

Note

Comparison of electron capture and electrolytic conductivity for the gas chromatographic detection of some perchloro derivatives of diethylstilbestrol

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Recently, we reported^{1,2} an empirical comparison of an electron-capture detector (ECD) with a Coulson electrolytic-conductivity detector (CECD) for the gas chromatographic analysis of some perfluoro derivatives of agricultural chemicals. It has been found that (i) the sensitivity of the derivatives increased with increasing fluorine content by both electron-capture and electrolytic-conductivity detection, (ii) electron capture was more sensitive than electrolytic conductivity and (iii) the heptafluoro and pentadecafluoro derivatives were preferred owing to their sensitivity and stability. Whereas electron-capture detection was dependent, in part, upon structure of the parent, the CECD response was directly related to the percent of fluorine by weight in the molecules. The present work extends this study of a comparison of the two detectors to a series of perchloro derivatives of diethylstilbestrol (DES) in order to assess the results for best application to routine analysis.

EXPERIMENTAL

Reagents

DES was dissolved in benzene at 1 mg/ml to effect equilibration of *cis* and *trans* isomers. The formation of the perchloro derivatives of DES was carried out as described below. The derivatization reagents were monochloroacetyl (MCA) chloride, dichloroacetyl (DCA) chloride, dichloroacetic anhydride, trichloroacetyl (TCA) chloride, trichloroacetic anhydride and, for further comparison, trifluoroacetic (TFA) anhydride.

Derivative formation

Hundred μg of DES in 0.1 ml of benzene (1 mg/ml stock solution) were added to a 15 ml centrifuge tube. Following this were added 0.4 ml of benzene, 0.5 ml of 0.1 M trimethylamine in benzene (prepared as described earlier²), and 50 μl of MCA chloride. The tube was stoppered, gently shaken and left for 60 min at 25°. After this, the contents were partitioned twice with 10 ml of distilled water. An aliquot of the benzene layer was injected directly, or after appropriate dilution, into the gas chromatograph. The same procedure was carried out for the other reagents mentioned above.

Gas chromatography

A Microtek MT 220 gas chromatograph equipped with a CECD in the halogen mode was used for the electrolytic conductivity results. The column (120 cm \times 4 mm I.D., glass) was packed with 3% OV-1 on Chromosorb W HP (80–100 mesh). Column temperature varied with the derivatives analysed. Operating conditions were: furnace temperature, 820°; transfer line, 210°; injection port, 225°; helium carrier flow-rate, 38 ml/min; hydrogen flow-rate, 32 ml/min.

The ECD data were obtained using a Hewlett-Packard Model 5713 gas chromatograph with a linearized ^{63}Ni ECD. The column was the same as that used for the CECD results. Chromatography conditions were: detector temperature, 250°; injection port, 250°; argon-methane (95:5) carrier flow-rate, 60 ml/min. The column temperature was varied depending upon the derivatives analysed.

RESULTS AND DISCUSSION

Table I records the gas chromatographic properties and detector responses to the MCA, DCA, TCA and TFA derivatives of DES on 3% OV-1. DES consistently produced two peaks, the *cis* and *trans* isomers, normally in a ratio (peak area) of 1:2.2 by ECD and 1:2.6 by CED, respectively. The *cis* peak was the first eluted in all cases. For the series MCA, DCA, TCA, and TFA, the ECD responses decreased although not by much at the 50-ng level of injection. This is in agreement with Donoho *et al.*³ who reported that TCA-DES was the least sensitive of the three chloroacetates.

TABLE I
RESPONSES OF DES DERIVATIVES

Derivative	Electron capture				Coulson			
	Sens.*	Retention (min)		Column temp. (°C)	Sens.**	Retention (min)		Column temp. (°C)
		<i>cis</i>	<i>trans</i>			<i>cis</i>	<i>trans</i>	
MCA	2.7	3.3	4.5	240	0.5	2.8	4.0	230
DCA	2.6	4.4	6.5	240	1.1	3.6	5.4	230
TCA	2.1	6.1	9.9	240	0.4	5.0	6.6	230
TFA	1.7	2.5	3.7	200	1.7	2.4	3.2	190

* Relative peak area (*cis* + *trans* peaks) (in.²)/50 ng DES injected, 128 \times attenuation.

** Relative peak area (*cis* + *trans* peaks) (in.²)/200 ng DES injected, 32 \times attenuation.

Landowne and Lipsky⁴ found that molar sensitivity by electron capture decreased in the same order as obtained in Table I for the monochloro-, dichloro-, trichloro- and trifluoroacetates of chloesterol.

Although the ECD results showed a decrease in sensitivity with increasing chlorine content, it was expected that the CECD sensitivity would increase and be approximately proportional to the number of chlorine atoms in the derivative. The CECD has been shown to provide a very consistent response/chlorine with several organochlorine compounds⁵. We also have found this to be reasonably consistent with chlorine- as well as fluorine-containing compounds². However, when the CECD

response decreased for the TCA derivative (Table I) relative to the other two chloroacetates, we suspected column decomposition since there was no adequate explanation for the decrease by considering electrolytic-conductivity detector mechanism. Further studies with calibration curves have shown this to be so. While the response decreased nearly linearly with smaller quantities injected of the MCA and DCA derivatives of DES by both ECD and CECD, there was a non-linear rapid decrease in response for the TCA derivative with smaller quantities injected. The non-linear decrease for TCA-DES appeared to be the result of degradation on the chromatographic column. In fact at low nanogram levels, it was necessary to inject at least five times before a consistent peak was obtained for the TCA derivatives. The decomposition of the TCA derivative varied somewhat with different conditions and from day to day especially when new chromatographic columns were used.

Some other support for gas chromatographic decomposition was found in the fact that when the injection port and column temperature were lowered using the CECD, an increase in TCA derivative sensitivity was observed. However, this was not very consistent and usually amounted to not more than 10% when the temperature was dropped to 200° while using a shorter column.

There were no significant differences between the reactions of the acid chlorides and the anhydrides for the DES derivatives.

The TFA derivative of DES was found to be the least sensitive by ECD of the four derivatives examined, even when column decomposition of the TCA derivative was ignored. However, it was most sensitive by CECD, although all were within the same order of magnitude.

CONCLUSIONS

Electron capture was found to be about 10–100 fold more sensitive than electrolytic conductivity for the detection of the chloroacetate derivatives of DES. The relatively low sensitivity of both detectors for the trichloroacetate was found to be due to decomposition during chromatography and not to lower detector response. The TFA derivative was the least sensitive by ECD but was the most sensitive by CECD.

REFERENCES

- 1 J. F. Lawrence and J. J. Ryan, *J. Chromatogr.*, 130 (1977) 97.
- 2 J. J. Ryan and J. F. Lawrence, *J. Chromatogr.*, 135 (1977) 117.
- 3 A. L. Donoho, O. D. Decker, J. Koester and J. R. Koons, *J. Ass. Offic. Anal. Chem.*, 54 (1971) 75.
- 4 R. A. Landowne and S. R. Lipsky, *Anal. Chem.*, 35 (1963) 532.
- 5 R. Norstrom, *9th Eastern Canada Seminar for Pesticide Residue Analysis, Montreal, 1977.*