QUANTITATIVE ANALYSIS OF POLYNITROAROMATIC COMPOUNDS IN COMPLEX MIXTURES BY COMBINATION THIN-LAYER CHROMATOGRAPHY AND VISIBLE SPECTROMETRY

JOHN C. HOFFSOMMER AND JOANNE F. McCULLOUGH

U.S. Naval Ordnance Laboratory, White Oak, Silver Spring, Md. 20910 (U.S.A.)

(Received September 20th, 1968)

SUMMARY

A quantitative method of analysis for polynitroaromatic compounds in complex mixtures by combination thin-layer chromatography and visible spectrometry is described. Examples of the utility of the method for thermal decomposition studies and product analysis in complex reaction mixtures are presented.

INTRODUCTION

Although reasonable success has been achieved for the quantitative thin-layer chromatographic analyses of certain aliphatic nitro compounds in explosive mixtures by the spot area and weight method[1], spot densitometry[2], combination TLC and U.V. methods[3,4], relatively little has been done in the field of the polynitroaromatic compounds. An excellent summary of the thin-layer chromatography of explosives has recently been presented by KIRCHNER[5]. A spot area and weight method[6] for the analyses of 1,3-diamino-2,4,6-trinitrobenzene and 2,2',4,4',6,6'-hexanitrostilbene[7] after subjection to both thermal heating and nuclear irradiation has only recently been described. This latter method involved visualizing the developed zones with methanolic potassium hydroxide and photographing the resulting red spots with transparency film. In order to obtain the desired areas, the transparencies were projected on graph paper, traced, and finally measured with a compensating polar planimeter. The method depends strongly on excellent control of experimental conditions, is somewhat time consuming, but has been shown to have an accuracy within 5 %. Ideally, in terms of both speed and accuracy, one would desire a quantitative method of analysis based on the TLC separation of the desired polynitroaromatic compound, followed by physical or chemical analysis of the pure compound.

A method has been recently developed by GLOVER AND KAYSER[8] for the quantitative spectrophotometric analysis of polynitroaromatic compounds as their "Meisenheimer" complexes in ethylenediamine (EDA)–dimethylsulfoxide (DMSO) solutions. The method is especially suitable for microgram quantities of materials which can be applied on a single TLC plate, and provides a relatively rapid means for the quantita-

tive separation and analysis of polynitroaromatic compounds occurring in complex mixtures. We wish to report this procedure and illustrate its usefulness in several applications.

EXPERIMENTAL

Preparation of thin-layer acid-washed plates

Since considerable blanks in the visible were obtained from the DMSO extractions of the Silica Gel G support and subsequent treatment with EDA, it was necessary to prepare special, acid-washed plates according to the following. Thirty grams of Silica Gel G, according to Stahl, containing 13 % $CaSO_4$ (Brinkman Inst., Co.) was thoroughly mixed with 200 ml of acetone containing 2 ml concentrated HCl for 10 min. The slurry was suction-filtered on a Buchner funnel, and the filter cake washed with five 30 ml portions of acetone, sucked dry, and dried overnight at 100°. Thirty grams of this acid-washed Silica Gel G, when slurried with 50 ml distilled water, made approximately ten 10 × 20 × 0.03 cm coated TLC plates with a Camag applicator. Plates were dried for 1 h at 125°, and stored in a closed container. Negligible blanks were obtained with this specially prepared Silica Gel G.

Developing solvents

A mixed solvent system consisting of toluene-benzene-n-pentane-acetone in the ratio 40:40:20:5, by volume, was used for the development of all the polynitroaromatic compounds studied.

Visualizing reagent

Developed zones containing the polynitroaromatic compounds were visualized and located by spraying the TLC plate with a mixture of DMSO and EDA, (1:1) by volume (see Table I).

TABLE I

R_F VALUES AND COLORS OF VARIOUS POLYNITROAROMATIC COMPOUNDS ON SILICA GFI. G PLATES VISUALIZED WITH DMSO-EDA

Compound	R_F value	Color
1,3,5-Trinitrobenzene	0.75	Red
3,5-Dinitroiodobenzene[9]	0.85	Purple
2,2',4,4'-Tetranitrobiphenyl[10]	0.36	Violet
2,2',6,6'-Tetranitrobiphenyl[11]	0.60	Blue-purple
3,3',5,5'-Tetranitrobiphenyl[12]	0.62	Purple
3',5',2,4,6-Pentanitrobiphenyl[a]	0.54	Red

[a] Prepared by Dr. Howard E. Ruskie of this Laboratory from 3,5-dinitrobromobenzene and picryl chloride in a mixed Ullman reaction.

Analytical procedure

About 0.1 g of the polynitroaromatic compound to be analyzed was accurately weighed, dissolved in acetone, and transferred into a 25 ml volumetric flask. Ten to thirty microliter aliquots of these solutions were spotted on a 10 × 20 × 0.03 cm acid-washed, Silica Gel G TLC plate in a series of two to six separate spots with the aid of a plastic spotting guide. A Hamilton 50 μl (No. 705) or 5 μl (No. 7005) syringe

was used to measure accurately the aliquots. The usual analytical care was exercised in filling and applying the aliquots on the TLC plate. For convenience in locating the developed zones, a "guide" spot of the polynitroaromatic compound to be determined was spotted separately at the origin at the same time. The spotted plate was allowed to air dry for about 1 min, then developed by an ascending technique in a 16 × 21 × 26 cm glass, rectangular developing chamber fitted with a ground glass cover.

At the completion of the chromatogram, or when the solvent front had traveled a distance of between 10 and 14 cm, the plate was removed from the developing chamber and allowed to air dry in a vacuum hood for about 10 min. The sample side of the TLC plate was then completely covered with a clean glass plate, while the "guide" side was left exposed. A small line was drawn in the silica gel at the edge of the protecting cover plate to prevent contamination of the sample side with spray reagent. Visualization of the "guide" spot was accomplished by spraying the exposed area with EDA–DMSO (1:1, by volume). The protecting cover plate was removed, the zone containing the polynitroaromatic compound marked, and scraped off the plate with a clean flat spatula into a small 5–10 ml beaker prior to extraction with DMSO.

The silica gel containing the polynitroaromatic compound was extracted at least four times with DMSO in 0.5 to 1.0 ml portions. For repetitive extractions and analyses, it was most convenient to suction filter the DMSO extracts from the silica gel through a 2 ml, medium porosity, sintered glass funnel directly into a calibrated, 10 ml, glass test tube (Fig. 1). After the necessary amount of EDA was added, the total volume was adjusted to 10 ml with DMSO, and the solution concentration was determined spectrophotometrically in the visible (Table II). Absorbancy readings were made with a Beckman spectrophotometer, model DU, using quartz cells with a 1 cm light path.

The EDA–DMSO solutions of 1,3,5-trinitrobenzene, 3,5-dinitroiodobenzene, 3,3',5,5'-tetranitrobiphenyl and 3',5',2,4,6-pentanitrobiphenyl were stable for periods up to a half hour, while solutions of 2,2',6,6'-tetranitrobiphenyl were stable for periods up to 5 min. Extrapolation of the straight line resulting from a plot of absorbance of the 2,2',4,4'-tetranitrobiphenyl complex in EDA–DMSO solution *versus* time, to time zero, was necessary for the quantitative analysis of this compound. Beer's law was found to be valid over the entire concentration range investigated, $0.7 × 10^{-5} M$ to $4.0 × 10^{-5} M$ for all compounds.

TABLE II

SPECTRAL DATA. VISIBLE MAXIMA AND EXTINCTION COEFFICIENTS OF POLYNITROAROMATIC "MEISENHEIMER" COMPLEXES IN DMSO–EDA SOLUTIONS

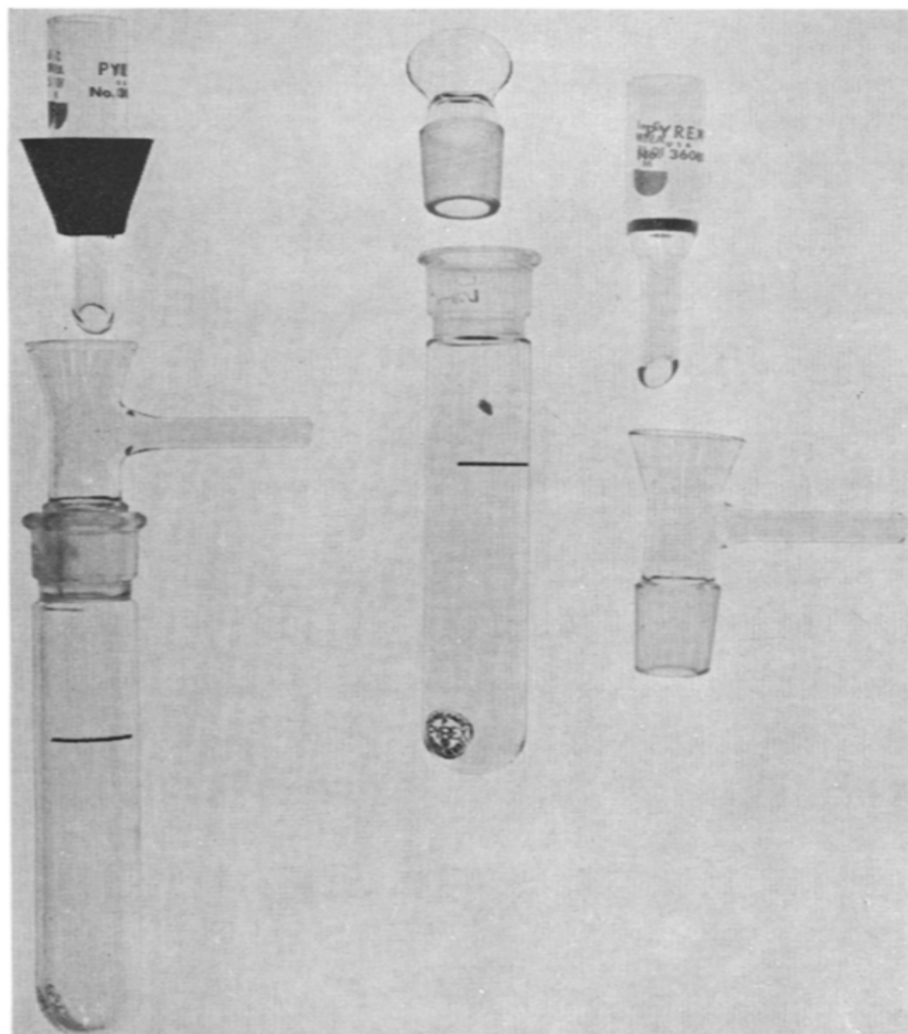| Compound	Milliliters EDA/10 ml solution	Max. in mμ; (ε)
1,3,5-Trinitrobenzene	0.1	540 (15,500); 455 (29,100)
3,5-Dinitroiodobenzene	1.0	535 (24,800); 363 (15,450)
2,2',4,4'-Tetranitrobiphenyl	2.0	545 (23,800); 355 (10,700)
2,2',6,6'-Tetranitrobiphenyl	5.0	550 (20,100); 350 (10,800)
3,3',5,5'-Tetranitrobiphenyl	1.0	550 (19,900); 450 (13,400)
3',5',2,4,6-Pentanitrobiphenyl	0.1	555 (13,600); 455 (29,100)

Fig. 1. Small-scale filtration apparatus for TLC extractions.

TABLE III

THIN-LAYER CHROMATOGRAPHIC–VISIBLE SPECTROPHOTOMETRIC ANALYSES OF KNOWN
QUANTITIES OF POLYNITROAROMATIC COMPOUNDS

	Micrograms applied[a]		Micrograms found[b]	
	Plate 1	Plate 2	Plate 1	Plate 2
Single compounds				
1,3,5-Trinitrobenzene	100.9	100.9	99.2	99.2
3,3',5,5'-Tetranitrobiphenyl	83.6	125.4	85.5	126.6
2,2',6,6'-Tetranitrobiphenyl	130.6	87.2	130.2	87.0
2,2',4,4'-Tetranitrobiphenyl	62.8	125.6	62.2	123.2
Mixture				
1,3,5-Trinitrobenzene	100.9	60.5	98.2	59.2
3',5',2,4,6-Pentanitrobiphenyl	53.3	32.0	51.8	30.4
2,2',6,6'-Tetranitrobiphenyl	101.8	61.1	97.8	57.8

[a] Stock acetone solutions contained approximately 0.1 g of compound accurately weighed into a 25 ml volumetric flask.
[b] After development and extraction of silica gel zone containing compound.

RESULTS AND DISCUSSION

Analyses of single compounds and synthetic mixtures

To test the accuracy and validity of the thin-layer analytical procedure for the determination of polynitroaromatic compounds, analyses were performed as outlined on solutions of known concentrations for both single compounds and a three-component mixture. The results are shown in Table III. To obtain maximum resolution and avoid tailing of sample, each spot applied was made no larger than 5 μl and contained no more than 20 μg of compound. Routinely, the extinction coefficients occurring at maxima between 535 mμ and 555 mμ were used for the analyses (Table II), while those maxima between 350 and 455 mμ provided additional identification and verification. As Table III shows, the accuracy is 1–2 % for single compounds and 2–5 % for a three-component mixture analyzed simultaneously on a single plate.

APPLICATIONS

Thermal degradation of explosives

The thermal degradation of explosives has most commonly been assessed by total gas evolution[13]. Since the decomposition processes forming these gases are understandably extremely complex, perhaps a more reliable measure of stability would be an analysis of residual explosive after a specified heat treatment. The thermal degradation of 2,2',6,6'-tetranitrobiphenyl (m.p. 219–220°) in the melt at 260° is illustrative (Table IV). Samples of 2,2',6,6'-tetranitrobiphenyl were weighed into 10 ml pyrex glass tubes which were evacuated and sealed prior to heating. Analysis of residual 2,2',6,6'-tetranitrobiphenyl was accomplished by dissolving or extracting the entire contents of the opened tube with acetone and quantitatively transferring to a 25 ml volumetric flask. Aliquots of this solution were spotted on TLC plates, the plates developed, and zones containing the chromatographed 2,2',6,6'-tetranitrobiphenyl extracted into 10 ml volumetric flasks and analyzed spectrophotometrically. The results obtained (Table IV) indicate that the 2,2',6,6'-tetranitrobiphenyl decomposed almost linearly at a rate of 0.9 %/h with an average deviation of 0.1 %/h as a neat liquid at 260° to about 95 % decomposition.

TABLE IV

THERMAL DECOMPOSITION OF 2,2',6,6'-TETRANITROBIPHENYL AS A NEAT LIQUID AT 260°

Initial tetranitro-biphenyl (g)	Heating period (h)	Residual tetranitro-biphenyl (g)[a]	
		Analysis 1	Analysis 2
0.1046	8.03	0.0980	0.0968
0.1061	15.42	0.0897	0.0850
0.1040	21.03	0.0840	0.0828
0.1045	68.50	0.0427	0.0449
0.1026	100.00	0.0161	0.0182
0.1039	138.20	0.0051	0.0055

[a] Calculated from: absorbancy/ε × 10 ml/aliquot (ml) × 25 ml/1000 ml × mol.wt. tetranitrobiphenyl.

Analyses of a complex reaction mixture. Ullman synthesis of 3,3',5,5'-tetranitrobiphenyl

The research synthetic organic chemist would often like to be able to analyze a difficultly separable reaction mixture for the desired product without gross isolation of product. Not only is isolation of product time consuming, but more important, the isolation is never complete. How much is actually isolated often depends largely on the chemist's patience and technique. The Ullman synthesis of 3,3',5,5'-tetranitrobiphenyl serves as another example of the utility of the TLC–spectrophotometric method of analysis for polynitroaromatic compounds.
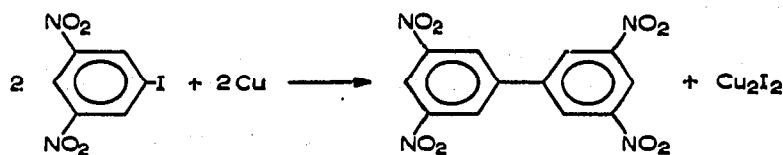
Twenty grams (0.0670 moles) of 3,5-dinitroiodobenzene and 12.0 (0.189 moles) Cu (U.S. Bronze Powder Incorp., Venus Brand, Natural Copper Fine, No. 44-F) in 150 ml dry nitrobenzene were placed in a three-necked, round-bottom flask equipped with a mechanical stirrer, reflux condenser, and thermometer. The mixture was heated for a total of 2.5 h at 208°, and appeared muddy, black-brown at the end of this period. After filtration of the inorganic salts, excess copper, and washing the filter cake with methanol, 200 ml of a dark-brown filtrate was obtained. Qualitative TLC indicated that the filtrate contained at least ten materials including the desired 3,3',5,5'-tetranitrobiphenyl and some unreacted 3,5-dinitroiodobenzene. Simultaneous analyses of the 3,3',5,5'-tetranitrobiphenyl and the 3,5-dinitroiodobenzene were accomplished as follows. A 2 ml aliquot of the dark-brown filtrate was diluted to 10 ml with acetone, and 3 ml of this solution was further diluted to 10 ml. Then, 15 μl and 30 μl aliquots of this final solution were spotted on two thin-layer plates and developed with benzene–toluene–*n*-pentane (50:10:5, by volume). The location, isolation, extraction, and spectrophotometric analyses of the zones containing the 3,3',5,5'-tetranitrobiphenyl and the 3,5-dinitroiodobenzene were accomplished by the general analytical procedure already given. The results of these analyses indicated the formation of 5.35 ± 0.01 g (0.0160 moles) of the 3,3',5,5'-tetranitrobiphenyl and 2.34 ± 0.03 g (0.00796 moles) of the starting 3,5-dinitroiodobenzene was still left unreacted.

On the basis of these TLC–spectrophotometric analyses, a 53 % yield of the 3,3',5,5'-tetranitrobiphenyl was actually formed under the conditions of the experiment and was based on the amount of 3,5-dinitroiodobenzene reacted. But, in the normal synthetic procedure after steam distillation, solvent extraction, and several carbon treatments followed by two recrystallizations, only 2.0 g (0.0060 moles) of the 3,3',5,5'-tetranitrobiphenyl was actually isolated and represented a 20 % yield on the same basis. A 13 % yield has been reported[12] for the formation of the 3,3',5,5'-tetranitrobiphenyl from 3,5-dinitroiodobenzene in the absence of solvent. These results clearly indicate the greater efficiency of separation by TLC as compared to conventional techniques.

ACKNOWLEDGEMENTS

REFERENCES

1 J. A. KOHLBECK, *Anal. Chem.* 37 (1965) 1282.
2 J. HANSSON, *Explosivstoffe*, 10 (1963) 73.
3 M. I. FAUTH AND G. W. ROECKER, *J. Chromatog.*, 18 (1965) 608.
4 D. F. DEBENHAM AND A. J. OWEN, *E. R. D. E. Rept.*, 1/BR/66, Waltham Abbey, England, January, 1966.
5 J. G. KIRCHNER, *Thin-Layer Chromatography, Techniques of Organic Chemistry*, Vol. 12, Interscience, New York, 1967, p. 684–686.
6 J. C. HOFFSOMMER, J. M. ROSEN AND J. S. FEIFFER, *Ind. Eng. Chem. Prod. Res. Develop.*, in press.
7 K. G. SHIPP, *J. Org. Chem.*, 29 (1964) 2620.
8 D. J. GLOVER AND E. G. KAYSER, *Anal. Chem.*, 40 (1968) 2055.
9 T. L. FLETCHER, M. J. NAMKUNG, W. H. WETZEL AND H. PAN, *J. Org. Chem.*, 25 (1960) 1342.
10 F. ULLMAN AND J. BIELECKI, *Ber.*, 34 (1901) 2177.
11 W. BORSCHE AND D. RANTSCHEFF, *Ann.*, 379 (1911) 176.
12 F. H. CASE, *J. Am. Chem. Soc.*, 64 (1942) 1848.
13 C. E. H. BAWN, in W. E. GARNER (Editor), *Chemistry of the Solid State*, Academic Press, New York, 1955, p. 254–267.