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SHORT COMMUNICATION

Prostaglandins and cannabis—V. Identification of *p*-vinylphenol as a potent inhibitor of prostaglandin synthesis

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A systematic examination of cannabis for substances which can alter prostaglandin (PG) biosynthesis has been undertaken in this laboratory. Thus far, it has been shown that Δ^1tetrahydro-cannabinol (THC) and several other natural cannabinoids are effective inhibitors *in vitro* of PG synthesis [1, 2]. Moreover, an examination of the non-cannabinoid-containing essential oil fraction has revealed the presence of two substances which show appreciable activity in our system *in vitro* [1, 2]. One of these has been isolated and identified as eugenol [3], a substance which was reported to be a cannabis constituent some years ago [4]. We would now like to report on the isolation and identification of the second active volatile component.

The extraction and preliminary fractionation of the cannabis sample were done as described previously [3]; synthetase inhibitory activity was also assayed as before [2] using a bovine seminal vesicle preparation. Briefly this involved steam distillation of a standardized marihuana preparation (National Institute on Drug Abuse) followed by ether extraction of the distillate. Silica gel thin-layer chromatography and high pressure liquid chromatography were utilized as described previously to resolve the extract into two active fractions. Activity was determined by measuring the extent of conversion of [^{14}C]arachidonic acid to PGE_2 in the presence of bovine seminal vesicle microsomes at various drug concentrations. Under the conditions of the assay, the possible conversion of endogenous precursors or the production of other PGs was not monitored. Once again two active components were found, one of which was identical to authentic eugenol.

The other active fraction, previously referred to as Fraction 5 [3], consisted of several components as seen on gas–liquid chromatography (6 ft; 5% OV-17; T = 120°). These were resolved into pure fractions by preparative g.l.c., and the location of the active compound was determined by enzymic assay [1, 2]. Mass spectral determination indicated that the nominal molecular weight of the substance was 120 with major fragments at *m/e* 119, 94 and 91.

The ultraviolet absorption spectrum (λ_{max}^{EtOH} 260, ϵ = 6000) of the unknown suggested that it was aromatic in nature and its solubility in mildly alkaline solution further suggested a phenolic substance. A careful search of the literature revealed that indeed a phenol with molecular weight 120, namely *p*-vinylphenol, had been isolated from cannabis smoke condensate [5].

The identity of our substance was confirmed by direct g.l.c. and mass spectral comparison with authentic *p*-vinylphenol. The latter was obtained by thermal decomposition of *p*-hydroxycinnamic acid (Aldrich Chemical Co.) as it is otherwise unavailable. It has been proposed [5] that *p*-hydroxycinnamic acid, which is also a cannabis constituent [6], is probably the source of the *p*-vinylphenol in smoke condensate (*vide supra*). Apparently steam distillation conditions such as those used in our procedure are sufficient to cause the decarboxylation to occur. It seems less likely that *p*-vinylphenol exists as such in cannabis, as it is reported to be a rather unstable substance [5].

Table 1 gives an indication of the potency of *p*-vinylphenol as an inhibitor of PGE_2 synthesis in the bovine seminal vesicle system [2]. For comparison purposes, aspirin was also assayed by us and, as can be seen in Table 1, *p*-vinylphenol is somewhat more potent. Since *p*-hydroxycinnamic acid is a likely source for the *p*-vinylphenol in cannabis distillates, we thought it of interest to test it as well. Surprisingly, it was a moderate stimulator of PGE_2 synthesis at doses where *p*-vinylphenol was inhibitory. Certain other substances have shown stimulation in our system, the most notable being morphine; some data are shown in Table 1 for comparison. The significance of PG synthesis stimulation is not well established at this time; however, Collier *et al.* [7] have also reported that

Table 1. Effects on PGE_2 synthesis by *p*-vinylphenol and comparison substances*

	Concentration		
	(μg/ml)	(M × 10^{-6})	% Change ± S.E.M.†
p-Vinylphenol	1	8.6	↓ 27 ± 3.3
	10	86	↓ 86 ± 0.72
p-Hydroxycinnamic acid	1	6.0	0 ± 2.9
	10	60	↑ 12 ± 3.7
	100	600	↑ 47 ± 7.0
Aspirin	10	56	↓ 47.8 ± 4.9
	100	560	↓ 85.0 ± 8.0
Morphine	1	3.5	↑ 22
	10	35	↑ 30 ± 7.8
	100	350	↑ 38 ± 15.2

* PGE_2 synthesis was assayed using a bovine seminal vesicle preparation as described previously in Ref. 2.

† Key to symbols: ↓ indicates inhibition, while ↑ denotes stimulation. All values are relative to control which consists of the identical system without drug. Control values ranged from 30 to 45 per cent conversion of [^{14}C]arachidonic acid to [^{14}C]PGE_2. N = 3.

morphine enhances PG production in a similar experiment, and our results are in good agreement with theirs.

It is very much a matter for speculation as to what the role of these phenolic substances in the overall pharmacology of cannabis may be. Activities other than mood alteration have recently been the subject of considerable study with a view toward possible therapeutic applications for cannabis [8]. There has been a good correlation between therapeutically active agents and substances which alter PG synthesis (or release) so that it is not unreasonable that constituents such as eugenol and *p*-vinylphenol may contribute to the overall activity of the drug. While the major activity of cannabis is attributable to its Δ^1-THC content, the contributions of other constituents (both cannabinoid and non-cannabinoid) should be considered in assessing the results of marihuana smoking.

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