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AUGMENTATION OF THE PRESSOR RESPONSE TO OCTAPRESSIN BY AUTONOMIC BLOCKING AGENTS IN THE PITHED RAT

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The effect of octapressin on blood pressure was studied in anesthetized pithed male rats, pretreated with autonomic blocking agents. Phenoxybenzamine and chlorpromazine induced an augmentation of the blood pressure response to octapressin, whereas atropine, propranolol and hexamethonium had no effect on the response to octapressin in the pithed rat.

It is concluded that phenoxybenzamine and chlorpromazine augment the blood pressure response to octapressin in the pithed rat by a direct effect on the peripheral vascular effector cells.

octapressin
autonomic blocking agents
blood pressure response
pithed rat
norepinephrine-infusion
peripheral vascular effects

1. INTRODUCTION

The mechanism by which autonomic blocking agents augment the pressor response to vasopressin had not yet been elucidated. This potentiating action has been attributed to the blockade of compensatory cardiovascular reflexes, the blocking of β-vasodilator receptors or to a direct peripheral effect on the vascular effector cells (Mantegazza et al., 1958; Prado and Carlini, 1959; Page and McCubbin, 1963).

De Jong and McLeod (1967) have shown that phenoxybenzamine, chlorpromazine, propranolol and atropine enhance the pressor effect of the synthetic lysine-vasopressin analogue octapressin (2-phenylalanine-8-lysine-vasopressin) in the intact rat. Because these authors did not find changes in cardiac output and central venous pressure at the peak of the pressor response to octapressin, it was concluded that the increase in blood pressure following octapressin is caused by an increase in the vascular resistance in the systemic circulation.

It was considered of interest to determine whether direct peripheral effects of these autonomic blocking agents played a role in the enhancement of the pressor response to octapressin. Therefore, the influence of these autonomic blocking agents on the blood pressure response to octapressin was studied in the pithed rat.

2. METHODS

2.1. *General*

Male white rats, weighing 160–230 g, were used. Half an hour before urethane anesthesia (1.25–1.50 g/kg i.p.) the animals of four of the experimental groups were injected s.c. with one of the following blocking agents: atropine sulfate, phenoxybenzamine (Dibenzyline ®), propranolol (Inderal ®), hexamethonium chloride, each at a dose of 10 mg/kg. The fifth group of rats received chlorpromazine hydrochloride (Largactil ®, s.c. 50 mg/kg) half an hour before anesthesia with pentobarbital sodium (35 mg/kg i.p.). These doses expressed in terms of the salt used, were

the same as those used in the previous studies by De Jong and McLeod (1967). Saline served as placebo for the control groups.

In one experiment a group of pithed rats received an infusion of norepinephrine (200–400 μg/hour, 200 μg/ml) into the femoral vein to maintain an approximately normal blood pressure

2.2. *Experimental procedure*

The animals treated as described above, were used in random order. Injections of octapressin (2, 6 and 18 mU in a volume of 0.1 ml) and of saline (0.1 ml) were administered via a lateral tail vein, and washed in with 0.05 ml saline. Doses were given to each animal every 30 min, in random sequence. Generally, 3–6 rats were studied simultaneously.

2.3. *Operative procedure*

Following anesthesia, bilateral cervical vagotomy and sympathectomy were performed, the carotid arteries were tied off and the trachea cannulated. Pithing was performed according to Shipley and Tilden (1947). Immediately after pithing respiration was maintained using a Braun respiratory pump, at 72 strokes/min.

Blood pressure and heart rate were monitored from a carotid artery, as described previously (De Jong and McLeod, 1967). The experiments were started 20–30 min after pithing, when the blood pressure had stabilized.

Body temperature of the rats was maintained between 36–38°C.

2.4. *Statistics*

Results are expressed as mean ± standard error of the mean (S.E.). Statistical analysis of the data was performed using a single variance analysis, followed by Duncan's multiple range test (Duncan, 1955) and a single variance analysis for related observations series with replications (De Jonge, 1964).

3. RESULTS

The combined effects of pithing and treatment with the autonomic blocking agents on blood pressure are shown in table 1. Blood pressure was lowered in the pithed rats. The decrease in blood pressure was slightly inhibited by chlorpromazine-pretreatment, as shown by a significantly ($p < 0.05$) higher blood pressure in response to pithing in this group than that seen in the saline-treated pithed rats. Comparing blood pressures in the pithed rats at the beginning and the end of the experiments showed that the blood pressure tended to rise during the experiments except in the propranolol-treated rats ($p < 0.05$). Following pithing mean heart rate dropped from 415 ± 13 to 325 ± 7 beats/min.

Table 1
Effect of pithing procedure and autonomic blocking agents on blood pressure before and after 3 octapressin injections

Pretreatment	Blood pressure in mm Hg before	after
Intact rats	105 ± 5*	99 ± 5
Pithed + saline	31 ± 4	41 ± 3
Pithed + atropine	38 ± 4	42 ± 3
Pithed + phenoxybenzamine	38 ± 4	41 ± 5
Pithed + propranolol	28 ± 4	28 ± 4
Pithed + hexamethonium	33 ± 3	37 ± 3
Pithed + chlorpromazine	45 ± 3	49 ± 5

* Mean ± standard error of the mean
In each group 9 rats were used

The effect of octapressin on the blood pressure of pithed rats is depicted in table 2. The pithing procedure caused an increased response to 6 and 18 mU octapressin, as compared to that of the intact control group ($p < 0.01$). This increase was also observed in the pithed rats treated with the various autonomic blocking agents. In addition to this augmentation, the responses to the three doses of octapressin were greater in the pithed rats treated with chlorpromazine or phenoxybenzamine than in those treated with saline ($p < 0.01$). The decrease in heart rate following the injection of octapressin in the intact rat was abolished by pithing

Since the increase in the pressor response to octapressin in the pithed rat might be partially related to the low basal blood pressure, the response to octapressin was also studied in pithed rats, which received a continuous infusion of norepinephrine (200–400 μg/hour) to maintain blood pressure comparable to that of non-pithed rats. Systolic blood pressure of the norepinephrine-infused pithed rats was 108 ± 3 mm Hg, while heart rate increased to 453 ± 10/min (intact controls 387 ± 20/min). The increased pressor

Table 2
Effect of octapressin on blood pressure in pithed rats treated with autonomic blocking agents.

Pretreatment	Increase in blood pressure in mm Hg			
	saline	2 mU	6 mU	18 mU
Intact rats	−1 ± 1*	12 ± 2	16 ± 2	34 ± 3
Pithed + saline	2 ± 1	14 ± 5	32 ± 5	77 ± 7
Pithed + atropine	0 ± 2	19 ± 4	38 ± 5	73 ± 8
Pithed + propranolol	3 ± 1	14 ± 3	28 ± 5	60 ± 8
Pithed + hexamethonium	3 ± 0	12 ± 2	31 ± 5	85 ± 3
Pithed + phenoxybenzamine	0 ± 1	25 ± 4	53 ± 4	94 ± 4
Pithed + chlorpromazine	3 ± 2	27 ± 4	59 ± 7	93 ± 5

* Mean ± standard error of the mean.
In each group 9 rats were used.

response to octapressin was observed in both groups of pithed rats, but the response was less in the norepinephrine treated group (table 3). Analysis of the results obtained in pithed norepinephrine-treated rats is complicated by the fact that the injection of 0.1 ml of saline also induced a significant increase in blood pressure ($p < 0.05$).

Table 3
Effect of octapressin on blood pressure of norepinephrine-infused pithed rats.

Pretreatment	Increase in blood pressure in mm Hg		
	saline	6 mU	18 mU
Intact rats	3 ± 3*	26 ± 4	41 ± 5
Pithed + saline	4 ± 1	39 ± 8	75 ± 5
Pithed + norepinephrine	11 ± 1	31 ± 4	60 ± 6

* Mean ± standard error of the mean.
In each group 7 rats were used.

4. DISCUSSION

The present study shows, that the pithing procedure makes the rat more sensitive to the higher doses (6 and 18 mU) of octapressin. Raising the low blood pressure of pithed rats with a norepinephrine-infusion to the level of non-pithed controls somewhat decreased this increased sensitivity. Pretreatment with the autonomic blocking agents chlorpromazine and phenoxybenzamine induced an increased pressor response to 2, 6 and 18 mU of octapressin in the pithed rat, but atropine, propranolol and hexamethonium did not augment the pressor effect of octapressin in the pithed rat. Chlorpromazine pretreatment induced an increase in blood pressure in the pithed rats. We have no explanation for this effect. It does not seem to be related to the effect of chlorpromazine on the octapressin-induced increase of blood pressure.

Enhanced pressor responses to lysine-vasopressin and angiotensin in the pithed hamster have been found by Lembeck and Daum (1967). The pressor response to epinephrine and angiotensin (Shipley and Tilden, 1947) and to norepinephrine (Vanov, 1963) is also augmented in pithed rats. This increased sensitivity can probably be explained by the loss of compensatory cardiovascular reflexes and perhaps also by the low blood pressure. According to the law of the initial value of Wilder (1962) pressor responses to vasoactive agents are related in a reciprocal way to the initial blood pressure. When the blood pressure of the pithed rats was augmented by infusion of norepinephrine to approximately 100 mm Hg, the increased sensitivity to octapressin was less, but still present. The remaining difference can be attributed to the loss of the above-mentioned reflexes. The experiments with the norepinephrine infusion in pithed rats are difficult to evaluate because of the significant pressor response to saline injections in these animals, and the high dose of norepinephrine needed to restore the blood pressure to the level of non-pithed controls.

Propranolol, atropine and hexamethonium did not potentiate the pressor response of pithed rats to octapressin as observed in intact rats (De Jong and McLeod, 1967) following atropine or propranolol.

This probably depends on inhibition of compensatory cardiovascular reflexes, and not on a direct sensitizing effect on blood vessel elements.

Although the pressor response to octapressin was augmented by the pithing procedure, pretreatment with the autonomic blocking agents phenoxybenzamine or chlorpromazine further potentiated the response, indicating a direct sensitizing effect on the vascular effector cells. This is substantiated by microcirculation experiments of Altura (1967) who showed that in the rat, phenoxybenzamine and chlorpromazine prevent the vasoconstriction due to topical or intravenous administration of catecholamines, and even potentiate the effect of octapressin.

In the present experiments a direct peripheral effect on vascular effector cells was found in the pithed rats for phenoxybenzamine and chlorpromazine, both α-adrenergic blocking agents. This effect may be related to their α-adrenergic receptor blocking capacity. Bartelstone and Nasmyth (1965) have implicated cyclic 3', 5'-AMP in the pressor response to vasopressin and catecholamines. It is suggested that an α-adrenergic receptor block as induced by phenoxybenzamine and chlorpromazine may eliminate the stimulating influence of catecholamines on cyclic 3',5'-AMP production and subsequently increase the amount of substrate available, probably resulting in a more sensitive effector system for vasopressin. However, Bartelstone and Nasmyth (1965) have shown that infusions of non-pressor doses of arginine-vasopressin potentiated the pressor responses to catecholamines. Such an effect obtained by non-pressor doses of vasopressin requires only a small amount of available substrate and may differ from the effects of high doses of octapressin as used in the present study. Therefore, this finding does not necessarily contradict our hypothesis.

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