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Anti-hypertensive Action of 17-Imidazolyl-steroids in the Rat. (23824)

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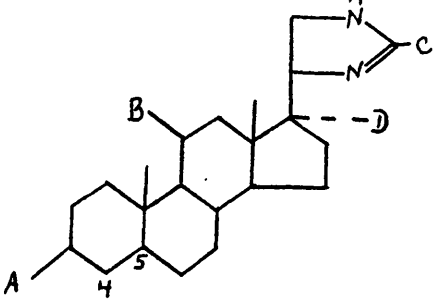
Metacorticoid hypertension is a self-sustaining cardiovascular disease attained in rats some 3 months after the implantation of a desoxycorticosterone acetate (DCA) pellet and the substitution of saline for drinking water. In many respects, this type of hypertension appears similar to essential hypertension in man(1) and responds to treatment with various drugs active in the human disease(2,3). In addition, certain steroids(4-6) and steroid-analogs(7) have been reported to exert transient depressor effects in metacorticoid hypertension. The present report describes similar activity in a new series of compounds, 17-imidazolyl-androstenes(8).

Acute experiments. This study utilized 70 male, Sprague-Dawley rats that had received subcutaneously a 40 mg wax pellet containing 20 mg DCA at least 3 months previously and that had been maintained on 0.86% NaCl solution and Rockland Rat Diet *ad libitum*. Saline and food were not removed during the course of the experiment. The systolic blood pressures of treated rats were estimated without anesthesia or heating using a photoelectric tensometer(9) before and 2, 4 and 6 hours after treatment, and were compared to simultaneous readings from propylene glycol-injected controls. Pressure readings on individual rats were made in ignorance of the injection the rat had received and of the previous pressure reading. Each test usually consisted of 4 treated and 4 control rats run simultaneously by one person on the same tensometer in a darkened room, with injections made by a different person. In order to minimize leakage, subcutaneous injections were always made at least 60 mm from the site of skin puncture with a 20 g hypodermic needle. The following 6 steroids were injected in a total of 35 rats as 1% solutions in propylene glycol at a dosage of 20 mg/kg body weight:



	A	B	C	D	Δ
SC-4200	OH	H	H	H	5
-4225	O	H	CH_3	H	4
-4577	O	H	H	OH	4
-6243	O	H	H	H	4
-6255	OH	H	CH_3	H	5
-6307	O	O	H	OH	4

Table I lists the mean pretreatment blood pressures and the changes observed after treatment with the 6 steroids; all control data are pooled. The probability values were read from Fisher's Table I(10) after applying the rank-sum test(11) to the blood pressure changes. On this basis, significant hypotensive effects were produced only by SC-4200

TABLE I. Blood Pressure Effects of 17-imidazolyl-steroids.

Compound	No. rats	B.P. before inj.	Change in B.P. after inj. 2 hr	4 hr	6 hr
SC-4200	8	188	−22*	−27†	−34‡
-6255	4	192	− 6	− 4	− 7
-6243	4	187	−13	−15	−14
-4225	7	190	−11	−15	−14
-4577	8	187	−26†	−32†	−23†
-6307	4	184	− 2	− 10	−20
Pooled controls	35	186	− 2	− 1	− 1

* P = 0.06.
† P <0.01.
‡ P <0.001 that treated = controls.

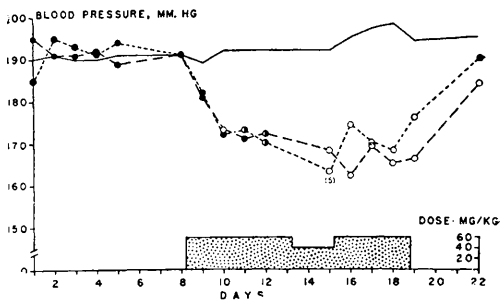


FIG. 1. Effect of SC-4200 (– – –), SC-4577 (----), and placebo (——) on blood pressure of metacorticoid hypertensive rats. Six rats per group unless otherwise noted. Significance of changes in blood pressure, compared to those of controls: P >0.05 (solid circles), <0.05 (halved circles), <0.01 (open circles).

and SC-4577. From consideration of the structures of the compounds, it appears that the methylation of the imidazole ring of SC-4200 decreases activity (SC-6255), while the activity of SC-4577 is reduced by 11-oxidation (SC-6307) or 17-dehydroxylation (SC-6243, SC-4225).

Chronic experiments. Eighteen additional metacorticoid rats were divided into 3 equal groups for the chronic administration of the 2 active compounds, SC-4200 and SC-4577, plus a corn oil-placebo. The experimental conditions and technics were in general the same as in the acute experiments. Six control blood pressure readings were taken over the 8 days before injection; individual rat blood pressures varied over a 26 mm range during this period, at absolute levels over 178 mm. Beginning the 8th day, while the controls received equivalent volumes (2-3 ml/kg/day) of the solvent, the treated groups received either SC-4200 or SC-4577 (2% in corn oil, subcutaneously) in the following doses: 20 mg/kg t.i.d. x 5 days, then 40 mg/kg q.d. x 2 days, and finally 20 mg/kg t.i.d. x 4 days; this was a total dosage of 620 mg/kg in 11 days. Blood pressures were taken 17-18 hours after the preceding injection (Fig. 1). All surviving rats were killed 4 days after the end of the treatment period.

Chronic treatment with either compound significantly lowered blood pressure and there was no apparent difference in potency. Both compounds exhibited a latent period of 3-4 days at the start of treatment and a fairly rapid (4-day) recovery of hypertensive levels after cessation of treatment. One of the SC-4577 rats died after 5 days of treatment; he was found to have severe periarteritis nodosa, nephrosclerosis, and infected lungs and pancreas. Most of the remaining rats had pneumonitis and subcutaneous residues of the compound, but no gross evidence of toxicity was found in any of the treated rats. Moreover, there were no significant losses of body weight and the relative organ weights of the 3 groups did not differ significantly (Table II). The mechanism of hypotensive action of these 2 compounds is unknown, but is not correlated with any glucocorticoid, androgenic, or anabolic activity, as determined in our laboratories by Dr. F. J. Saunders.

Summary. Six imidazolyl-steroids were tested for depressor activity in metacorticoid hypertensive rats. Two compounds were active acutely and chronically: 17β-(4[5]-imidazolyl)-5-androsten-3β-ol and 17α-hydroxy-

TABLE II. Organ Weights at Necropsy, %.*

Group	No. rats	Wt, g	△ Wt, g	B.P., mm	Adrenals	Thymus	Heart	Kidneys	Testes
SC-4200	6	413	+ 7	184	.018	.102	.388	.919	.865
SC-4577	5	391	+ 3	187	.016	.087	.411	.899	.898
Placebo	6	380	+18	197	.019	.098	.384	.830	.915

* P >0.05 that treated = controls.

17(4|5]-imidazolyl)-4-androsten-3-one. Activity was apparently reduced by methylation of the imidazole ring of the former and by 11-oxidation or 17-dehydroxylation of the latter.

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Regeneration of Liver in Lysine-Deficient, Partially Hepatectomized Rats.* (23825)

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Coincident with starvation or maintenance on nitrogen-free diet, there occurs a rapid and extensive loss of nitrogen from liver of rats (1,2). Upon realimentation with protein of adequate nutritional quality, the nitrogen content of liver returns to normal levels in a few days. This accumulation of nitrogen during realimentation of protein-depleted rats has been used for nutritional evaluation of proteins(1,3), of amino acid mixtures and of protein hydrolyzates(4). Similarly, after partial hepatectomy of rats, proliferation of the remaining liver occurs to some extent when they are post-operatively starved(5) or fed protein-free diet(6), and progressively more so as the diet furnishes increased amounts of proteins(6) of adequate nutritional quality(7).

It has been clearly demonstrated that adult rats fed a lysine-deficient amino acid mixture incur loss of appetite and body weight(8), negative nitrogen balance(9) and marked decrease of total circulating serum protein(10). In view of the importance of liver for formation of plasma proteins(11), abnormalities in composition of liver induced by imbalance of dietary amino acids or proteins(12) and increasing evidence that lysine can be a limiting factor in the nutritive value of some diets (13), it was of interest to investigate the effect of lysine deficiency on regeneration of liver.

Methods. Mature female rats of hooded strain, maintained in our colony for many years, weighing initially about 200 g were housed individually in cages with screen bottoms. They had access to food and water *ad libitum*. Records were kept of food consumption and each animal received a daily allotment of the ration (Table I) estimated to exceed slightly the

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