CHROM. 5510

Gas chromatography of ergot alkaloids

The use of gas chromatographic (GC) procedures for the specific and sensitive analysis of drugs and biological amines has increased rapidly and comprehensive methods are available, *e.g.* for the forensic identification of narcotic drugs (*e.g.* refs. 1 and 2). Relatively few reports have appeared concerning the GC separation of alkaloids but have included peyote[3,4] and *Amaryllidaceae*[5] alkaloids. GC methods have also been published for some groups of indole alkaloids such as simple tryptamines[6,7] and *Strychnos*[8] and *Voacanga*[9] alkaloids. Although thin-layer chromatography (TLC) has been used extensively (*e.g.* refs. 10 and 11) for the separation and identification of ergot alkaloids, GC has been applied only to the detection and identification of lysergic acid diethylamide (LSD)[12,13].

The main objective of the present study was to test the applicability of GC for the identification of ergot alkaloids, particularly using the combination GC–mass spectrometry.

Experimental

All separations were carried out isothermally using a Varian Aerograph Model 204 (FID) or Model 2100 (FID) gas chromatograph. The column supports, 100–120-mesh Gas-Chrom P or Gas-Chrom Q, were acid-washed and silanized. The stationary phases used and other conditions are given in Figs. 1 and 2. Glass columns, 6 ft. × 1/8 in., were used. Nitrogen flow rate was 25 ml/min. Flash heater and detector temperatures were maintained 20–30° above the column temperature.

Results and discussion

The GC results are given in Fig. 1.

The ergot alkaloids consist of about forty ergoline derivatives and are commonly divided into two groups. One group is a series of simpler ergoline derivatives, the clavine alkaloids, and the other group may be considered as amides of lysergic acid. LSD does not occur in nature but is structurally very closely related to the simple lysergic acid amides. The clavine alkaloids (general structure, Fig. 1) have low molecular weights (mol. wt. 238–260; fumigaclavine A, mol. wt. 299), whereas the amide-type alkaloids have such high molecular weights that only the simpler amides such as lysergic acid amide, lysergic acid methyl carbinolamide, LSD (mol. wt. 323) and ergometrine might be expected to pass a GC column.

All clavine alkaloids (compounds 1–20, Fig. 1) could be chromatographed with satisfactory results on JxR and SE-30 columns and most satisfactory also on XE-60 (Figs. 1 and 2). The peaks were generally acceptable (Fig. 2) but the alkaloids containing hydroxyl groups showed limited tailing. Attempts to overcome this by the formation of suitable derivatives (trimethylsilyl, heptafluorobutyryl or trifluoroacetyl) according to different procedures yielded no adequate results with extracted alkaloid mixtures.

The JxR and SE-30 columns which separate primarily on the basis of molecular weight gave similar results. The JxR column resolved the clavine alkaloids into groups (Fig. 1): firstly compounds with a C-17 methyl group and no exocyclic double

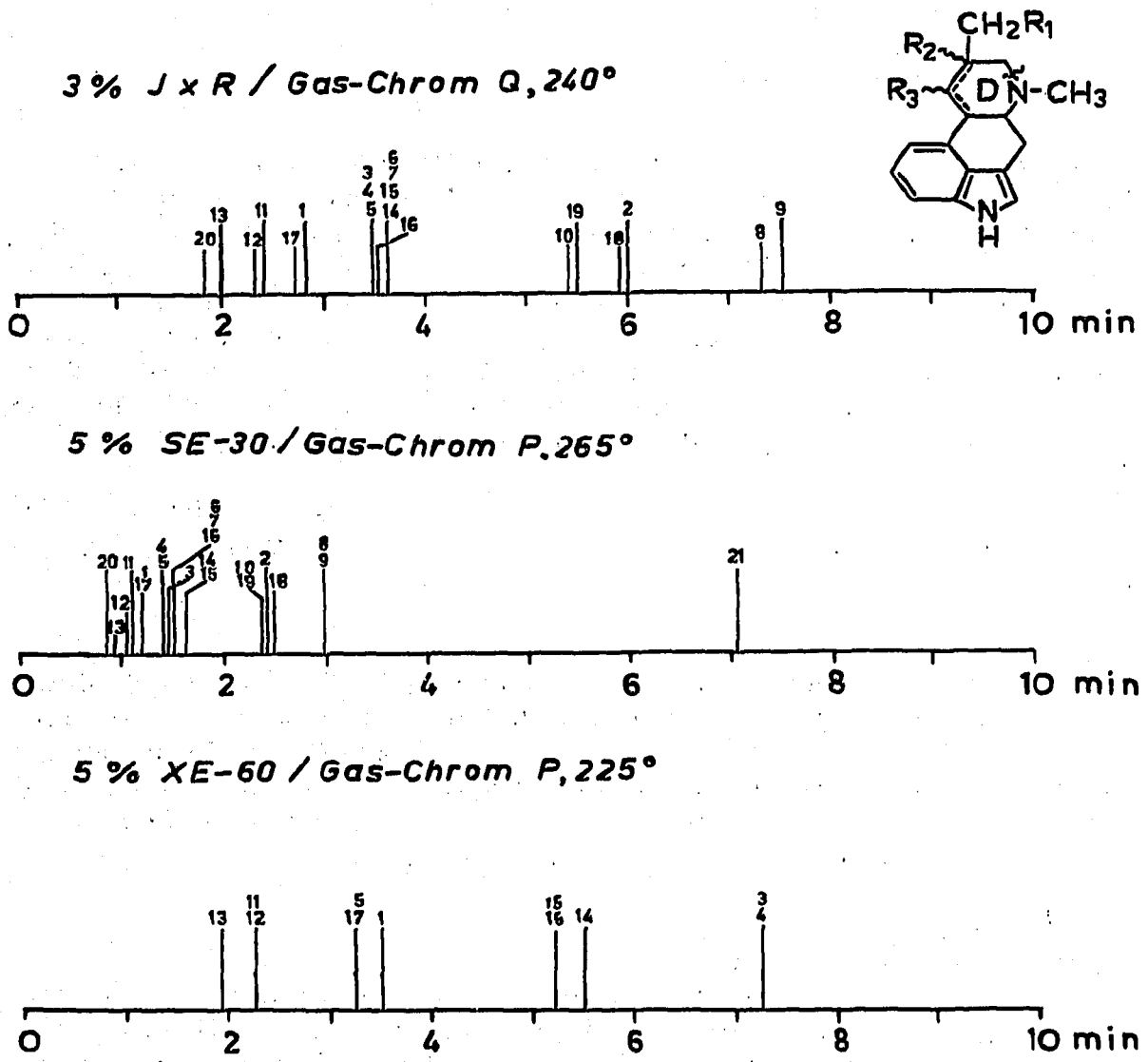

Fig. 1. General structure of clavine alkaloids. Retention times of reference compounds on 3% JxR, 5% SE-30 and 5% XE-60 columns. Numbers refer to the following compounds: 1 = agroclavine, 2 = elymoclavine, 3 = chanoclavine-I, 4 = chanoclavine-II, 5 = isochanoclavine-I, 6 = setoclavine, 7 = isosetoclavine, 8 = penniclavine, 9 = isopenniclavine, 10 = α-dihydrolysergol, 11 = festuclavine, 12 = pyroclavine, 13 = costaclavine, 14 = fumigaclavine B, 15 = fumigaclavine A, 16 = lysergene, 17 = lysergine, 18 = lysergole, 19 = isolysergole, 20 = cycloclavine, 21 = LSD. For formulae, see refs. 15–17.

bond (compounds 20, 13, 12 and 11) followed by those with a double bond (compounds 1 and 17); the next group (compounds 3–5) consisted of secondary amines with an open D-ring; then compounds with a C-17 methyl group and a secondary (compound 14) or tertiary (compounds 6, 7) hydroxyl function. Compounds having a C-17 hydroxymethyl group (compounds 10, 19, 18 and 2) had similar retention times and compounds with both a primary and a tertiary hydroxyl group (compounds 8 and 9) had the longest retention times of the clavine alkaloids.

The more polar XE-60 column showed no advantage over the previous columns except that the secondary amines chanoclavine-I and -II (compounds 3 and 4), as expected, showed increased retention times. The stereoisomer isochanoclavine-I had

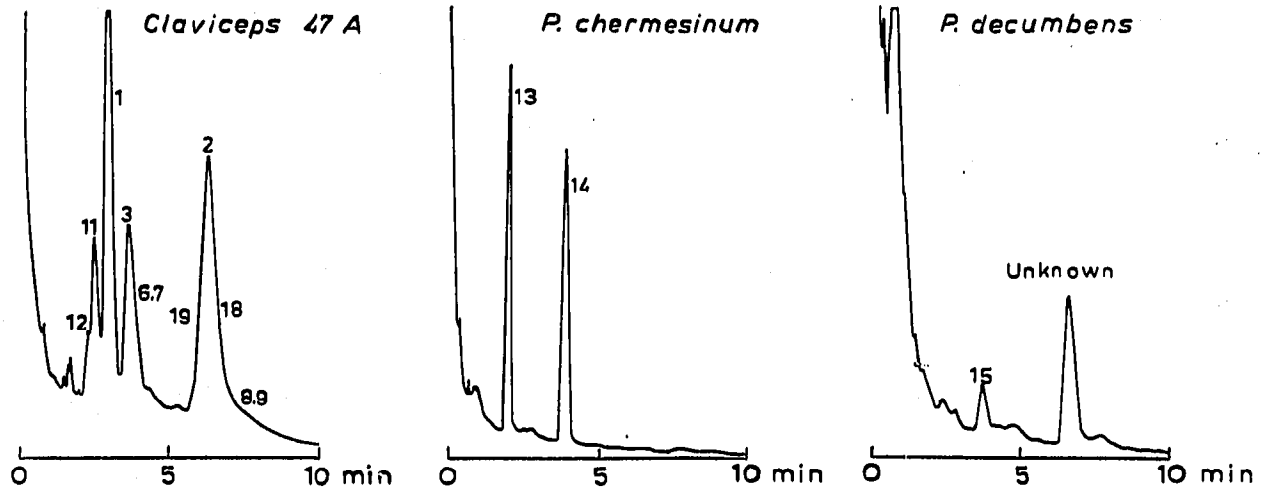

Fig. 2. Separation of ergot alkaloids from *Claviceps* strain 47A[14], *Penicillium chermesinum*[14] and *Penicillium decumbens* on 5% SE-30/Gas-Chrom P at 225°. Numbers refer to compounds as in the legend to Fig. 1.

a comparatively short retention time but this can be contributed to its stereochemistry. Dihydroxylic compounds (compounds 8 and 9) chromatographed poorly.

OV-17 and OV-1 columns can also be used to separate ergot alkaloids.

In contrast to TLC (ref. 10), none of the columns was able to separate stereoisomers.

Of the lysergic acid derivatives, only LSD, lysergic acid amide and lysergic acid methyl carbinolamide—the last after pyrolysis in the injector to lysergic acid amide—could be chromatographed on SE-30 and JxR columns and at comparatively high temperatures. Thus, lysergic acid amide had a retention time of 14.7 min on 3% JxR at 240° (Fig. 1).

In this connection, it may be mentioned that the hallucinogenic indole psilocin is also chromatographed easily on JxR and SE-30 and that its phosphorylated analogue, psilocybin, readily hydrolyzes in methanolic solution in the injector to yield psilocin.

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