Penicillin-induced convulsions and inhibition of glutamate decarboxylase

C.B. CHARINGTON & P.V. TABERNER

Department of Pharmacology, University of Bristol, University Walk, Bristol BS8 1TD

Penicillins administered topically to the brains of animals produce epileptiform convulsions possibly by the antagonism of GABA-mediated inhibition (Hill, Simmonds & Straughan, 1973). However, both GABA synthesis and uptake have been shown to be reduced in epileptogenic foci induced by penicillin (Gottesfeld & Elazar, 1975). In view of the fact that inhibitors of GABA synthesis also produce convulsions (Wood, 1975) and that some penicillins produce convulsions after a longer delay than might be expected from a direct interaction at GABA receptors, the present study has compared the relative potencies of a range of penicillins as glutamate decarboxylase (GAD) inhibitors and as convulsants.

Adult LACG mice were used throughout. CD_{50} values were determined following direct intracerebroventricular injection as described previously (Taberner, 1976). GAD activity was assayed in partially purified extracts of whole brain using essentially the radioisotopic method of Roberts & Simonsen (1963) in the presence of excess pyridoxal phosphate. Inhibitor constant (Ki) values were subsequently determined from Dixon plots using varying concentrations of inhibitor and substrate.

The penicillins tested fell into three groups in terms of CD_{50}. Group I (ampicillin, amoxycillin and 6-aminopenicillanic acid) failed to produce convulsions at doses up to the limit of their solubility (>220 nmoles). Group II (penicillin G, penicillin V, flucloxacillin and cloxacillin) were convulsant, with CD_{50}'s between 50 and 80 nmoles. Group III (D- and L-Penicillamine) were far less potent with CD_{50}'s of 1737 and 2139 nmoles respectively. All the compounds listed were inhibitors of GAD activity and competitive with respect to glutamate. The Ki values all fell within the range of 1–10 mM and showed no correlation with relative convulsant potency. Mean latency to convulsions varied between 1.6 min (penicillin V) and 2.8 min (cloxacillin), except D-penicillamine which had a mean latency of 43.5 minutes.

All group I and II compounds contain the β-lactam and thiazolidine rings of the penicillin nucleus whereas D-and L-penicillamine (ββ-dimethylcysteine) consist only of the uncondensed thiazolidine ring in a straight chain form. Since the penicillins and penicillamines were approximately equipotent GAD inhibitors it is unlikely that the β-lactam ring is required for GAD inhibition. The low convulsive potency of Group I compounds may reflect their poor solubility which prevents them from penetrating to their site of action *in vivo*. The average concentration of the group II penicillins in the brain after i.c.v. injection will be well below their Ki values for GAD inhibition, and it is therefore very unlikely to contribute significantly to the convulsant action of these compounds. However, the low potency of D- and L-penicillamine and the long latency exhibited by D-penicillamine suggest that, in this case, GAD inhibition may be responsible for their convulsant activity.

CBC was supported by a Canadian M.R.C. fellowship.

References

GOTTESFELD, Z. & ELAZAR, Z. (1975). GABA synthesis and uptake in penicillin focus. *Brain Res.* **84**, 346–50.

HILL, R.G., SIMMONDS, M.A. & STRAUGHAN, D.W. (1973). A comparative study of some convulsant substances as γ-aminobutyric acid antagonists in the feline cerebral cortex. *Br. J. Pharmac.*, **49**, 37–51.

ROBERTS, E. & SIMONSEN, D.G. (1963). Some properties of L-glutamic decarboxylase in mouse brain. *Biochem. Pharmac.* **12**, 113–134.

TABERNER, P.V. (1976). The anticonvulsant activity of the dissociative anaesthetic ketamine against seizures induced in mice by pentylene-tetrazol and mercaptopropionic acid. *Eur. J. Pharmac.* **39**, 305–312.

WOOD, J.D. (1975). The role of γ-aminobutyric acid in the mechanism of seizures. *Prog. in Neurobiol.* **5**, 77–95.

Some sub-cellular effects of an organophosphorus insecticide, Abate

M.A. ENNIN & C.S. FRANKLIN

Department of Pharmacology, Chelsea College, University of London

Organophosphorus compounds are characterized by their anticholinesterase effects; in addition, they have been found to affect microsomal drug metabolizing enzymes of the liver (Stevens, Stitzel & McPhillips, 1972). In this study the interactions of OOO'-O'-tetramethyl-OO'-thiodi-*p*-phenylene phosporothioate, Abate, with rat liver, brain and blood have been investigated. Rats of the CFHB-remote Wistar strain were given Abate in arachis oil daily by intraperitoneal injection at doses between 10 and 300 mg/kg body weight over periods of 4, 7 and 10 days. Changes in the levels of various enzymes in liver homogenates and sera have been used as indicators of hepatic injury. Of the enzymes determined, Aspartate transaminase, Alanine transaminase, acid phosphatase and the ratio of Lactate dehydrogenase (2oxobutarate:Pyruvate) did not show any significant changes over all dose periods (P=0.05). There were slight reductions in the levels of Glutamate dehydrogenase and aminopyrine demethylase at all dose periods and in the P_{450} in the 7

and 10 days. P_{450} levels were found to increase in the 4 day period which included the higher doses. The blood and brain cholinesterase levels were found to fall consistently with increased dose of Abate.

Reference

STEVENS, J.T., STITZEL, E.R. & MCPHILLIPS, J.J. (1972). Effects of anticholinesterase insecticides on hepatic microsomal metabolism. *J.Pharm. xp. Therap.*, **181**, 576–583.

Metabolites of intraduodenally instilled histamine after pretreatment with monoamine oxidase inhibitors (MAOI)

P.R. IMRIE, E. MARLEY & D.V. THOMAS

Department of Pharmacology, Institute of Psychiatry, De Crespigny Park, London SE5

Ingested histamine was reported to be metabolized in cats by N-methylation followed by oxidation by monoamine oxidase to *t*-methyl-imidazoleacetic acid (Schayer, 1956). The absorption therefore of large amounts of histamine following its intraduodenal instillation in cats pretreated with a hydrazine MAOI was unexpected (Blackwell & Marley, 1966). Subsequently, instilled- and by implication ingested histamine was found (Imrie, Marley & Thomas, 1978) to be metabolized predominately and equally to *t*-methylimidazoleacetic acid and to imidazoleacetic acid, indicating diamine oxidase (DAO) to be as important as N-methylation for metabolizing ingested histamine. Since hydrazine MAOI inhibit DAO (Burkard, Gey & Pletscher, 1962), an action possibly accounting for Blackwell & Marley's (1966) findings, the effect of non-selective and selective MAOI including MAOA and MAOB inhibitors was examined with both a small and a large dose of histamine. [^{14}C]-Histamine and its metabolites were assayed by scintillation spectrometry following paper chromatography (Thomas & Marley, 1978).

After pretreatment with mebanazine or nialamide (Table 1), the concentrations of histamine and *t*-methylhistamine compared to controls were elevated in portal venous blood with both doses of histamine, slow rates of absorption of ^{14}C-compounds occurring with the large dose (rate of absorption also appears to determine type of metabolite; slow rates giving rise to acid metabolites and a negligible proportion of histamine). In contrast, the consequences of tranylcypromine, deprenyl (MAOB-inhibitor) or clorgyline (MAOA-inhibitor) pretreatment depended on the amount of histamine instilled, the blood concentrations of histamine and *t*-methylhistamine decreasing with the small dose but increasing with the large dose. Unlike the other MAOI, clorgyline enhanced the rate of absorption of ^{14}C compounds. Intestinal 5HT and β-phenethylamine oxidation (Robinson, Lovenberg, Keiser & Sjoerdsma, 1968) at the end of experiments were reduced by deprenyl ($n = 6$) to 68 ± 11.9 and 66 ± 12.6% of control values while the corresponding values with clorgyline ($n = 4$) were 33.6 ± 7.3 and 65.6 ± 11.3%.

In conclusion, the non-selective MAOI led to increased circulating histamine and *t*-methylhistamine with both doses of histamine while the selective MAOI enhanced circulating histamine only with the large dose of histamine.

Generous support came from The Wellcome Trust (D.V.T.) and The Bethlem Royal and Maudsley Hospital Research Fund (P.R.I.).

Table 1 [^{14}C]-Histamine and its metabolites in portal venous (PV) and cranial mesenteric arterial (CMA) blood for control cats and those pretreated with MAOI

MAOI		*ImAA*	*MeImAA*	*MeHis*	*His*	*Rate of Absorption* (nmol/min)*	*n*
			(pmol/ml blood)				
[^{14}C]-Histamine (5μCi with 1.7 μmol/kg)							
———	PV	398	344	99	98	16	6
	CMA	255	189	66	53		
Mebanazine	PV	183	241	223	231	27.5	3
(120 μmol/kg)	CMA	94	151	138	25		
Nialamide	PV	392	531	1,327	377	49.5	2
(80 μmol/kg)	CMA	234	309	987	67		
Tranylcypromine	PV	247	229	14	17	40.5	2
(14 μmol/kg)	CMA	78	81	7	8		
Deprenyl	PV	211	149	59	22	10.5	2
(4.5 μmol/kg)	CMA	177	112	40	7		
Clorgyline	PV	605	326	45	11	20.5	2
(24.5 μmol/kg)	CMA	379	191	37	9		