Porphyrogenic activity of methsuximide and its demethylated metabolite

The antiepileptic drug, methsuximide (*N*-2-dimethyl-2-phenylsuccinimide) has been implicated in the precipitation of acute intermittent porphyria in an epileptic patient (Birchfield & Cowger, 1966).

To provide further information about this unwanted effect the porphyrinogenic activity of methsuximide and its metabolites, 2-methyl-2-phenylsuccinimide and hydrolysed methsuximide (Nicholls & Orton, 1971, and unpublished observations) was investigated in the developing chick embryo. Hydrolysed methsuximide refers to a mixture of *N*-2-dimethyl-2-phenyl and *N*-3-dimethyl-3-phenylsuccinamic acids prepared by hydrolysis of methsuximide in alkali.

Fertilized hen eggs (C. V. Bartlett Ltd., Chepstow, Monmouthshire) were incubated at 38° and 90% humidity in a Gallenkamp Humidity Oven for 17 days. The compounds (8 mg/egg) were dissolved in 0·1 ml absolute ethanol. The egg-shell was pierced above the air sac and the solution injected through the chorioallantois into the fluids surrounding the embryo. The opening in the shell was covered with sterile Cellophane tape (Scotch Tape) and incubation of the eggs continued for 30 h. The chick embryo was then killed by decapitation and the liver removed for extraction of porphyrins. The porphyrin content of chick embryo liver was estimated by the method of Racz & Marks (1969). Porphyria-inducing activity was expressed as μg coproporphyrin I/g wet liver. Absolute ethanol was not porphyrinogenic when compared with normal saline controls. The dosage and incubation time for a well-defined porphyrinogenesis was determined initially with methsuximide. The concentration of porphyrins in chick embryo liver after 24 h incubation with various doses of methsuximide (1 to 16 mg/egg) is in Table 1. At each dose the liver porphyrin content varied although it was possible to demonstrate a form of dose response relation using the mean values obtained at each dose.

Using a constant dose of 8 mg methsuximide, a 50-fold increase in the level of

Table 1. *Effect of duration of incubation of fertile hens eggs on the accumulation of porphyrins in chick embryo liver caused by methsuximide, 8 mg/egg, and in different doses after 24 h incubation.*

Incubation time[a] (h) after 8 mg dose	Porphyria inducing activity (μg porphyrins/g liver)[b]	Dose[a] (mg/egg)	Porphyria inducing activity (μg porphyrins/g liver)[b]
0	0·20 (0·15–0·25)	Control	0·19 (0·15–0·23)
18	0·38 (0·25–0·58)	1 mg	0·21 (all values identical)
24	2·38 (0·70–5·08)	2 mg	1·38 (0·38–3·25)
30	10·50 (0·83–20·00)	4 mg	1·68 (0·58–2·88)
34	10·68 (2·82–17·38)	8 mg	2·43 (1·30–4·18)
41	4·45 (1·05–10·30)	16 mg	5·05 (0·33–16·75)

[a] Methsuximide (dose/0·1 ml absolute ethanol) was injected into 17-day old fertile hens' eggs incubated at 38°/90% humidity.

[b] Expressed as coproporphyrin I/g wet weight liver.

The results are the mean of at least six eggs per time.

The extreme values are given in parentheses.

liver porphyrins was observed when the incubation time was extended to 30 h. A similar response was seen after 34 h incubation although after 41 h the response was reduced but still marked (Table 1).

Thus a well-defined porphyrinogenesis could be demonstrated in the liver after the incubation of chick embryos with methsuximide. This supports the finding of Racz & Marks (1969) with this drug. 2-Methyl-2-phenylsuccinimide, a pharmacologically active anticonvulsant metabolite of methsuximide in the rat and man (Nicholls & Orton, 1971), increased the porphyrin content of chick embryo liver approximately six-fold (1·2 μg porphyrins/g liver) which is about eight times less than that observed after administration of methsuximide (10 μg porphyrins/g liver). Hydrolysed methsuximide, which does not possess anticonvulsant properties (Nicholls & Orton, 1971), had no porphyrinogenic activity.

It, therefore, appears that 2-methyl-2-phenylsuccinimide not only contributes to the pharmacological anticonvulsant activity of methsuximide but also to the porphyrinogenic activity.

Welsh School of Pharmacy,
UWIST, Cardiff, CF1 3NU, U.K.

T. C. ORTON
P. J. NICHOLLS

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Simultaneous measurement of skeletal muscle, pulmonary mechanical and vascular responses to bronchodilators in the cat

Lands, Arnold & others (1967) and Lands, Luduena & Buzzo (1967) proposed two types of β-adrenoceptors. Bronchodilatation and vasodepression produced by β-adrenoceptor stimulants are believed to be mediated via β_2-adrenoceptors. These drugs can also enhance normal physiological tremor in man (Marsden, Foley & others, 1967) by a mechanism believed to involve β_2-adrenoceptors in skeletal muscle (Bowman & Nott, 1970). Inhalation therapy with β-stimulants is free from effects on skeletal muscle, but effective bronchodilatation after oral administration carries with it the side effect of muscle tremor in some patients (Legge, Gaddie & Palmer, 1971). Lands' proposal is based on results obtained from experiments in different species and so far no work has been reported in which simultaneous recordings have been made for the effect of β-stimulants on skeletal and bronchial muscle in the same animal. Some such experiments are now reported.

Cats of either sex, 2·2–4·0 kg, were anaesthetized with chloralose (80 mg/kg intravenously) after induction with a 3% halothane nitrous oxide 3 litre min^{-1} oxygen 1 litre min^{-1} mixture. Arterial blood pressure (mm Hg) was monitored from a cannula in a common carotid artery. The left soleus muscle was prepared (Bowman & Nott, 1970) and submaximal tetanic contractions were elicited by rectangular pulses of 50 μs duration at twice the voltage necessary to elicit a maximal twitch. Tetani (6–10 Hz) were produced for 1 s every 10 s and peak tension (g) recorded using an Ether UFI 32 oz strain gauge connected to a Devices M–4 pen recorder. Area ($g\ s^{-1}$) under the tetanus was obtained by integration of the strain