J. Pharm. Pharmac., 1977, 29, 143–146 Received October 29, 1976

Hyperpyrexic interaction between debrisoquine and pethidine in rabbits

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Pethidine injection into rabbits treated with debrisoquine either acutely or chronically resulted in severe interaction and fatal hyperpyrexia. Pretreatment of rabbits with *p*-chlorophenylalanine, chlorpromazine, or cyproheptadine protected them against the interaction, while α-methyl-*p*-tyrosine was ineffective. In addition the administration of debrisoquine into 5-HTP pretreated rabbits produced a severe interaction and hyperpyrexia. The hepatic *N*-demethylation of pethidine was significantly inhibited by debrisoquine pretreatment both *in vivo* and *in vitro*. The debrisoquine-pethidine interaction could be due to 5-HT potentiation or prevention of uptake. Alternatively it could be due to inhibition of biotransformation of pethidine by debrisoquine. However, neither mechanism by itself alone could be held responsible as the sole explanation of the interaction.

Debrisoquine, an antihypertensive agent similar in action to guanethidine is also a selective inhibitor of intraneuronal monoamine oxidase (MAO) in man (Pettinger, Korn & others, 1969) both *in vitro* and *in vivo*. But its inhibition *in vitro* is in excess of that concentration in the clinical use of the drug unless it is concentrated in specific tissues. However interaction between debrisoquine and some drugs in man related to MAO inhibition has been reported (Skinner, Coull & Johnston, 1969; Amery & Deloof, 1970; Allum, Aminu & others, 1974).

We have examined the possible interaction between debrisoquine and pethidine in the rabbit to find if this could be modified by drugs which affect the synthesis of brain amines. The interaction observed in man when pethidine is given to patients treated with MAO inhibitors is generally assumed to be caused by inhibition of demethylation of pethidine (London & Milne, 1962; Jacobson, 1965). We have therefore investigated the effect of debrisoquine on pethidine metabolism in rabbits both *in vivo* and *in vitro*.

METHODS

Rabbits of local strains and of either sex 1·5–2·0 kg were treated with debrisoquine sulphate (20 mg kg^{-1}, i.p.) either as a single injection and followed 1 h later by pethidine or injected daily for 4 successive days. Control rabbits received 1 ml of 0·9% w/v NaCl (i.p.). The following day temperatures were recorded at 15 min intervals electrically (Ellab electric universal thermometer type T.E 3 inserted 5–7 cm into the rectum). After the temperature had stabilized for 30 min, pethidine HCl (5 mg kg^{-1}) was slowly administered via a marginal ear vein. Experiments were made at 21–22° and were repeated in debrisoquine-pretreated rabbits with the addition of one of the following pretreatments: chloropromazine HCl or cyproheptadine HCl (5 mg kg^{-1}, i.v.) 30 min before debrisoquine; *p*-chlorophenylalanine (PCPA) (125 mg kg^{-1}, i.p.) 66, 42, and 18 h before debrisoquine and α-methyl-*p*-tyrosine (80 mg kg^{-1}, i.p.) 36, 24 and 12 h before pethidine. Finally, debrisoquine (20 mg kg^{-1}, i.v.) was administered to rabbits pretreated 1 h earlier with 5-hydroxytryptophan (5-HTP) (60 mg kg^{-1}, i.v.).

Pethidine N-*demethylation*. Rabbits of either sex 1·5–2·0 kg were used. For *in vivo* studies debrisoquine was either injected as a single dose (20 mg kg^{-1}, i.p.) and one h later the animals were killed, or it was injected in a dose of 20 mg kg^{-1}, intraperitoneally, daily for 4 successive days and the animals were killed 24 h later. Hepatic *N*-demethylation of pethidine was measured by determining the amount of formaldehyde formed according to the method of Axelrod (1956) as modified by Eade & Renton (1970). Enzyme activity was expressed as μ mol formaldehyde formed h^{-1} g^{-1} of liver. The influence of *in vitro* addition of various concentrations of debrisoquine on hepatic *N*-demethylation of pethidine was also examined.

RESULTS

Debrisoquine-pethidine interaction in rabbits

Rabbits treated with debrisoquine, pethidine, PCPA, α-MT, cyproheptadine, or 5-HTP did not show any significant change in their body temperature compared with saline-treated control animals.

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Debrisoquine (20 mg kg^{-1}, i.p.) did not result in the appearance of any abnormal symptoms or behaviour. However, when pethidine (5 mg kg^{-1}, i.v.) was injected one h later, an immediate interaction was precipitated characterized by motor restlessness, shivering like tremor, hyperexcitability, tachypnoea, dilated pupils and hyperpyrexia which reached an average of 2·80 + 0·23° 1 h after pethidine injection (Fig. 1). These symptoms persisted for nearly 3 h after which the temperature returned to normal and none of the animals died in hyperpyrexia. Control rabbits treated with either debrisoquine or pethidine and injected 1 h later with saline did not show any interaction or hyperpyrexia.

The injection of pethidine in rabbits pretreated chronically with debrisoquine (20 mg kg^{-1}, i.p. daily for 4 days) resulted in severe interaction and hyperpyrexia which reached an average of 3·20 + 0·28° within 75 min of injection (Fig. 2). Three out of five rabbits died within 75 min of being injected with pethidine.

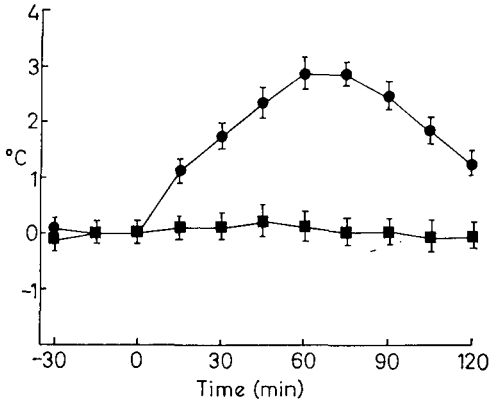

FIG. 1. The debrisoquine-pethidine interaction as measured by rectal temperature changes (°C) in rabbits. Debrisoquine (20 mg kg^{-1}, i.p.) was administered as a single dose in both groups. 1 h later and at zero time either pethidine (5 mg kg^{-1}, i.v.) was injected in one group of six rabbits (●) or saline (1 ml, i.v.) in another group of five rabbits (■). Vertical lines indicate s.e.m.

Debrisoquine produces similar interactions with imipramine and dextromethorphan but not with mepyramine (unpublished observations). Treatment of rabbits with guanethidine (20 mg kg^{-1}, i.p.) either acutely or chronically did not result in any interaction or hyperpyrexia when pethidine was injected.

The debrisoquine-pethidine interaction in rabbits was prevented by the administration of either

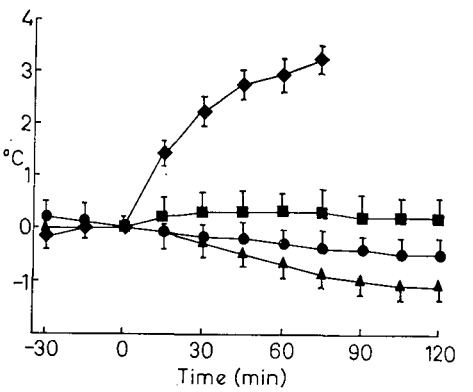

FIG. 2. The debrisoquine-pethidine interaction and its antagonism as measured by rectal temperature changes (°C) in rabbits. Debrisoquine (20 mg kg^{-1}, i.p.) was administered daily for 4 days in all groups and pethidine (5 mg kg^{-1}, i.v.) was injected at time zero. In addition the following drugs were administered at the times and doses indicated in the text: chlorpromazine (▲), cyproheptadine (●), and PCPA (■). Each point represents the mean ± s.e. from 4 animals except (◆), where 5 animals were used.

chlorpromazine or cyproheptadine 30 min before pethidine. None of the animals pretreated with either chlorpromazine or cyproheptadine died in hyperpyrexia. Similarly, pretreatment with PCPA protected the rabbits against the debrisoquine-pethidine interaction and all the animals survived (Fig. 1).

The injection of 5-HTP (60 mg kg^{-1}, i.v.) resulted in a slight increase in body temperature but when debrisoquine was injected 1 h after 5-HTP, severe symptoms of interaction and hyperpyrexia caused the death of 2 out of 4 animals (Fig. 3).

There was no protection against the debrisoquine-pethidine interaction following pretreatment of the rabbits with α-MT and 2 out of 4 rabbits died in hyperpyrexia 60 min after pethidine injection (Fig. 3).

Effect of debrisoquine on hepatic N-*demethylation of pethidine*

Debrisoquine significantly inhibited hepatic *N*-demethylation of pethidine (Table 1). There was no marked sex difference in the rate of *N*-demethylation. The average *in vivo* inhibition of *N*-demethylation of pethidine was about 35% following a single injection of debrisoquine (20 mg kg^{-1}, i.p.), and about 48% after 4 days treatment.

The *in vitro* addition of debrisoquine (0·5–5 mM) to the incubation mixture produced a dose dependent inhibition. The correlation coefficient for

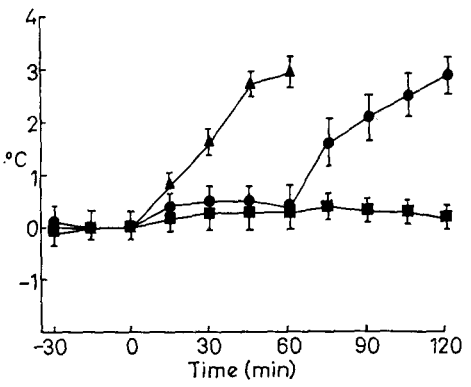

FIG. 3. The effect of α-MT on the debrisoquine-pethidine interaction in rabbits and interaction of debrisoquine with 5-HTP. In group (▲) doses of debrisoquine and pethidine are the same as in Fig. 2, in addition α-MT (80 mg kg^{-1}, i.p.) was injected at times 36, 24, and 12 h before test. 5-HTP (60 mg kg^{-1}, i.v.) was administered at time zero in group (●) and group (■). At time 60 either debrisoquine (●) or saline (■) was injected intravenously. Each point represents the mean ± s.e. from 4 animals. ordinate—Temperature change (°C).

in vitro incubation showed decreasing *N*-demethylation of pethidine with increasing concentrations of debrisoquine with a regression coefficient r = 0·9622 which was statistically significant ($P < 0·05$).

DISCUSSION

Debrisoquine differs from conventional MAOI drugs in that it is a weak and not a general or systemic MAOI nor does it inhibit intestinal MAO.

Table 1. *Effect of debrisoquine on hepatic* N-*demethylation of pethidine in rabbits.* Mean concentration (μ mol) ± s.e. HCHO h^{-1} g^{-1} liver.

	Control	Single injection of debrisoquine	4 days injections of debrisoquine
Male rabbits	1·91 ± 0·14 (6)	1·26* ± 0·11 (6)	0·96* ± 0·06 (6)
Female rabbits	1·94 ± 0·22 (5)	1·24* ± 0·05 (5)	1·04* ± 0·02 (5)

* Significantly different from control at $P < 0·05$. Figures in parentheses indicate the number of animals used in each experiment.

In addition it inhibits the uptake of 5-HT by the human platelets both *in vitro* and *in vivo* (Solomon, Ashley & others, 1969).

Signs of hyperexcitability, motor restlessness and hyperpyrexia seen in rabbits when pethidine was administered either after a single injection of debrisoquine or after chronic treatment for four days were similar to those produced in rabbits treated with MAOI's and injected with pethidine (Nymark & Nielsen, 1963; Loveless & Maxwell, 1965; Penn & Rogers, 1971; Fahim, Ismail & Osman, 1972; Sinclair, 1972, 1973; Eltayeb & Osman, 1975).

That 5-HT might be involved in the debrisoquine-pethidine interaction is supported by facts that pretreatment of rabbits with PCPA, which selectively inhibits tryptophan hydroxylase (Jequier, Lovenberg & Sjoerdsma, 1967) protected the animals against the fatal hyperpyrexic interaction. Also both chlorpromazine and cyproheptadine effectively antagonized the interaction. On the other hand, the injection of debrisoquine 1 h after 5-HTP resulted in severe interaction and fatal hyperpyrexia although in this dose range 5-HTP has little effect on the temperature of normal rabbits (Sinclair, 1973). Depletion of brain catecholamine by pretreatment with α-MT has no effect on the debrisoquine-pethidine interaction.

The MAO inhibiting properties of debrisoquine could be responsible for the fatal hyperpyrexic interaction with pethidine. However, this, in the acute stage, is unlikely. It might be due to 5-HT potentiation through prevention of its uptake as was shown by Solomon & others (1969). Pettinger & others (1969) reported a temporary supersensitivity after an intravenous injection of tyramine in patients within 1 h of receiving the first 20 mg dose of debrisoquine. This supersensitivity was independent of MAO inhibition as indicated by the full platelet MAO activity. They indicated that during long term treatment with debrisoquine the MAO inhibition was responsible for the tyramine supersensitivity.

Brownlee & Williams (1963), and Gessner (1973), reported that the acute toxicity of pethidine in mice was increased by the prior administration of MAOI's. Recently, Fuller & Snoddy (1975) showed that in mice the toxicity of a compound, Lilly 110140 (3-[p-trifluoromethylphenoxy]-*N*-methyl-3-phenylpropylamine hydrochloride), which was far more effective as an inhibitor of 5-HT uptake than pethidine, was not significantly potentiated by prior treatment with the MAOI tranylcypromine. These

findings are inconsistent with the hypothesis that blockade of 5-HT uptake by pethidine is responsible for the toxic interaction with MAOI's. So it is unlikely that 5-HT uptake inhibition by pethidine is the sole mechanism responsible for this toxic interaction.

The toxic interaction between debrisoquine and pethidine could be due to impairment of the biotransformation of pethidine. We found a single injection of debrisoquine results in about 35% inhibition of hepatic *N*-demethylation of pethidine while after four days treatment there was about 48% inhibition. However, as the acute interaction after a single injection occurred within minutes of pethidine administration, the time could be too short to allow a significant accumulation of the analgesic drug. After chronic treatment biotransformation of pethidine could be impaired which would be in agreement with earlier results of other workers (Clark, 1967; Jounela, 1968; Eade & Renton, 1970).

As the rabbit has been used for predicting potentially dangerous interactions in man especially those involving MAOI's and pethidine type drugs (Nymark & Nielsen, 1963; Loveless & Maxwell, 1965; Penn & Rogers, 1971; Sinclair, 1972). There is a possible hazard in administering pethidine to patients being chronically treated with debrisoquine.

Acknowledgements.

We are grateful to Mr Mahdi El-helo and to Mr S. Abdel Rahim for skilful technical assistance. We also wish to thank Messers F. Hoffmann-La Roche & Co. Ltd, Basle, Switzerland for the generous gift of debrisoquine sulphate (Declinax).

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