Table 1 Relationship between α-GP levels, body weight changes and histological evidence of ulceration following indomethacin treatment

Treatment Indomethacin (mg/kg)	*α-GP level (area of diffusion ring mm²) Means + s.e. Time (days) after single treatment with indomethacin*				*Body weight (g) Means*				*Histology scored 0–10 in arbitrary scale for severity of ulceration (total score for group of 4 rats)*			
	0	*3*	*5*	*10*	*0*	*3*	*5*	*10*	*0*	*3*	*5*	*10*
5	15 ± 1	18 ± 3	17 ± 2	16 ± 1	165	207	217	256	0	0	0	0
10	16 ± 1	111 ± 14	87 ± 20	24 ± 4	164	173	187	231	0	12	10	0
15	16 ± 2	164 ± 12	112 ± 19	48 ± 24	164	152	167	221	0	24	28	4
20	14 ± 1	146 ± 7	167 ± 11	100 ± 20	166	147	143	177	0	26	38	12*
Vehicle control	16 ± 1	13 ± 1	16 ± 2	13 ± 1	162	206	220	258	—	—	—	—

* Two rats died in this group.

damage produced by indomethacin, as determined histologically and in body weight changes (Table 1) and thus proved to be a useful means of following, non-invasively, the time course of the ulcerative process.

References

BILLINGHAM, M.E.J. & GORDON, A.H. (1976). The role of the acute phase reaction in inflammation. *Agents and Actions*, **6**, 195–200.

CROCKSON, R.A. RATCLIFFE, A.P. PAYNE, C.J. & SOOTHILL, J.F. (1966). Time sequence of acute phase reactive proteins following surgical trauma. *Clin. Chem. Acta*, **14**, 435–441.

GLENN, E.M., BOWMAN, B.J. & KOSLOWSKE, T.C. (1968). The systemic response to inflammation. *Biochem. Pharmac.* Suppl. March. 27–49.

LOWE, J.S. (1964). Serum protein changes in rats with arthritis induced by mycobacterial adjuvant. *Biochem. Pharmac.* **13**, 633–641.

MCCONKEY, B., CROCKSON, R.A. CROCKSON, A.P. & WILKINSON, A.R. (1973). The effects of some anti-inflammatory drugs on the acute phase proteins in rheumatoid arthritis. *Quart. J. Med.* **42**, 785–791.

MANCINI, G., CARBONARA, A.O. & HEREMANS, J.F. (1965). Immunochemical quantitation of antigens by single radial immunodiffusion. *Immunochem.*, **2**, 235–254.

Effects of β-adrenoceptor antagonists on the hepatic mixed-function oxygenases in the rat

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Following reports that the β-adrenoceptor blocking agents, pronethalol and propranolol exhibited tumorigenic properties in certain strains of mice (Paget, 1963; Alcock & Bond, 1964; Howe, 1965; Smith & Butler, 1978), some concern has been expressed over the potential carcinogenicity of this whole group of drugs.

It is known that chemical carcinogens may initiate malignancy through alkylation of DNA, or may potentiate malignant cell transformations by epigenetic mechanisms. Whereas there are many mutagenic tests for monitoring chemical damage to DNA, there are very few short-term tests for studying epigenetic mechanisms. Chemical carcinogens modify the liver microsomal haemoproteins, leading to the formation of cytochrome P-448 which catalyses the de-ethylation of ethoxyresorufin (Burke & Mayer, 1975), and the 2-hydroxylation of biphenyl (Burke & Mayer, 1975; Atlas & Nebert, 1976). In the present study, the effects of 5 β-adrenoceptor blocking agents (propranolol, practolol, pronethalol, acebutolol, atenolol) on the activities of rat liver microsomal biphenyl-2-hydroxylase, ethoxyresorufin de-ethylase and other mixed-function oxidase enzymes were investigated.

Male Wistar albino rats received single daily oral doses (5, 50 and 150 mg kg^{-1} day^{-1}) of a β-adreno-

ceptor blocking drug for three days and were sacrificed 24 h after the last administration. Enzyme activities and cytochrome content were determined in liver microsomes (105,000 g pellet resuspended in 1.15% KCl).

Pretreatment of animals with carcinogens leads to preferential enhancement of the cytochrome P-448-mediated 2-hydroxylation of biphenyl, whereas pretreatment with non-carcinogens enhances only the cytochrome P-450-mediated 4-hydroxylation (Creaven & Parke, 1966; Bridges *et al.*, 1973; McPherson, Bridges & Parke, 1974). None of the β-adrenoceptor blocking agents, at any dose level stimulated the 2-hydroxylation of biphenyl with the exception of propranolol which, at the highest dose of 150 mg/kg, caused a significant increase in the 2-hydroxylation (45%), as well as in the 4-hydroxylation (30%) of biphenyl. The activity of ethoxyresorufin de-ethylase was elevated following pretreatment with propranolol and pronethalol and this appeared to be dose-dependent, the increase being 2-fold at the highest dose (150 mg/kg) for both drugs. Acebutolol also stimulated the activity of this enzyme (65%) at this dose. The carcinogen 3-methylcholanthrene caused a very marked induction of this enzyme (180-fold) (Burke, Prough & Mayer, 1977), many orders of magnitude higher than that observed with the β-adrenoceptor blocking agents in this study.

None of the compounds, at any dose level, affected the demethylation of ethylmorphine, or the concentrations of cytochrome P-450 and cytochrome b_5, or cytochrome c reductase, demonstrating that none of the β-adrenoceptor blocking agents is a potent inducer of the liver microsomal mixed-function oxidase system.

Pronethalol and propranolol show a dose-response inductive effect of ethoxyresorufin de-ethylase in the rat, and propranolol at high dosage also induces biphenyl 2-hydroxylation, suggesting that both drugs might act as weak promoters of carcinogenesis when administered in prolonged high dosage. However, at the normal therapeutic dose (5 mg/kg) there is no inductive effect, although one must bear in mind that rates of drug oxidation in man are generally some 10-fold slower than in smaller animals, such as the rat.

References

ALCOCK, S.J. & BOND, P.A. (1964). Observations on the toxicity of Alderlin (pronethalol) in laboratory animals. Some factors affecting drug toxicity. *Proc. Eur. Soc. Study Drug Toxic.*, **4**, 30–39.

ATLAS, S.A. & NEBERT, D.W. (1976) Genetic association of increases in naphthaline, acetanilide and biphenyl hydroxylations with inducible aryl hydrocarbon hydroxylase in mice. **175**, 495–506.

BRIDGES, J.W., MCPHERSON, F.J., KING, L.J. & PARKE, D.V. (1973). The influence of various chemicals on hepatic drug metabolism systems. *Proc. Eur. Soc. Study Drug Toxic.*, **15**, 98–104.

BURKE, M.D. & MAYER, R.T. (1975). Inherent specificities of purified cytochromes P-450 and P-448 towards biphenyl hydroxylation and ethoxyresorufin de-ethylation. *Drug Metab. Dispos.*, **3**, 245–253.

BURKE, M.D., PROUGH, R.A. & MAYER, R.T. (1977). Characteristics of a microsomal cytochrome P-448-mediated reaction. Ethoxyresorufin O de-ethylation. *Drug Metab. Dispos.*, **5**, 1–8.

CREAVEN, P.J. & PARKE, D.V. (1966). The stimulation of hydroxylation by carcinogenic and non-carcinogenic compounds. *Biochem. Pharmac.*, **15**, 7–16.

HOWE, R. (1965). Carcinogenicity of 'Alderlin' (pronethalol) in mice. *Nature (Lond.)*, **207**, 594–595.

MCPHERSON, F.J., BRIDGES, J.W. & PARKE, D.V. (1974). *In vitro* enhancement of hepatic microsomal biphenyl 2-hydroxylation by carcinogens. *Nature (Lond.)*, **252**, 488–489.

PAGET, G.E. (1963). Carcinogenic action of pronethalol. *Br. Med. J.*, **2**, 1266–1267.

SMITH, B. & BUTLER, M. (1978). The effects of long-term propranolol on the salivary glands and intestinal serosa of the mouse. *J. Path.*, **124**, 185–187.

Quantitative analysis of noradrenaline clearance

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Interindividual variations in plasma noradrenaline may be due to variations in clearance of noradrenaline (NA) as well as to different levels of sympathetic nervous activity (FitzGerald, Davies & Dollery, 1979). The aim of this study was to quantify the relative contributions of uptake$_1$ (UP_1), uptake$_2$ (UP_2), catechol-*O*-methyl transferase (COMT) and monoamine oxidase (MAO) to NA clearance, with a view to establishing whether a defect in any one pathway could lead to significant reduction in clearance. Selective blockade of each pathway was provided by, respectively, desimipramine, metanephrine, pyrogallol and pargyline. Total systemic clearance was measured in 8 rabbits; (−)-NA (3.5 μg kg^{-1} min^{-1}) was infused for 1 h into the left ventricle, and timed samples were then