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The effects of corticosteroids on the reduction of α-adrenoceptor blockade produced by β-adrenoceptor blocking agents

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The inhibition of the pressor effects of noradrenaline by phenoxybenzamine was reduced by propranolol and sotalol in the normal and acutely adrenalectomized, but not in the chronically adrenalectomized, rat. The administration of a glucocorticoid, but not a mineralocorticoid, to the chronically adrenalectomized rat, produced a preparation in which the phenoxybenzamine-produced α-adrenoceptor blockade was reduced by the β-adrenoceptor blocking agents, demonstrating that this action is glucocorticoid-dependent.

The reduction of α-adrenoceptor blockade produced by β-adrenoceptor blocking agents *in vivo*, is postulated to be due to the unmasking of residual α-activity not previously blocked by α-adrenoceptor blocking agents (Hull, Eltherington & Horita, 1960; Olivares, Smith & Aronow, 1967; Yamamura & Horita, 1968; Davis, 1971). The actions of β-adrenoceptor stimulants or their antagonists are thought to be mediated through the stimulation or inhibition of 3′5′-AMP production (Sutherland & Rall, 1960; Sutherland & Robison, 1966). In rats, which have been chronically adrenalectomized, the ability of catecholamines to stimulate 3′5′-AMP production is impaired (Brodie, Davies & others, 1966).

This paper presents the results of studies into the effects of bilateral acute and chronic adrenalectomy on the reduction of phenoxybenzamine produced α-adrenoceptor blockade by propranolol and sotalol.

METHODS

Rats (Wistar strain), 180–225 g, were anaesthetized with urethane (lg/kg). The trachea was cannulated and the blood pressure, monitored from the left carotid artery by means of a Condon manometer, recorded on a smoked kymograph. Drugs were injected through a polythene cannula inserted in the left femoral vein. The rats were heparinized with 500 units/kg and drugs were administered in a volume of 0·1 ml and washed through the cannula with 0·1 ml of saline.

Responses to noradrenaline were obtained and phenoxybenzamine (2·5 mg/kg) was injected slowly into the rat in 0·5 ml of saline. The effect of the phenoxybenzamine was allowed to develop over a period of 60 min and the responses to noradrenaline again noted. Propranolol (1·0 mg/kg) or sotalol (5·0 mg/kg) was then administered slowly in 0·25 ml of saline and its effect allowed to develop over 10 min at which time the responses to noradrenaline were again observed.

Acute adrenalectomy

Rats were anaesthetized with urethane and a transverse incision was made in the abdomen just below the diaphragm. The adrenal glands were removed along with

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associated fatty tissue. The abdominal wall was sutured and the preparation set up and the experiment performed as above.

Chronic adrenalectomy

Bilateral adrenalectomy was performed under ether anaesthesia using a dorsal midline approach. The adrenal glands and associated fatty tissue were removed, the abdominal wall sutured and the wound closed with Auto clips. Some chronically adrenalectomized rats were treated with cortisone acetate (6 mg/kg) or deoxycorticosterone acetate (doca) (2·5 mg/kg) given daily by the intramuscular route and commencing immediately after adrenalectomy. The doca-treated rats were maintained on tap water, whilst those which were untreated or treated with cortisone were given 0·9% NaCl solution to drink. Experiments were performed as previously described 3 days after adrenalectomy. Immediately before an experiment, the cortisone and doca-treated animals were given the maintenance dose of the adrenocorticoid.

α-Adrenoceptor blockade

To determine whether acute or chronic adrenalectomy or replacement therapy in chronically adrenalectomized rats modified the α-adrenoceptor blocking activity of phenoxybenzamine, a second series of experiments was performed. The preparation was set up as previously described and a dose response curve to noradrenaline was obtained. Phenoxybenzamine (0·25 mg/kg), a dose which produced only a partial α-adrenoceptor block was then administered and its effect allowed to develop for 60 min, at which time the responses to noradrenaline were again determined.

The drugs used were: propranolol hydrochloride; sotalol hydrochloride; phenoxybenzamine hydrochloride; (−)-noradrenaline bitartrate; cortisone acetate; deoxycorticosterone acetate. All doses are expressed in terms of the base.

RESULTS

The pressor responses produced by noradrenaline in the different preparations are shown in Table 1. The inhibition of the pressor effects produced by phenoxybenzamine (2·5 mg/kg), was partially reduced by propranolol (1·0 mg/kg) or sotalol (5·0 mg/kg) in the normal, acutely adrenalectomized and cortisone treated chronically adrenalectomized rats but was not reduced in the untreated or doca-treated chronically adrenalectomized preparations. The results are shown in Table 1. In the presence of phenoxybenzamine, the responses of the rat blood pressure to 24 and 48 μg/kg noradrenaline were greatly reduced but never totally abolished. In the untreated and doca-treated chronically adrenalectomized rats, the initial doses of noradrenaline administered after propranolol or sotalol in the presence of phenoxybenzamine produced a greater pressor effect than after phenoxybenzamine alone. However, with repeated doses of noradrenaline, the responses decreased in magnitude illustrating the development of tachyphylaxis (see Fig. 1). The responses to noradrenaline shown in Table 1 for the untreated and doca-treated chronically adrenalectomized rats after the administration of propranolol or sotalol are the mean values of three observations in six experiments taken after the first three responses to noradrenaline were discarded. No tachyphylaxis was observed in the responses of the normal, acutely adrenalectomized and cortisone-treated chronically adrenalectomized rats to noradrenaline (see Fig. 1). The data were statistically analysed using the *t*-test ($P < 0.05$). There was a significant difference in the pressor effects produced by

Table 1. *The change in blood pressure (mean ± s.e.) produced by noradrenaline before and after phenoxybenzamine (2·5 mg/kg) and after propranolol (1·0 mg/kg) or sotalol (5·0 mg/kg). (No. of experiments in parentheses.)*

Preparation	Dose (μg/kg)	Responses to Noradrenaline mm Hg			
		Before Phenoxybenzamine	After Phenoxybenzamine	After Propranolol	After Sotalol
Control	1·5	36 ± 1·4	0	0	0
	3·0	49 ± 2·8	0	0	0
	6·0	67 ± 2·5	0	11 ± 0·9	10 ± 1·0
	12·0	92 ± 3·1 (12)	0 (12)	26 ± 0·7 (6)	22 ± 1·8 (6)
	24·0	96 ± 3·4	3·1 ± 0·7	36 ± 1·6	30 ± 1·6
	48·0	101 ± 3·6	7·6 ± 0·9	43 ± 2·3	38 ± 2·2
Acute adrenalectomized	1·5	31 ± 2·0	0	0	0
	3·0	46 ± 3·7	0	0	0
	6·0	64 ± 4·2	0	10 ± 1·0	12 ± 1·6
	12·0	87 ± 4·9 (12)	0 (12)	23 ± 1·6 (6)	20 ± 3·4 (6)
	24·0	95 ± 3·2	3·0 ± 0·7	31 ± 1·6	30 ± 2·9
	48·0	98 ± 2·4	6·0 ± 0·6	44 ± 2·2	39 ± 2·6
Chronic adrenalectomized	1·5	32 ± 2·8	0	0 **	0 **
	3·0	48 ± 3·8	0	0	0
	6·0	65 ± 4·6 (12)	0 (12)	0 (6)	0 (6)
	12·0	84 ± 7·1	0	0	0
	24·0	92 ± 4·2	2·6 ± 0·4	3·0 ± 0·5	2·0 ± 0·6
	48·0	94 ± 3·1	4·0 ± 1·0	7·3 ± 0·9	4·3 ± 0·6
Chronic adrenalectomized + cortisone acetate	1·5	30 ± 2·1	0	0	0
	3·0	48 ± 2·1	0	0	0
	6·0	66 ± 2·8 (12)	0 (12)	12 ± 0·7 (6)	9 ± 1·6 (6)
	12·0	84 ± 6·0	0	23 ± 1·8	16 ± 2·0
	24·0	92 ± 4·2	3·0 ± 0·4	32 ± 1·8	27 ± 2·5
	48·0	97 ± 1·6	6·0 ± 1·0	43 ± 2·4	36 ± 3·1
Chronic adrenalectomized + doca	1·5	35 ± 1·6	0	0 **	0 **
	3·0	52 ± 3·1	0	0	0
	6·0	70 ± 4·8 (12)	0 (12)	0 (6)	0 (6)
	12·0	90 ± 4·6	0	0	0
	24·0	96 ± 2·3	2·0 ± 0·5	2·0 ± 0·5	3·0 ± 0·8
	48·0	101 ± 2·6	7·3 ± 0·8	4·5 ± 0·5	6·5 ± 0·6

** indicates a significant differene in the responses of the test groups compared with those of the control group following β-adrenoceptor blockade. ($P < 0·001$).

noradrenaline after the administration of propranolol or sotalol in the presence of phenoxybenzamine to the untreated and doca-treated chronically adrenalectomized rats compared with those produced in normal rats.

The mean resting blood pressure (mmHg ± standard error) for each group of rats was: control 79 ± 8·0; acute adrenalectomized 82 ± 7·0; chronic adrenalectomized 79 ± 9·0; cortisone-treated chronically adrenalectomized 79 ± 6·5; doca-treated chronically adrenalectomized 84 ± 9·2. After the administration of phenoxybenzamine (2·5 mg/kg) the blood pressures were lowered to: control 37 ± 4·4; acute adrenalectomized 35 ± 3·9; chronic adrenalectomized 40 ± 6·8; cortisone-treated chronically adrenalectomized 37 ± 4·4; doca-treated chronically adrenalectomized 35 ± 3·0, and remained at this lower pressure for the remainder of the experiment. When propranolol (1·0 mg/kg) or sotalol (5·0 mg/kg) (Fig. 1) was administered, the drug produced a rise in blood pressure which was not well maintained, the pressure returning to its original level within 10 min. The mean pressor effect (± standard error) was most marked in the normal 32 ± 4·0; acutely adrenalectomized 30 ± 2·5; and cortisone-treated chronically adrenalectomized rats 35 ± 5·2; and least marked

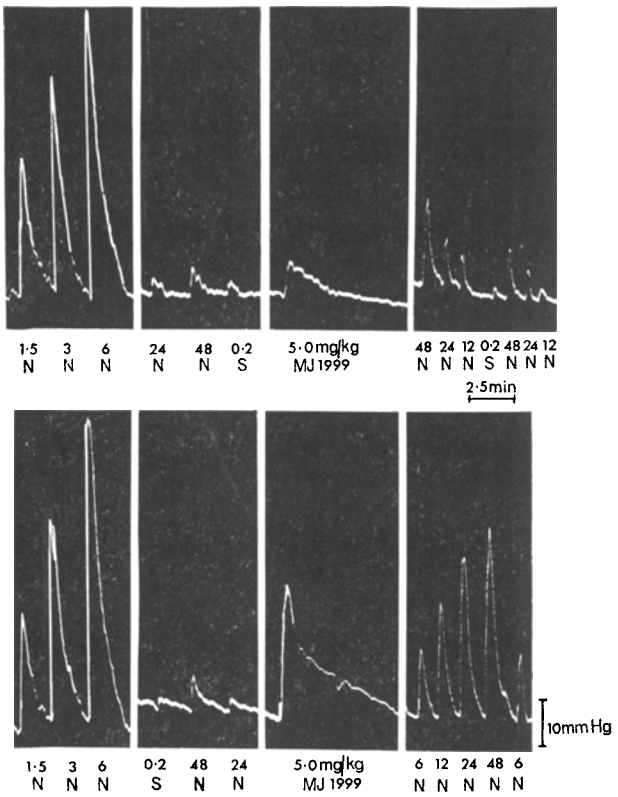

FIG. 1. The upper tracing shows the blood pressure responses of a chronically adrenalectomized rat and the lower, the blood pressure responses of a cortisone treated chronically adrenalectomised rat. The first panel shows the control pressor responses to noradrenaline. The second panel shows the responses to noradrenaline obtained 60 min after the administration of phenoxybenzamine (2·5 mg/kg). The third panel shows the pressor response produced by sotalol (MJ 1999, 5·0 mg/kg) in the phenoxybenzamine treated preparation. The fourth panel shows the pressor responses produced by noradrenaline in the presence of phenoxybenzamine and sotalol. The values for noradrenaline (N) are in μg/kg and for saline (S) in ml.

in the doca-treated 24 $\pm$ 2·5 and untreated chronically adrenalectomized rats 12 $\pm$ 3.

Acute or chronic adrenalectomy or replacement therapy with cortisone acetate or doca to chronically adrenalectomized rats did not modify the partial antagonism of the noradrenaline-elicited pressor effects produced by phenoxybenzamine (0·25 mg/kg). The data were analysed using *t*-test and there was no significant difference between the results of the different preparations ($P < 0{\cdot}05$).

DISCUSSION

In the urethane anaesthetized rat, the antagonism of the pressor effects of noradrenaline by the α-adrenoceptor blocking agent phenoxybenzamine was reduced by the β-adrenoceptor blocking agents propranolol and sotalol. The doses of β-adrenoceptor blocking agents required to produce this effect were similar to those for propranolol (Olivares & others, 1967) and sotalol (Smith & Nash, 1969) used to reduce the effects of phenoxybenzamine in the dog. Propranolol and sotalol reduce the effects of phenoxybenzamine in the acutely adrenalectomized preparation, there being no significant difference between the reduction of α-adrenoceptor blockade in the normal or acutely adrenalectomized preparations. These results indicate that

propranolol and sotalol do not produce the reduced α-adrenoceptor blockade indirectly, by causing the release of catecholamines from the adrenal glands.

The experiments on chronically adrenalectomized rats were performed 3 days after adrenalectomy to allow for the depletion of the corticosteroids (Fortier, 1959). The β-adrenoceptor blocking agents were unable to reduce the α-adrenoceptor blockade produced by phenoxybenzamine. After propranolol or sotalol the response to the initial dose of noradrenaline was potentiated though less than in the normal preparation, but with repeated doses of noradrenaline the responses decreased in magnitude, illustrating a tachyphylactic effect. The inability of the α-adrenoceptor blocking agents to reduce the phenoxybenzamine-produced β-adrenoceptor blockade was not due to an initial decrease in the responsiveness of the blood vessels to noradrenaline or to an increase in the α-adrenoceptor blocking activity of phenoxybenzamine.

The doses of cortisone acetate and doca used were similar to those found by Kuizanga, Nelson & Cartland (1940), to maintain the well being and growth of young rats. Cortisone acetate, a steroid with a marked gluco- and weak mineralocorticoid activity (Dorfman, 1954), given to chronically adrenalectomized rats, restored the ability of propranolol and sotalol to reduce the phenoxybenzamine-produced α-adrenoceptor blockade. Replacement therapy with doca, which has a marked mineralo- and weak glucocorticoid activity (Dorfman, 1954), did not facilitate the reduction of the α-adrenoceptor blockade by propranolol or sotalol and the responses to noradrenaline showed tachyphylaxis similar to that seen in the untreated chronically adrenalectomized preparation. The reduction of phenoxybenzamine-produced α-adrenoceptor blockade produced by propranolol and sotalol is glucocorticoid dependent.

In all preparations pretreated with phenoxybenzamine, propranolol and sotalol produced a pressor effect which was not well maintained, the blood pressure returning to its original level within 10 min. The pressor effect was most marked in preparations in which the reduction of the phenoxybenzamine-produced α-adrenoceptor blockade was obtained. The significance of this pressor effect in relation to the reduction of the phenoxybenzamine produced α-adrenoceptor blockade is not clear. The observation that propranolol produces a pressor effect in the presence of phenoxybenzamine agrees with that of Yamamoto & Sekiya (1969).

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