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CENTRAL CARDIOVASCULAR ACTIONS OF γ-AMINOBUTYRIC ACID

BY

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The vasomotor and cardiac effects of γ-aminobutyric acid (GABA) administered by different routes were studied in cats and dogs. The cat was resistant to the action of GABA administered intravenously or into a vertebral artery. Intrathecal injection of GABA into the cat depressed the vasomotor reponse to spinal compression. In dogs, intravenous, intrathecal or intraventricular injection, or topical application of GABA to the floor of 4th ventricle consistently produced hypotension and depressed the reflex and direct excitability of the vasomotor neurones, located at the supraspinal and spinal levels. Bradycardia observed after intravenous and intraventricular injections of GABA into dogs was abolished by stellate ganglionectomy but not by vagotomy. It has been attributed to depression of the central sympathetic neurones.

Several investigators have reported that intravenous injection of γ-aminobutyric acid (GABA) produces hypotension and bradycardia in anaesthetized dogs, rabbits and guinea-pigs (Takahashi, Tiba, Iino & Takayasu, 1955; Takahashi, Tiba, Yamzaki & Noguchi, 1958; Elliott & Hobbiger, 1959; Stanton & Woodhouse, 1960; Stanton, 1963). In conscious man, also, intravenous GABA has been reported to cause transient hypotension and bradycardia (Elliott & Hobbiger, 1959). The response to intravenous GABA in cats may be hypotension or hypertension, or there may be no effect (Takahashi *et al.*, 1958; Stanton, 1963). Not only species differences but the type of anaesthetic are known to modify the effects of GABA (Elliott & Hobbiger, 1959).

The depressor response to intravenous GABA in the dog is always preceded by a transient pressor response and respiratory stimulation attributed to stimulation of the carotid and aortic chemoreceptors (Stanton & Woodhouse, 1960). Stanton (1963) believes that a peripheral ganglion-blocking action of intravenous GABA is the cause of the depressor response. Takahashi *et al.* (1958) have suggested a central action of GABA on the vasomotor areas in the medulla oblongata. In a preliminary communication from this laboratory a central vasomotor depressant action of intraventricularly administered GABA in dogs has been reported (Bhattacharya, Kishor, Saxena & Bhargava, 1964).

In the present study are reported the cardiovascular effects of GABA administered by intravenous, intrathecal and intraventricular routes into cats and dogs. In a few cats the effects of intravertebral arterial injection and topical application of GABA to the exposed medulla oblongata were studied.

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METHODS

Forty-nine dogs (4 to 17 kg) and eleven cats (1.9 to 3.1 kg) of either sex were employed in the study. For vasomotor studies thirty-three dogs and eleven cats were anaesthetized with pentobarbitone sodium (40 mg/kg, intravenously or intraperitoneally). The animals were vagotomized (except where mentioned) and the trachea was intubated for positive-pressure artificial ventilation. The blood pressure was recorded from a carotid artery by means of a mercury manometer writing on a kymograph. The effect of GABA on the heart rate was studied in sixteen dogs anaesthetized with chloralose (100 mg/kg, intravenously). The heart rate was obtained from the electrocardiogram (lead II) recorded on a Grass Polygraph. The blood pressure was measured with a Statham transducer (P23) and recorded at the same time on the Polygraph.

GABA was administered intravenously through a polyethylene cannula introduced into a femoral vein. Intravertebral arterial injections (into cats) were made according to the technique of Tangri & Bhargava (1960). Intraventricular injections (into dogs) were made through a polyethylene cannula (Bhargava & Tangri, 1959). The drug was administered in a volume of 0.25 ml. followed by 0.25 ml. of 0.9% saline, the total volume of intraventricular injection not exceeding 0.5 ml.

The Horsley-Clark stereotaxic technique in dogs was employed to elicit vasomotor responses by direct electrical stimulation of the vasomotor areas in the medulla. The electrode placement was aided by parameters described by Lim, Liu & Moffitt (1960). The parameters of stimulation varied from 1.5 to 10 V, 1 msec duration, and 30 to 120 shocks/sec for a period of 5 to 10 sec. Rectangular wave pulses were delivered from a Grass Electronic stimulator through a bipolar needle electrode insulated except at its tip.

The spinal cord was ligated at C7 within the meninges. A pressure bottle connected in parallel with a mercury manometer was used for transmitting fluid compression through a hypodermic needle introduced into the theca at the lumbo-sacral articulation. Spinal fluid compression (100 to 150 mm Hg) for a period of 10 to 15 sec consistently elicited a vasomotor response which is initiated from the vasoactive neurones in the spinal cord (Bhargava & Kulsreshtha, 1959, 1960).

Reflex vasomotor responses were elicited by occlusion of the noncannulated common carotid artery for 30 sec and by electrical stimulation of the central cut end of the cervical vagosympathetic trunk. The cut end of the trunk was passed through a 2 cm long polyethylene tube in which silver wire electrodes were embedded for stimulating the nerve. Rectangular wave pulses were obtained from a Grass stimulator (30 to 120 shocks/sec, 1 msec duration, 3 to 15 V for 5 to 15 sec).

Noradrenaline (5 to 15 μg) was used to induce pressor responses as controls for peripheral adrenergic action. The nictitating membrane response to preganglionic nerve stimulation was employed for detecting ganglion-blocking activity.

For stellate ganglionectomy an incision was made in the third intercostal space. The intercostal muscles and pleura were carefully cut. On retracting the lung the sympathetic chain was exposed close to the vertebral column. The stellate ganglion, which lay over the head of the first rib, was removed along with about 3 cm of the chain.

RESULTS

Studies on blood pressure

Cats. GABA injected intravenously (two cats) and into a vertebral artery (three cats) in a dose range of 2.5 to 10 mg/kg produced no significant change in blood pressure or in the reflex pressor response to carotid occlusion.

Dogs (*Table I*). Fig. 1 shows the effects of GABA (1 mg/kg) given to three dogs by intravenous and intraventricular routes. The two left-hand panels show the effect of intravenous GABA in a normal dog. A transient fall and a rise of blood pressure followed by a prolonged hypotension is characteristic. In dogs in which both the

TABLE 1

THE EFFECT OF GABA ON HEART RATE AND BLOOD PRESSURE OF DOGS

I.c.v.=intracerebroventricular ; i.v.= intravenous ; n = number of observations. Values for heart rate and blood pressure are mean changes with standard errors

State of dog	Route	Dose (mg/kg)	n	Heart rate (beats/min)	Blood pressure (mm Hg)
Stellate ganglionec-tomized	I.c.v.	1	1	+30	−15
	I.c.v.	10	5	+24± 4·8	−30± 5·4
Intact	I.c.v.	1	4	−26± 2·8	−16± 3·1
	I.c.v.	10	4	−50±15·2	−29± 7·1
	I.v.	1	5	−41±10·1	−72± 9·0
	I.v.	10	2	−27± 3·0	−18± 2·5
Vagotomized	I.c.v.	1	1	−48	−60
	I.c.v.	10	3	−30± 7·0	−27± 6·7
	I.v.	1	3	−43± 7·5	−70±10·5

carotid sinus nerves and the vagi were cut the response to intravenous GABA was modified (middle panels), in that the transient fall and rise of blood pressure were not observed and the later depressor phase was more prolonged. Injection of GABA through an intraventricular cannula in a normal dog (right-hand panels) produced only a prolonged hypotension similar to the response to intravenous GABA in a dog in which the buffer nerves had been cut. It may, therefore, be concluded that the

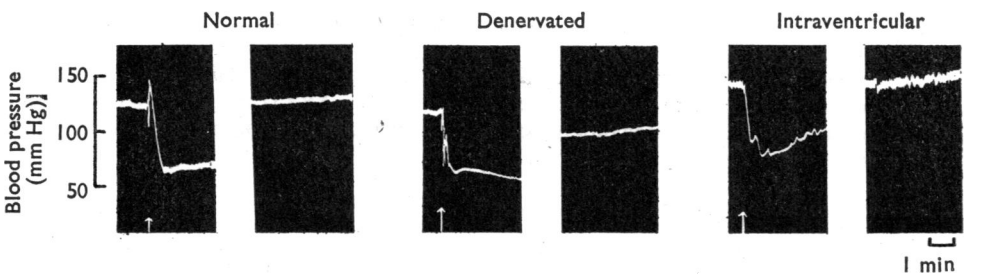

Fig. 1. Arterial blood pressure records of three dogs anaesthetized with pentobarbitone sodium, showing effects of GABA (1 mg/kg, injected at arrows). Left-hand panels : normal dog, GABA injected intravenously. Middle panels : vagotomized dog with carotid sinuses denervated, GABA injected intravenously. Right-hand panels : normal dog, GABA injected intraventricularly.

transient fall and rise of blood pressure on intravenous injection of GABA must be due to peripheral actions, possibly on the aortic and carotid sinus receptors. In sixteen dogs GABA was injected intravenously in doses of 0.01, 0.03, 0.5, 1.0, 2.0 and 5 mg/kg. With the two smallest doses there was only a transient fall in blood pressure. With the other doses there was the characteristic transient fall and rise of blood pressure followed by the prolonged depressor phase. The peak effect was reached within 5 min and gradual recovery occurred in an average period of 60 min (range 30 to 100 min).

Fig. 2 shows the typical effects of intravenous GABA (1 mg/kg) on the pressor response due to carotid arterial occlusion and the response of the nictitating membrane to electrical stimulation of the central cut end of the vagosympathetic trunk. The former response was depressed at 15 min, and recovery occurred at 90 min after injection. The nictitating membrane response was unaffected throughout. The pressor response to noradrenaline (not illustrated) was also unchanged.

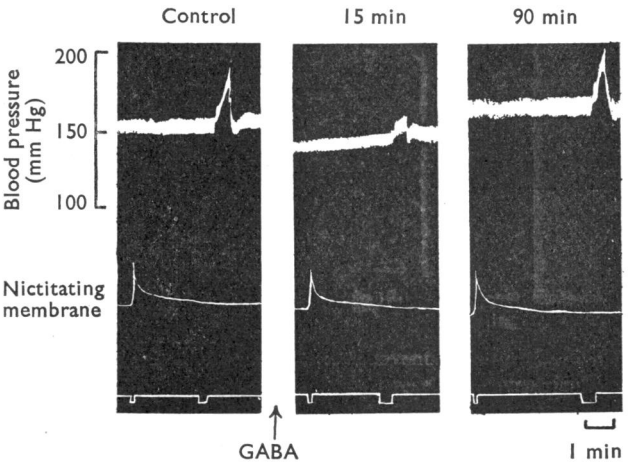

Fig. 2. Bilaterally vagotomized dog, pentobarbitone sodium (35 mg/kg) anaesthesia. Records of blood pressure (uppermost tracing), nictitating membrane contraction (middle tracing) and signal marker (lowest tracing). In the first (control) panel, the first response is of the nictitating membrane to preganglionic stimulation and the second response is to 30 sec of occlusion of a carotid artery. After 15 min from intravenous injection of GABA (1 mg/kg) the blood pressure and the carotid occlusion vasomotor response were reduced. The nictitating membrane response was unchanged. Recovery had occurred at 90 min.

The effect of intraventricular injection of GABA has been studied in twenty-six dogs, in the dose range of 0.1 to 10 mg/kg. There was consistent fall in blood pressure with doses above 0.5 mg/kg. In all experiments, the response was purely hypotensive without a transient rise of blood pressure. The peak effect was reached within 2 min with a slow recovery extending over 1 hr. The effects of intraventricular injection of GABA (1 mg/kg) in a typical experiment are shown in Fig. 3. Here also, the carotid occlusion response was blocked at 15 min and the nictitating membrane response was unaffected. The effect of intraventricularly injected GABA on the carotid occlusion response might be due to a central vasomotor depressant action of the agent.

Fig. 4 demonstrates the central vasomotor depressant action of GABA on pressor responses evoked by direct electrical stimulation, by means of the stereotaxic technique, of the medulla oblongata in a dog. In all panels, the " threshold " and " optimal " pressor responses (first and second responses) were evoked by stimulation with 60 shocks/sec, and 5 and 10 V respectively. The carotid occlusion response (third response) was not prominent in this dog. The uppermost panel shows the control responses and the effects of intravenous GABA are shown in the

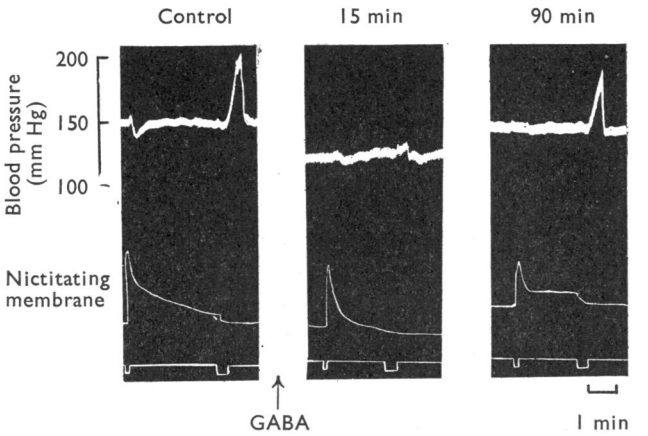

Fig. 3. Record showing the effect of intraventricular injection of GABA (1 mg/kg) on the nictitating membrane response to preganglionic stimulation and the blood pressure response to carotid arterial occlusion in a dog anaesthetized with pentobarbitone sodium and vagotomized. The first panel shows the control responses. In the middle panel 15 min after administration of GABA, blood pressure has fallen and the carotid occlusion response is depressed but there is no change in the nictitating membrane response. The last panel shows the recovery of blood pressure and the carotid occlusion response at 90 min.

middle panel. After GABA (1 mg/kg intravenously) the " threshold " medullary response was blocked whereas the " optimal " medullary response and the carotid occlusion responses were only depressed. Recovery of the responses is shown in the lowest panel. Furthermore, GABA when applied topically to the floor of the 4th ventricle in the medulla oblongata depressed the excitability of the vasomotor centre to direct electrical stimulation.

Effect of GABA on spinal compression vasomotor response

The vasomotor responses evoked by compression of the spinal cord were depressed by intrathecal administration of GABA (0.5 and 1 mg/kg in two cats and 1 mg/kg in one dog).

Studies on heart rate (Table I)

Intraventricular injection of GABA (1 to 10 mg/kg) into eight dogs consistently produced bradycardia and hypotension. Similarly intravenous GABA (1 mg/kg) consistently produced bradycardia and hypotension in five dogs.

Bilateral stellate ganglionectomy, to exclude the sympathetic supply to heart, was carried out in six dogs. Injection of GABA (1 and 10 mg/kg) into a cerebral ventricle produced tachycardia, instead of the usual bradycardia, in five out of six such dogs.

Bilateral vagotomy was performed before administration of GABA. Intraventricular GABA (1 mg/kg, one dog, and 10 mg/kg, three dogs) as well as intravenous GABA (1 mg/kg, three dogs) consistently produced bradycardia as in the dogs with intact vagus nerves.

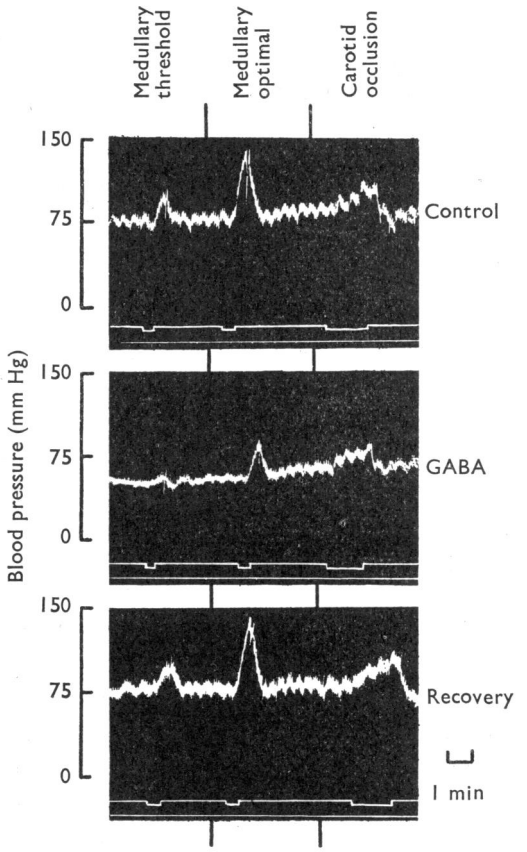

Fig. 4. Record of arterial blood pressure of a dog (vagotomized) showing the effect of intravenous GABA on the direct pressor response to electrical stimulation of the medulla oblongata and on the reflex pressor response to carotid arterial occlusion. In all the panels the first response is the medullary threshold response (5 V, 60 shocks/sec) ; the second is the medullary optimal response (10 V, 60 shocks/sec) ; and the last is the carotid occlusion response (30 sec). The uppermost panel shows control responses. The middle panel 10 min after intravenous GABA (1 mg/kg), shows a fall in blood pressure, inhibition of the medullary threshold response, and diminution of the medullary optimal response and the carotid occlusion pressor response. The lowest panel shows the recovery of all three responses at 30 min.

DISCUSSION

Cats were resistant to GABA administered intravenously or into a vertebral artery. In the dog, however, intravenous GABA produced hypotension and bradycardia similar to the effect of intraventricular GABA. It seems that GABA does penetrate the blood-brain barrier in this species. Similar species variation has been reported by Takahashi *et al.* (1958), Elliott & Hobbiger (1959) and Stanton (1963).

In the normal dog the initial transient fall and rise of blood pressure upon intravenous administration of GABA must be a peripheral action, possibly on aortic and carotid receptors. Stanton & Woodhouse (1960) have also reported a similar action

of intravenous GABA. Intraventricular GABA does not elicit the transient vasomotor effects and these effects are not observed in the dog in which buffer nerves were cut.

The more prolonged hypotensive effect of GABA seems to originate from a central site of action. Recently Stanton (1963) has attributed the hypotension to a transient ganglion-blocking action of GABA. The results of the present study on transmission across the cervical sympathetic ganglion (nictitating membrane experiment) do not support this contention. No evidence of a cervical sympathetic ganglion-blocking action of intravenous GABA could be obtained. No change in the pressor response to noradrenaline was observed after intravenous or intraventricular injection of GABA. This result excludes an adrenergic neurone blocking activity of GABA. The depression of the carotid arterial occlusion response in the prolonged hypotensive phase indicates a central vasomotor depressant action of this agent. This is confirmed by a similar hypotensive effect observed after intraventricular injection of GABA. The central vasomotor depressant action of GABA is further indicated by the inhibition of the pressor responses evoked by direct electrical stimulation of the medullary vasomotor centre. Furthermore, a similar depressant action of intravenous or intrathecal GABA was observed on the spinal compression vasomotor response in the dog with ligated spinal cord (C7).

Inhibition of central sympathetic neurones is probably responsible for the prolonged hypotension and bradycardia produced by GABA. The elimination of the sympathetic supply to the heart (stellate ganglionectomy without vagotomy) eliminates the bradycardia produced by intraventricular GABA. However, it seems that GABA produces nonspecific inhibition of neurones in the central nervous system (Elliott, 1958; Eccles, 1962). The tachycardia observed in the stellate ganglionectomized dog with vagi intact may thus be the result of inhibition of vagal tone.

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