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THE EFFECT OF RESERPINE ON THE PRESSOR RESPONSE TO INJECTION OF 2-HALOGENOETHYLAMINES

BY

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Dibenamine-like compounds sometimes caused an increase in blood pressure when injected intravenously in mammals. This response varied with species, with the preparation, and with the structure of the compound. The response became smaller after repeated injections. In spinal atropinized rats injected with hexamethonium 5 mg/kg this pressor effect produced by injection of the adrenaline antagonist, compound AT3 [ethylfluoren-9-yl(2-iodoethyl)amine hydriodide], was reduced by prior treatment for five days with 1 mg/kg reserpine. Acute adrenalectomy also reduced this effect of the adrenaline antagonist; reserpine plus adrenalectomy abolished it. Subsequent injection of adrenaline and noradrenaline restored it.

It is well known that compounds like phenoxybenzamine [benzyl-(2-chlorethyl)(1-methyl-2-phenoxyethyl)amine] antagonize the pressor response to injected catechol amines and lower the blood pressure for prolonged periods. Less attention has been paid to the acute pressor response which may be caused by intravenous injection of the 2-halogenoethylamine itself. A study of this phenomenon may reveal an aspect of the activity of these compounds which is an essential preliminary to their antagonistic effect.

METHODS

Demonstration of the pressor action. Carotid blood pressure was recorded manometrically from the following preparations: (*a*) dogs anaesthetized with 40 mg/kg pentobarbitone sodium intravenously and injected with 5 mg of atropine sulphate subcutaneously, (*b*) atropinized spinal cats, (*c*) atropinized spinal rats with or without injection of 5 mg/kg of hexamethonium hydrobromide and with and without acute bilateral adrenalectomy. Three 2-halogenoethylamines were tested, (2-bromoethyl)ethyl(naphth-1-ylmethyl)amine hydrobromide (SY28 or J11, Graham & Lewis, 1953), (*p*-chlorobenzyl)(2-chloroethyl)ethylamine hydrochloride (JFA8) and ethylfluoren-9-yl(2-iodoethyl)amine hydriodide (AT3, Graham, 1960). In all cases the compounds were dissolved from the crystalline state in minimal volumes of acetone, diluted with neutral saline, and injected intravenously at once in a fixed volume in a dosage of 10 μg/kg of compound AT3 to 1.0 mg/kg of compound J11. For rats this was 0.1 ml. per injection, washed in with 0.2 ml. of saline. Control observations were always made with the same volume of solvent similarly injected. The injections were repeated three times at intervals of 15 min with or without the interspersion of injections of 4 μg/kg adrenaline or noradrenaline (calculated as base) in cats and dogs, 1 μg/kg in rats.

Isolated preparations. Doses of 1 μg to 1 mg of each of the three compounds were injected into the fluid perfusing standard preparations of (*a*) isolated rabbit hearts (each drug was tested on three preparations) and isolated perfused hearts of young cats and (*b*) perfused vessels of the rabbit ear.

Effect of reserpine on the pressor action. Twenty-five male hooded rats of approximately 300 g were treated with 1 mg atropine, a spinal preparation was made and hexamethonium

hydrobromide 5 mg/kg injected intravenously. The effect on the blood pressure of compound AT3 10 μg/kg in 0.1 ml. of solution was recorded and the injection repeated three times. Ten rats treated in this way were subjected to acute bilateral adrenalectomy at the time of making the spinal preparation. Thirty more were pretreated with reserpine 1 mg/kg daily subcutaneously for five days and 10 of them adrenalectomized at the time of making the spinal preparation.

RESULTS

Pressor action. In anaesthetized atropinized dogs the pressor response to injection of any of the three 2-halogenoalkylamines was more dramatic than in spinal cats or rats, despite the greater sensitivity of the latter preparations to injected adrenaline or noradrenaline. Fig. 1 shows a particularly striking example of the pressor response in a dog to injection of compound JFA8. As the response to injected adrenaline or noradrenaline declined or was reversed, so did the response to injected JFA8 alter.

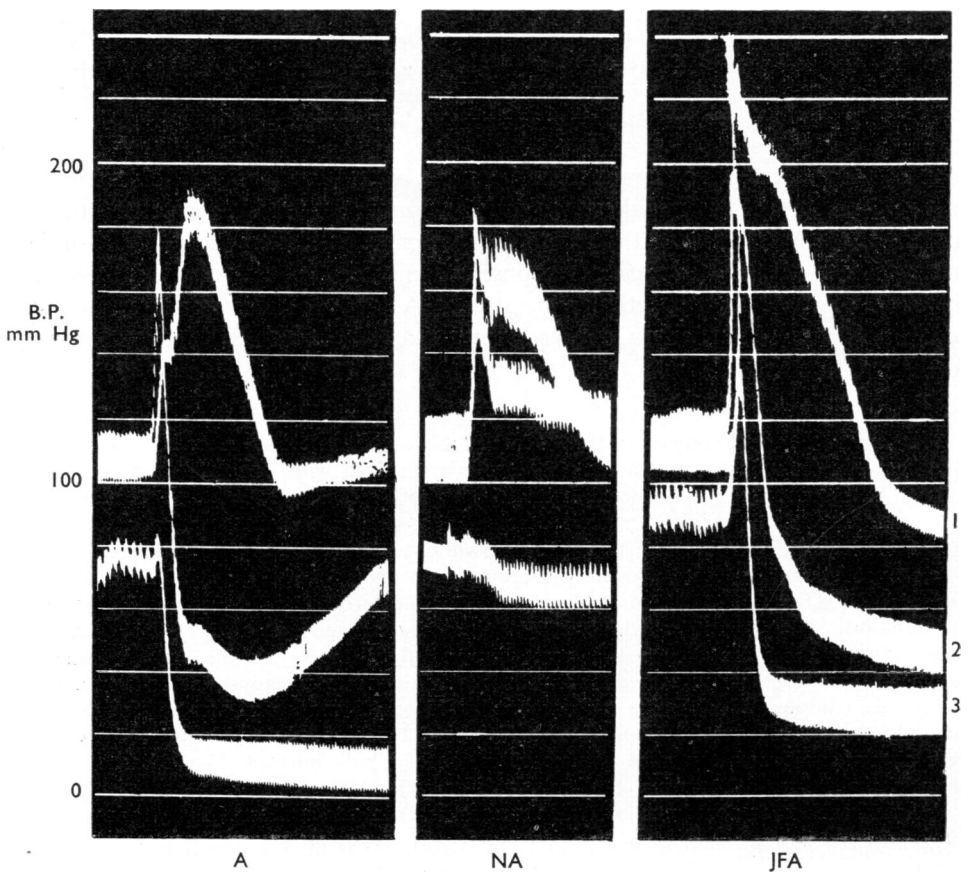

Fig. 1. Dog, 16.3 kg, anaesthetized with pentobarbitone sodium 40 mg/kg intravenously and treated with atropine sulphate 2 mg. Effect on arterial blood pressure of adrenaline (A) 4 μg/kg, noradrenaline (N) 2 μg/kg and compound JFA8 (JFA) 1 mg/kg, injected in that order and repeated twice (1,2,3).

In spinal cats and rats this acute pressor effect was variable. In rats the response to a standard dose of compound AT3 (which is the difference between the pressor response in mm Hg to the paired injections of saline and of compound) was slightly greater when hexamethonium has been given ($0.3>P>0.2$). This may be due to the lower initial level of blood pressure following injection of hexamethonium. In 25 such rats the mean response was 4.7 ± 4.9 mm Hg (range 0–16).

Acute bilateral adrenalectomy reduced the response but did not abolish it entirely in rats given hexamethonium, but if adrenalectomy was performed and hexamethonium not administered there was no detectable pressor response.

Repetition of the injections of 2-halogenoalkylamines resulted in a reduction of the pressor response (see Fig. 1) and in some experiments the response was either abolished or reversed. There was an apparent parallelism between the degree of antagonism of the injected catechol amine exerted by the compound and the reaction of the blood pressure to an injection of the compound itself. Thus the response to injection of compound AT3 for the first time in the 25 rats was a pressor response of 4.7 mm Hg, on the fourth injection it was a depressor response of 2.4 ± 3.2 mm Hg.

Action on isolated preparations. In small doses (10 μg of compound AT3 to 100 μg of compound J11) these compounds caused a transient stimulation of the isolated perfused heart of cat and rabbit. In doses 10 times greater they produced a strong and prolonged depression. When administered in small doses (10 μg of compound AT3, 100 μg of compound J11) they also caused a small and transient constriction of the perfused vessels of the rabbit ear.

Effect of reserpine. Pretreatment of rats with reserpine reduced the pressor response to injected compound AT3 to a very low level, and additional adrenalectomy abolished it. This reduction of the response after reserpine was statistically significant (see Table 1). If adrenaline and noradrenaline were injected into

TABLE 1

PRESSOR RESPONSES IN ATROPINIZED SPINAL RATS

The response was measured before and after hexamethonium hydrobromide (C6) 5 mg/kg, after additional treatment with reserpine 1 mg/kg for 5 days and after subsequent injections of adrenaline and noradrenaline (A–NA) 1 μg/kg

Expt. no.	No. of rats	Treatment	Pressor response to AT3	Difference
1	25	Nil	Present	—
2	25	C_6	Greater	$0{\cdot}2>P>0{\cdot}2$
3	20	Reserpine+C_6	Less	$0{\cdot}01>P$
4	20	Reserpine+C_6+A-NA	Greater	$0{\cdot}02>P>0{\cdot}01$

acutely prepared rats pretreated with reserpine before giving the compound AT3, the effect of the reserpine was not apparent and compound AT3 gave a pressor response. If compound AT3 was injected into acutely prepared rats pretreated with reserpine before and again after this restorative injection of catechol amine, the difference in response was very apparent. Fig. 2 illustrates the recovery of the pressor reaction to compound AT3 in this way, and Table 1 records the degree of statistical significance found in the series of trials performed.

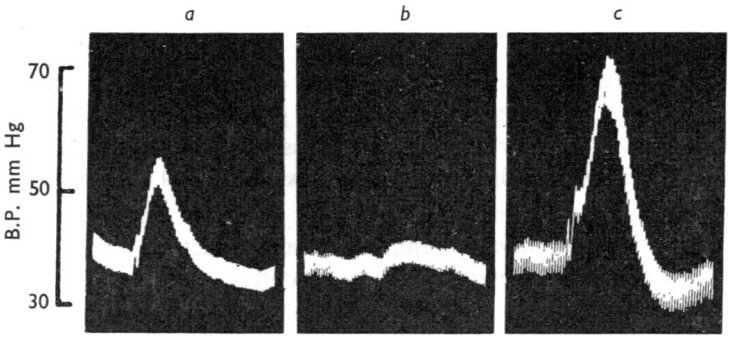

Fig. 2. Pressor responses of spinal atropinized rats to injections of 10 μg/kg of compound AT3 in 0.1 ml. of solvent, washed in with 0.2 ml. of saline. All three preparations received hexamethonium hydrobromide 5 mg/kg. (*a*) Preparation without any other treatment, (*b*) preparation given reserpine 1 mg/kg on 5 successive days, (*c*) preparation given reserpine as in (*b*) and an additional injection of adrenaline and noradrenaline, 1 μg/kg.

DISCUSSION

The transient rise in blood pressure which occurs after injection of 2-halogenoethylamines might be attributed to a central stimulant action affecting the sympathetic nervous system. It may in part be due to a direct cardiac stimulant effect. Millar, Keener & Benfey (1959) have shown that intravenous injection of phenoxybenzamine raises the plasma levels of adrenaline and noradrenaline in dogs and the plasma level of noradrenaline in adrenalectomized dogs, and Benfey, Ledoux & Melville (1959) proved that increased plasma levels of adrenaline and noradrenaline result in an increased urinary excretion of catechol amines. These effects they attributed largely to nervous stimulation, reflex or direct, of adrenal medullary secretion because administration of hexamethonium prevented them.

The three compounds used in the present experiment cause more depression than stimulation of the central nervous system on intravenous injection into mice; the bromo-analogue of compound AT3 is a sedative but is not synergistic in this respect with reserpine (Graham, 1960). The pressor phenomenon is seen in spinal preparations where the influence of central actions is less likely to predominate. The adrenal medulla must be an important source of the pressor effect since adrenalectomy reduces it, but the effect of hexamethonium in the spinal rat is equivocal. These results suggest a direct release of pressor amines from the adrenal medulla while nervous influence is reduced by the ganglion blocker. Brown & Gillespie (1957) and Brown, Davies & Gillespie (1958) have shown that administration of phenoxybenzamine causes a greater release of amine from a tissue when the sympathetic nerve supply is stimulated than takes place without the influence of this 2-halogenoethylamine, while Schapiro (1958) demonstrated an increased urinary excretion of noradrenaline and a loss of catechol amines from the heart, adrenals and spleen after giving this compound.

Reserpine is known to disperse noradrenaline from various tissues including artery walls (Burn & Rand, 1958). The present experiments indicate that the transient pressor response in rats to 2-halogenoethylamine is reduced by treatment with reserpine and restored by infusion of catechol amines.

It may be surmised that 2-halogenoethylamines release catechol amines from tissue stores, including the adrenal medulla, by direct action. This might account for a number of their actions, such as the initial pressor response which may follow injection, the initial increase in sensitivity to injected adrenaline (Graham & Lewis, 1953), the rise in plasma levels of adrenaline and noradrenaline, increased urinary excretion and the fall in tissue content of catechol amines and the reduced excitability of cardiac muscle in response to hydrocarbon anaesthetics.

Presumably the increased catechol amine excreted after treatment with phenoxybenzamine has not been subjected to *O*-methylation.

The relation between possible release of pressor amines by these compounds from the tissue store, occupation of the receptor and the normal inactivation of catechol amine is close.

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