The Effects of Salicylate on the Activity of Tryptophan Pyrrolase in Rat Liver

By A. A.-B. BADAWY and M. J. H. SMITH. (*Department of Biochemical Pharmacology, King's College Hospital Medical School, Denmark Hill, London S.E.5, U.K.*)

The intraperitoneal injection of sodium salicylate (400mg/kg body wt.) in the rat causes a decrease in the tryptophan pyrrolase activity in the liver at 30 min. The enzyme activity is restored to normal 2 h after injection, increases to sixfold after 5 h and returns to the basal value at 12 h. The initial fall in the pyrrolase activity may be due to an inhibitory action of the drug, because salicylate, in concentrations of 0.05 mM and above, significantly decreases the formation of kynurenine from tryptophan by homogenates of rat liver *in vitro*. The inhibition is abolished by adding haematin to the reaction mixtures and therefore resembles that caused by globin, which interacts with iron protoporphyrins (Feigelson & Greengard, 1961).

The later activation of the enzyme by salicylate *in vivo* is not prevented by pretreatment with actinomycin D, nor is the increased activity inhibited by salicylate *in vitro*. The effects of the combined injection of salicylate and cortisol are additive whereas those of salicylate plus tryptophan are not. The increased enzyme activity caused by salicylate therefore resembles that produced by tryptophan (Knox, 1966) and not that by cortisol (Civen & Knox, 1959; Schimke, 1969). Salicylate has been reported to release L-tryptophan from its binding sites to circulating proteins (McArthur & Dawkins, 1969). The injection of salicylate (400 mg/kg body wt.) in the rat causes the content of tryptophan in the liver to increase approximately twofold at 15 min and to remain constant for at least 3 h. It is suggested that an increased availability of tryptophan to the liver may be a factor involved in the enhanced activity of tryptophan pyrrolase caused by salicylate *in vivo*.

We thank Dr A. Andrew for a gift of actinomycin D and the Nuffield Foundation for financial support.

Civen, E. & Knox, W. E. (1959). *J. biol. Chem.* **234**, 1787.
Feigelson, P. & Greengard, O. (1961). *J. biol. Chem.* **236**, 153.

Knox, W. E. (1966). In *Advances in Enzyme Regulation*, vol. 4, p. 287. Ed. by Weber, G. Oxford: Pergamon Press Ltd.

McArthur, J. N. & Dawkins, P. D. (1969). *J. Pharm. Pharmac.* **21**, 744.

Schimke, R. T. (1969). *Curr. Top. cell. Regulation*, **1**, 77.

Effects of Propyl Gallate and other Antioxidants on Rat Liver Microsomal Nicotinamide-Adenine Dinucleotide Phosphate-Linked Cytochrome c Reductase

By T. F. SLATER* and M. V. TORRIELLI. (*Department of Biochemical Pathology, University College Hospital Medical School, London WC1E 6JJ, U.K., and Institute of General Pathology, Corso Raffaello 30, Turin 10125, Italy*).

During a study of the attenuating effects of a number of antioxidants on the hepatotoxicity of carbon tetrachloride in the rat it was observed that low concentrations of propyl gallate inhibited drug metabolism by liver endoplasmic reticulum *in vitro*. For example, 30 μ M-propyl gallate inhibited the demethylation of aminopyrene (present in 5 mM concentration as substrate) by approx. 25% during a 20 min incubation. This effect of propyl gallate did not result from a competition with aminopyrene for the binding site near to cytochrome P-450; moreover, studies of the difference spectrum by using the procedure of Schenkman, Remmer & Estabrook (1967) suggested that the interaction of propyl gallate with the microsomal binding site was relatively weak. Low concentrations of propyl gallate also inhibited NADPH-neotetrazolium reductase to a similar extent to its action on the demethylation of aminopyrene; neotetrazolium is believed to couple with the NADPH electron-transport chain in the region of the rate-limiting factor X₁ (Dallner, Siekevitz & Palade, 1965).

The proximal flavoprotein of the NADPH-cytochrome P-450 chain can interact directly with added cytochrome c; the activity of NADPH cytochrome c reductase was measured by the procedure of Slater & Sawyer (1969). Propyl gallate (10–20 μ M) strongly inhibited the reductase and the inhibition was not competitive with NADPH or with cytochrome c. A number of other antioxidants were studied (promethazine, butylated hydroxytoluene, vitamin E, *NN*-diphenyl-*p*-phenylenediamine, cysteine), but none had a similar inhibitory action to that shown by propyl gallate. The observation that propyl gallate decreases drug metabolism by reaction with proximal flavoprotein is of general pharmacological interest, and is of value in interpreting the protective action of propyl gallate *in vivo* in a number of cases of acute liver injury.

Dallner, G., Siekevitz, P. & Palade, G. E. (1965). *Biochem. biophys. Res. Commun.* **20**, 135.

Schenkman, J. B., Remmer, H. & Estabrook, R. W. (1967). *Molec. Pharmac.* **3**, 113.

Slater, T. F. & Sawyer, B. C. (1969). *Biochem. J.* **111**, 317.

* Present address: Department of Biology, Brunel University, Kingston Lane, Uxbridge, Middlesex, U.K.