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Anorexic Properties of Three Monoamine Oxidase Inhibitors

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WILLIS, G. L. AND G. C. SMITH. *Anorexic properties of three monoamine oxidase inhibitors*. PHARMAC. BIOCHEM. BEHAV. 17(6) 1135–1139, 1982.—The anorexic properties of 3 monoamine oxidase (MAO) inhibitors, pargyline, tranylcypramine and nialamide were determined 24 hr after their intra-peritoneal administration. The duration of anorexia occurring after the peripheral administration of the dose of pargyline commonly used in conjunction with 6-hydroxydopamine (6-OHDA) to enhance its dopamine-depleting properties was also determined. Body weight regulation and the control of food and water intake were severely impaired 24 hr after the administration of the three highest doses of pargyline, tranylcypramine and nialamide employed. Impairment in the ability to regulate body weight lasted for as long as 6 days in some animals injected with 50 mg/kg of pargyline. These findings bring to light the problems in interpretation of results from studies which employ MAO inhibitors and 6-OHDA simultaneously to enhance dopamine depleting properties of 6-OHDA.

Catecholamines MAO inhibitors Consummatory behaviour Catecholamine agonists Catecholamine accumulation

MONOAMINE oxidase (MAO) is an intracellular enzyme which regulates the intraneuronal levels of catecholamines (CA). MAO inhibitors are drugs which decrease the activity of this enzyme and are frequently used in conjunction with the specific neurotoxin 6-hydroxydopamine (6-OHDA) to enhance its dopamine (DA) depleting properties [1, 5, 14]. The suggested mode of action by which this is accomplished is prevention of the rapid deamination of 6-OHDA, thereby prolonging its neurotoxic effect. The degree of MAO inhibition achieved is an important consideration since it has been demonstrated that the dopamine depleting properties of 6-OHDA are potentiated only when MAO activity is inhibited by more than ninety percent [13].

Several investigators have employed this drug combination in an attempt to demonstrate the functional significance of the ascending dopaminergic projections in the regulation of consummatory behaviour. When 6-OHDA treatment is preceded by MAO inhibition both the magnitude and duration of the behavioural deficits are more severe than when 6-OHDA is injected on its own [5, 24, 17, 18]. However, since the peripheral [7] and central [8, 9, 10] injection of CA and their agonists can also inhibit consummatory behaviour, MAO pretreatment may be contributing to the enhanced severity of consummatory deficits seen in such experiments, by temporarily increasing intracerebral CA levels rather than enhancing DA depletion.

Although, in support of this hypothesis, there is some recognition of the anorexogenic properties of the MAO inhibitors [8, 9, 10] it is claimed that they have no effect on food and water intake when they are injected on their own [17, 18]. The object of this experiment was to determine the dose-dependent anorexogenic properties of the 3 MAO inhibitors, at doses commonly used in conjunction with 6-OHDA treatment to enhance DA depletion, and to assess the effects on brain CA, as determined by fluorescence histochemistry.

METHOD

In the first study 120 male Sprague-Dawley rats ranging in weight from 225 to 275 g were housed individually in wire cages and allowed ad lib access to powdered rat cubes (Clarke King, Melbourne) from dishes specially designed to minimise spillage (<1 g/day). Tap water was made available from 100 ml calibrated cylinders attached to the front of each cage. A 12 hr light/dark cycle was maintained throughout the experiment with lights on at 0700 hr. Room temperature was maintained at 22°C (±°C) throughout the experiment. Food and water intake and body weight were measured each day immediately after the onset of the light cycle. Food and water intake and body weight were measured for at least 5 days prior to injection to allow food and water intake to stabilize. The rats were then randomly divided into 12 groups of 10 rats per group. Each of the 3 MAO inhibitors employed was injected at 4 different dosage levels; pargyline HCl (Sigma Chemicals) 15, 30, 50 or 100 mg/kg; tranylcypromine sulphate (Smith, Kline and French) 1, 2.5, 5 or 10 mg/kg; nialamide (Sigma Chemicals) 20, 50, 100 or 166 mg/kg. Each dose was administered to 10 rats and one dose only to each rat. Distilled water was the vehicle used for dissolving all drugs. Nialamide was injected as a suspension by adding a few drops of Tween 80. Animals receiving the same drug were all injected on the same day between 1000 hr and 1100 hr. Twenty-four hours later food and water intake and body

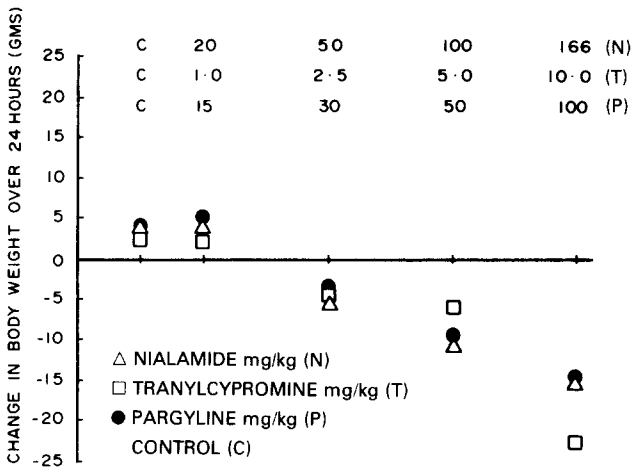

FIG. 1. The mean change in body weight 24 hr after the intraperitoneal injection of various doses of pargyline, tranylcypromine or nialamide.

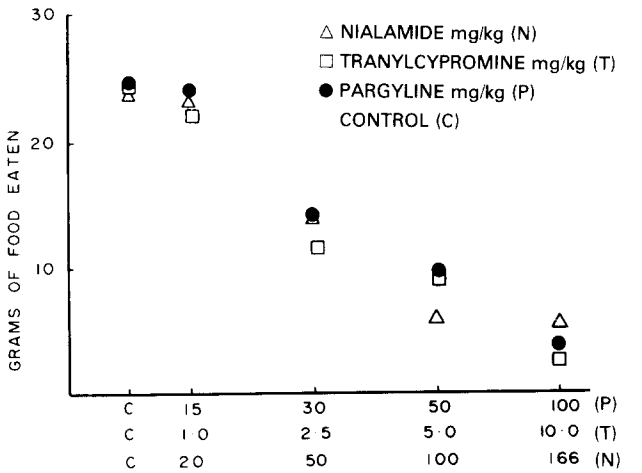

FIG. 2. The mean quantity of food eaten 24 hr after the intraperitoneal injection of various doses of pargyline, tranylcypromine or nialamide.

weight measurements were taken and animals were randomly selected from each drug group, decapitated and prepared for fluorescence histochemical examination using either the method described by Burnstock *et al.* [3], or Furness *et al.* [6]. Evaluation of changes in fluorescence in the hypothalamus and forebrain were made using a single blind method.

In a second experiment 16 male Sprague Dawley rats were maintained as described above and their body weight measured for 5 days. Eight of the animals were then injected IP, with 50 mg/kg of pargyline HCl and the remaining 8 animals were injected IP with 10 mg/kg of the same drug. Body weight of each animal was measured once each day after injection until it returned to the preinjection body weight. All data were analysed using Student's *t*-test.

RESULTS

As shown in Fig. 1 body weight was significantly reduced 24 hr after the injection of 30 ($p<0.02$), 50 ($p<0.001$) or 100 ($p<0.001$) mg/kg of pargyline. Tranylcypromine also produced a significant loss in body weight at the doses 2.5 ($p<0.001$), 5.0 ($p<0.001$) and 10.0 ($p<0.001$) mg/kg. Similar dose-dependent reductions in body weight were observed after the injection of 50 ($p<0.001$), 100 ($p<0.001$) or 166 ($p<0.001$) mg/kg of nialamide. Body weight regulation was not significantly different from that of controls after the injection of 15 mg/kg of pargyline, 1.0 mg/kg or tranylcypromine or 20 mg/kg of nialamide.

Food consumption (Fig. 2) decreased after the injection of 30 ($p<0.001$), 50 ($p<0.001$) and 100 ($p<0.001$) mg/kg of pargyline in a dose-dependent manner. Similarly, tranylcypromine injection reduced food intake at the doses of 2.5 ($p<0.001$), 5 ($p<0.001$) and 10 ($p<0.001$) mg/kg. Nialamide also reduced food intake in a dose-dependent manner at the doses of 50 ($p<0.001$), 100 ($p<0.001$) and 166 ($p<0.001$) mg/kg. Animals injected with 15 mg/kg of pargyline, 1.0 mg/kg of tranylcypromine and 20 mg/kg of nialamide displayed food intake which was not significantly different from that seen in controls.

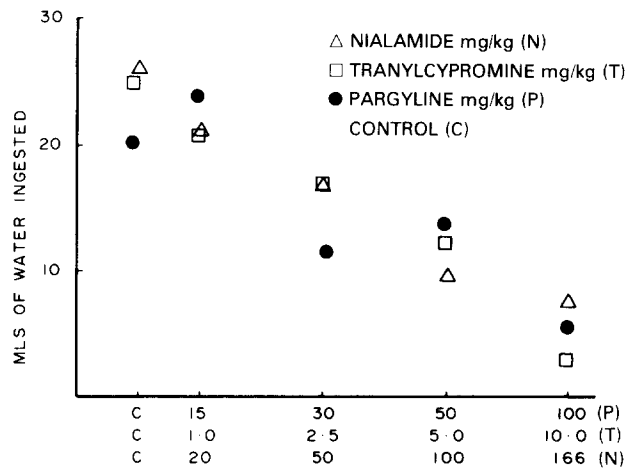

FIG. 3. The mean volume of water consumed 24 hr after the intraperitoneal injection of various doses of pargyline, tranylcypromine or nialamide.

Water intake (Fig. 3) was reduced in a dose-dependent manner after 30 ($p<0.001$), 50 ($p<0.001$) and 100 ($p<0.001$) mg/kg of pargyline. Only the 5.0 ($p<0.001$) and 10.0 ($p<0.001$) mg/kg doses of tranylcypromine significantly reduced water intake. Nialamide injection reduced water consumption at the doses of 50 ($p<0.02$), 100 ($p<0.001$) and 166 ($p<0.001$) mg/kg. No significant changes in water intake were seen after the injection of 15 mg/kg of pargyline, 20 mg/kg of nialamide or 1.0 and 2.5 mg/kg of tranylcypromine.

When brains were examined histochemically dose related increases (Table 1) in the specific fluorescence of numerous

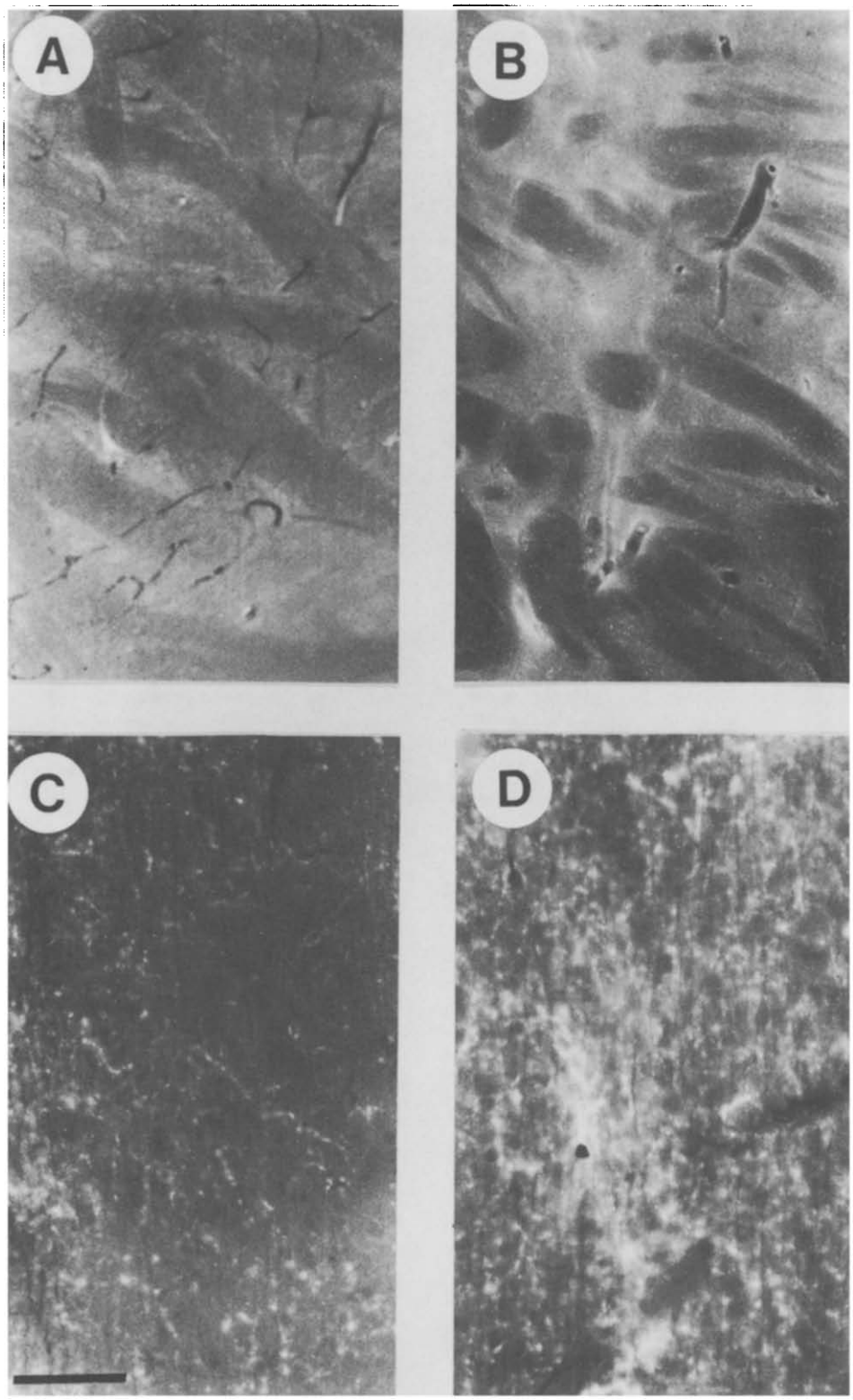

FIG. 4. Fluorescence photomicrographs of amine fluorescence in the forebrain after the intraperitoneal injection of various monoamine oxidase inhibitors. (A) Normal fluorescence of the N. caudatus putamen after 15 mg/kg of pargyline. (B) Increased fluorescence in the N. caudatus putamen after 50 mg/kg of pargyline. (C) Normal fluorescence of the medial aspect of the lateral hypothalamus adjacent to the periventricular nucleus 24 hr after 2.5 mg/kg of tranylcypromine. (D) Increased fluorescence in the medial aspect of the lateral hypothalamus adjacent to the periventricular nucleus 24 hr after 10 mg/kg of tranylcypromine.

TABLE 1

DOSES OF MAO INHIBITOR WHICH PRODUCED AN INCREASE IN THE FLUORESCENCE OF THE STRIATUM AND HYPOTHALAMUS 24 HR AFTER INJECTION

	No Change in Fluorescence Observed	Increased Fluorescence
Pargyline (mg/kg)	15, 30	50, 100
Tranylcypromine (mg/kg)	1, 2.5, 5	10
Nialamide (mg/kg)	25, 50	100/166

TABLE 2

THE NUMBER OF DAYS REQUIRED FOR PARGYLINE TREATED ANIMALS TO RETURN TO THEIR PREINJECTION BODY WEIGHT AFTER RECEIVING AN IP INJECTION OF 10 or 50 MG/KG OF PARGYLINE

10 mg/kg	50 mg/kg
1	4
1	2
1	2
1	1
4	6
1	6
1	5
1	3
mean=1.3 Days	mean=3.6 Days

structures on the hypothalamus and forebrain were observed and are illustrated in Fig. 4.

The mean number of days required for pargyline injected animals to return to their preinjection body weight are expressed in Table 2. While 10 mg/kg of pargyline depressed body weight for slightly more than 24 hr those injected with 50 mg/kg did not return to their preinjection body weight until 3.6 days after injection ($p<0.01$).

DISCUSSION

These results demonstrate that like many other CA agonists [7, 8, 9, 10], the injection of MAO inhibitors can impair the ability to regulate food and water intake and body weight shortly after their injection. Furthermore, impairment in the ability to regulate body weight produced by the injection of 50 mg/kg of pargyline can last for many days after this drug is injected. Although it has been recognized that MAO inhibitors do have anorexogenic potential [8, 9, 10] a thorough study of this problem has not been attempted previously. That MAO inhibitors can cause deficits in consummatory behaviour is important to know since they are commonly used in conjunction with the specific neurotoxin 6-OHDA to enhance its DA depleting properties. When the deficits in consummatory behaviour which occur after 6-OHDA injection are made more severe by pretreating with an MAO inhibitor, it must now be regarded as unclear whether it is the enhanced DA depletion or the combined but individual effects of both the 6-OHDA injection and MAO inhibition which potentiate the resulting behavioural impairment. Although some experimenters allow the animals to recover from the acute treatment effects before behavioural testing [2, 4, 18], many compare the effects of 6-OHDA injection with and without MAO pretreatment during the first few days after injection, when both drugs could be producing their individual anorexogenic effects [2, 7, 11, 17, 18].

The results from this study are compatible with our previous work which describes a role for increased CA in degenerating axons proximal to the site of 6-OHDA injection in producing the behavioural deficits seen after central CA depletion [12, 15, 18]. Potentiation of the anorexic properties of 6-OHDA by MAO inhibition suggests to us that both treatments may work to increase hypothalamic CA levels. The observation that the highest doses of MAO inhibitor employed in this study cause marked increases in catecholamine fluorescence stands as further support for such a hypothesis. This interpretation is supported by other reports which describe anorexia resulting from the injection of CA and CA agonists via a peripheral route [7] or directly into the hypothalamus [8, 9, 10].

The difficulty in explaining an anorexic effect of drugs which both increase [8, 9, 10] and decrease [2, 17, 18] DA activity in the brain has been duly recognised [7]. However, in light of the results from this study and from our previous work [12, 15, 16] it is possible that the common biochemical effect that both DA depleting treatments and DA agonists produce is to cause an increase in the levels of hypothalamic CA. Consequently, assessment of the role of the ascending CA systems has been made purely on the basis of the occurrence of CA depletion when other events such as localized amine build up have been occurring simultaneously and yet have not been considered as a possible contributor to behavioural deficits which commonly result. This demonstrates the importance of taking into account all biochemical changes which any particular experimental procedure produces before ascribing a function to any single neuronal system.

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