The metabolism of 4-ethynyl biphenyl

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4-Ethynyl biphenyl was designed and tested as a potential anti-inflammatory drug (Allen & Hanbury Ltd.). It was found to possess anti-inflammatory activity but further development was prevented because of its toxic ulcerogenic behaviour in animal trials. Nevertheless, it was decided to investigate the metabolism of the compound because of its interesting structure, a hydrocarbon molecule possessing a terminal acetylene group. To this end [^{14}C]-4-ethynyl biphenyl was synthesized and its metabolism studied in rat and rabbit.

When [^{14}C]-4-ethynyl biphenyl was dosed orally to rats (40 mg/kg, male Wistar albinos, 200 g) no unchanged parent drug was excreted in the urine or faeces. After 24 h 52% of the radioactivity had been excreted (43% urine, 9% faeces), and after 48 h 80% (60% urine, 20% faeces). Small amounts of radioactivity were excreted for a further 3 days. The excretion pattern in rabbits following oral dosing (40 mg/kg, male Dutch rabbits, 2.5 kg) was different, although again no unchanged parent drug was excreted. Excretion of radioactivity was more rapid, 75% of the dose in 24 h (70% urine, 5% faeces), and after 48 h greater than 95% of the dose was accounted for. No radioactivity was excreted after the third day.

The significant amount of radioactivity in rat faeces and the prolonged excretion of the compound suggested that biliary excretion and entero-hepatic circulation was occurring and this was confirmed by bile-duct cannulation studies. Radioactive bile from one animal was injected into the duodenum of a second animal fitted with a bile-duct cannula. Biliary excretion of [^{14}C] was detected (up to 30% of dose in 6 h). In similar experiments where the second animal had been treated with antibiotics or 1-4-saccharo-lactone the biliary excretion of [^{14}C] fell to 5% and 8% respectively.

The major metabolite in rat and rabbit urine was 4'-OH-4-biphenylyl acetic acid (greater than 95%) although small amounts of biphenylyl acetic acid were also found (0–2% in rat, 2–4% in rabbit). In the rat the urinary metabolites were not conjugated but in the rabbit about 50% of urinary radioactivity was present as conjugates. Five significant conjugates were observed, one of which was identified as the glucuronide of 4'-OH-4-biphenylyl acetic acid. Faecal metabolites in rat and rabbit and biliary metabolites in rat were acid-hydrolyzed to free 4'-OH-4-biphenylyl acetic acid and 4-biphenylyl acetic acid, these compounds accounting for greater than 95% of the metabolites.

The major urinary and faecal metabolite, 4'-OH-4-biphenylyl acetic acid was inactive as an inhibitor of prostaglandin synthetase whereas 4-biphenylyl acetic acid showed moderate activity as an inhibitor.

Metabolism of 4-ethynyl biphenyl was studied in rat liver microsomes, both normal and induced with phenobarbitone or 3-methylcholanthrene. The major metabolite in all cases was 4-biphenylyl acetic acid (>95% total metabolites), whilst small amounts of 4'-OH-4-biphenylyl acetic acid were observed, as well as several other minor metabolites. The metabolism of 4-ethynyl biphenyl, or more specifically, of the acetylene group was greatly induced by both phenobarbitone and 3-methylcholanthrene.

The gastrointestinal activity of the parent compound and its metabolites is now being studied.

Metabolism studies with practolol

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A small proportion of patients treated with practolol have been reported to develop adverse reactions termed collectively the oculomucocutaneous syndrome, the occurrence of which does not appear to be related to the pharmacology of practolol (Nicholls, 1976). Practolol metabolism has been re-examined to investigate any abnormality in its disposition in affected subjects; to search for animal species in which extensive metabolism, actually atypical of man, might result in toxic signs in an 'animal model'; and to study the possible formation of metabolites which bind irreversibly to protein.

[^{14}C]-(Acetyl) and [^{14}C]-(phenyl) practolol (100 mg, orally) was given to eight patients, six with skin reactions (Reeves, Case, Felix, Fluke, Holt, Jepson, McCormick, Nicholls & Zacharias, 1978). Peak blood levels (*ca* 1 μg/ml) at 3 h decayed monoexponentially ($T_{\frac{1}{2}}$: 12–16 h). Elimination rates and metabolic patterns were similar in all subjects, 74–90% dose was eliminated in urine, primarily unchanged. Collec-

of oxygen consumption in the presence of a NADPH regenerating system. Intraperitoneal injection of 3-amino-1,2,4-triazole which decreases the catalase content in the liver (Heim, Appleman & Pyfrom, 1956) as well as the content of cytochrome P450 (Baron & Tephly, 1969) increased the rate of oxygen consumption as did the addition of azide *in vitro*. Therefore, H_2O_2 seems to be the direct reduction product of oxygen. This is in accordance with the increased oxidation of methanol to formaldehyde in the microsomal system in the presence of PQ which also has been observed by others (Ilett *et al.*, 1974). This agrees with the observed NADPH/O_2 ratios.

The effect of PQ on microsomal electron transport is directly comparable to that of menadione which increases NADPH oxidation (Gillette, Brodie & La Du, 1957), O_2 uptake (Sato, Nishibayashi & Omura, 1962) and methanol oxidation with the same NADPH/O_2 ratio as PQ does. Menadione, however, is not NADPH-specific. Both substances divert the electrons from the flavoproteins to oxygen, keeping the flavoproteins in the oxidized state. Thereby they inhibit the mixed function oxidations. The inhibition of MDA formation by PQ in this system with a high rate of oxygen reduction does not support the intermediate formation of superoxide anions.

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The production of periportal necrosis by allyl alcohol in the rat

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Daily dosing of rats with allyl alcohol or its esters produces a characteristic liver lesion, periportal necrosis, which has been found to regress by d.28 in spite of continued administration. Butterworth, Carpanini, Gaunt, Grasso & Lloyd (1975) have found a correlation between the hepatotoxicity of such esters and the rate of their hydrolysis *in vitro* to allyl alcohol. The periportal necrosis produced is believed to depend on the metabolism of the alcohol to acrolein (Reid, 1972; Serafini-Cessi, 1972). The abilities of allyl alcohol and acrolein to induce hepatic lesions have been compared when given by intraportal infusion to minimize the effects of differences in the absorption of allyl alcohol and acrolein. Preliminary experiments to investigate the development of tolerance to the administration of allyl alcohol have also been performed.

Groups of 3 male Wistar-derived weanling rats were anaesthetized with pentobarbitone sodium (120 mg/kg i.p.). Either allyl alcohol or acrolein was injected as a solution in saline (0.9% w/v NaCl soln.) through heparin-primed polyethylene tubing tied into a mesenteric vein. An infusion of 0.1 ml was made over a 10 s period, the vein ligated and the animal allowed to recover. After 24 h the rats were killed by cervical dislocation, the incidence of macroscopic liver lesions determined and the tissues examined histologically.

Allyl alcohol (8.5, 17.0 or 25.5 mg/kg) produced no liver lesions, but acrolein (0.425, 0.85 or 1.70 mg/kg) produced periportal necrosis in 0, 2 and 3 rats respectively. No hepatotoxicity was produced in the control rats given saline intraportally. These results show that acrolein is effective in producing liver lesions when administered by intraportal infusion, in contrast to the allyl alcohol. These lesions were histopathologically similar to those observed after repeated oral administration of allyl alcohol or its esters.

In a second series of experiments, groups of 10 rats were given corn oil (5 ml/kg) with or without allyl alcohol (25 mg/kg), daily by intubation for 28 days. The absence of macroscopic liver lesions in any of the alcohol-treated animals at d.28 was taken as evidence that tolerance to the alcohol had developed.