CHROM. 5614

Minor components of cannabis resin

I. Their separation by gas chromatography, thermal stability, and protolytic properties

Gas chromatography (GC) was introduced for analysis of cannabis resin in the early 1960's (see ref. 1 and the earlier literature cited in it). To-day it is known that the composition of cannabis resin is very complex. Some twenty components have been described including cannabidiol (CBD), tetrahydrocannabinol (THC), cannabinol (CBN) and their carboxylic derivatives. The present GC method was developed both for forensic purposes and for mass spectrometric analysis, which is now in process of development. In this study, the method was applied to two materials, of the hashish and marihuana type, respectively. Some of the protolytic properties were studied for about forty minor components of the hashish material, while for the marihuana material, the behaviour of some fifty minor components under smoking conditions was investigated.

Experimental

Apparatus. The gas chromatograph used was a Perkin-Elmer F 11 with a No. 4 analyser unit (all-glass dual-channel system), linear temperature programmer, and flow control unit. The recorder used was a Perkin-Elmer 165. The GC conditions were as follows:

Columns: O.D. 6 mm (0.25 in.), I.D. 2-mm glass tubes of 1.9-m length and 130-mm coil diameter, packed with Gas-Chrom Q (80–100 mesh), coated with 3 % OV-17.

Carrier flow: 30 ml of nitrogen per minute.

Temperatures: injector, 275°; column, initially 100° and finally 300°.

Programme: minutes 0–2, isothermal (100°); 2–18, 8°/min; 18–30, isothermal (228°); 30–39, 8°/min; 39–45, isothermal (300°).

Hydrogen inlet pressure: 1.3 atm.

Air inlet pressure: 2.0 atm.

Chart speed: 5 mm/min.

Recorder sensitivity: 1 mV f.s.d.

Procedure. The marihuana material (cut flowers and leaves) had been seized by the police and was of unknown origin. According to GC analysis (not shown in this report) it contained 0.5 % CBD, 3 % Δ^1-THC, and 0.5 % CBN. For the purpose of chromatography two cigarettes were rolled from the material. One was put in a smoking apparatus. The condensate was collected on a glass fibre filter. The filter and the remaining cigarette were extracted in a Soxhlet apparatus with petroleum ether (b.p. 30–60°) for 6 h. After evaporation of the extracts to about 200 μl, 1 μl of each sample was injected into the gas chromatograph (see Figs. 1 and 2).

The seized hashish material consisted of sticky "soles" about 1 cm thick. Its origin was unknown. The contents of CBD, Δ^1-THC, and CBN were found to be 8 %, 3 %, and 3 %, respectively. For the purpose of chromatography 8 g of the hashish

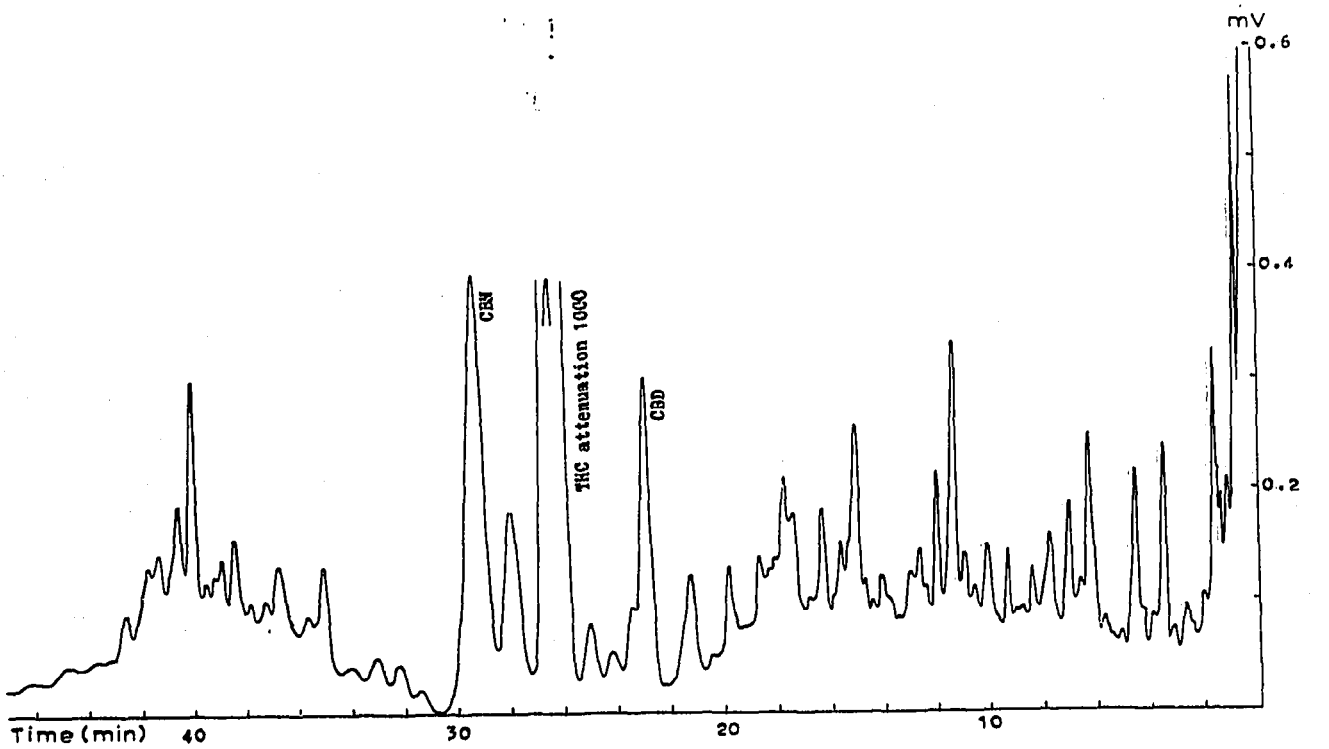

Fig. 1. Gas chromatogram of a petroleum ether extract of a marihuana cigarette. Attenuation × 500.

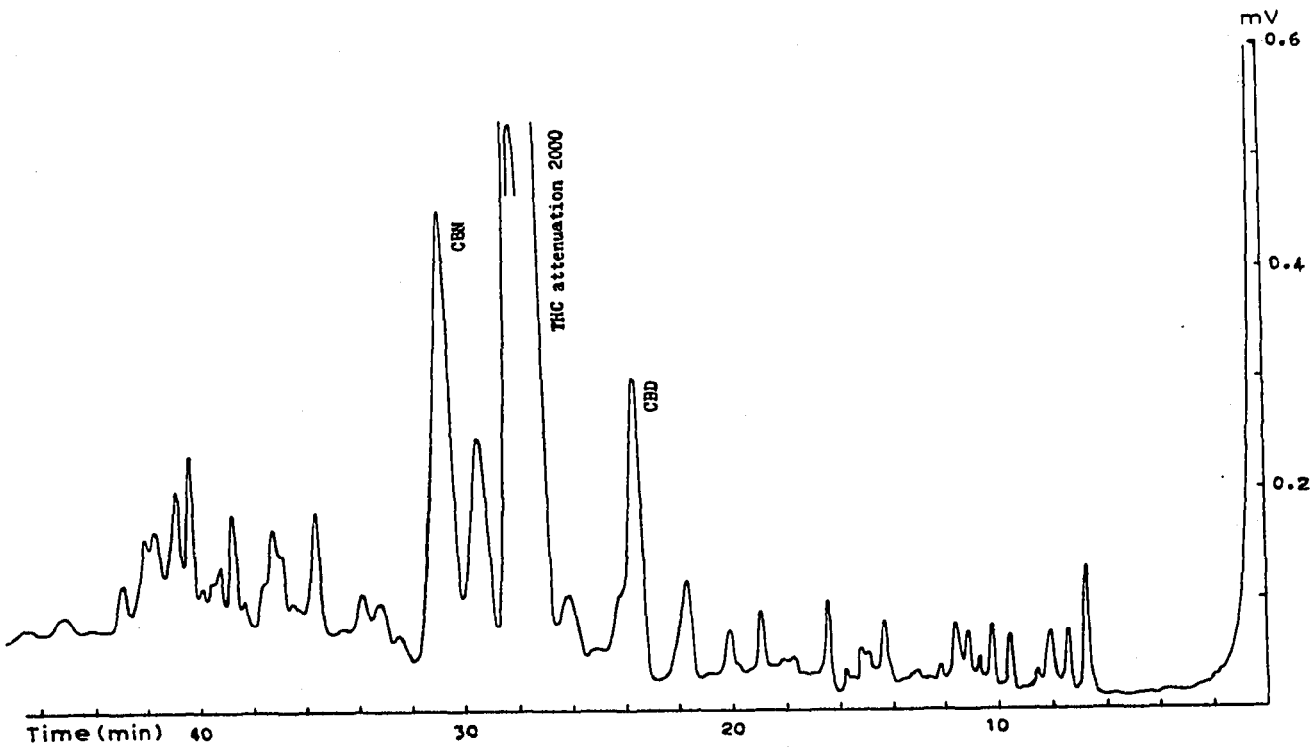

Fig. 2. Gas chromatogram of a petroleum ether extract of a smoke condensate. Attenuation × 200

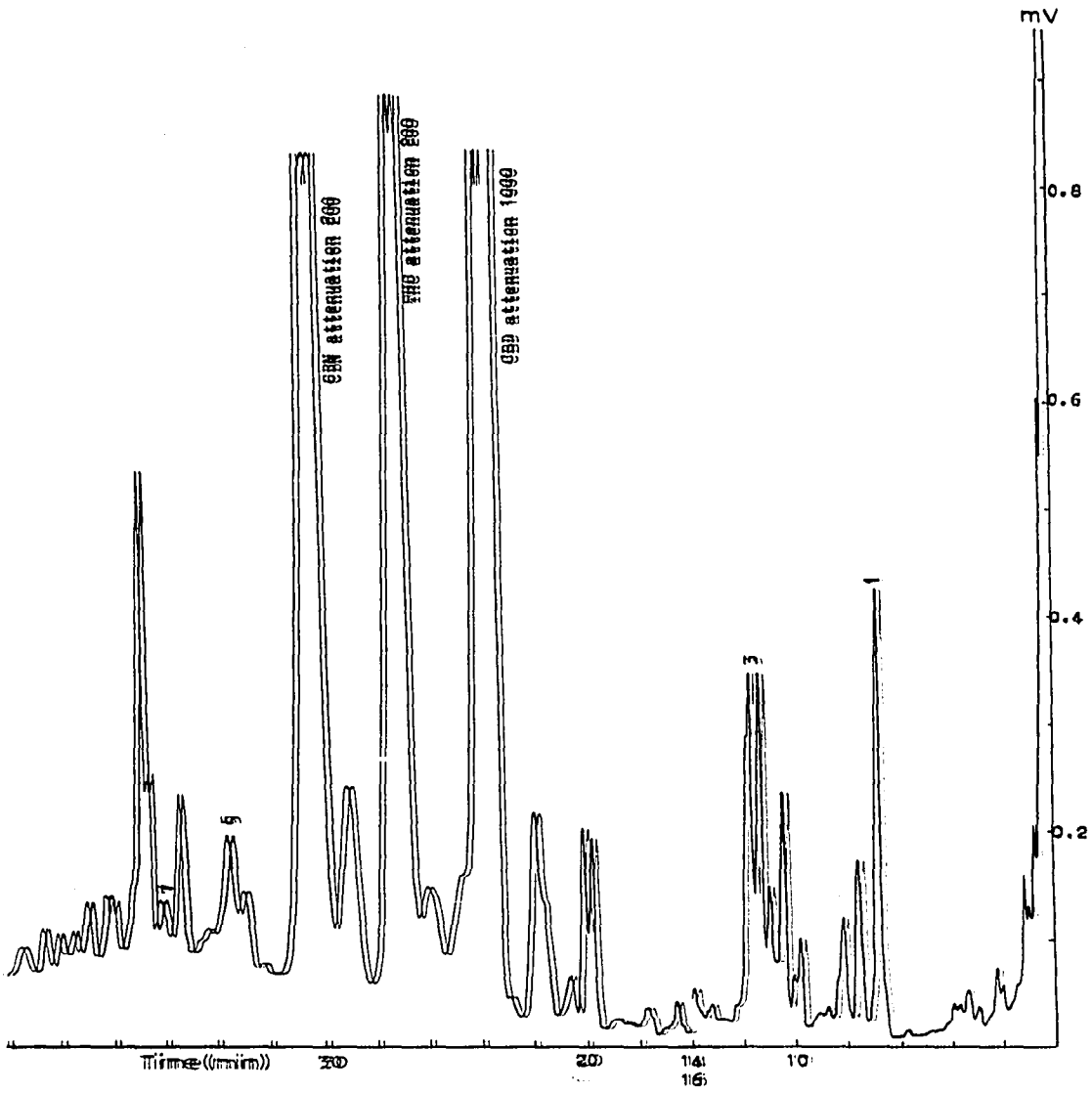

Fig. 3. Gas chromatogram of a methanolic extract of hashish partitioned between petroleum ether and water. 1 μl of organic phase was injected. Attenuation × 100.

were ground to a fine powder and extracted in a Soxhlet apparatus with methanol for 6 h. The extract was made up to 25 ml. Three aliquots of 4 ml were partitioned between 100 ml of petroleum ether (b.p. 30–60°) and 100 ml of an aqueous phase, which was neutral in the first case, consisted of 0.1 *M* potassium hydroxide in the second, and of 0.1 *M* sulphuric acid in the third. A fourth aliquot was added to 20 ml of 0.5 *M* methanolic potassium hydroxide and refluxed for 30 min. It was then diluted with 75 ml of water and extracted with 100 ml of petroleum ether. The four organic layers were evaporated to 2 ml and 1 μl of each was injected into the gas chromatograph (see Figs. 3–6).

Results and discussion

When the hashish extract was chromatographed at varying injection temperatures (300°, 250° and 200°), essentially identical chromatograms were obtained. At

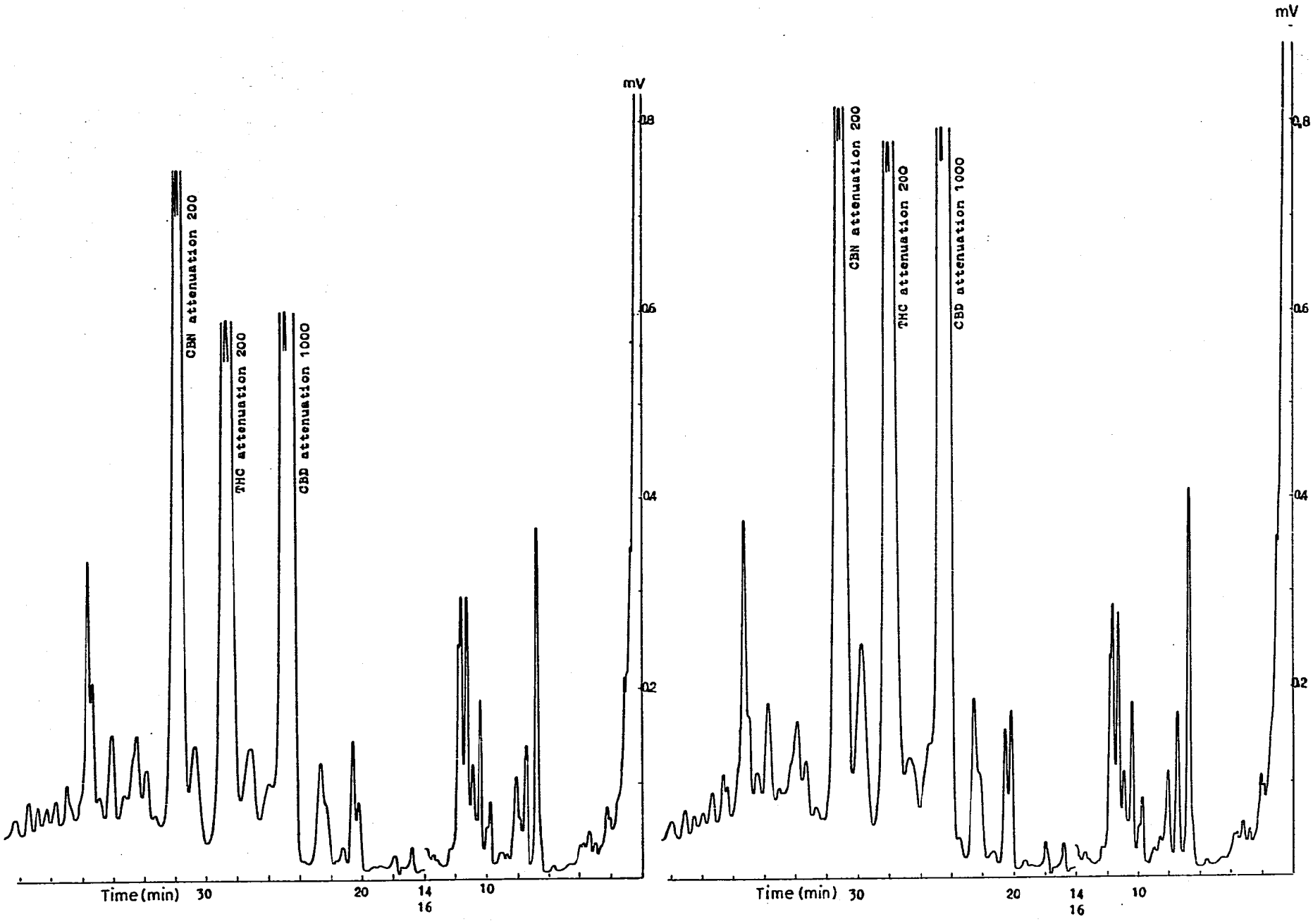

Fig. 4. Gas chromatogram of a methanolic extract of hashish partitioned between petroleum ether and 0.1 *M* aqueous potassium hydroxide. 1 μl of organic phase was injected. Attenuation × 100.

Fig. 5. Gas chromatogram of a methanolic extract of hashish partitioned between petroleum ether and 0.1 *M* aqueous sulphuric acid. 1 μl of organic phase was injected. Attenuation × 100.

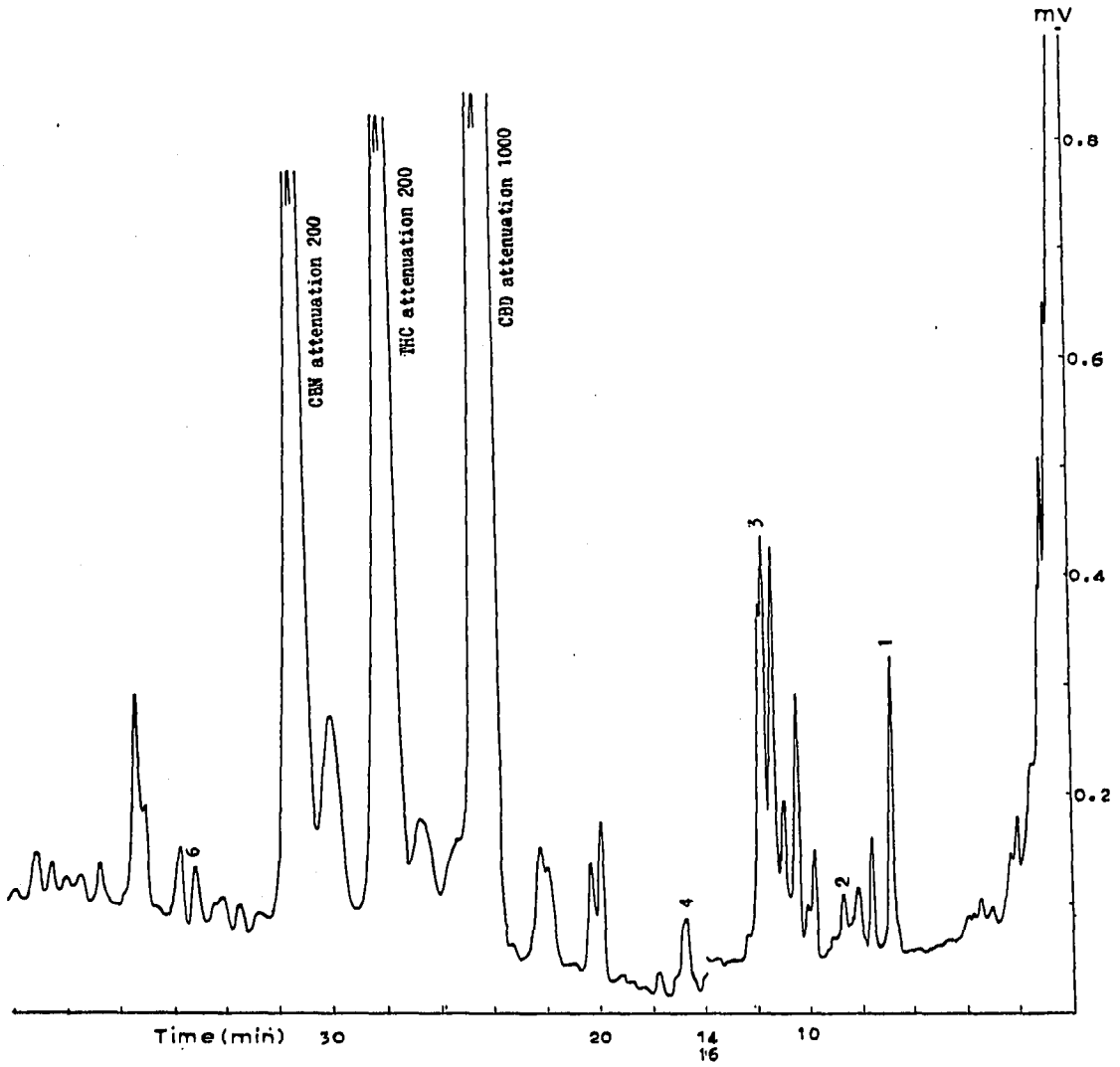

Fig. 6. Gas chromatogram of a methanolic extract of hashish treated with 0.5 *M* methanolic potassium hydroxide and partitioned between petroleum ether and 0.1 *M* aqueous potassium hydroxide. 1 μl of organic phase was injected. Attenuation × 100.

150°, asymmetric peaks did not appear until CBN was eluted. These results indicate that the minor components are present in the extract and are not artefacts formed in the injection region of the chromatograph.

The stability of the minor components of the marihuana material is demonstrated by the smoke condensate analysis (Fig. 2). As compared with Fig. 1 (the gas chromatogram of the unsmoked marihuana extract), the following effects are observed: The heavier part (including the cannabinoids, eluted between 18 and 45 min) remains essentially unchanged. Some of the lighter components seem to have been partially decomposed, giving some twenty new components, probably representing cracking products.

Figs. 3–5 show only slight variations of peak heights with varying pH conditions, indicating the absence of pronounced acidic or basic volatile components in the extract. However, a comparison of Figs. 3 and 6 shows some more pronounced differences, such as the appearance of peaks 2, 4 and 6 (Fig. 6), the decrease of peaks 5

and 7 (Fig. 3), and the inverted ratio of intensities of peaks 1 and 3. In the 14- to 16-min interval ghost peaks appear, probably due to column bleeding. (These are deleted in Figs. 3–6.)

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The National Laboratory of Forensic Science LARS ERIK STRÖMBERG
(*Statens Kriminaltekniska Laboratorium*),
Fack, 171 20 Solna 1 (*Sweden*)

1 C. G. FARMILO AND T. W. M. DAVIS, *J. Pharm. Pharmacol.*, 13 (1961) 767.

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