Synaptic vesicle magnesium-adenosine triphosphatase as a possible site of action of anticonvulsant drugs

J.C. GILBERT & M.G. WYLLIE*

Department of Pharmacology, University of Aberdeen

Phenytoin and ethosuximide inhibit a magnesium-activated adenosine triphosphatase (Mg-ATPase) located in the vesicle-containing fraction derived from nerve terminals prepared from rat cerebral cortex (Gilbert & Wyllie, 1974a,b). Mg-ATPase activities in the membrane and mitochondrial fractions derived from the nerve terminals were not influenced by the drugs.

We have recently tested the effects of diazepam and the anticonvulsants phenobarbitone, trimethadione, acetazolamide, sodium valproate and SC 13504 (Searle) on the Mg-ATPase activity of synaptic vesicles using techniques similar to those used previously to prepare synaptosomes, to disrupt them by osmotic shock, to recover the sub-synaptosomal fractions and to assay ATPase activities (Gilbert & Wyllie, 1974a,b; 1975).

All the anticonvulsants and diazepam, when added to preparations *in vitro*, inhibited the Mg-ATPase contained in the fraction enriched in synaptic vesicles but not the enzyme contained in the other fractions prepared from the synaptosomes. Other drugs tested, including convulsants, did not inhibit the enzyme in the vesicle fraction. Administration of phenytoin (50 mg/kg and 100 mg/kg) or phenobarbitone (25 mg/kg and 100 mg/kg) *in vivo* to rats and subsequent preparation of the vesicle-containing fractions showed that the Mg-ATPase was inhibited to the maximum degree which had been observed in the experiments involving the addition of the drugs *in vitro*.

Dose response curves indicated that the relative potencies of the anticonvulsants with regard to inhibition of the enzyme were compatible with their relative potencies in preventing seizure activity in mice.

Studies of the distribution of phenytoin using the assay technique of Dill & Glazko (1972) suggested that the drug was present in the synaptosome cytoplasm and vesicle fractions.

This work suggests that the Mg-ATPase of nerve terminal vesicles may be implicated in the mechanism of action of anticonvulsants.

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References

DILL, W.A. & GLAZKO, A.J. (1972). Fluorimetric assay of diphenyhydantoin in plasma or whole blood. *Clin. Chem.*, **18**, 675-676.

GILBERT, J.C. & WYLLIE, M.G. (1974a). The effects of the anticonvulsant ethosuximide on adenosine triphosphatase activities of some subcellular fractions prepared from rat cerebral cortex. *Br. J. Pharmac.*, **52**, 139-140P.

GILBERT, J.C. & WYLLIE, M.G. (1974b). The effects of phenytoin on adenosine triphosphatase activities of synaptosomes and their components. *Br. J. Pharmac.*, **52**, 445P.

GILBERT, J.C. & WYLLIE, M.G. (1975). Effects of Prostaglandins on the ATPase activities of synaptosomes. *Biochem. Pharmac.*, **24**, 551-556.

Interaction of carbenoxolone sodium with other drugs bound to plasma proteins

S. GOTTFRIED, D.V. PARKE, P.J. SACRA & P.C. THORNTON*

Department of Pharmacology, Biorex Laboratories Ltd., London, and Department of Biochemistry, University of Surrey

Displacement from plasma protein binding sites has been suggested as one of the causative factors in increased efficacy or toxicity during concomitant administration of two or more drugs (O'Reilly & Levy, 1970; Mason & McQueen, 1974; Pugh, Geddes & Yeoman, 1975). Carbenoxolone sodium is an ulcer-healing drug which is highly bound to plasma proteins. Its efficacy, mode of action and low toxicity may even depend upon this binding (Parke & Lindup, 1973).

We have carried out a series of *in vitro* experiments with the fluorometric probe 1-anilino-8-naphthalene sulphonic acid using the method of Jun, Luzzi & Hsu (1972). Carbenoxolone sodium was studied together with several other protein-bound drugs which may be clinically encountered in carbenoxolone therapy. These included, phenylbutazone, flufenamic acid, prednisolone, aspirin, warfarin, tolbutamide, phenytoin and imipramine. The parent triterpenoid moeity of