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Deprenil: loss of selectivity for inhibition of B-type MAO after repeated treatment

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Monoamine oxidase (monoamine: O_2 oxidoreductase, EC 1.4.3.4; MAO) has been shown to exist in mammals in at least two forms with different substrate specificities [1–3]. The form which preferentially deaminates serotonin, noradrenaline [1, 4] and, at least in the rat [5, 6] and the mouse [7], dopamine has been termed A-type MAO, and the other form, which preferentially deaminates phenethylamine and benzylamine [8], B-type MAO [1]. Both forms are present in varying proportions in most tissues [9], though not necessarily in the same compartments. There is evidence that in the superior cervical ganglion and in the pineal gland, for instance, MAO A is associated with adrenergic neurones whereas MAO B is not [10].

A number of MAO inhibitors inactivate the one or other form of the enzyme selectively, *in vitro* and after administration to animals [2]. Thus, deprenil [11, 12] and, at lower doses, pargyline [2], inhibit MAO B more effectively than MAO A. The reverse holds true for clorgyline [1, 12]. Other MAO inhibitors, such as iproniazid or tranylcypromine, do not display a differential effect on the two forms of MAO [2].

All the drugs mentioned above with the exception of tranylcypromine block the enzyme irreversibly [13]. Restoration of MAO activity after its inhibition with these drugs is due to enzyme resynthesis, which occurs at different rates in different tissues [13], and possibly also in different species.

In the rat brain, the normalization of MAO activity after acute treatment with an MAO inhibitor takes about 3 weeks, corresponding to a half-life of the enzyme of about 10 days [13]. This average value applies to both forms of the enzyme [14]. In the rat liver, MAO activity is restored faster, within 10 days, corresponding to a half-life of about 4 days [13]. It is therefore, conceivable that chronic treatment with irreversible MAO inhibitors, which after a single application inhibit one form of the enzyme preferentially, but not entirely selectively may lead to progressive inhibition of the other form. In other words, the selectivity observed after acute treatment might be lost as a result of chronic administration.

An indication of such an effect was obtained by Long *et al.* [15], who studied the MAO inhibitory properties of a number of substituted 5-phenyl-3-(*N*-cyclopropyl)ethylamine-1,2,4-oxadiazoles. Such a loss of selectivity would clearly have an important bearing on the interpretation of the results of animal and clinical studies involving repeated treatment with MAO inhibiting agents and designed to elucidate the role of certain transmitter amines in psychiatric and neurological disorders.

MATERIALS AND METHODS

Groups of four female albino rats of a Sprague–Dawley strain (Tierfarm Sisseln, Switzerland) weighing 160–220 g were treated with 1, and 10 mg/kg s.c. of both deprenil and the reversible A-type inhibitor harmaline [16], acutely or once daily for 2, 4, 7 or 14 days. The animals were decapitated 2 hr after the last dose, and MAO activity in the brains and livers determined as described by Wurtman and Axelrod [17].

[^{14}C]5-hydroxy-tryptamine binoxalate (5-HT; 51 mCi/m-mole, New England Nuclear, Boston, MA), [^{14}C]-phenethylamine-HCl (PEA; 51 mCi/m-mole, NEN), and [^{14}C]tyramine-HCl (TYR; 55 mCi/m-mole, Radiochemical Centre, Amersham, U.K.) were used as substrates at a concentration of 20.8 μM (10 nCi per sample) at pH 7.9. The final volume of the incubation mixture was 0.3 ml, and the reaction was carried out with 2.5 and 0.5 mg of brain and liver tissues. After incubation at 37° for 20 min, the labelled deaminated metabolites were extracted into 6 ml ethyl acetate. Aliquots of 4 ml were counted after the addition of 1 ml ethanol and 10 ml scintillator.

RESULTS AND DISCUSSION

After 4 days treatment with 1 mg/kg s.c. deprenil, deamination in the brain of all three substrates was inhibited to a greater extent than after acute treatment. This inhibition continued progressively at a slower rate until the last day of treatment, when there was no longer much difference in the inhibition of deamination of the three substrates. Inhibition of 5-HT deamination, initially barely detectable, and of TYR deamination, initially about 30 per cent, was increased more markedly than that of PEA, which was initially already blocked by 70 per cent.

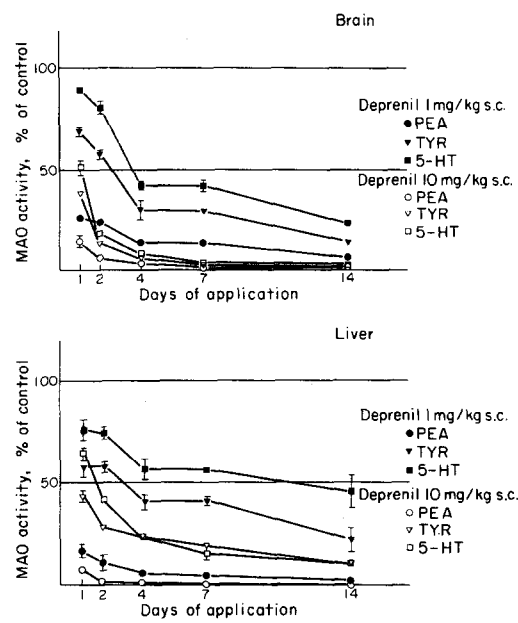

Fig. 1. Effect of acute and repeated treatment with deprenil on MAO activity in rat whole brain (upper panel) and liver (lower panel). Groups of four rats were treated acutely or once daily for 2, 4, 7 and 14 days with 1 or 10 mg/kg s.c. deprenil. Two hr after the last dose the brains and livers were removed and MAO activity determined using 5-HT, TYR, and PEA as substrates. The points represent $\bar{x} \pm$ S.E.M. in percent of controls.

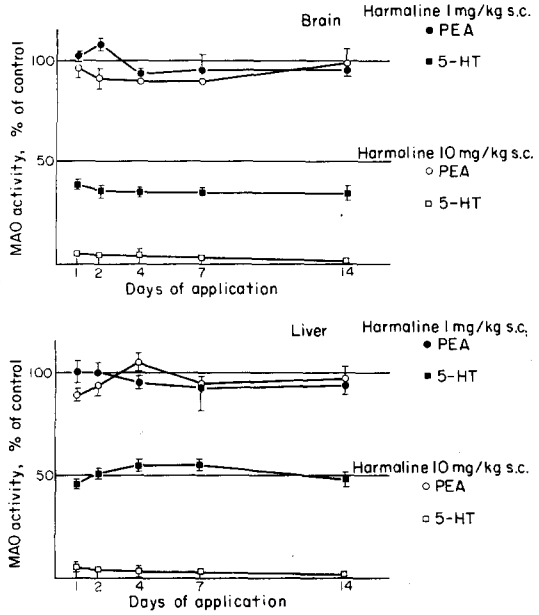

Fig. 2. Effect of acute and repeated treatment with harmaline on MAO activity in rat whole brain (upper panel) and liver (lower panel). Groups of four rats were treated acutely or once daily for 2, 4, 7 and 14 days with 1 or 10 mg/kg s.c. harmaline. Two hr after the last dose the brains and livers were removed and MAO activity determined using 5-HT and PEA as substrates. The points represent $\bar{x} \pm$ S.E.M. in percent of controls.

The results obtained with 10 mg/kg s.c. were very similar. Owing to the higher initial degree of inhibition of deamination of all three substrates, selectivity was already completely lost after 4 days (Fig. 1 upper panel).

In the rat liver, a similar phenomenon was observed after both doses. The extent by which deamination decreased in the course of repeated treatment seemed to be somewhat less than in the brain, however. Nevertheless, after a dose of 10 mg/kg s.c. selectivity for inhibition of PEA deamination was reduced to much greater degree after 14 days treatment than after acute treatment, in the liver also (Fig. 1, lower panel).

In contrast, the extent of MAO inhibition, measured with 5-HT and PEA as substrates, remained constant throughout the 2-week treatment with 1 or 10 mg/kg s.c. of the reversible A-type inhibitor harmaline, in the brain (Fig. 2, upper panel) and in the liver also (Fig. 2, lower panel).

These results demonstrate that in the rat, the selectivity for inhibition of MAO B (PEA deamination) observed after acute treatment with the irreversible MAO_B inhibitor deprenil diminishes progressively in the course of repeated treatment, as MAO A (5-HT deamination) becomes more and more effectively inhibited compared to MAO B, which is initially much more strongly inhibited. This effect seems to be more pronounced in the brain than in the liver, probably owing to the lower rate of enzyme resynthesis in cerebral tissue.

A similar decrease in selectivity during repeated treatment has also been observed with the irreversible inhibitors pargyline, which preferentially inhibits B-type MAO at low doses, and clorgyline, which preferentially inhibits A-type MAO (unpublished results).

The reversible inhibitor harmaline, which has a duration of action of less than 24 hr, did not give rise to such a phenomenon, which underlines that it is the accumulation due to irreversible inactivation of the enzyme by the classical MAO inhibitors that is responsible for this effect.

These results have a practical bearing on the interpretation not only of pharmacological experiments involving repeated administration of "selective" MAO inhibitors, but also of clinical studies, e.g. in Parkinsonism treated with combinations of an MAO inhibitor (deprenil) and L-DOPA [18, 19]. The clinical use of selective MAO inhibitors, provided they are of the reversible type, might also help to avoid the problem of tyramine potentiation [10] since tyramine, being a mixed substrate [2] could be metabolized by the uninhibited form of the enzyme.

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