CENTRAL COMPONENT OF THE CARDIO-ACCELERATOR ACTION OF ANGIOTENSIN II IN DOGS

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Summary: Injection of angiotensin II into a lateral cerebral ventricle (I.C.V.) or into a peripheral vein of anaesthetized dog elicited a rise in blood pressure and transient bradycardia followed by sustained tachycardia. Spinal transection at C_2 and bilateral vagotomy abolished the central cardiovascular effect of I.C.V. angiotensin. However, in spinal transected dogs the usual pressor response to intravenous angiotensin was observed. Since the transient bradycardia was absent in bilaterally vagotomized dogs or in dogs with their blood pressure stabilized by means of a mechanical buffer device it must be reflex in origin. The tachycardia was more marked in vagotomized dogs. Prior administration of a beta adrenergic receptor blocking agent propranolol, blocked the tachycardia, but the pressor response was unaffected.

The cardiovascular responses to centrally administered angiotensin were practically abolished by prior treatment of dogs with reserpine or by extirpation of both adrenal glands. Thus it may be concluded that ICV angiotensin induces a centrogenic release of catecholamines from the adrenal medulla which is responsible for the cardiovascular responses.

Key words: Angiotensin II cardioacceleration lateral cerebral ventricle central cardiovascular dog

INTRODUCTION

Angiotensin II exerts a powerful pressor response which has generally been attributed to a direct vasoconstrictor action on vascular smooth muscle (7,8,4). More recently it was demonstrated that angiotensin can exert a neurally induced pressor response which appeared to be due to activation of central sympathetic structures. However, the corresponding effects on heart rate are variable. Marked bradycardia (6,9), no change (18,19) or tachycardia (14,21) was observed when angiotensin II was administrated intravenously.

Accordingly various explanations have been offered for the mechanism of action of angiotensin on the heart rate. A stimulant action of symphathetic centers in the central nervous system was suggested by Nishith *et al.* (21). More recently Krasney *et al.* (14) reported that the cardiac acceleration produced by angiotensin was due to stimulation of adrenergic mechanisms.

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Angiotensin induced bradycardia is generally believed to be of reflex origin, but the mechanism of the cardioacceleration is not yet settled. The present work was undertaken to determine the central component of cardioaccelerator action of angiotensin by localizing the compound in cerebral ventricles of dogs anaesthetized with chlroralose and eliminating the reflex effects by employing mechanical blood pressure stabilizing device. In order to further work out the central mechanism of cardiovascular actions of angiotensin, the effects were studied after surgical interruptions of the neural pathways and bilateral adrenalectomy, as well as after prior treatment with propranolol and reserpine.

MATERIAL AND METHODS

Fifty mongrel dogs of either sex, weighing between 8-16 *kg* were employed in the study. The animals were anaesthetized by injecting 80-120 *mg/kg* of chloralose (British Drug House) intravenously. The animals were maintained on positive pressure artificial respiration. Systemic arterial pressure was recorded from right femoral artery by means of mercury manometer writing on smoked kymograph paper. The heart rate was determined from Lead II of the electrocardiogram. Intravenous administration of drug solutions was done through a polythene cannula inserted into the right femoral vein. In some experiments the blood pressure was stabilized by using a mechanical device "Buffer System", as described by Varma *et al.* (25). The intracerebroventricular (ICV) injections of angiotensin were made through a cannula placed in the lateral cerebral ventricle according to the technique used by Bhargava and Tangri (3). Aspiration of clear cerebrospinal fluid indicated the correct placement of the cannula which was comfirmed at autopsy. To interrupt the neural pathways bilateral vagotomy was done by cutting the vagi high in the neck. Laminectomy was done to transect the spinal cord at C_2 level.

Prior extirpation of the adrenal glands was performed by lumbar route in some experiments and nearly 2 hr were allowed for stablization of blood pressure before angiotensin administration. Catecholamine depletion was achieved in dogs by injecting reserpine (0.5 *mg/kg*/ i.p.) on two successive days. In some animals propranolol (2.5 *mg/kg*), a beta adrenergic receptor blocking agent was administered slowly intravenously 30 min prior to injection of angiotensin. The intravenous dose of angiotensin (Hypertensin CIBA) used was 1 *µg/kg* and the intracerebroventricular (ICV) dose was 4 *µg* (total dose). The volume of drug solution injected into the cerebral ventricle did not exceed 0.25 *ml*.

RESULTS

Intravenous angiotensin (Table I) :

In 14 dogs an intravenous injection of 1 *µg/kg* of angiotensin induced an average maximum increase of 60 ± 11.0 mm Hg in blood pressure and an increase in heart rate of 33±5.7 beats/min. The rise of blood pressure appeared within 20 sec, reached a peak at 1 min and

lasted for about 30 min. Concomitant with the increase in blood pressure there was a transient bradycardia of 15±10.2 beats/min which lasted for only 1 min and was followed by a prolonged tachycardia.

TABLE I: Effects of intravenously administered angiotensin (1 *μg/kg*) on heart rate and blood pressure of dogs.

State of animal	*Number of experiments*	*Max. B.P. change (mm Hg)*	*Initial decrease in heart rate (beats/min)*	*Maximum increase in heart rate (beats/min)*
Normal	14	+60±11.0	15±10.2	33±5.7
Stabilized blood pressure	15	+11± 5.5	None	38±7.8
Bilateral vagotomy	5	+76±13.8	None	37±6.7
Bilateral vagotomy + transected spinal cord (C_2)	5	+58±14.3	None	6±1.5

Repeated administration of angiotensin at interval of every 30 min produced similar magnitude of pressor response and tachycardia. Thus, the factor of tachyphylaxis was excluded in our studies.

In 5 dogs the reflex influences on heart secondary to a rise of arterial blood pressure were eliminated by using a mechanical device "Buffer system" which prevented the rise of blood pressure to a large extent. In these animals intravenous injection of angiotensin (1 *μg/kg*) induced a rise of blood pressure of 11±5.5. mm Hg. However, the increase in heart rate averaged 38 ± 7.8 beats/min. The transient bradycardia was not observed in these dogs. Thus, when the increase in arterial blood pressure was prevented to a large extent, the initial cardiac slowing was abolished. Bilateral vagotomy in 5 dogs, abolished the initial bradycardia without significantly affecting the tachycardia.

In 5 bilaterally vagotomized and spinal cord (C_2) transected dogs, intravenous injections of angiotensin (1 *μg/kg*) elicited the usual pressor response (+58±14.3 mm Hg) but there was insignificant increase in heart rate (6±1.5 beats/min). Initial cardiac slowing was also abolished.

II. Intracerebroventricular (ICV) administration of angiotensin.

a. *Effects of various surgical procedures (Table II):*

Equal volume of saline as a control was injected into a lateral cerebal ventricle of 12 normal anesthetized dogs to rule out the volume effects on the cardiovascular responses. The saline (0.25 *ml*) administration produced a slight increase in arterial blood pressure (21.2 mm Hg) and a transient mild bradycardia of 6 ±1.1 beats/min.

TABLE II: Effects of various procedures on ICV angiotensin induced cardiovascular responses.

State of animal	*Dose of ICV angiotensin (μg)*	*Number of experiments*	*Max. B.P. change (mm Hg)*	*Max. change in heart rate (beats/min)*
Normal	Saline control	12	+2±1.2	— 6±1.1
Normal	4.0	20	+35±9.9	+30±7.6
Stabilized blood pressure	4.0	3	+4	+39
Bilateral vagotomy	4.0	5	+39±4.1	+38±3.0
Bilateral vagotomy + transected spinal cord (C_2)	4.0	4	+0.5±1.0	+0.5±1.0
Bilateral adrenalectomy	4.0	3	+5	+6

Injection of 4 μg of angiotensin into a lateral cerebral ventricle of 20 normal anaesthetized dogs elicited a rise of blood pressure and tachycardia. Effects of a typical experiment are shown in Fig 1. The average maximum increase in blood pressure was 35±9.9 mm Hg and the increase in heart rate was 30±7.6 beats/min. The tachycaridia appeared within 10 sec and lasted for about 30 min. However, in 5 dogs an initial transient bradycardia of 7±3.9 beats/min was observed. This was considered to be merely a volume effect since equal volume of normal saline injected into lateral cerebral ventricle also elicited a similar cardiac slowing.

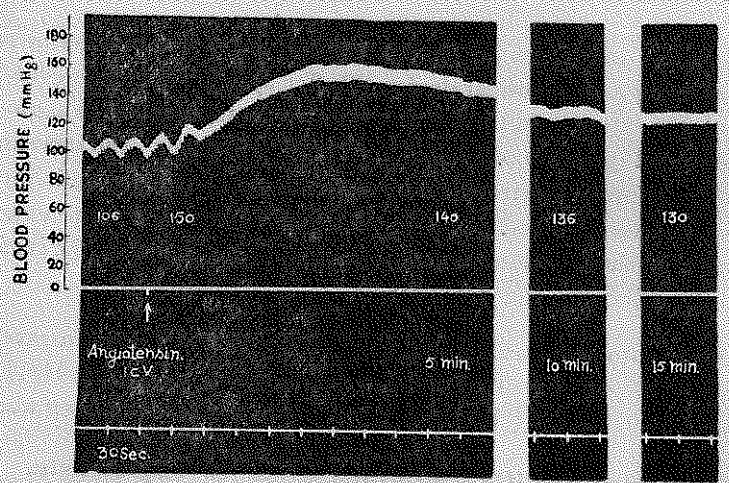

Fig. 1: Time course of pressor response to an injection of angiotensin (4 μg) into the lateral cerebral ventricle of dog. The numbers below the blood pressure tracing refer to the heart rate (beats/min.)

In 3 dogs angiotensin was administered while blood pressure was stabilized by the buffering device. In these dogs the rise in arterial blood pressure was largely prevented and the initial cardiac slowing was also absent. The average increase in heart rate was 39 beats/min.

In 5 bilaterally vagotomized dogs the injection of angiotensin elicited an increase in arterial blood pressure of 39 ± 4.1 mm Hg, and an increase in heart rate of 38±3.0 beats/min. Initial bradycardia was not observed after bilateral vagotomy (Fig 2).

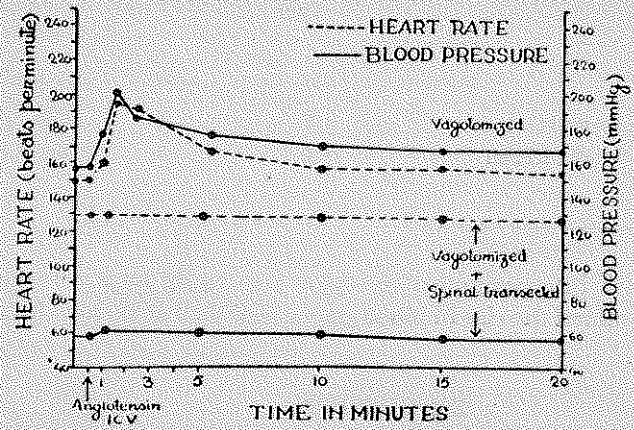

Fig. 2: Effect of angiotensin injection (ICV) on heart rate and blood pressure of 1. Vagotomized 2. Vagotomized spinal cord transected dog.

In 4 bilaterally vagotomized and spinal cord transected dogs, the cardiovascular responses following angiotensin administration were practically abolished (Table II, Fig. 2,3).

In 3 dogs bilateral adrenalectomy was performed. Angiotensin (4 *μg* ICV) in these dogs elicited an insignificant rise (5 mm Hg) of blood pressure and increase in heart rate (6 beats/min). Results of a typical experiment are shown in Fig 3.

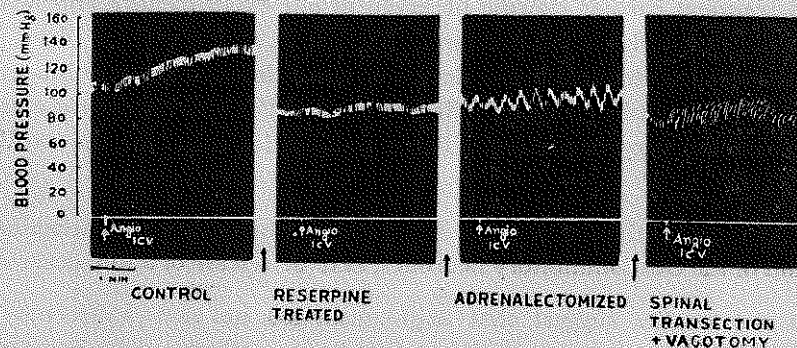

Fig. 3: Effect of angiotensin injection (ICV) on blood pressure of dogs following prior treatment with reserpine, bilateral adrenalectomy and bilateral vagotomy + spinal section.

II. b. *Effects of prior drug treatment (Table III):*

In 3 dogs prior administration of propranolol (2.5 *mg/kg* I.V.), was effective in blocking the tachycardiac response (mean increase of 4 beats/min) to ICV injection of angiotensin. However, the rise in blood pressure was unaffected.

TABLE III: Effects af drug treatment on ICV angiotensin induced cardiovascular responses.

State of animal	*Dose of ICV angiotensin* (μg)	*Number of experiments*	*Max. B.P. change* (*mm Hg*)	*Max. change in heart rate* (*beats/min*)
Normal	4.0	20	+35±9.9	+30±7.6
Propranolol treated (2.5 *mg/kg*)	4.0	3	+38	+4
Reserpine treated (0.5 *mg/kg* I.P. on two successive days)	4.0	5	+6 ±1.6	+2 ±1

Pretreatment with intraperitoneal reserpine (0.5 *mg/kg* for 2 consecutive days) in 5 dogs, to a great extent prevented the rise in blood pressure as well as tachycardia to centrally injected angiotensin (Fig 3).

DISCUSSION

Normally, when the systemic arterial blood pressure is raised, the heart rate is reflexly decreased. In the case of intravenous angiotensin there was always a tachycardia associated with a marked rise in blood pressure, though transient bradycardia often preceded the prolonged tachycardiac response.

The pressor effect has been attributed to a peripheral (7,8) as well as central action of the polypeptide (20,22,24). However, the mechanism of changes in heart rate induced by angiotensin have not been clearly defined.

The initial transient cardiac slowing observed in our experiments seems to be of reflex origin due to a rise in arterial blood pressure. Because in animals where blood pressure was stabilized by the use of mechanical buffer device or when the animals were bilaterally vagotomized intravenous angiotensin (1 *μg/kg*) failed to elicit the initial bradycardia and the heart rate incteased from the very beginning. Three possibilities exist for the cardioaccelerator action of angiotensin:

1. direct myocardial stimulation; 2. central stimulation of the cardioaccelerator neurones and/or inhibition of the cardioinhibitory neurones, and 3. release of a humoral substance.

Results of our studies suggest that angiotensin does not exert a direct action on the myocardium, since its intravenous administration had no effect on the heart rate of spinal transected and vagotomized dogs. Further, Haney *et al.* (12) failed to observe cardiac acceleration following intravenous administration of angiotensin in denervated dog's heart *in situ*. Therefore, the tachycardia induced by angiotensin may be mediated via the nerves innervating the heart. A central cardiovascular action of angiotensin has been suggested by Bickerton & Buckley (5) and Laverty (15). It appears that the polypeptide does cross the blood brain barrier to some extent.

In order to further work out the central mechanism of the cardioaccelerator action of angiotensin, the agent was introduced into the lateral cerebral ventricle of dogs to localize the drug action on the neurones and synapses within the central nervous system. Angiotensin (4 μg) administered into a lateral cerebral ventricle produced a marked pressor response and tachycardia lasting for about 30 min. The occasional transient initial bradycardia observed is of reflex origin since it was abolished by bilateral vagotomy or stabilization of arterial blood pressure. The dose of angiotensin required to produce tachycardia of equal magnitude was much less when administered into the lateral cerebral ventricles than when it was injected intravenously. Also the onset of tachycardia was quicker (within 10 seconds) by the intracerebroventricular route as compared to the intravenous route (20 seconds.) The observations support the contension that cardioacceleration produced by angiotensin is of central origin. The inhibition of cardioinhibitory neurones is not a major factor in the production of cardioacceleration by angiotensin is evident from the observation that tachycardia was not significantly affected by bilateral vagotomy. The rise in the blood pressure and tachycardia induced by centrally administered angiotensin must be central in origin because spinal transection abolished it and that angiotensin must be exciting the central cardioaccelerator neurones. A facilitatory effect of angiotensin on peripheral synapses has been reported by Lewis and Reit (16,17). Furthermore, Bickerton and Buckley (5) and Halliday and Buckley (11) demonstrated that angiotensin induced pressor response in dogs by activation of central sympathetic structures. Evidences for the centrally mediated pressor response of angiotensin is well documented (13,15,20,23,24).

The cardiovascular response of centrally administered angiotensin appears to be primarily due to centrogenic release of catecholamines. This was confirmed by the use of certain pharmacological and surgical procedures. Propranolol, a beta adrenergic receptor blocking agent was effective in blocking only the tachycardiac response to intraventricular injection of angiotensin. The rise of blood pressure was not affected. Pretreatment with reserpine prevented the rise of blood pressure as well as tachycardia to centrally injected angiotensin. Reserpine is known to deplete catecholamines from tissues including the CNS and the adrenal medulla.

Release of catecholamines from adrenals appears to be responsible for tachycardia produced by ICV angiotensin because it was abolished by bilateral adrenalectomy. Benetato *et al.* (2) observed that central administration of angiotensin in cross circulation experiments was capable of releasing catecholamines from the peripheral stores. Furthermore, Feldberg and Lewis (10) have shown angiotensin to cause release of adrenomedullary catecholamines.

Thus we conclude from our studies that angiotensin produces tachycardia by exerting a stimulant action on the central cardioaccelerator neurones and that centrogenic release of adrenal catecholamines is largely responsible for angiotensin induced tachycardia.

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