.9–15.6 min. (Attenuation 256).

Table III Analysis of SRM 1650 for selected compounds

	Found (μg/g)	NBS analysis[a] (μg/g)	Per cent recovery
1-Nitropyrene	18.0	19	95
7-Nitrobenz(a)anthracene	0.8	2.8	29
2-Nitrofluorene	0.3	0.3	100
6-Nitrobenzo(a)pyrene	1.9	1.6	119
9-Nitroanthracene	4.1	N.R.[b]	
9-Nitrophenanthrene	N.Q.[c]		
6-Nitrochrysene	N.Q.		
9-Methyl, 10-Nitroanthracene	N.Q.		
4-Methyl, 3-Nitropyrene	N.Q.		

[a]Reported in the NBS certificate of analysis for SRM 1650.
[b]N.R.: Not Reported.
[c]N.Q.: Identified, but not quantitated.

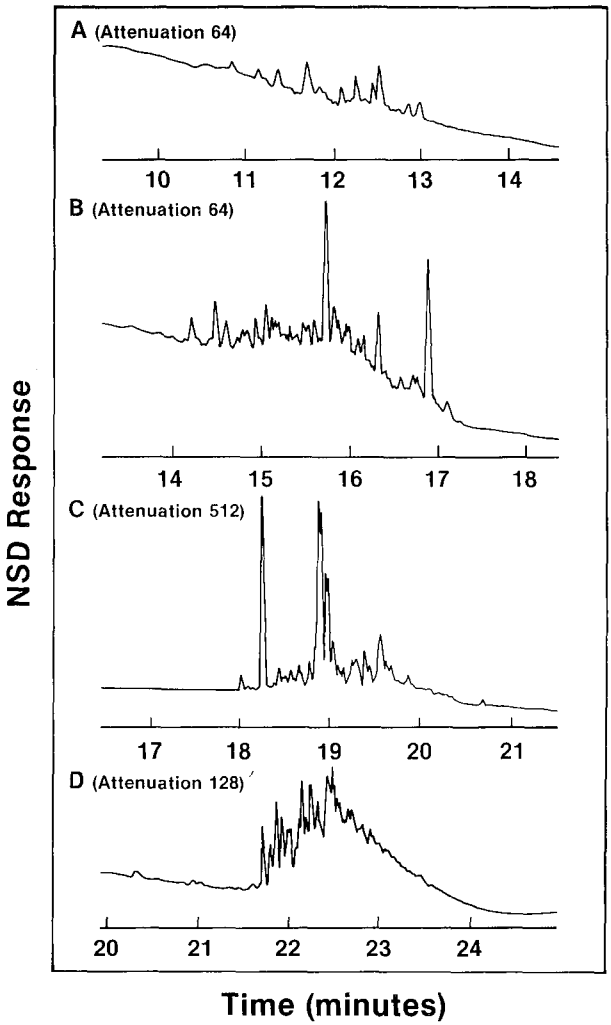

Figure 4 NSD chromatograms of toluene eluate II of SRM 1650: (A) cut at 5.9–7.9 min; (B) cut at 7.9–9.9 min; (C) cut at 9.9–11.9 min; (D) cut at 11.9–13.9 min.

Results of the nitro-PAH analysis of SRM 1650 were in good agreement with the National Bureau of Standards values, except for 7-nitrobenz(a)anthracene (Table III). The poor agreement between the 7-nitrobenz(a)anthracene values may be the result of the poor

quality of the NSD chromatogram containing the 7-nitrobenz(a)-anthracene, which was in sharp contrast to that containing the 1-nitropyrene (Figure 3). The 7-nitrobenz(a)anthracene was contained in a weak hump of poorly resolved peaks, which made identification of the peak and determination of the chromatographic base line very difficult. Those compounds which were identified but not quantitated were present at too low a concentration for accurate measurement, or there was no quantitative standard available.

3-nitrofluoranthene was selected as an internal standard, since none was detected in a preliminary analysis of SRM 1650. It was added to the initial extraction mixture to give a concentration of 145 ng/ml. 93% of the 3-nitrofluoranthene added was recovered, indicating the absence of any significant handling losses, even though this compound is very susceptible to photodegradation.

CONCLUSIONS

MDGC coupled with an NSD provided highly reproducible retention times and sensitive detection of nitro-PAHs in complex mixtures. Front column retention times and NSD/FID response ratios can, under favorable circumstances, provide additional information for highly reliable identification and measurement of analytes in diesel particulate extracts.

Efficient extraction of nitro-PAHs from diesel particulate samples was obtained with ultrasonic extraction using a 3:1 mixture of toluene and methylene chloride. Clean-up of the disel extract with a short Florisil column was sufficient to prevent GC column deterioration. However, further sample fractionation is desirable to simplify the NSD and FID chromatograms and reduce ambiguities in compound identification.

Although the conformation of the MDGC analysis system employed in this study is not well-suited for routine, extensive analyses of complex mixtures, it can be useful when monitoring such mixtures for selected nitro-PAHs, which may serve as surrogates or markers in environmental samples.

This article has not been subjected to Agency review and does not necessarily reflect the views of the Agency. Mention of trade names or commercial products does not constitute endorsement or recommendation for use.

References

1. I. Salmeen, R. A. Gorse, Jr., T. Riley and D. Schuetzle, in M. Cooke and A. J. Dennis (eds.), *Polynuclear Aromatic Hydrocarbons: Formation, Metabolism and Measurement* (Battelle Press, Columbus, Ohio, U.S.A., 1056–1066, 1983).
2. G. Nishioka, B. A. Petersen and J. Lewtas, in M. Cooke and A. J. Dennis (eds.), *Polynuclear Aromatic Hydrocarbons: Physical and Biological Chemistry* (Battelle Press, Columbus, Ohio, U.S.A., 603–613, 1982).
3. T. C. Pederson and J.-S. Siak, *J. Appl. Toxicol.* **1,** 54–60 (1981).
4. J. Siak, T. L. Chan, T. L. Gibson and G. T. Wolff, *Atmos. Environ.* **19,** 369–376 (1985).
5. M. C. Paputa-Peck, R. S. Marano, D. Schuetzle, T. L. Riley, C. V. Hampton, T. J. Prater, L. M. Skewes, T. E. Jensen, P. H. Ruehle, L. C. Bosch and W. P. Duncan, *Anal. Chem.* **55,** 1946–1954 (1983).
6. R. A. Gorse, Jr., T. L. Riley, F. C. Ferris, A. M. Pero, and L. M. Skewes, *Environ. Sci. Technol.* **17,** 198–202 (1983).
7. T. Ramdahl, G. Becher and A. Bjørseth, *Environ. Sci. Technol.* **16,** 861–865 (1982).
8. H. Y. Tong, J. A. Sweetman and F. W. Krasek, *J. Chromatogr.* **264,** 231–239 (1983).
9. J. A. Sweetman, F. W. Karasek and D. Schuetzle. *J. Chromatogr.* **247,** 245–254 (1982).
10. D. Schuetzle, T. L. Riley, T. J. Prater, T. M. Harvey and D. F. Hunt, *Anal.*
11. P. A. D'Agostino, D. R. Narine, B. E. McCarry and M. A. Quilliam, in M. Cooke and A. J. Dennis (eds.), *Polynuclear Aromatic Hydrocarbons: Formation, Metabolism and Measurement* (Battelle Press, Columbus, Ohio, U.S.A. 365–377, 1983).
12. W. M. Draper, *Chemosphere*, **15,** 437–447 (1986).
13. B. A. Tomkins, R. S. Brazell, M. E. Roth and V. H. Ostrum, *Anal. Chem.* **56,** 781–786 (1984).
14. T. Nielsen, B. Seitz and T. Ramdahl, *Atmos. Environ.* **18,** 2159–2165 (1984).
15. T. Nielsen, B. Seitz, A. M. Hansen, K. Keiding and B. Westerberg, in M. Cooke and A. J. Dennis (eds.), *Polynuclear Aromatic Hydrocarbons: Formation, Metabolism and Measurement* (Battelle Press, Columbus, Ohio, U.S.A., 961–970, 1983).
16. W. C. Yu, D. H. Fine, K. S. Chiu and K. Biemann, *Anal. Chem.* **56,** 1158–1162 (1984).
17. R. M. Campbell and M. L. Lee, *Anal. Chem.* **56,** 1026–1030 (1984).
18. T. R. Henderson, J. D. Sun, R. E. Royer, C. R. Clark, A. P. Li, T. M. Harvey, D. H. Hunt, J. E. Fulford, A. M. Lovette and W. R. Davidson, *Environ. Sci. Technol.* **17,** 443–449 (1983).
19. W. Lindner, W. Posch, O. S. Wolfbeis and P. Tritthard, *Chromatographia* **20,** 213–218 (1985).
20. T. R. Henderson, J. D. Sun, A. P. Li, R. L. Hanson, W. E. Bechtold, T. M. Harvey, J. Shabanowitz and D. F. Hunt, *Environ. Sci. Technol.* **18,** 428–434 (1984).
21. J. F. K. Huber, E. Kenndler and G. Reich, *J. Chromatogr.* **172,** 15–30 (1979).
22. W. V. Ligon, Jr. and R. J. May, *J. Chromatogr.* **294,** 77–86 (1984).
23. G. Schomburg, H. Husman and E. Hubinger, *J. High Resolut. Chromatogr. Chromatogr. Commun.* **8,** 395–400 (1985).
24. D. W. Wright, K. O. Mahler and L. B. Ballard, *J. Chromatogr. Sci.* **24,** 60–65 (1986).
25. W. Bertsch. *J. High Resolut. Chromatogr. Chromatogr. Commun.* **1,** 85–90 (1978).
26. F. Muller and U. K. Goekeler, *American Laboratory* **17,** 25–38 (May, 1985).