ml^{-1}. By concentrating the final hexane extract it was possible to readily detect 0·1 $\mu g\ ml^{-1}$ in plasma.

Phenylbutazone was well absorbed after topical application in an ointment containing DMSO (Table 1). In all plasma samples, the drug could be measured readily. The half-life in Wistar rats is 6 h (Burns & others, 1953). Since the concentration of phenylbutazone in plasma from the rats increased during the 3 h of the experiment this would indicate that absorption of the drug occurred throughout the 3 h. From these results the possibility of using ointments containing phenylbutazone for the treatment of localized musculoskeletal lesions deserves further investigation.

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Identification of hexadecanamide in cannabis resin

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More than three hundred non-cannabinoid compounds have been identified in various parts of the cannabis plant or in cannabis resin, essential oil or smoke. These compounds include alkaloids, alkanes, amines, amino acids, carbohydrates, carboxylic acids, cyclitols, phenols, sterols and terpenes (Slatkin, Doorenbos & others, 1971; Skelton & Witschi, 1974; Burstein, Varanelli & Slade, 1975; El-Feraly & Turner, 1975; Hendriks, Malingré & others, 1975; Jones & Foote, 1975; Lotter, Abraham & others, 1975; Lee, Novotny & Bartle, 1976; Novotny, Lee & others, 1976a,b; Turner, Hsu & others, 1976). Earlier work has been reviewed by Mechoulam, McCallum & Burstein (1976).

We wish to report the identification of hexadecanamide in a sample of cannabis resin thought to be of Pakistan origin. The parent acid, palmitic acid, has previously been identified in cannabis smoke (Fentiman, Foltz & Kinzer, 1973; Jones & Foote, 1975). One other amide, *N*-(*p*-hydroxy-β-phenylethyl)-*p*-hydroxy-*trans*-cinnamamide, and its parent acid, *p*-hydroxycinnamic acid (*p*-coumaric acid; melilotic acid), have been found in cannabis root and leaves respectively (Bate-Smith, 1962; Slatkin & others, 1971).

Hexadecanamide was present in the resin in a trace quantity. Since it has a low ultraviolet absorbance, it was not observed on high-pressure liquid chromatography (h.p.l.c.) of a resin extract (even a moderately concentrated solution of the pure compound could not be detected). It was first noticed on gas-liquid chromatographic (g.c.) examination of a concentrated fraction prepared from a methanol–chloroform (9:1) extract of the resin (0·5 g resin in 1 ml solvent). The extract was fractionated by h.p.l.c. using methods previously described (Smith, 1975). When the fraction from the cannabidiol-cannabinol region of the chromatogram was examined by g.c. on OV-17 at 240°, a peak with a retention of 0·29 relative to Δ^1-tetrahydrocannabinol was detected. This was further examined by gas chromatography–mass spectrometry (g.c.–m.s.). The electron impact spectrum, obtained under conditions described previously (Smith, 1975), had a base peak of 59 and a second most abundant peak of 72, features characteristic of an amide. The molecular weight was found to be 255 by chemical ionization mass spectrometry using a source temperature of 160°, an electron energy of 100 eV, an emission current of 200 μA, an accelerating voltage of 4 kV and isobutane as the reactant gas. The electron impact and chemical ionization spectra corresponded to literature data for hexadecanamide (*Eight Peak Index of Mass Spectra*, 1974) and the identity of the compound was confirmed by synthesizing a sample of hexadecanamide and examining it by g.c.–m.s. The g.c. retention time and the mass spectral data matched those of the compound found in cannabis resin.

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Interference by endogenous *p*-hydroxyphenylacetic acid with estimation of *N*-acetyl-*p*-aminophenol in urine by gas chromatography

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Prescott (1971) described a gas liquid chromatographic method for the assay of phenacetin and its metabolite *N*-acetyl *p*-aminophenol (acetaminophen or paracetamol) in biological fluids after conversion to trimethylsilyl (TMS) derivatives using *N*-trimethylsilyl-imidazole (TMSI) or *N*, *O*-bis(trimethylsilyl) acetamide (BSA). This method was modified by Thomas & Coldwell (1972) by substituting the more powerful silylating reagent bis(trimethylsilyl) trifluoroacetamide (Regisil) and a more selective extractant (diethyl ether). Using this modified method we observed that extracts of normal human urine regularly show a peak with an identical retention time to *N*-acetyl-*p*-aminophenol (NAPA). The peak was considerably accentuated after ingestion of ethanol.

A Hewlett-Packard Model 5750 gas chromatograph with flame ionization detector was used with helium as the carrier gas at a flow rate of 50 ml min^{-1}. Hydrogen and air flow rates were 30 and 300 ml min^{-1} respectively. The glass column 6 ft × 6 mm was packed with 3% OV-1 on 80/100 mesh chromosorb W H.P. and run at 160°. The injector and detector temperatures were 180° and 210° respectively.

Samples of urine and serum were collected before and 6 to 8 h after ethanol ingestion by healthy volunteers. Over 1 h, nine volunteers received 1 litre of beer each, and three received 50 ml ethanol in orange juice. These volunteers had no previous history of ingesting phenacetin, NAPA or any other drug for at least two weeks. Urine or plasma samples were extracted by the method of Thomas & Coldwell (1972), except that extraction was performed twice with diethyl ether. The two extracted ether layers were combined and dried with nitrogen. The residue was dissolved in 50 μl of Regisil (Regis Chemical Co., Chicago, Illinois) and kept at 50° for 30 min. After cooling, 1 μl of the sample was injected for chromatography. The retention times for *p*-bromoacetanilide, phenacetin and NAPA were 3·1, 3·5 and 4·1 min respectively.

Fig. 1 shows a typical gas chromatogram of urine extracts before and after ingestion of ethanol. Peak III with a retention time identical to NAPA, was found in each case when extracts of urine collected before ingestion of ethanol were chromatographed. Analysis of the extracts of urine collected before and after ethanol ingestion by coupled gas-liquid chromatography mass spectrometry (g.l.c.-ms) revealed this peak to be the TMS derivative of *p*-hydroxyphenylacetic acid (*p*-HP AA). Reanalysis of the derivatized extracts after spiking

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