Journal of Chromatography, 251 (1982) 165–174
Chromatographic Reviews
Elsevier Scientific Publishing Company, Amsterdam — Printed in The Netherlands

CHROM. 14,524

HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY–MASS SPECTROMETRY WITH TRANSPORT INTERFACES

NICHOLAS J. ALCOCK, CHRISTINE ECKERS, DAVID E. GAMES*, MARGUERITE P. L. GAMES, MICHAEL S. LANT, MARK A. McDOWALL, MICHAEL ROSSITER, RICHARD W. SMITH, STEVEN A. WESTWOOD and HEE-YEN WONG

Department of Chemistry, University College, P.O. Box 78, Cardiff CF1 1XL (Great Britain)

CONTENTS

1. INTRODUCTION

In the interfacing of a high-performance liquid chromatograph to a mass spectrometer (HPLC–MS) there are three fundamental problems to be overcome.

(1) How to make the mass spectrometer (which can handle 20 cm^3 min^{-1} of gas if configured for chemical ionization (CI)), compatible with solvent flow-rates of the order of 1 cm^3 min^{-1}, which result in gas volumes in the range of 150–1200 cm^3 min^{-1}, depending on the solvents used.

(2) Introduction of the solute into the mass spectrometer so that mass spectral information can be obtained from it and it does not undergo thermal decomposition.

(3) Coupling of the high-performance liquid chromatograph with the mass spectrometer so that there is no loss of chromatographic performance.

There have been a large number of different approaches to the problem which have been described in a number of review articles[1–11] and in this volume. This paper reviews the progress that has been made with one of these approaches, *i.e.* interfaces of the transport type.

2. HPLC–MS INTERFACES OF THE TRANSPORT TYPE PROVIDING ELECTRON IMPACT AND CHEMICAL IONIZATION DATA

The development of interfaces of the transport type originated with Scott *et al.*'s studies using a moving-wire system[12–14], which was based on the Pye Unicam moving-wire HPLC detector. The interface consisted of a moving stainless steel wire

0378-4355/82/0000–0000/$02.75

which sampled the column eluent, solvent was removed in two differentially pumped chambers through which the wire passed. Solute was vaporized into the combined electron impact (EI)/CI source of a quadrupole mass spectrometer by electrical heating of the wire. The system was shown to work well, providing both EI and CI mass spectral data from complex mistures, and showing minimal band dispersion. There were two problems with the system: relatively poor sensitivity, because less than 1% of the column eluent was taken up by the wire, and the fact that the transport system was not continuous.

McFadden *et al.*[15] modified the approach by the use of a continuous moving belt and were able to achieve improved transfer efficiencies and hence better sensitivity. Initially a stainless steel belt was used; this was later replaced by a Kapton® belt. The former system, although exhibiting better wetting properties for aqueous solvent systems, caused thermal decomposition of samples. Kapton being a more inert surface provided excellent spectra from low volatility and thermally unstable molecules, since the samples were flash-vaporized into the ion source[5], the effect being similar to that observed in direct probe studies using rapid heating of sample from inert surfaces such as PTFE[16]. Problems were encountered with the early design of this interface in handling more polar solvents at the flow-rates used for HPLC. This was partially overcome by the introduction of an infrared (IR) heater in the area prior to the first vacuum lock[17]. Even with this facility a low dead volume splitter is necessary for most aqueous solvent mixtures since the system is for example unable to handle all the column eluent at flow-rates of 1 cm^3 min^{-1}, from systems containing more than 20% water. A further problem which occurs is that with aqueous solvent systems containing in excess of 50% water, beading of the solvent occurs on the belt causing pressure fluctuations in the ion source and hence providing poor mass spectral data. A number of solutions to this problem have been developed and will be discussed later. A further improvement to the system has been the provision of a scrubber immediately after the belt exits from the first vacuum chamber[7,18]. Solvent is continually fed onto two pads which remove residual materials from the belt, and the facility is particularly useful for studies using non-volatile buffers and ion-pairing reagents.

There are now numerous publications which testify to the utility of this type of HPLC–MS interface. Excellent maintenance of chromatographic integrity is observed, particularly if the eluent is fed directly onto the belt interface rather than being passed through a ultraviolet (UV) detector prior to feeding onto the belt. In fact we have recently observed improved resolution using microbore HPLC when the HPLC column is adjusted so that the eluent feeds directly onto the belt, thus ensuring a virtually zero dead volume connection. Both EI and CI mass spectral data can be obtained from the system using a wide variety of solvent systems, including reversed-phase systems containing volatile and involatile buffers. Using conventional liquid chromatographic columns full EI or CI spectra are obtainable from 200 ng of sample injected on-column and selected-ion monitoring (SIM) enables low nanogram amounts of sample to be detected.

Although microbore HPLC was initially utilized[15] with this system it has had little subsequent usage with interfaces of this type[19]. Because this type of HPLC uses much lower solvent flow-rates than conventional HPLC it has had extensive usage for HPLC–MS with systems of the direct liquid introduction[4,20–25] jet[26] and nebulizing[27]

types, because all the HPLC eluent can be fed into the mass spectrometer. With one exception[22] little attention has been paid in these studies to optimisation of chromatographic performance, and the efficiencies obtained appear to be considerably inferior to those obtained with conventional HPLC. Our initial experiences[19] with a JASCO microbore HPLC system were disappointing in that poor efficiencies were obtained. Recently we have modified the column-packing technique and reduced the dead volumes in the connection to the UV detector and have been able to obtain chromatographic data more comparable with that obtained from conventional HPLC systems[28,29]. One advantage of the moving belt systems over other types of HPLC–MS interfaces is that when microbore HPLC is used an effectively zero dead volume coupling can be obtained since the column can be placed almost in contact with the belt system. Extremely low dead volume couplings are essential with chromatographic systems of this type if chromatographic performance is to be maintained. With interfaces of the direct liquid introduction type, a novel way of overcoming the problem has been to introduce the microbore column into the interface[22]. We have found that utilization of LC systems of this type with our moving belt interfaces results in considerable improvement in sensitivity, enabling full scan spectra to be obtained at the low nanogram injected on column level. However the technique suffers from the fact that column overload occurs at the 15–20 μg sample injected on column level, resulting in loss of resolution. We have developed a manual column switching technique to assist with problems of this type[28,29].

With regard to the Finnigan system's ability to handle thermally labile and low volatility compounds, it appears to fall between the direct insertion probe and desorption chemical ionization[30]. Mono- and di-saccharides provide relative molecular mass information from on-line HPLC–MS with this interface[31]; however, even using chloride attachment negative ion CI we have failed to obtain such data from trisaccharides[19], although there are structurally useful fragment ion data present in the spectra. Data recently reported[32] for sucrose using a direct liquid introduction interface indicate that this approach provides more abundant ions in the relative molecular mass region than we have been able to obtain with the Finnigan interface under similar ionization conditions. The Finnigan system's ability to provide HPLC–MS data from more difficult compounds is illustrated by studies of nucleosides[21], peptides[8,19] and antibiotics[8,18,33]. One feature which we have consistently observed is that the spectra obtained from on-line HPLC–MS do not provide such abundant ions in the relative molecular mass region as those obtained by spotting solutions of the same samples onto the interface. We are presently unable to explain this effect, and we have also observed in studies of ergot alkaloids that the EI data obtained from minor components when compared with those obtained from the direct insertion probe indicate that thermal decomposition has occurred[34,35]. This phenomenon appears to be absent in the CI spectra at the same concentration and in the EI spectra at higher concentrations.

The main problems with this first commercial transport interface are:

(1) Limitations in the system's ability to handle highly volatile compounds which may be lost in the pumping system or distilled off by the IR heater.

(2) Because of the relatively high background, mass spectral data are not available below m/z 110.

(3) Difficulties in handling solvent systems containing a high proportion of water.

(4) Relatively poor sensitivity compared with gas chromatography (GC)–MS.

(5) Limited ability to provide relative molecular mass data from more difficult compounds.

Little can be done to overcome the first problem. It does not normally cause difficulties since HPLC is used mainly for compounds of lower volatility than would be lost on the interface. Microbore HPLC assists in the solution of the second problem and full scan spectra can be obtained which show good correlation with spectra obtained from the direct insertion probe, although if lower mass data are required then more sample has to be injected onto the LC and in order to obtain good reconstructed total ion current traces the computer must be adjusted to neglect the lower mass ions contributions.

Difficulties experienced with handling aqueous solvent systems have resulted in the development of a variety of different approaches for more effective handling of solvent systems of this type. The simplest approach is the addition of a water-miscible solvent, *e.g.* 2-propanol, to the belt prior to the aqueous eluent from the liquid chromatograph[36]. Alternatively the wetting characteristics of the belt can be improved by use of Snoop[18]. Improvement of the mass spectrometer pumping by use of turbo molecular pumps, incorporation of heaters in the differentially pumped chambers, increasing the belt speed and tunnelling of the isolation ports results in more dramatic improvements in the system's performance[37,38]. An aerosol deposition method using a preheated stream of argon fed into the HPLC eluent appears to be a very effective method for removal of solvent from interfaces of the belt type and is reported as allowing water to be handled at flow-rates of 1 cm^3 min^{-1} (ref. 39). An alternative approach[40–42] is to remove the necessity of feeding aqueous solvent systems onto the interface. This has been effected by use of a modified segmented-flow extractor between the liquid chromatograph and the mass spectrometer interface. The solute is extracted into methylene chloride or other suitable solvent, which is then delivered to the belt system of the interface. The technique also has the advantage of enabling ion pairing reagents to be more readily used for HPLC–MS. One problem with this type of approach is that inevitably there must be loss of sensitivity and chromatographic resolution during the extraction process. We have found[38,39] that the use of microbore HPLC offers an equally effective solution for the handling of high percentage aqueous solvent systems, *e.g.* solvent systems consisting of water–acetonitrile (80:20) can be readily handled if a flow-rate of 0.2 cm^3 min^{-1} of ethanol is fed onto the belt system prior to the microbore column eluent.

The remaining problems with moving-belt interfaces are more difficult to overcome. As was mentioned earlier, enhanced sensitivity can be obtained by use of microbore HPLC since background is reduced and the necessity for splitting high percentage aqueous solvent systems is removed. It may well prove that aerosol deposition of eluent[39] improves sensitivity for many classes of compounds since there is less danger of sample distillation or vaporization or sputtering off the belt, and the necessity for splitting the eluent is avoided. Introduction of the interface into the source should also result in enhanced sensitivity and lend assistance in obtaining relative molecular mass data from more difficult compounds. Two recently developed commercial systems, which work on the same principles as the Finnigan system, have adopted this latter approach. The system designed by VG analytical[43] is available on both magnetic and quadrupole instruments, and its availability on instruments of the

former type enables high-resolution and linked-scan data to be obtained. Sensitivity data reported to date indicate that the system is more sensitive than systems which do not enter the ion source of the mass spectrometer, it is also capable of handling higher solvent flow-rates than the earlier type of system. As yet there is no indication that the system has any particular advantages in handling more difficult compounds. More attention appears to have been given to the positioning of the interface in the ion source and the provision of controlled rapid heating in the system designed by Finnigan MAT[44]. Again better handling of aqueous solvent systems is reported and sensitivity appears to be improved, a full ammonia CI spectrum of caffeine being obtained from 1 ng of compound injected on column. Considerable improvement in the ability to obtain mass spectral data from difficult molecules is indicated by the spectrum obtained from deoxyadenosine 5-phosphate using water as a CI reagent gas. The spectrum exhibits an $(M + 1)^+$ ion at m/z 332 which shows 20% intensity relative to the base peak in the spectrum at m/z 136. The trisaccharide raffinose shows an abundant $(M + NH_4)^+$ ion at m/z 522 and equally impressive data is reported for α-acetyldigoxin and the flavanoid glycoside rutin. Although this data does not appear to have been obtained on-line it indicates that similar data should be obtainable from on-line HPLC–MS studies. Once again the system is available on both magnetic and quadrupole instruments and appears to represent a considerable improvement in systems of the transport type.

3. HPLC–MS INTERFACES OF THE TRANSPORT TYPE WITH OTHER FORMS OF MASS SPECTRAL IONIZATION

A moving-wire transport system has been utilized to interface an HPLC to a plasma chromatograph[45]. Iodo-, chloro- and bromo-benzene were studied with the system and it does not appear to have been further developed. This type of approach would not appear to be the most effective way of developing interfaces of this type and studies using an atmospheric pressure ion source where all the LC eluent is fed into the ion source appear to have been more effective[46]. Recent studies in this area which enable more difficult samples to be handled[47,48] and enable fragmentation behaviour to be controlled[48] may lead to the development of a useful system.

Another earlier approach to HPLC–MS used a continuous stainless steel belt, which was perforated with small holes thus enabling solvent to be entrained[49]. Solvent was removed in an evaporator and the remaining solute transported to a reactor, where it was converted into hydrocarbons by catalytic reduction with hydrogen. The hydrocarbons produced are characteristic of the solutes and were swept into the ion source of mass spectrometer operating in the CI mode. Impressive analyses of complex lipid mixtures, where the system was used as an HPLC detector, were reported. The main disadvantages were that conventional EI and/or CI spectra were not obtained and that extensive studies would have to be undertaken of a wide range of compounds to ascertain the types of spectrum that they produce before the system could be utilized for structural studies.

One advantage of the transport type of interface is that it readily lends itself to use with surface ionization techniques, which could considerably extend the range of compounds amenable to HPLC–MS. Patents have appeared for transport systems which could be used for laser desorption[50,51], and preliminary results have been

reported[52] using this technique with a Finnigan system which has been modified so that a stainless-steel belt passes through the ion source. Laser-desorbed neutrals are ionized under CI conditions using an argon discharge. The system has only as yet been used in the off-line mode. A further system designed for laser desorption together with EI and SIM has been reported[53–55] but as yet has only been used in the latter ionization modes. Data reported to date, with this system do not indicate that it has any particular advantages over existing systems. Preliminary data has also been reported for another transport system using SIM[56]. Insufficient data are available to assess the potential of this system. An interesting patent[57] describes a transport system which has a belt constructed from a high work function metal. Ionization is effected by use of a 10^5 V cm^{-1} field. Unfortunately, no examples of spectra of compounds or HPLC–MS are given and hence its potential cannot be assessed. Currently the most exciting possibility in this area is the use of fast atom bombardment (FAB) MS[58]. The technique has been shown to have an impressive ability to provide both relative molecular mass data and structurally significant fragment ions from compounds that cannot be handled by either mass spectral or ionization techniques except ^{252}Cf desorption mass spectrometry. One problem is that the sample has to be loaded in a matrix of glycerol. However, FAB spectra can be obtained from samples not loaded in this way, but the spectra only last for a relatively short time. Thus use of transport system which should provide a continued source of sample may be viable for HPLC–MS with FAB.

4. APPLICATIONS OF HPLC–MS USING INTERFACES OF THE TRANSPORT TYPE

Interfaces of this type have been used in a wide range of studies within the constraints mentioned earlier, and Table I summarizes where the technique has been applied and areas of potential application.

Recently there has been considerable interest in MS–MS techniques for the examination of crude mixtures. It has been indicated by some groups that the technique is preferable to HPLC–MS or GC–MS for studies of this type[77,78]. Other groups advocate a combined approach[79–81]. We have undertaken a comparative study of the two approaches, using both HPLC–MS and B/E linked scans[82] with and without collision-activated dissociation of protonated molecular ions formed from the mixture under CI conditions to study a crude extract of ergot[34,35]. Whilst we found the latter approach to be relatively easy to use we were unable to resolve isomeric mixtures. Although individual isomers in pure form could be readily differentiated we had problems in spectral interpretation due to artefact peaks and difficulties in interpretation of spectra due to interferences from fragment ions from other components of the mixture. HPLC–MS did not have these problems and also provided ready-made conditions for the isolation of components of interest. In addition we found that source contamination problems were not as severe in the HPLC–MS studies. As a result of this study we feel that MS–MS provides a useful screening technique for the location of known compounds in complex mixtures, although one must beware of matrix effects which could result in the production of false negatives. The technique is not really suitable by itself for the identification of unknown compounds, but should be extremely useful if used in combination with HPLC–MS and GC–MS in providing additional structural information and in differentiating isomers.

TABLE I

APPLICATIONS OF HPLC–MS USING INTERFACES OF THE TRANSPORT TYPE

Compound class	*References*
Aflatoxins	6, 17
Amaryllidaceae alkaloids	33
Antibiotics	8, 18, 33, 59
Aromatic acids	40, 60
Bile acids and their conjugates	8, 61
Carbamate pesticides	8, 33, 40, 60, 62, 63
Chinchona alkaloids	33
Chlorinated phenols in urine	36, 83
Coal liquefaction products	64, 65
Dinitrophenyl hydrazones	66
Drugs	8, 13, 14, 17, 41, 42, 67, 68
Effluent analysis	69
Ergot alkaloids	34, 35
Glycosides	31
Herbicides	70
Lipids	49
Liquid crystals	71
Natural coumarins	8, 10, 33, 67, 72
Nucleosides	8, 19, 31, 61
Peptides	8, 19
Pesticides	15
Petroporphyrins	73
Polychlorinated biphenyls and their metabolites	74
Polynuclear aromatics	37, 64, 65, 66, 75
Rotenoids	19, 76
Steroids	8, 13, 14, 43, 67
Sugars	11, 19, 31
Triglycerides	17
Waxes	6, 73

One area which has received comparatively little attention is quantitative studies. The viability of transport systems in this context has been shown[65], and recent improvements in the senstivities of interfaces of this type may lead to more exploitation of the technique in this area.

5. CONCLUSIONS

Combined HPLC–MS using moving-belt interfaces has been shown to be a viable technique. Extremely complex samples can be studied and the systems show a high degree of reliability. Recent developments have resulted in improved sensitivity and extended the range of compounds which can be studied. The ability of the system to be readily adapted for surface ionization techniques augurs well for future developments. Amongst the advantages of this approach to HPLC–MS are the ability to provide both EI and CI data, less care has to be taken with the quality of LC solvents, and the systems can be readily adapted to new ionization techniques. Use of the approach with MS–MS techniques should also enhance its ability to assist in the solution of problems.

6. ACKNOWLEDGEMENTS

We are indebted to the Royal Society of Chemistry Analytical Trust Fund (N.J.A.), SRC and Lilly Research (C.E.), SRC and ICI Pharmaceuticals (M.S.L.), SRC and Beecham Pharmaceuticals (M.A.M.), SRC and Esso Research (R.W.S.) ARC (S.A.W.), University of Wales and ICI Plant Protection (H.-Y.W.), G. D. Searle and A. H. Robins for their financial support. The SRC and the Royal Society are thanked for their financial assistance in the purchase of MS and HPLC equipment. We thank Finnigan MAT and VG Analytical for their assistance in our studies.

7. SUMMARY

The utility of various types of transport interface for high-performance liquid chromatography–mass spectrometry is reviewed, together with areas in which the technique has been applied. The advantages of using microbore high-performance liquid chromatography systems with interfaces of this type are described, and the relative merits of mass spectrometry–mass spectrometry and liquid chromatography–mass spectrometry for studies of complex mixtures are discussed.

REFERENCES

1 R. P. W. Scott, in A. Weissberger (Editor), *Contemporary Liquid Chromatography*, Wiley-Interscience, New York, 1976, p. 272.
2 B. G. Dawkins and F. W. McLafferty, in K. Tsuji and W. Morozowich (Editors), *GLC and HPLC Determination of Therapeutic Agents*, Marcel Dekker, New York, 1978, p. 259.
3 E. Kenndler and E. R. Schmid, in J. F. K. Huber (Editor), *Instrumentation of High-Performance Liquid Chromatography*, Elsevier, Amsterdam, 1978, p. 163.
4 P. J. Arpino and G. Guiochon, *Anal. Chem.*, 51 (1979) 682A.
5 P. J. Arpino, in M. Klem, A. V. Kruegel and S. P. Sobol (Editors), *Instrumental Applications in Forensic Drug Chemistry*, G.P.O., Washington, DC, 1978, p. 151.
6 W. H. McFadden, *J. Chromatogr. Sci.*, 17 (1979) 2
7 W. H. McFadden, *J. Chromatogr. Sci.*, 18 (1980) 97.
8 D. E. Games, C. Eckers, J. L. Gower, P. Hirter, M. E. Knight, E. Lewis, K. R. N. Rao and N. C. Weerashinghe in A. M. Lawson, K. Lim and W. Richmond (Editors), *Current Developments in the Clinical Application of HPLC, GC and MS*, Academic Press, London, 1980, p. 97.
9 D. E. Games, *Anal. Proc.*, 17 (1980) 110.
10 D. E. Games, *Anal. Proc.*, 17 (1980) 322.
11 D. E. Games, *Biomed. Mass Spectrom.*, 8 (1981) 454.
12 R. P. W. Scott, C. G. Scott, M. Munroe and J. Hess, Jr., in R. Porter (Editor), *The Poisoned Patient: The Role of the Laboratory*, Elsevier, New York, 1974, p. 155.
13 R. P. W. Scott, C. G. Scott, M. Munroe and J. Hess, Jr., *J. Chromatogr.*, 99 (1974) 395.
14 R. P. W. Scott, in H. S. Hertz and S. N. Chester (Editors), *Trace Organic Analysis: A New Frontier in Analytical Chemistry*, N.B.S., Washington, DC, 1979, p. 637.
15 W. H. McFadden, H. L. Schwartz and S. Evans, *J. Chromatogr.*, 122 (1976) 389.
16 R. J. Beuhler, E. Flanigan, L. J. Green and L. Friedman, *J. Amer. Chem. Soc.*, 96 (1974) 3990.
17 W. H. McFadden, D. C. Bradford, D. E. Games and J. L. Gower, *Amer. Lab.*, (1977) 55.
18 P. E. Kelley, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 276.
19 D. E. Games, P. Hirter, W. Kuhnz, E. Lewis, N. C. A. Weerasinghe and Steven A. Westwood, *J. Chromatogr.*, 203 (1981) 131.
20 J. D. Henion and G. A. Maylin, *Biomed. Mass Spectrom.*, 7 (1980) 115.
21 J. D. Henion, *J. Chromatogr. Sci.*, 19 (1981) 57.
22 J. J. Brophy, D. Nelson and M. K. Withers, *Int. J. Spectrom. Ion Phys.*, 36 (1980) 205.

23 K. H. Schäfer and K. Levsen, *J. Chromatogr.*, 206 (1981) 245.
24 E. Yamauchi, T. Mizuno and K. Azuma, *Shibak. Shitsuryo Bunseki*, 28 (1980) 227.
25 J. D. Henion, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 474.
26 T. Takeuchi, Y. Hirata and Y. Okumara, *Anal. Chem.*, 50 (1978) 659.
27 S. Tsuge, Y. Hirata and T. Takeuchi, *Anal. Chem.*, 51 (1979) 166, Y. Yoshida, H. Yoshida, S. Tsuge, T. Takeuchi and K. Mochizuki, *J. High Resolut. Chromatogr. Chromatogr. Commun.*, 3 (1980) 16; S. Tsuge, Yu. Yoshida, T. Takeuchi, K. Mochizuki, N. Kokubun and K. Hibi, *Chem. Biomed. Environ Instrum.*, 10 (1980) 405.
28 D. E. Games, M. S. Lant, S. A. Westwood and B. J. Woodhall, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 476.
29 S. A. Westwood, D. E. Games, M. S. Lant and B. J. Woodhall, *Anal. Proc.*, in press.
30 U. Rapp, G. Dielmann, D. E. Games, J. L. Gower and E. Lewis, in A. Quayle (Editor), *Advances in Mass Spectrometry*, Vol. 8, Heyden, London, 1980, p. 1660.
31 D. E. Games and E. Lewis, *Biomed. Mass Spectrom.*, 7 (1980) 433.
32 P. J. Arpino, P. Krien, S. Vajta and G. Devant, *J. Chromatogr.*, 203 (1981), 117.
33 C. Eckers, D. E. Games, E. Lewis, K. R. N. Rao, M. Rossiter and N. C. A. Weerasinghe, in A. Qualye (Editor), *Advances in Mass Spectrometry*, Vol. 8, Heyden, London, 1980, p. 1396.
34 D. E. Games, C. Eckers, B. P. Swann and D. N. B. Mallen, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 484.
35 C. Eckers, D. E. Games, D. N. B. Mallen and B. P. Swann, *Anal. Proc.*, in press.
36 L. H. Wright and T. R. Edgerton, *27th Annual Conference on Mass Spectrometry and Allied Topics, Seattle, 1979*, 1979, p. 742.
37 D. Hilker and P. P. Dymerski, *27th Annual Conference on Mass Spectrometry and Allied Topics, Seattle, 1979*, 1979, p. 625.
38 P. P. Dymerski, *28th Annual Conference on Mass Spectrometry and Allied Topics, New York, 1980*, 1980, p. 624.
39 R. D. Smith and A. L. Johnson, *Anal. Chem.*, 53 (1981) 739.
40 B. L. Karger, D. P. Kirby, P. Vouros, R. L. Foltz and B. Hidy, *Anal. Chem.*, 51 (1979) 2324.
41 D. P. Kirby, P. Vouros and B. L. Karger, *Science*, 209 (1980) 495.
42 D. P. Kirby, P. Vouros, B. L. Karger, B. Hidy and B. Petersen, *J. Chromatogr.*, 203 (1981) 139.
43 D. S. Millington, D. A. Yorke and P. Burns, in A. Quayle (Editor), *Advances in Mass Spectrometry*, Vol. 8, Heyden, London, 1980, p. 1819.
44 C. Brunnee, L. Delgmann and G. Dielmann, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 280.
45 F. W. Karasek and D. W. Denney, *Anal. Lett.*, 6 (1973) 993.
46 D. I. Carroll, I. Dzidic, R. N. Stillwell, K. D. Haegele and E. C. Horning, *Anal. Chem.*, 47 (1975) 2369; E. C. Horning, D. I. Carroll, I. Dzidic, K. D. Haegele, M. G. Horning and R. N. Stillwell, *J. Chromatogr.*, 99 (1974) 13; *J. Chromatogr. Sci.*, 12 (1974) 725; E. C. Horning, D. I. Carroll, I. Dzidic, K. D. Haegele, S.-N. Lin, C. U. Oertli and R. N. Stillwell, *Clin. Chem.*, 23 (1977) 13; E. C. Horning, D. I. Carroll, I. Dzidic and R. N. Stillwell, *Pure Appl. Chem.*, 50 (1978) 113.
47 B. A. Thomson and J. V. Iribarne, *J. Chem. Phys.*, 71 (1979) 4451.
48 M. Tsuchiya and T. Taira, *Int. J. Mass Spectrom. Ion. Phys.*, 34 (1980) 351.
49 W. L. Erdahl and O. S. Privett, *Lipids*, 12 (1977) 797, O. S. Privett and W. L. Ehrdahl, *Chem. Phys. Lipids*, 21 (1978) 361.
50 H. G. Noeller, H. D. Polaschegg and R. Wechung, *German Pat.*, 2,837,799 (1980).
51 R. Wechung, *German Pat.*, 2,837,715 (1980).
52 D. F. Hunt, W. M. Bone and J. Shabanowitz, *28th Annual Conference on Mass Spectrometry and Allied Topics, New York, 1980*, 1980, p. 620.
53 R. D. Smith and A. L. Johnson, *Anal. Chem.*, 53 (1981) 1120.
54 R. D. Smith, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 165.
55 R. D. Smith and A. L. Johnson, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 482.
56 A. Benninghoven, A. Ercke, M. Junack, W. Sichtermann, J. Krizek and H. Peters, *Org. Mass Spectrom.*, 15 (1980) 459.
57 C. Brunnee, F. Franzen and S. Meyer, *German Pat.*, 2,654,057 (1978).

58 M. Barber, R. S. Bordoli, R. D. Sedgwick and A. N. Tyler, *J. Chem. Soc. Chem. Commun.*, (1980) 325.
59 P. C. Tway and W. B. Caldwell, *29th Annual Conference on Mass Spectrometry and Allied Topics, Minneapolis, 1981*, 1981, p. 278.
60 B. L. Karger, D. P. Kirby and P. Vouros, *J. Chromatogr. Sci.*, 18 (1980) 111.
61 M. A. Quilliam and E. Y. Osei-Twum, *28th Annual Conference on Mass Spectrometry and Allied Topics, New York, 1980*, 1980, p. 612.
62 L. H. Wright and E. O. Oswald, *26th Annual Conference on Mass Spectrometry and Allied Topics, St. Louis, 1978*, 1978, p. 47.
63 D. E. Games and N. C. A. Weerasinghe, *J. Chromatogr. Sci.*, 18 (1980) 106.
64 W. A. Dark, W. H. McFadden and D. L. Bradford, *J. Chromatogr. Sci.*, 15 (1977) 454.
65 W. A. Dark and W. H. McFadden, *J. Chromatogr. Sci.*, 16 (1978) 289.
66 F. L. DeRoos and R. L. Foltz, *27th Annual Conference on Mass Spectrometry and Allied Topics, Seattle, 1979*, 1979, p. 358.
67 D. E. Games, J. L. Gower, M. G. Lee, I. A. S. Lewis, M. E. Pugh and M. Rossiter, in E. Reid (Editor), *Blood Drugs and Other Analytical Challenges*, Ellis Horwood, Chichester, 1978, p. 185.
68 D. E. Games, E. Lewis, N. J. Haskins and K. A. Waddell, in A. Quayle (Editor), *Advances in Mass Spectrometry*, Vol. 8, Heyden, London, 1980, p. 1233.
69 A. D. Thurston, Jr. and J. M. McGuire, *Biomed. Mass Spectrom.*, 8 (1981) 47.
70 R. F. Skinner, Q. Thomas, J. Giles and D. G. Crosby, *J. Chromatogr. Sci.*, 18 (1980) 108.
71 T. I. Martin and W. E. Hass, *Anal. Chem.*, 53 (1981) 593A.
72 D. E. Games, J. L. Gower, M. G. Lee, I. A. S. Lewis, M. E. Pugh and M. Rossiter, *Proc. Anal. Div. Chem. Soc.*, (1978) 101.
73 W. H. McFadden, D. C. Bradford, G. Eglington, S. K. Kajlbrahim and N. Nicolaides, *J. Chromatogr. Sci.*, 17 (1979) 518.
74 P. Dymerski, M. Kennedy and L. Kaminsky, in H. S. Hertz and S. N. Chester (Editors), *Trace Organic Analysis: A New Frontier in Analytical Chemistry*, N.B.S., Washington, DC, 1979, p. 685.
75 D. J. Stalling, J. D. Petty, G. R. Dubay and R. A. Smith, *J. Chromatogr. Sci.*, 18 (1980) 108.
76 S. A. Westwood, D. E. Games and L. Sheen, *J. Chromatogr.*, 204 (1981) 103.
77 R. W. Kondrat and R. G. Cooks, *Anal. Chem.*, 50 (1978) 81A.
78 D. F. Hunt, J. Shabanowitz and A. B. Giordani, *Anal. Chem.*, 52 (1980) 386.
79 F. W. McLafferty, *Acc. Chem. Res.*, 13 (1980) 33.
80 P. F. Bente III and F. W. McLafferty, in C. Merritt Jr. and C. N. McEwen (Editors), *Practical Spectroscopy Series, Vol. 3, Mass Spectrometry Part B*, Marcel Dekker, New York, 1980, p. 253.
81 F. W. McLafferty, *Biomed. Mass Spectrom.*, 8 (1981) 446.
82 A. P. Bruins, K. R. Jennings and S. Evans, *Int. J. Mass Spectrom. Ion Phys.*, 26 (1978) 395
83 L. H. Wright, T. R. Edgerton, S. J. Arbes Jr. and E. M. Lores, *Biomed. Mass Spectrom.*, 8 (1981) 475.