EFFECT OF GAMMA AMINO-BUTYRIC ACID ON SPINAL VASCULAR REFLEX MECHANISMS

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Summary : Stimulation of sciatic nerve produced a rise in arterial blood pressure followed by a fall with an increase in the rate and depth of respiration in dogs under pentobarbitone anaesthesia. Application of cotton swabs soaked with GABA in 0.9% saline to the spinal cord at L_1 and L_2 segments did not change the basal arterial pressure and respiratory rate, but elicited only the pressor effect of sciatic nerve stimulation without changing the respiratory rate and depth. Injection of GABA at the same level in spinal cord had a similar effect. Control applications or injections with normal saline showed no effect. The effects of GABA were temporary and lasted for 45 minutes only. The depressor effect returned earlier than respiratory reflex changes which became prominent with the increase of depressor effect on stimulation of sciatic nerve. Section of spinal cord at the level of L_2 abolished the changes in arterial pressure and respiration on stimulation of sciatic nerve.

Key words : GABA spinal vascular reflexes

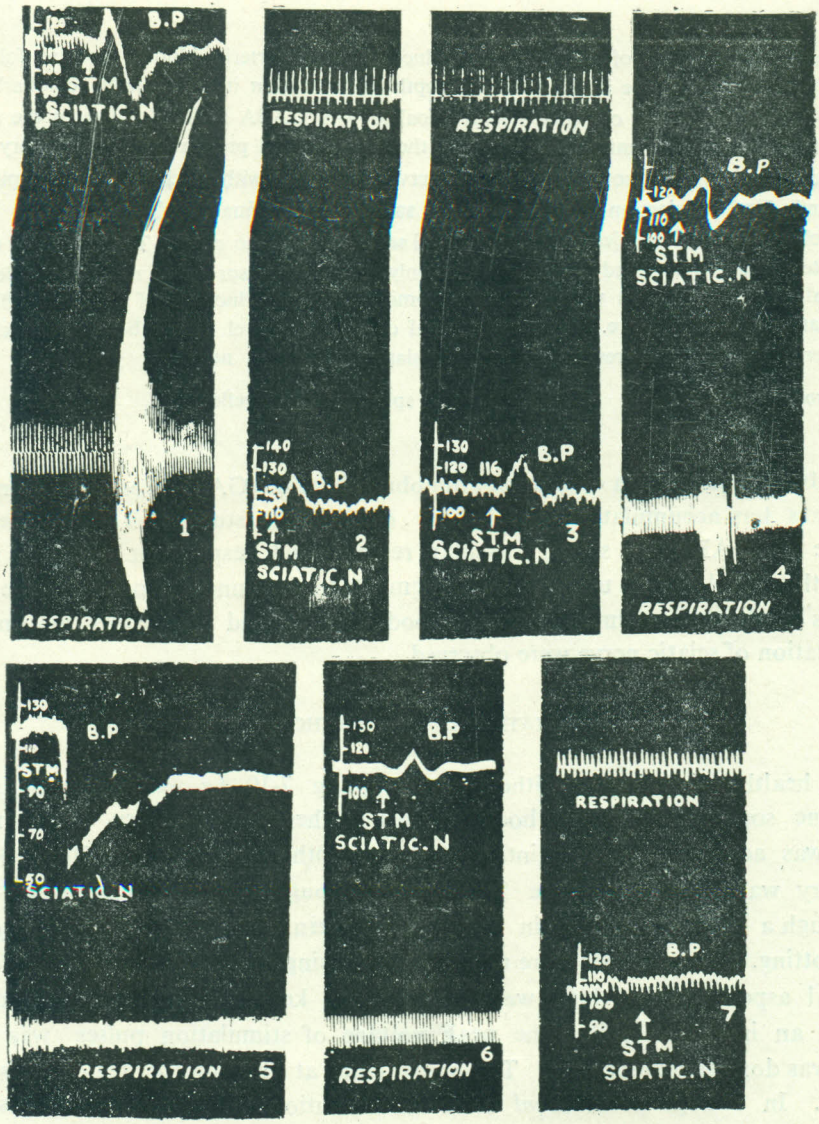
Literature on the action of gamma-aminobutyric acid (GABA) on motor neurones and stretch reflexes has accumulated considerably (1,2). This study was undertaken to observe the influence of GABA on spinal vascular reflexes and respiration. GABA was applied topically on the spinal cord at upper lumbar segments in dogs under pentobarbitone anaesthesia, and its effects on changes in femoral arterial blood pressure and respiratory movements induced by the stimulation of sciatic nerve were observed.

MATERIALS AND METHODS

Stray, healthy adult dogs of either sex, weighing 7-10 *kg* were anaesthetised with i.v. pentobarbitone sodium (30 *mg/kg* body weight). Whenever necessary extra amount of the anaesthetic was administered to maintain the same depth of anaesthesia. Blood pressure of the femoral artery was recorded on a kymograph through mercury manonmeter. Fluids were injected through a cannula inserted in the femoral vein. Heparin (250 units/*kg.*) was used to prevent clotting. Respirations were recorded by passing a tracheal tube. Sciatic nerve in the posterolateral aspect of the thigh was exposed and kept moist to be stimulated with faradic shocks from an induction coil. The peak voltage of stimulation pulses was 6 volts and stimulation was done for 20 seconds. The spinal cord at the lumber region was exposed by laminectomy. In 7 dogs about 2 *ml* of GABA solution (10 $\mu g/ml$ in normal saline at body temperature) soaked in thin folds of cotton was applied all around the spinal cord at L_1 — L_2

after gently raising the spinal cord with glass hooks. In 8 dogs 2 *ml.* of GABA solution was slowly injected in spinal cord at L_1 — L_2 level. The effects of these procedures on sciatic nerve stimulation response were observed.

Complete section of spinal cord was made between L_1 and L_2. After the animal recovered from shock and the blood pressure got stabilised, the effects of stimulation of sciatic nerve were noted again.



RESULTS

Stimulation of sciatic nerve increased the arterial blood pressure, followed by a fall which soon returned to normal. There was increase in rate and depth of respiration but only during the fall of blood pressure (Graph—I).

On application of cotton pledgets soaked in GABA the basal levels of blood pressure and respiration remained constant. Stimulation of sciatic nerve 2 - 5 minutes after the application of GABA resulted in a slight rise in blood pressure without a fall (Graph—2). After 20 minutes this rise in pressure was maximum (Graph—3). In 45 minutes the blood pressure changes due to stimulation of sciatic nerve started to return to the initial pattern (Graph—4). With stimulation of sciatic nerve, though change in blood pressure was noticed throughout, there was no change in rate and depth of respiration after application of GABA. After 30 minutes the respiration gradually changed over to the original pattern. It was observed that the depressor effect of blood pressure returned earlier than respiratory reflex changes, which became prominent with the increase of depressor effect. In about 45 minutes the respiration also returned to the original pattern.

On injecting GABA in spinal cord at L_1 — L_2 there was a transient fall in blood pressure by 30 — 50 mm Hg and there was very slight decrease in depth of respiration while the rate remained almost constant and (Graph—5). Blood pressure increased immediately but remained below the original level. On stimulation of sciatic nerve after the blood pressure had settled, there was a similar change in blood pressure, as in the previous case of applying GABA externally to the cord, though the rise in blood pressure was less prominent. No change in respiration was noticed (Graph—6). Increase in amounts of GABA injected from 2—4 *m* or more into spinal cord did not alter the pattern of response. Repeating the experiments with normal saline instead of GABA showed no effect on blood pressure and respiratory responses. Stimulation of sciatic nerve after the section of spinal cord between L_1 and L_2 did not produce any response (Graph—7). Stimulation was done after the animals recovered from spinal shock which was usually within 30—45 minutes after the section.

DISCUSSION

Gamma-amino butyric acid (GABA) was first identified as a product of biological interest by Ackermann (3), and Ackermann and Kustcher (4). Abderhalden *et al.* (5) later confirmed it and demonstrated it in various bacteria, yeast, other organisms and higher plant tissues. Roberts, Frankel and Harman (6) and Awapara *et al* (7) discovered that the mammalian brain contains large amount of GABA. Curtis and Johnston (8) in their review on aminoacid transmitters have discussed the mode of action of GABA in central and peripheral nervous system especially on motor neurones.

It seems to be a possible inhibitory transmitter in the cerebral cortex and other nervous tissues including the spinal cord, particularly the dorsal horn cells. Florey and McLennan (9)

found that the mammalian brain principle i.e. Factor-I of Florey (10) when applied to the spinal cord of cat abolished monosynaptic tendon jerks completely within 5 seconds while polysynaptic flexor reflex was enhanced. Although GABA apparently mimics the action of Factor–I upon crayfish neuron, it does not seem to have a similar action in a number of other situations. Thus, McLennan (11) and Honour and McLennan (12) noted that topical application of this material on spinal cord even in higher concentrations was without any effect on the knee jerk reflex and on the transmission process through the mesentric ganglion.

The sciatic nerves are known to contain 2 types of afferent fibres which influence the spinal vascular centres differently. In anaesthetized animal one fibre group induces depressor response for relatively low activation and low frequency while another group of fibres with high activation threshold is responsible for pressor effects (13). According to Ranson and Billingsley (14) the fibres transmitting the pressor effect enter the cord in lateral division of posterior roots and end in the tract of Lissauer. These impulses leave the cord by the lateral and anterior rami. The depressor impulses are known to ascend in the lateral column of spinal cord in close relation to the spinothalamic tracts. It must be mentioned that not all the pressor or depressor impulses ascend to the medulla but reflex arcs exist within the spinal cord itself (15).

Injection of GABA into the spinal cord or topical application abolished the depressor component of the reflex while the pressor component appeared within 2—5 minutes. Transection of cord abolished both pressor and depressor effect of stimulation of sciatic nerve. These observations suggested that blood pressure variation due to stimulation of sciatic nerve is not due to an axon reflex but is a reflex of spinal origin. It is possible that GABA inhibits the dilator neurones of spinal cord completely resulting in the elicitation of a pure pressor response on sciatic nerve stimulation.

It was also observed that the respiratory rate and depth increased with the stimulation of sciatic nerve only when there was a fall in blood pressure and not during the rise indicating that the respiratory changes may be secondary due to a reflex initiated by the fall in blood pressure. It is known for example that the fall in systemic pressure can produce hyperpnoea through the excitation of baroreceptors (16). Recent work, however, has centered upon the role played by the sympathetic nervous system in the mediation of circulatory and respiratory responses evoked by somatic afferent nerve stimulation. Biscoe and Purves (17) observed that passive movement of hind limbs of anaesthetised cats caused an impressive rise in sympathetic and chemoreceptor activity associated with increased respiration within 5 seconds of the onset of movement. This reflex reaction was abolished with the section of sciatic nerve, femoral or post-ganglionic cervical sympathetic. Cunningham (18) however, has questioned this aspect of increased ventilation.

The fall in blood pressure after the injection of GABA into spinal cord, is not associated with increased ventilation. This may be due to its blocking action on ascending

ways to the supra-segmental areas affecting the sympathetic discharge or the respiratory res. For the reflex change in respiration due to hypotention the integrity of reflex ciation between the spinal neurones and supra segmental chemo or baroreceptor activity haps is necessary,

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