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QUANTITATIVE ANALYSIS OF MIXTURES OF DITERPENE HYDROCARBONS BY GAS CHROMATOGRAPHY-MASS SPECTROMETRY

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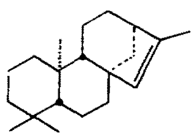
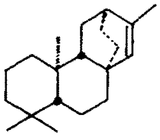
Abstract—The use of combined gas chromatography-mass spectrometry in the quantitative analysis of hydrocarbon mixtures containing components inseparable by gas chromatography is described.

THE direct combination of a gas chromatograph with a mass spectrometer has been successfully used for several years as a tool for qualitative analysis of complex mixtures of organic compounds.[1-4] Earlier attempts to overcome the difficulties due to different working pressures of the two instruments consisted either in splitting the effluent stream from the chromatograph so that only about one per cent entered the ion source of the mass spectrometer[1] or in the use of capillary columns.[3] The technique has recently been greatly improved by the introduction of "molecule separators" by Ryhage[4] and by Watson and Biemann.[5] These devices preferentially remove the bulk of the carrier gas (He) at the outlet of the gas chromatograph, thus allowing a larger proportion of the sample to be introduced into the ion source. The question arises as to whether the total ion current records (gas–liquid–chromatograms) of instruments equipped with separators can be used for quantitative analysis in the conventional manner, as the principal of operation undoubtedly results in differential losses of samples of varying molecular weights in the pumping lines of the separators. This problem would appear to be only of academic interest in the majority of cases as quantitative analyses can easily be performed by re-running the mixture on a gas–liquid chromatograph equipped with a conventional detector. However, this method generally requires a good separation of the individual components of the mixture.

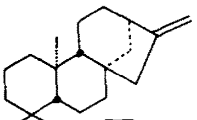
During a recent investigation of the acid-catalysed rearrangements of various diterpene hydrocarbons[6] it was found that isokaurene (I), isoatisirene (II), kaurene (III) and atisirene (IV) often occurred together as products of the reaction. Of these the pairs I and II and III and IV could not be resolved by GLC even on a diverse range of stationary phases. Thus quantitative analysis of such product mixtures was impossible by conventional methods. Lindeman and Annis,[7] using the older effluent splitting technique of gas chromatography-mass spectrometry have shown that quantitative analyses of unresolved chromatograms may be obtained by making use of mass spectral sensitivity data. No such data are available for the compounds mentioned above and as the mass spectrometer used was equipped at the time only with a gas-chromatographic inlet system it would have been impossible to measure relative sensitivities in the usual manner.[8] However, as was first shown by Otvos and Stevenson,[9] hydrocarbon isomers on electron bombardment in the mass spectro-



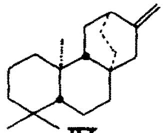
I



II



III



IV

meter ion source produce the same total ion current (ΣI) per mole of sample. This can be used in the analysis of a mixture of isoatisirene and isokaurene, e.g. as follows:

In the mass spectra of isokaurene and isoatisirene (Fig. 1) suitable peaks for the quantitative analysis of a mixture of the two[10] occur at m/e 123 and m/e 137. These have the further advantage that, being close together in m/e value, their relative heights are unaffected by pressure changes in the ion source during the mass scan—an unavoidable occurrence if several mass spectra have to be recorded during the elution time of a gas–liquid chromatograph peak. m/e 123 is 2·15% of ΣI for isokaurene and is 0·90% ΣI for isoatisirene. This means that in the mass spectrum of a mixture of equal parts of isokaurene and isoatisirene the relative contributions to the mixture peak height at m/e 123 are 2·15 and 0·90 respectively, i.e.

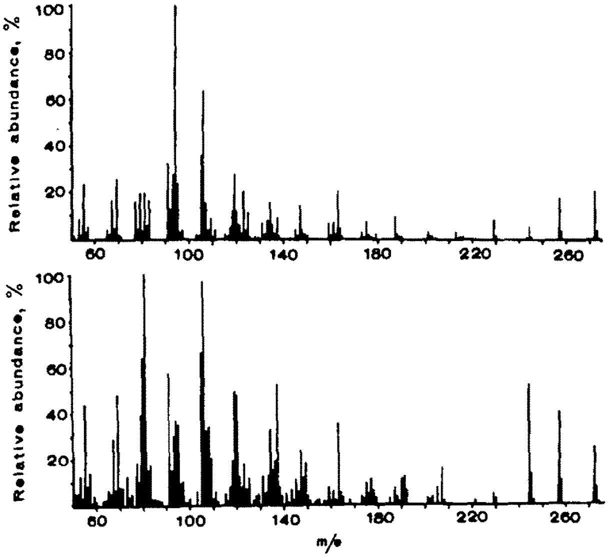


FIG. 1 Mass spectra of isokaurene (upper trace) and isoatisirene.

$$H_{123} = 2{\cdot}15C_{K} + 0{\cdot}90C_{A} \tag{1}$$

where H_{123} is the mixture peak height at *m/e* 123 (measured in arbitrary units) and C_K and C_A are the respective relative concentrations of isokaurene and isoatisirene in the mixture. Similarly at *m/e* 137

$$H_{137} = 0{\cdot}95C_K + 2{\cdot}63C_A \tag{2}$$

C_K and C_A can then be found by solution of Eqns 1 and 2. Isokaurene and isoatisirene are, however, partially separated on GLC and a single mass spectrum taken during the elution of the mixture will give only the concentration at the time of scanning. This may be overcome by recording a large number of spectra during the elution of the GLC peak, finding C_K and C_A for each spectrum and plotting the percentage of each constituent in the mixture on the appropriate ordinate of the total ion current record. Drawing a smooth curve through each of the two sets of points results in a "derived" GLC peak for each component of the mixture from whose areas the overall composition can be obtained (Fig. 2).

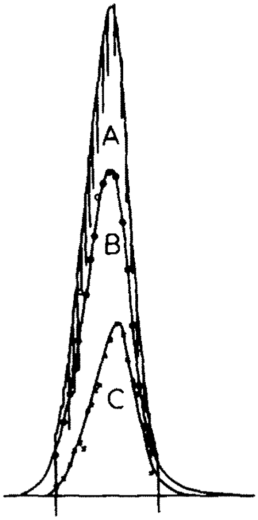


FIG. 2 A. Total ion current peak for mixture of isokaurene and isoatisirene. Vertical lines are caused by deflexion of the recorder pen during the mass scan thus giving an accurate record of the times of scanning.
B and C. Replotted peaks for isoatisirene and isokaurene, respectively.

Solution of many sets of simultaneous equations is tedious and may be avoided as follows:

From Eqns 1 and 2, in an equimolar mixture of isokaurene and isoatisirene the ratio of the peak heights at *m/e* 137 and *m/e* 123 is given by

$$\frac{H_{137}}{H_{123}} = \frac{0{\cdot}95 + 2{\cdot}63}{2{\cdot}15 + 0{\cdot}90}$$

Similarly for a mixture containing 75% of isokaurene

$$\frac{H_{137}}{H_{123}} = \frac{3 \times 0{\cdot}95 + 2{\cdot}63}{3 \times 2{\cdot}15 + 0{\cdot}90}$$

A few such simple calculations enables the graph of Fig. 3 to be drawn. If H_{137}/H_{123} in any mixture spectrum is measured then the composition can be read directly from the graph.

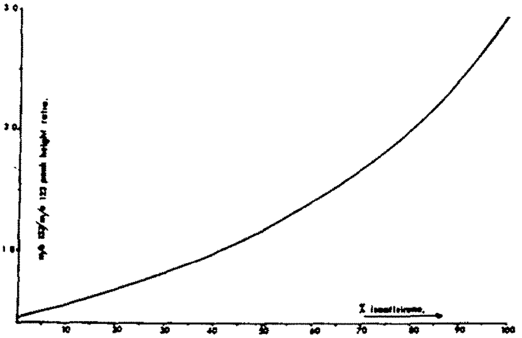


FIG. 3 Calculated ratio of peak heights at *m/e* 137 and *m/e* 123 versus composition for mixtures of isokaurene and isoatisirene.

A typical result is illustrated in Fig. 2 which was obtained from a synthetic mixture of isokaurene and isoatisirene containing 31·3% of the former. The method described gave the composition as 32·7% isokaurene.

A similar approach to the problem of determining unresolved gas–liquid chromatograph peaks has recently been described by Sweeley *et al.*[11] These authors use a modified mass spectrometer which enables the changing intensity of ion peaks characteristic of each component to be monitored. This has the advantage that the greater sensitivity of the electron multiplier detector is used and subsequent data handling is simplified. However the difficulty in tuning the instrument to record particular *m/e* values prior to the analysis and the fact that further modification is required to deal with mixtures of more than two components is pointed out. The method here described requires no modification of the mass spectrometer and is not restricted in the complexity of the mixture which can be analysed, provided reference spectra of the constituents can be obtained. A further advantage of recording spectra over a short range of *m/e* values is that other mass peaks are then available as a check on the analysis, e.g. *m/e* 81 and *m/e* 94 in the mass spectra of isokaurene and isoatisirene (Fig. 1).

From the results obtained in this investigation together with those reported in the earlier communication[11] it can be concluded that the modern gas chromatograph-mass spectrometer equipped with a carrier gas separator can provide useful quantitative as well as qualitative data. By a combination of this technique and conventional GLC the qualitative and quantitative analysis of small amounts of complex mixtures can be carried out.[6]

EXPERIMENTAL

The instrument used was an L.K.B.9000-A gas chromatograph-mass spectrometer (LKB Produkter AB, Stockholm). Total ion current records were recorded at 20 eV to avoid ionization of the He carrier gas. Mass spectra were recorded at 70 eV (The instrument incorporates an automatic switch from one eV setting to the other on starting the mass scan.) Reference spectra were obtained from compounds carefully purified by GLC and TLC on Ag^+/silica.[6] Samples were introduced via a 10 ft. × $\frac{1}{8}$ in. i.d. glass column containing S.E.30(1 %) on Gas Chrom P (Applied Science Laboratories, Inc., State College, Pa). Plotting of the data on the total ion current record is facilitated if broad GLC peaks are produced. Suitable results were obtained by using a column temperature of 120° and a paper speed of 15 in./hr for the total ion current recorder.

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