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Commission on Pesticide Residue Analysis

Working Paper On
Mass Spectrometry of Organo Chlorine Compounds
and
Attempts to reach 'Positive Identification'

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by

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Sirs:

In view of the well-known limitations of ECD, TID, FID and MCD decectors[x] now in use in the gas-chromatographic analysis of pesticide residues, our research group at the Institute of Analytical Chemistry, University of Stockholm, is investigating various alternatives.

We have specified the requirements of this new detector as follows:

1. It should achieve the same sensitivity, or better, than does the ECD for chlorinated pesticides. In general, 100 pg of sample injected into the gas chromatograph should be detectable.
2. This sensitivity should be applicable to all types of compounds which can be analysed by gas chromatography.
3. This general sensitivity should also be selective in an adjustable way, so as to discriminate between the major and minor components of the sample mixture, and to aid peak identification.

A detector which fulfills the above requirements may be too advanced, and hence expensive, to replace those now used in general pesticide-residue analysis; simple detectors, such as the ECD, will long be used, particularly for routine and quantitative work of this kind. On the other hand, improved GC-detection systems are needed in research and development laboratories to complement the less versatile instruments generally available. To illustrate the need for research on new detectors one may cite the reluctance of residue analysts to consider currently-available analytical proofs as legal evidence.

Our first step in developing the new system was to draw up the flow chart shown in Figure 1 (page 2). Exhaustive methods of cleaning up and of separation were considered, so that in conjunction with simple GC detectors a known pesticide of a given chemical class could be identified. However, although work along these lines has led to some new and useful methods of confirmation, essentially-chemical methods cannot provide a general solution to the problem of positive identification.

As Fig. 1 clearly indicates, of the instrumental identification methods available at present, only mass spectrometry (MS) provides sensitivity comparable to that of the ECD. Although aromatics are detectable by MS at much lower levels than most aliphatics, the differences in MS response factors are not as drastic as those of the ECD. The mass spectrometer may

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x
ECD, TID, FID and MCD are respectively, the electron capture, thermionic, flame ionisation, and micro coulometric detector.

FIGURE 1.

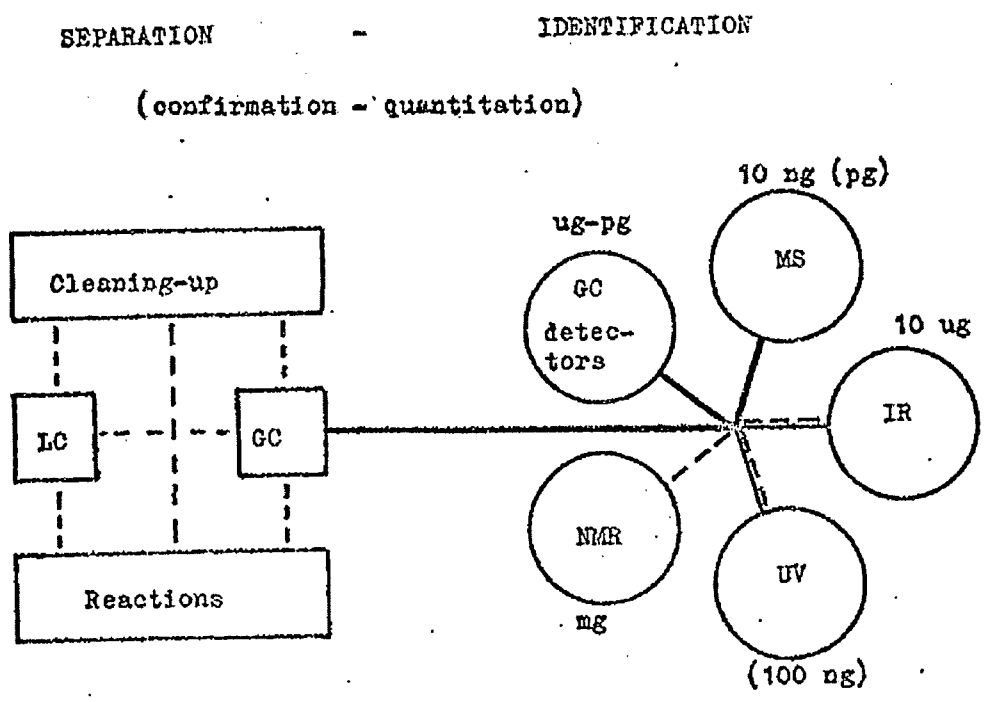

Figure 1. Flow chart of separation and identification processes in pesticide-residue analysis. Full line indicates that on-line combinations are available (thick line indicated that they are in common use); dotted line, manipulations.

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be classified as a general detector.

By using a mass spectrometer focused at a certain mass number as a GC detector, while interposing slow-acting filters to reduce the noise level, 100 pg of eluant is generally detectable, and in special cases this amount may be very much less (e.g. 1 pg of toluene). The use of two or more mass numbers is a move in the direction of positive identification. This technique requires alternation (at 1 - 10 cps) between the selected mass numbers so that, unfortunately, slow-acting filters must be excluded. Sensitivity is hence reduced, but the method undoubtedly affords an elegant means of overcoming certain gas chromatographic limitations, as shown by Sweeley, Elliot, Fries and Ryhage (1); see also Schomburg and Henneberg (2). In the form of 'mass fragmentography' the refinement has been used by Hammar, Holmstedt and Ryhage (3) to determine drug metabolites in human blood.

The most likely mass-spectrometric method of identifying the compound which produced a given GC peak is a recording of the entire spectrum. However, scanning mass spectrometry usually gives more information than is conveniently handled, and a correct recording of intensities requires either slow scanning or amounts of material not always available in pesticide analysis. The latter limitation is illustrated in Figure 2 (page 4); samples of lindane, respectively 1 ug and 25 ng, were injected into a combination GC/MS instrument (LKB-9000). Maximum and minimum intensities from ten scanning mass spectra are marked, and show that for the 25 ng injection the variation is too great to allow correct recognition of characteristic isotope distribution patterns.

Figure 2 indicates that in the latter case too few ions are reaching the detector. This assumption is in accord with a report of McFadden and Day (4); see also Littlewood (5). Thus, repetitive scanning of a small part of the mass spectrum is required to separate the true isotope 'fingerprint' from the background noise. At our Institute, we have recently coupled to our mass spectrometer a novel recording system, consisting mainly of a multi-channel analyser (Intertechnique DIDAC-4000) which allows on-line digitized recording, preferably while at the maximum of a GC peak. We can now scan repetitively up to ten mass units at a rate of up to ten sweeps per second. Alternatively, using a mass marker, we can repetitively record the whole mass spectrum. The instrumentation also allows subtraction of the bleeding, and then normalisation, and print out in digitized or plotted form.

Figures 3-6 (page 5) illustrate an application of this apparatus which is particularly pertinent to our pollution studies. The isotopic pattern for the

FIGURE 2.

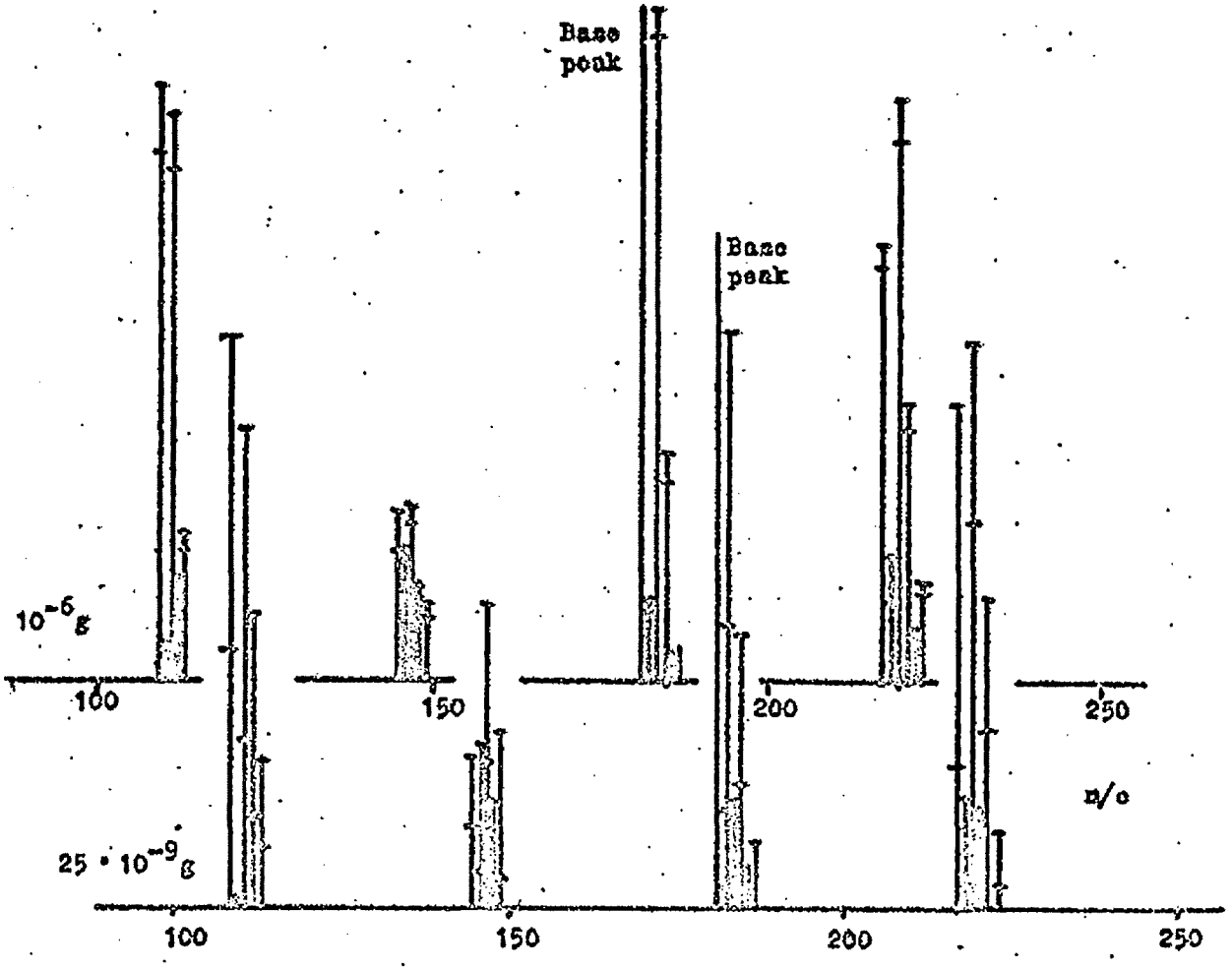

Figure 2.
Mass spectra of 10^{-6} gm (upper trace) and 25 x 10^{-9} gm (lower) of lindane injected into gas chromatograph. All spectra have been normalized to base peak. Horizontal bars represent max mum and minimum values from ten recordings of each spectrum

GC/MS Analysis of tetraethyl lead.

Spectra recorded when using multi-channel analyser (DIDAC-800, Intertechnique) on-line to GC/MS (LKB-9000).

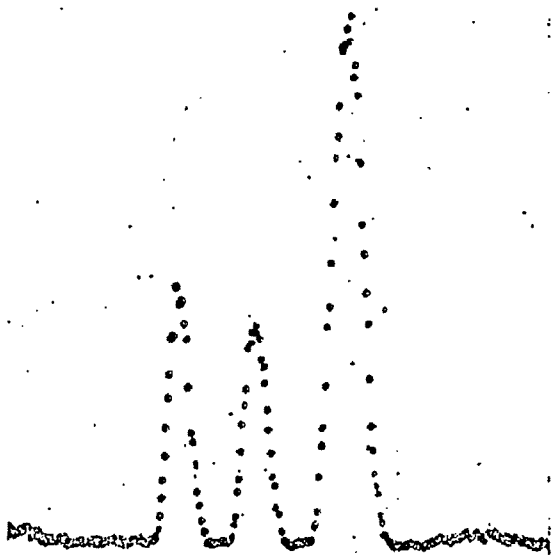

Figure 3. Mass spectrum in m/e range 235-237 ($PbEt^+$ ion) from 28 x 10^{-9} gm injection of tetraethyl lead into gas chromatograph (LKB-9000); 30 sweeps at 2 sweeps/sec.

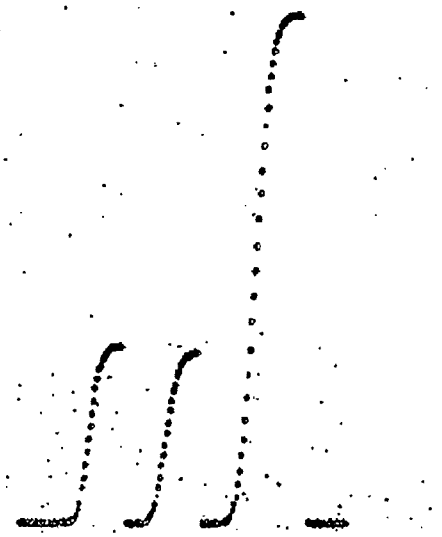

Figure 4. Integrated version of pattern shown in Figure 3.

Figure 5. Mass spectrum in m/e range 235-237 from 0.3 x 10^{-9} gm injection of tetraethyl lead; 30 sweeps at 2 sweeps/sec.

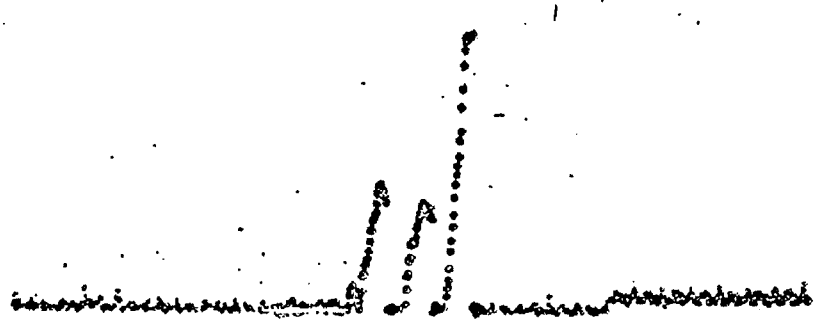

Figure 6. Integrated version of trace in Figure 5.

$PbEt^+$ ion can be correctly recognised from only 2$\cdot$ ng of tetraethyl lead (Figure 3 - normal spectrum; Figure 4 - integrated spectrum). Even if the weight of tetraethyl lead injected into the gas chromatograph is reduced by two orders of magnitude to 0.3 ng, an almost 'true' isotopic pattern, while invisible in the normal spectrum (Figure 5), is reproduced in the integrated version (Figure 6).

Although these improved methods of the recording of isotopic patterns are undoubtedly of great value as an aid to the chemist in the recognition of compounds present in pesticide residues, the rationale of the method can be criticised. It is possible that a wholly fortuitous combination of mass spectral peaks could give rise to the pattern then assumed to be the result of a certain isotope distribution. One could argue, for example, that a pattern even as specific as that of the CH_3Hg^+ ion could arise by chance. Further proof of compound identity, sufficient in fact for legal purposes, but still not absolute, may be provided if some more characteristic patterns deriving from the same compound are found in the same mass spectrum.

Further proof of identity by MS in residue analysis can be provided by making use of atomic mass defects; for this purpose a precision of the order of 0.001 mass units is required in the measurement of mass number. In fact, modern high-resolution mass spectrometers usually allow more accurate mass-number measurement than is required for the determination of the elementary composition of the ions, but need more sample than is often available in residue analysis. Accuracy just sufficient to allow calculation of elementary composition may, however, be obtained by the use of peak-matching methods at medium resolution, with concomitant smaller sample size. Although a growing need is evident, not only among residue analysts, for methods of rapid elementary analysis of fractions eluted from a gas chromatograph, neither of the above techniques can yet be readily used during the short time available as the compound emerges from the column.

If mass-peak positions are to be measured with sufficient accuracy at medium resolution, the Gaussian shape of the recorded traces must not be distorted. Since simple repetitive scanning - see Figure 7 - very much improves the recorded shapes of mass-peaks, we think that facilities for "instant peak-matching' will soon be commercially available.

Even if such instruments gave only several alternative combinations for an elementary composition, it may be that all but one of these could

FIGURE 7

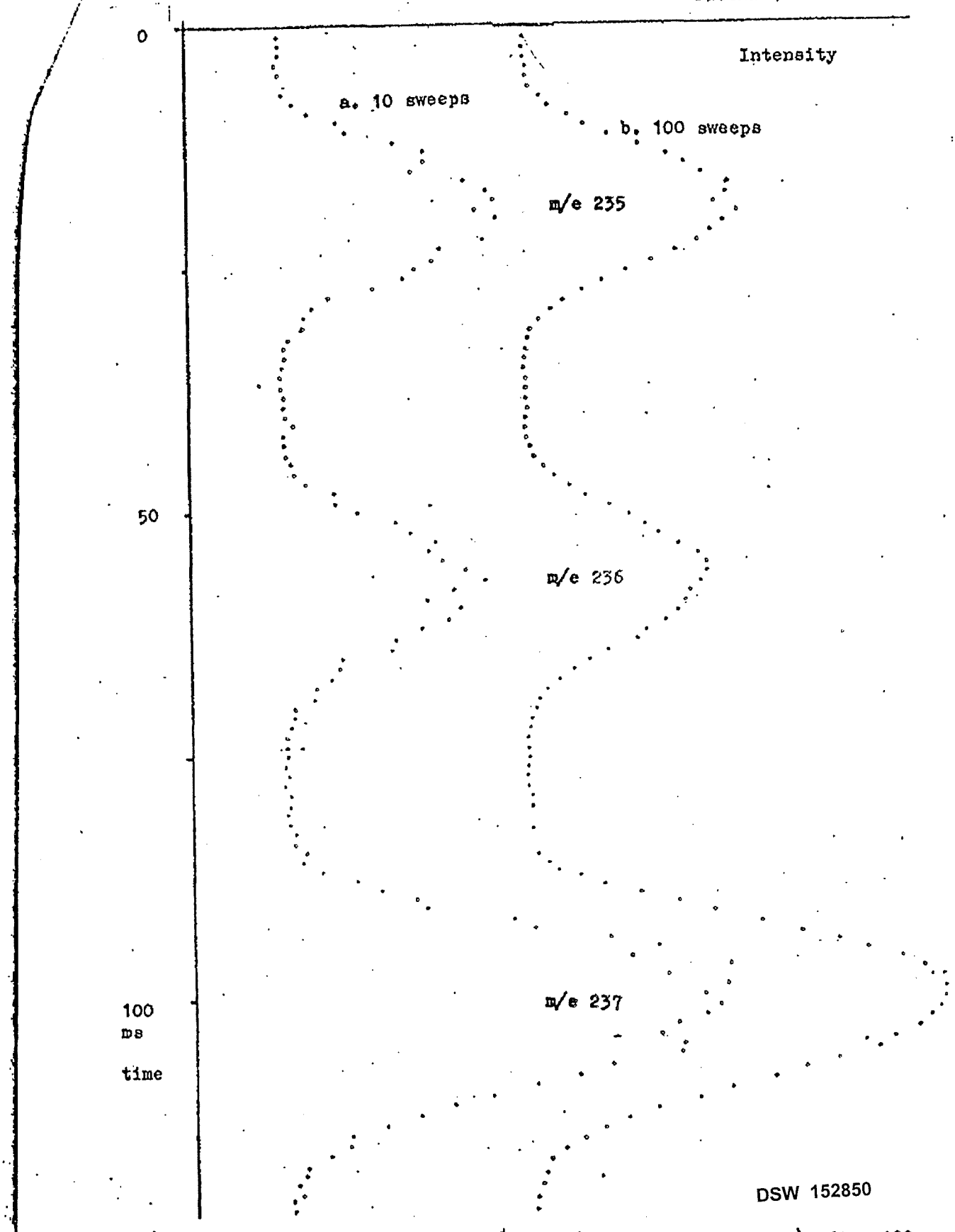

Figure 7. Mass spectrum of $PbEt^+$ ion a) after 10 sweeps, b) after 100 sweeps. Medium resolution mass spectrometer (IKB-9000) used.

be eliminated from a consideration of other available information.

Data which lead to the determination of elementary composition come under the heading of positive information, but do not, of course, allow identification of a given compound. At the same time, a knowledge of the molecular formula provides the desired 'platform' for further work, which begins in the library. Moreover, this approach conforms to that of 'classical' organic chemistry in organizing the large amount of data which may be obtained in the future from more refined mass spectrometry linked with more sensitive versions of other identification methods as listed in Figure 1.

Finally, in spite of our title, it is perhaps worthwhile to ask whether in the general sense, 'positive identification' is ever possible in analytical chemistry.

References

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(5) Littlewood, A.B., Chromatographia 1, 37 (1968).