SEROTONIN INVOLVEMENT
IN THE BLOCKADE OF
BULBOSPINAL AND RECURRENT INHIBITION
OF THE
MONOSYNAPTIC REFLEX

by

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ABSTRACT

The monoamine uptake blocking agents, imipramine HCl (5 mg/kg i.v.) and desipramine HCl (4.8 mg/kg i.v.), and the monoamine oxidase inhibitor, pargyline HCl (30 mg/kg i.v.) antagonized bulbospinal inhibition (BSI) of the monosynaptic reflex (MSR) in unanaesthetized cats decerebrated at the mid-collicular level. The effect of imipramine was quantitatively more on BSI of the quadriceps (QUAD)-MSR compared to that on BSI of the posterior biceps-semitendinosus (PBST)-MSR. Imipramine's action on this inhibition was also quantitatively greater compared to that of the equimolar dose of desipramine. Pretreatment of the animals with the tryptophan hydroxylase inhibitor, DL-p-Chlorophenylalanine (p-CPA) (300 mg/kg i.p. for 3 consecutive days) completely eliminated the blocking action of imipramine. However, pretreatment of the animals with the tyrosine hydroxylase inhibitor, DL-α-Methyl-p-tyrosine methyl ester HCl (α-MPT) (126 mg/kg i.p. given 16 and 4 hours before the recording) had no effect on imipramine's action. These findings strongly suggest that a 5-hydroxytryptamine (5-HT, serotonin) system antagonizes BSI of the MSR. They do not support the proposal of Clineschmidt and Anderson (1970) that the bulbospinal inhibitory pathway involves a 5-HT interneurone in the spinal cord.

Imipramine HCl (5 mg/kg i.v.) and pargyline HCl (30 mg/kg i.v.) blocked recurrent inhibition (RI) of the MSR evoked by stimulation of a dorsal root. Imipramine blocked RI of the QUAD-MSR but had no effect on RI of
the PBST-MSR. Pretreatment of the animals with either p-CPA or \( \alpha \)-MPT prevented the blocking action of imipramine on RI. Application of a 'cold block' which potentiated RI of the QUAD-MSR also eliminated the blocking action of imipramine on this inhibition. These observations suggest that a supraspinal monoaminergic system which involves 5-HT and noradrenaline links has a tonic inhibitory effect on RI of the QUAD-MSR.

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INTRODUCTION

Recently, several investigators have suggested that the putative neurotransmitters in the central nervous system (CNS), 5-hydroxytryptamine (5-HT, serotonin) and noradrenaline (NA) are involved in motor control (Cranmer et al., 1959; Fuxe, 1965; Anden et al., 1966; Anderson, 1972).

Chronic transection of the spinal cord depletes 5-HT and NA caudal to the transection (Carlsson et al., 1963; Magnusson and Rosengren, 1963). Stimulation of the spinal cord gives rise to a release of 5-HT and NA (Anden et al., 1964, 1965). These observations suggest that the spinal cord contains descending 5-HT and NA fibres of supraspinal origin.

The raphe nuclei are located along the mid-sagittal plane in the midbrain, pons and medulla oblongata (Taber et al., 1960). Histochemical (Dahlstrom and Fuxe, 1964, 1965) and pharmacological (Fuxe, 1965) studies indicate that almost the entire 5-HT neuronal population in the CNS is located in the raphe nuclei. Neurones containing NA are most densely located in the medulla oblongata but also exist in the pons and midbrain. Noradrenergic cells have a more scattered distribution than 5-HT cells.

Most of the descending 5-HT and NA fibres originate in the medulla oblongata (Dahlstrom and Fuxe, 1965). These monoaminergic fibres descend in the dorsolateral and ventromedial funiculi of the spinal cord and terminate in the substantia gelatinosa in the dorsal horn and on the ventrolateral and dorsolateral motor nuclei in the ventral horn (Fuxe, 1965).
Existence of the descending monoaminergic systems raises the question of their functional significance in the spinal cord. In unanaesthetized cats with an acute spinal transection, L-tryptophan, 5-hydroxytryptophan (5-HTP) and L-3,4-dihydroxyphenylalanine (L-dopa) increased the MSR (Anderson and Shibuya, 1966; Baker and Anderson, 1970a). Pretreatment of the cats with pargyline potentiated the effects of 5-HTP, L-tryptophan and L-dopa on the MSR (Anderson et al., 1967). In cats with a chronic spinal transection, pargyline and L-tryptophan did not alter the MSR (Shibuya and Anderson, 1968). Moreover, imipramine, a 5-HT neuronal uptake blocking agent, potentiated the effect of 5-HTP and pargyline on the MSR in cats with an acute spinal transection but not in cats with a chronic spinal transection (Clineschmidt et al., 1971; Clineschmidt, 1972). Pretreatment of the animals with the tryptophan hydroxylase inhibitor, DL-p-chlorophenylalanine (p-CPA), blocked the effects of 5-HTP on the MSR (Taber, 1971). These observations indicate that a descending 5-HT system in the spinal cord has a facilitatory action on the MSR.

Bulbospinal inhibitory systems were also suggested to exist. In decerebrate-decerebellate cats, stimulation in the nucleus raphe magnus produced long latency negative dorsal root potentials (DRPs) in the lumbosacral spinal cord. These DRPs were blocked by the 5-HT antagonists, methysergide and cinanserin (Proudfit and Anderson, 1973). Similarly, bulbospinal inhibition of the MSR, as first described by Magoun and Rhines (1946), was also blocked by the 5-HT antagonists, methysergide, d-lysergic acid diethylamide (LSD), 2-bromo-LSD (BOL) and cinanserin (Clineschmidt and Anderson, 1970). These authors found, using close-arterial injections to the brain stem and the spinal cord, that LSD and methysergide act at the spinal cord level. Hence, they suggested that
the bulbospinal inhibitory system contains a 5-HT interneurone in the spinal cord.

To investigate further the possibility that the latter bulbospinal inhibitory system contains a 5-HT link it is reasoned that, if the hypothesis of Clineschmidt and Anderson (1970) is correct, drugs which increase the availability of 5-HT in the synaptic clefts should increase bulbospinal inhibition (BSI) of the MSR. Imipramine preferentially blocks the uptake of 5-HT whereas desipramine preferentially blocks the uptake of NA (Ross and Renyi, 1969; Carlsson et al., 1969; Shaskan and Snyder, 1970). In the spinal cord, pargyline elevates the levels of 5-HT but does not significantly increase the levels of NA and the effects of this drug on the MSR are very likely mediated through 5-HT (Anderson et al., 1967). Therefore, in the present study, imipramine, desipramine and pargyline are tested on BSI of the MSR.

Stimulation of the ventromedial mesencephalic reticular formation, ventrolateral bulbar reticular formation, ventral thalamus (Fields of Forel and Zona Incerta) and pericruciate cortex decreased the rate of Renshaw cell discharges evoked by antidromic activation of the motor axons (Koizumi et al., 1959; Haase and Meulen, 1961; Mac Lean and Leffman, 1967). In a similar study by Haase and Meulen (1961), stimulation of the anterior lobe of the cerebellum was found to facilitate the Renshaw cell discharges. These findings indicate that the Renshaw cell discharges are influenced by supraspinal inputs.

In decerebrate cats, imipramine blocked recurrent inhibition (RI) of the MSR (Von Tan and Henatsch, 1968). However, this drug had no effect on the inhibition in cats with the spinal cord transected at the thoracic level (Von Tan and Henatsch, 1969). These authors suggested that a mono-
aminergic pathway has a strong curbing effect on RI of the MSR. The pre-
sent investigation is an extension of the above work and is designed to
determine whether RI of the flexor and extensor MSRs are equally affected
by imipramine and whether the drug's effects are mediated through a
supraspinal 5-HT or NA system.
Organization of the Lumbosacral Spinal Cord of the Cat

The cell bodies of the lumbosacral afferent fibres are located in the dorsal root ganglia. All central processes of these cells enter the spinal cord. The peripheral processes innervate various structures such as skeletal muscles, skin, etc., and convey impulses centrally. These fibres are classified into various groups according to their thickness, myelination and function. Group I afferents are 12 - 20 \(\mu\)m in diameter and are heavily myelinated. These fibres are subclassified into Ia and Ib afferents. The Ia fibres convey impulses from the annulospiral endings of the muscle spindles, whereas, the Ib fibres convey impulses from the Golgi tendon organs. The conduction velocity of the group I fibres is high (about 120 m/sec) compared to that of the other fibres. Group II (diameter 6 - 12 \(\mu\)m) and group III (1 - 6 \(\mu\)m) fibres are called high threshold muscle afferents. They are thinly myelinated and their conduction velocity is fairly low (about 5 - 30 m/sec). The thinly myelinated cutaneous alpha (6 - 17 \(\mu\)m) and delta (1 - 6 \(\mu\)m) fibres convey impulses from cutaneous origin. Both muscle nerves and cutaneous nerves contain unmyelinated group IV fibres (Ruch and Patton, 1960).

Rexed (1952, 1954, 1964) subdivided the spinal gray matter into nine laminae and a tenth region surrounding the central canal. This type of subdivision of the cell groups is based on the appearance of the cells and the boundaries are only zones of transition. Lamina I is a thin sheath that caps the surface and bends around the margins of the dorsal horn.
Lamina II is situated immediately ventral to the lamina I. It contains tightly packed small cells and is traversed by many strands of spinal afferent fibres. This lamina corresponds to the substantia gelatinosa. Lamina III is a band of large neurones scattered across the dorsal horn and is situated ventral to lamina II. Lamina IV is the broadest of the first four layers. It is a heterogenous zone with small to large cells of variable shapes. Lamina V is a broad zone extending across the neck of the dorsal horn and is subdivided into medial and lateral zones. Many fibres pass through the lateral zone to give it a reticulated appearance. Lamina VI is located at the base of the dorsal horn. It is subdivided into the small medial and the larger lateral zones. The medial zone is less compact and contains large triangular or star shaped neurones. Descending supraspinal pathways project to cells in the lateral zone. Lamina VII occupies most of the intermediate zone of the gray matter. The ventral part of this lamina extends into the ventral horn. Medium sized cells predominate in this zone. The dorsal nucleus of Clarke, the intermediolateral and intermediomedial nuclei are seen in this layer. The ventral part of this layer includes Renshaw cells, interneurones and γ-motoneurones. Lamina VIII contains a heterogeneous mixture of small and medium sized cells with scattered large neurones; and is not sharply separated from lamina VII. Lamina VIII is confined to the medial part of the ventral horn. Vestibulospinal and reticulospinal fibres, and the medial longitudinal fasciculus terminate in this zone. Lamina IX is composed of the largest cells of the spinal cord, the α-motoneurones, situated in the ventrolateral region of the ventral horn. The medial nuclear masses have a diffuse border with lamina VIII.

A considerable number of the thickly myelinated afferent fibres terminate in the dorsal nucleus of Clarke (Grant and Rexed, 1958). The
collateral branches of the afferent fibres that are distributed to parts of the anterior horn become concentrated mostly in the central part of the lamina VI. These collaterals in numerous small bundles pass into lamina IX where they arborize about the soma and dendrites of the motorneurones (Sprague and Ha, 1964). The dorsal root fibres also give off collaterals which pass into lamina VII. Since, collaterals of dorsal root fibres passing to lamina VII and IX traverse broad regions of lamina VII, some fibres end upon interneurones in this zone, as well as on dendrites of motor nuclei which extend beyond the limits of lamina IX (Sprague and Ha, 1964). Group Ib, II and III muscle nerve afferents and cutaneous afferents generate synaptic potentials in the central parts of laminae V, VI and VII (Sprague and Ha, 1964).

The motoneurones are organized according to their functional innervation. The cells that innervate the extensor muscles are located in the ventrolateral horn lateral to those that innervate the flexor muscles. The flexor motoneurones are arranged in a number of subgroups such that each group of cells innervate muscles which move a particular joint lie in the same horizontal plane in the ventral horn; the more distal the muscle, the more dorsal the position of the cells (Romains, 1964).

The Segmental Monosynaptic Reflex

When a spinal dorsal root is stimulated with minimal intensities of current, a small ventral root discharge can be observed. As the stimulus strength is increased, the amplitude of the early ventral root discharge increases and reaches a maximal value and upon further increase in the stimulus strength late discharges are observed. The early sharp spike reflects the synchronous activation of α-motoneurones through Ia afferents and is referred to as the monosynaptic reflex (MSR). The late
asynchronous discharges reflect the firing of motoneurones through polysynaptic reflex (PSR) pathways mediated by activation of high threshold afferent fibres. The latency between stimulation of the dorsal root and recording the MSR from the ventral root includes the conduction time in the Ia fibres, one synaptic delay and the conduction from the motoneurone cell bodies to the ventral root recording electrode. The total time to the peak of MSR is about 2 msec. The central delay in transmission across a single synapse is approximately 0.5 msec (Eccles, 1961).

The predominant feature of the afferent fibres is divergence. That is, a single afferent fibre branches and participates in firing of many motoneurones. However, for a motoneurone to fire, many presynaptic knobs impinging on the motoneurone must be activated. The amplitude of the MSR elicited by stimulation of a dorsal root is an index of the number of motoneurones that are recruited into the discharge zone. But, many motoneurones are depolarized only to a subthreshold level and are said to be excited subliminally. These motoneurones do not contribute to the amplitude of the MSR. During the process of facilitation, however, the motoneurones that are excited subliminally and those that are not excited by the test stimulus will be available for recruitment into the discharge zone. Hence, as more motoneurones are recruited, the amplitude of the MSR increases. During inhibition of motoneurones, the excitability of these cells is reduced and they are eliminated from the discharge zone, hence, the amplitude of the MSR decreases.

Postsynaptic Inhibition in the Spinal Cord

Postsynaptic inhibition is an index of depression of the neuronal excitability which occurs independently of the excitatory synaptic activity (Brock et al., 1952; Coombs et al., 1955a, b). This process involves
inhibition of a neurone by direct synaptic impingement. Examples of post-synaptic inhibition include reciprocal (direct, Ia) inhibition (Lloyd, 1941) and recurrent (antidromic) inhibition (Renshaw, 1941). Reciprocal inhibition is exerted by Ia afferents activating inhibitory interneurones which impinge on α-motoneurones of antagonistic muscles (Eccles et al., 1956). Recurrent inhibition is brought about by volleys in the motor axon collaterals which activate inhibitory interneurones known as Renshaw cells, which in turn, inhibit the motoneurones (Eccles et al., 1954).

The membrane potential of a mammalian motoneurone is about -70 mV and is called the resting membrane potential (RMP) (Frank and Fourtes, 1955). During inhibition of motoneurones a hyperpolarization of the motoneurone membrane occurs which is known as the inhibitory postsynaptic potential (IPSP) (Brock et al., 1952). The equilibrium potential for the IPSP is about -80 mV (Brock et al., 1952).

During reciprocal inhibition, the IPSP in the motoneurone is observed about 1.5 to 2.0 msec after its onset. The decay of the IPSP is about 3.0 msec (Brock et al., 1952a). During hyperpolarization the inhibitory transmitter increases the membrane permeability to ions having a hydrated diameter less than 1.4 times that of potassium ion. Thus, there is said to be a decrease in membrane resistance or an increase in membrane conductance. It was suggested (Coombs et al., 1955a, b) that during the IPSP there is an inward diffusion of chloride ions and an outward diffusion of potassium ions through the neuronal membrane. However, Lux et al. (1970), Lux (1971) and Llinas and Baker (1972) proposed that the IPSP is generated by a selective permeability increase to chloride ions in the outward direction. They also reported that a potassium ion permeability change is probably not significantly involved in this process.

When the membrane potential is lowered (depolarized), the IPSP
increases, the IPSP decreases or reverses when the membrane is hyperpolarized by passage of cathodal current. The IPSP may also be reversed following an iontophoretic injection of chloride ions into the neurone (Coombs et al., 1955a).

a. Reciprocal Inhibition

Discharges in group Ia afferents not only excite the motoneurones of the synergistic muscles but also inhibit the motoneurones of the antagonistic muscles through an interneurone. The inhibitory transmitter released from the Ia inhibitory interneurone axon terminal is suggested to be glycine (Werman et al., 1968; Curtis et al., 1968). When glycine was administered iontophoretically in the vicinity of the motoneurone, this amino acid hyperpolarized and reduced the resistance of the neuronal membrane. Prior hyperpolarization of the membrane reduced or reversed the effect of glycine. Comparison of the equilibrium potentials of the ionic events associated with the IPSP and the hyperpolarization produced by glycine indicate that they were similar (Werman et al., 1968; Curtis et al., 1968). Strychnine, which blocks the Ia inhibitory pathway, specifically blocks the effects of glycine on the motoneurone (Curtis et al., 1971).

Hultborn et al. (1971) found that impulses in the motor axon collaterals inhibit the interneurones of the Ia inhibitory pathway. Thus, they suggested that increased motoneuronal firing inhibits the discharges from the Ia inhibitory interneurones to the antagonistic motoneurones. Hultborn and Udo (1972) have shown that the disynaptic inhibitory effects on the motoneurones from the descending cortico-, rubro- and vestibulospinal tracts involve the Ia inhibitory interneurones. Thus, there seems to be a convergence of supraspinal and Ia afferent inputs exciting the
Ia inhibitory interneurones.

b. Recurrent Inhibition of the Spinal Motoneurones

Volleyes in the motor axon collaterals synaptically activate the inhibitory interneurones, Renshaw cells, which inturn impinge on motoneurones. Renshaw cells, activated by ventral root stimulation, discharge in a characteristic burst with an initial frequency of greater than 1000 spikes per second (Eccles et al., 1954). The reason for such a high frequency of firing of these neurones was attributed to a convergence of excitatory input from the collaterals of many motoneurone axons (Eccles et al., 1956a; Ryall et al., 1970). When synaptically activated, the duration of Renshaw cell discharge is about 50 msec (Eccles et al., 1956a).

Ryall (1970) found that in cats anaesthetized with chloralose, antidromic volleys in the motoneurone axons may evoke a postsynaptic inhibition of Renshaw cells instead of excitation. The latency observed for this inhibition suggested that the effect is brought about by a disynaptic pathway and the other interneurone involved in this pathway was found to be a Renshaw cell (Ryall, 1970). Thus, some Renshaw cells inhibit the discharges of other Renshaw cells and this is probably responsible for recurrent facilitation. Renshaw cells are also involved in inhibiting the Ia inhibitory interneurones as discussed in the previous section, but there seems to be no input from Ia inhibitory interneurones to Renshaw cells (Ryall and Piercey, 1971). Stimulation of ipsilateral afferent fibres in the group II and III muscle nerves and cutaneous afferents have excitatory input to Renshaw cells through polysynaptic chains. However, stimulation of the same afferents on the contralateral side brought about inhibition of these cells without excitation (Ryall and Piercey, 1971). Renshaw cells are more strongly excited by the collate-
rals of large phasic $\alpha$-motoneurone axons than the small tonic motoneurone axons. However, the small tonic motoneurones rather than the large phasic motoneurones are more effectively inhibited by Renshaw cells (Ryall et al., 1972).

Several pharmacological and physiological studies indicate that the chemical transmitter released at the motor axon collateral-Renshaw cell synapse is acetylcholine (Curtis and Ryall, 1966a, b, c). Dihydro-$\beta$-erythroidine, which blocks cholinergic transmission at the nicotinic receptors, depressed the response of Renshaw cells to synaptic stimulation (Eccles et al., 1956a; Curtis and Ryall, 1966b). Eserine, an anticholinesterase drug, greatly prolonged the discharges of Renshaw cells induced by synaptic excitation (Eccles et al., 1954, 1956a). Intra-arterial injection of acetylcholine or nicotine excites Renshaw cells (Eccles et al., 1956a; Curtis and Ryall, 1966a). The excitatory action of acetylcholine, but not nicotine, is increased by eserine while dihydro-$\beta$-erythroidine decreased the excitatory action of both the substances (Eccles et al., 1956a). The inhibitory transmitter released at the Renshaw cell-motoneurone synapse was suggested to be glycine (Eccles, 1966; Werman et al., 1968; Curtis et al., 1968; Curtis, 1969; Curtis et al., 1971).

**Presynaptic Inhibition in the Spinal Cord**

Presynaptic inhibition in the spinal cord was first shown by Barron and Matthews (1938). Stimulation of a dorsal root produces a potential difference along the stimulated or the adjacent dorsal root. The potential developed is called negative dorsal root potential (DRP). The DRP represents a primary afferent depolarization (PAD), and generation of this DRP in the spinal cord induces a net inhibitory action on the motoneuronal output due to depression of the presynaptic excitatory impulses.
(Renshaw, 1946; Brooks et al., 1948).

Powerful presynaptic inhibition can be obtained in cats anaesthetized with pentobarbital and usually this inhibition is less intense in decerebrate cats (Eccles et al., 1963b). In decerebrate cats volleys in Ia and Ib afferents of flexor muscle nerves depolarize group Ia fibres of both flexor and extensor muscle nerves. But group Ia and Ib volleys in extensor muscle nerves, except those of the quadriceps muscle, do not have any depolarizing effects on the flexor or extensor Ia afferents (Eccles, 1964). There was no effect from Ia afferents of any muscle nerves on group Ib (Eccles et al., 1963a) and cutaneous afferents (Eccles et al., 1963c). Group Ib and II volleys from all muscle nerves and cutaneous volleys exert a presynaptic inhibition on group Ib afferents of flexor or extensor origin. Of those, the most powerful effect was found to be from group Ib afferents (Eccles et al., 1963a). Cutaneous volleys to a greater extent and group Ib and II volleys from muscle nerves to a lesser extent produce a DRP on the cutaneous afferents (Eccles et al., 1963c).

In producing presynaptic inhibition on Ia afferents by activating Ia and Ib afferents, there is a latent period of about 4 msec between the conditioning stimulus and onset of the PAD. The PAD reaches its maximum at about 20 msec and persists for about 200 msec or more (Eccles et al., 1962). Since the synaptic delay in the mammalian central nervous system (CNS) is about 0.5 msec (Eccles, 1961), it was postulated that the central delay during presynaptic inhibition involves transmission through at least two serially arranged interneurones (Eccles et al., 1962).

Eccles (1963, 1964) suggested that the PAD is due to a chemical synaptic transmitter released near the primary afferent terminal from the last interneurone in the chain. He also postulated that depolarization of the presynaptic terminals reduce the magnitude of the potential in
these terminals, thereby limiting the output of the synaptic transmitter (Eccles, 1963).

Picrotoxin (Eccles et al., 1963a) and bicuculline (Curtis et al., 1970, 1971a) were found to reduce the DRP. Since these agents selectively block the effects of \( \gamma \)-aminobutyric acid (GABA), this amino acid was proposed as the neurotransmitter mediating the PAD (Eccles, 1964a). Consistent with this proposal, Barker and Nicoll (1972) showed a sodium ion-dependent depolarizing action of GABA on the primary afferent terminals and a specific blockade of its effect by bicuculline in the isolated spinal cord of the frog.

Recently, Krnjevic and Morris (1972) showed that the negative DRP produced in the lumbar spinal cord of the cat during stimulation of the spinal afferent fibres is associated with an increase in the extracellular potassium ion concentration. These authors suggested that this rise in the extracellular potassium ion concentration is either due to the activity of the unmyelinated nerve terminals or to a release of this ion from the postsynaptic structures.

The Raphe Nuclei

The raphe nuclei are situated along the mid-sagittal plane in the midbrain, pons and medulla oblongata (Taber et al., 1960). These cells are separated from other cellular aggregations by fibre masses. The rostral end of the raphe complex is found in the rostral mesencephalon and the caudal end in the caudal half of the medulla oblongata. Depending upon the location and type of cells, the raphe nuclei are classified as follows: nucleus raphe obscurus, nucleus raphe pallidus, nucleus raphe magnus, nucleus raphe pontis, nucleus centralis superior, nucleus linearis intermedius and nucleus linearis rostralis (Taber et al., 1960).
The nucleus raphe obscurus is located mid-sagittally in the caudal medulla. This nucleus extends rostrally upto the caudal pole of the inferior olivery complex. In the dorsoventral plane, the nucleus is situated primarily on the dorsal side. The cells are medium to small in size. The nucleus raphe pallidus is situated ventral to nucleus raphe obscurus and extends from the level of facial nucleus to the caudal medulla, slightly rostral to the caudal end of raphe obscurus nucleus. The nucleus raphe pallidus is bordered by the pyramidal tract ventrally and divided into dorsal and ventral masses at the caudal part.

The nucleus raphe magnus extends from the rostral end of the nucleus pallidus to the level of trapazoid body. The raphe magnus consists of a comparatively large mass of cells which extend laterally into the reticular formation. At many levels, these cells are separated from the reticular formation by longitudinally running fibres. The nucleus raphe pontis occupies the mid-sagittal part of the pons; however, some cells are found more laterally. Most of the cells of the nucleus raphe pontis are separated by dorsoventrally running fibres.

The nucleus centralis superior is bordered by the decussation of brachium conjunctivum in the dorsolateral plane and the nucleus interpeduncularis is situated ventral to these cells. The nucleus raphe dorsalis is situated in the ventral part of the periaqueductal gray and extends from the level of the dorsal tegmental nucleus to the caudal pole of the occulomotor nucleus. Ventral to the nucleus raphe dorsalis, the medial longitudinal fasciculus is situated on either side. The nucleus linearis intermedius is composed of scattered cells of small size located in the caudal midbrain. The nucleus linearis rostralis includes cells of large, medium and small size and is situated medial to the occulomotor outflow.
The Descending Monoaminergic Fibres in the Spinal Cord

In rabbits, after chronic spinal transection at the second thoracic level, both 5-hydroxytryptamine (5-HT) and noradrenaline (NA) levels decrease caudal to the transection (Carlsson et al., 1963; Magnusson and Rosengren, 1963). In mice and frogs, stimulation of the spinal cord in vitro induces a release of 5-HT and NA (Anden et al., 1964, 1965). Thus these authors concluded that 5-HT and NA are associated with the descending neuronal pathways.

Using histochemical fluorescence techniques, Dahlstrom and Fuxe (1964, 1965) showed the existence of monoaminergic neurones in the CNS of rats, guinea pigs, rabbits and cats. They found two distinct types of nerve cells showing either yellow or green fluorescence. The cells exhibiting yellow fluorescence are medium to large in size and round to oval in shape. The distribution of these cells that give rise to descending axons is almost entirely limited to the caudal raphe nuclei (Raphe obscurus, raphe pallidus and raphe magnus). The cells showing green fluorescence are scattered as small masses composed of neurones small to medium in size, multipolar and round to oval in shape. Of these, cells that give rise to descending fibres are located mostly in the medulla oblongata from the rostral end of the inferior olivery complex to the level of pyramidal decussation.

Pharmacological studies (Fuxe, 1965) indicate that cells showing yellow fluorescence are those containing 5-HT whereas those exhibiting green fluorescence are those containing NA. The evidence is based on the following observations: The monoamine depleting agent, reserpine, almost completely removes both the yellow and green fluorescence; the monoamine oxidase (MAO) inhibitor, nialamide, enhances the yellow fluorescence; and the tyrosine hydroxylase inhibitor, α-methyl-DL-tyrosine, selectively
reduces the green fluorescence.

Fluorescence microscopic studies revealed that 5-HT and NA nerve terminals in the region of raphe nuclei make contacts with cells of each other as well as with some nonfluorescent cells (Fuxe, 1965). The 5-HT axons from the caudal raphe nuclei run in a ventrolateral direction and almost reach the ventral surface of the brain stem lateral to the pyramidal tract (Dahlstrom and Fuxe, 1965). Spinal lesion studies revealed that the 5-HT fibres descend in the dorsolateral and ventromedial funiculi of the spinal cord (Dahlstrom and Fuxe, 1965). Some of these fibres cross to the other side of the spinal cord. Most of the fibres descending dorsolaterally in the lumbosacral spinal cord terminate in the substantia gelatinosa, and the ventromedially descending fibres terminate in the ventrolateral and dorsolateral motor nuclei of the lamina IX. In cats, the density of termination of 5-HT fibres in substantia gelatinosa and the motornuclei is almost the same.

The NA fibres also run ventrally in the brain stem to reach the ventrolateral part. In the spinal cord, these fibres descend in the ventrolateral and dorsolateral funiculi with some fibres crossing to the opposite side. The NA terminals were found to be most dense in substantia gelatinosa and dense in the ventrolateral and dorsolateral motor nuclei of lamina IX. However, regions other than the above also receive NA terminals (Fuxe, 1965). Both the descending 5-HT and NA fibres are unmyelinated and of 0.3 to 1.0 μm in diameter (Dahlstrom and Fuxe, 1965).

The observation that monoamine neurones of the caudal brain stem give rise to descending pathways raises the question of their functional significance. In unanaesthetized cats with an acute spinal transection, 5-hydroxytryptophan (5-HTP) (75 mg/kg i.v.), L-tryptophan (100 mg/kg i.v.) and L-3,4-dihydroxyphenylalanine (L-dopa) (30 mg/kg i.v.) increased the
MSR to 310 %, 172 % and 212 % of the control levels respectively (Anderson and Shibuya, 1966; Baker and Anderson, 1970a). The 5-HT levels in the spinal cord of the cat increase after treatment with 5-HTP (Anderson and Shibuya, 1966). Four hours after the injection of pargyline (30 mg/kg i.v.) the levels of 5-HT were elevated by 70 % but the NA levels were not significantly increased in the cat's spinal cord (Anderson et al., 1967). Pargyline also increased the MSR and its effects are blocked by the 5-HT antagonists but not by α-adrenergic blocking agents; thus, the effects of this MAO inhibitor on the MSR are very likely mediated through 5-HT (Anderson et al., 1967). However, pretreatment of cats with pargyline potentiated the effects of L-dopa as well as 5-HTP and L-tryptophan on the MSR (Anderson et al., 1967). But, the action of L-dopa on the MSR was blocked by the 5-HT antagonists (Anderson and Banna, 1968).

In cats with a chronic spinal transection, pargyline and L-tryptophan did not alter the MSR; however, 5-HTP still enhanced the MSR in these animals (Shibuya and Anderson, 1968). In the spinal cord, caudal to a chronic transection, 25 % of the control 5-HT levels (Shibuya and Anderson, 1968), 20 % of the dopa decarboxylase activity (Anden et al., 1964) and 17 % of the tryptophan hydroxylase activity remained (Clineschmidt et al., 1971a). Thus, Shibuya and Anderson (1968) suggested that the 5-HTP enhancement of the MSR in cats with a chronic spinal transection might be due to the presence of 5-HT interneurones in the spinal cord. However, evidence exists to show that 5-HT synthesis from 5-HTP can occur extraneuronally (Kuhar et al., 1971). Furthermore, 5-HTP can either enter the adrenergic terminals and displace catecholamines (Ng et al., 1972) or directly activate the adrenergic receptors (Innes, 1962). These findings offer alternative explanations for the work of Shibuya and Anderson (1968) which shows a 5-HTP enhancement of the MSR in chronic
spinal animals. Clineschmidt et al. (1971) and Clineschmidt, (1972) showed that the neuronal uptake blocking agent, imipramine, potentiated the effect of 5-HP and pargyline in cats with an acute spinal transection but not in the animals with a chronic spinal transection. Thus, a descending 5-HT system exists in the spinal cord of the cat whose overall effect on the MSR is facilitation.

Pretreatment of the cats with the tryptophan hydroxylase inhibitor, DL-\(\beta\)-chlorophenylalanine (\(\beta\)-CPA) (300 mg/kg i.p. for 2 consecutive days) blocks the effects of 5-HP on the MSR (Taber, 1971). Hence, Taber suggested that, although 5-HP was converted into 5-HT in these animals, 5-HT is taken up by the empty synaptic vesicles but did not overflow into the synaptic cleft and activate the receptors. The increase of the MSR induced by 5-HP in cats with an acute spinal cord transection was also blocked by the following 5-HT antagonists: methysergide, \(d\)-lysergic acid diethylamide (LSD), 2-bromo-LSD (BOL), cinanserin and cyproheptadine (Banna and Anderson, 1968; Clineschmidt et al., 1971).

**Bulbospinal Inhibition of the Monosynaptic Reflex**

Magoun and Rhines (1946) reported that the ventromedial bulbar reticular formation contains a descending neuronal system which exerts a general inhibitory influence on the segmental MSR. They also observed a concomitant melting of the decerebrate rigidity. The caudal raphe nuclei fall within the inhibitory area described by Magoun and Rhines (1946).

The mechanism of bulbospinal inhibition (BSI) of the MSR was studied by Llinas (1964a, b), Llinas and Terzuolo (1964, 1965) and Jankowska et al. (1968). These studies involved stimulation in the bulbar reticular formation while recording the MSR and the intracellular mem-
brane potential of a participating α-extensor motoneurone. Under these conditions, the MSR was reduced, the membrane was hyperpolarized, the resistance of the motoneurone membrane was reduced and the soma-dendritic (SD) component of the action potential was blocked when the motoneurone was activated antidromically. These findings indicate that BSI of the MSR is of the postsynaptic type. When chloride ions were iontophoretically injected into the motoneurone the hyperpolarization produced during stimulation of the bulbar reticular formation was reversed (Llinas and Terzuolo, 1964). Thus the ionic mechanisms responsible for BSI are similar to those of the reciprocal inhibition.

Bulbospinal inhibition of the α-flexor motoneurones also involves a synaptic inhibitory impingement on these motoneurones (Llinas and Terzuolo, 1965; Jankowska et al., 1968). Llinas and Terzuolo (1965) concluded that the inhibitory synapses of the pathway are on the dendritic tree of the motoneurone, far from the soma. This conclusion was based on the observation that injection of chloride ions into the motoneurone did not reverse the hyperpolarization produced during BSI. However, Jankowska et al. (1968) did not observe any difference between flexor and extensor motoneurones in this regard. The difference in these two studies may be due to the difference in the experimental preparations. Jankowska et al. (1968) used decerebrated cats with a contralateral hemisected and an ipsilateral dorsal transected spinal cord. The spinal cord was intact in the study of Llinas and Terzuolo (1965).

The bulbospinal inhibitory pathway descends in the ventral quadrant of the spinal cord. This pathway most likely has a disynaptic linkage and its conduction velocity is high (Jankowska et al., 1968).

Llinas (1964b) found that strychnine (0.15 and 0.5 mg/kg i.v.) decreased the hyperpolarization of the extensor motoneurone membrane.
caused by BSI but this drug did not block the inhibition of the MSR. He also found that picrotoxin (1 mg/kg i.v.) and mephenesin (120 mg/kg i.v.) did not block BSI and suggested that BSI is probably not of presynaptic type. However, picrotoxin blocks the segmental DRP but does not block the heterosegmental and heterosensory DRPs (Besson and Abdelmoumene, 1970; Besson et al., 1971; Benoish et al., 1972). Thus the possibility for a presynaptic type of BSI to exist cannot be entirely ruled out. Although neither strychnine nor mephenesin blocked BSI when administered individually, a combination of low doses of these two drugs blocked the inhibition (Llinas, 1964b). In an attempt to explain these findings, Llinas (1964b) suggested the following two possible mechanisms: 1. Strychnine may increase activity in the inhibitory pathway while depressing the inhibitory action. Mephenesin may antagonize this increased activity in the pathway. Thus, the blocking effect of the combined injection can be observed. 2. When strychnine blocks BSI it may increase the background excitatory impingement on the motoneurone and mephenesin may block this excitatory influence.

Stimulation of the medial reticular formation in the caudal brain stem 1 mm below the floor of the fourth ventricle (V about -5 to -6 in the Stereotaxic Atlas of Snider and Niemer, 1964) produced negative DRPs on Ia afferents of both flexor and extensor muscle nerves (Carpenter et al., 1966). These reticulospinal fibres descend in the ventromedial spinal cord. Carpenter et al. (1966) also reported that stimulation of the ventral caudal bulbar reticular formation (about 4 mm below the floor of the fourth ventricle) did not produce negative DRPs on Ia afferents. However, Chan and Barnes (1972) reported that stimulation of the ventral caudal bulbar reticular formation (2 mm lateral from the mid-sagittal line) produced both short and long latency negative DRPs.
on la afferents. Thus, a possible involvement of a presynaptic type of BSI can not be ruled out. It is not understood why there is a controversy between the above two reports.

Several pharmacological studies have been carried out on BSI of the MSR. The following drugs were shown to block BSI: Strychnine (0.05 mg/kg i.v), dichloroisoproterenol (7 mg/kg i.v.) and reserpine (0.5 mg/kg i.p) (McLennan, 1961); mephenesin (20 - 30 mg/kg i.v.) (Kaada, 1950); morphine and meperidine (0.5 - 16 mg/kg i.v.) (Sinclair, 1973). Bicuculline, a specific GABA antagonist, blocked BSI of the flexor MSR but had no effect on BSI of the extensor MSR (Huffman and McFadin, 1972). The 5-HT antagonists: methysergide (0.5 mg/kg i.v.), LSD (0.25 mg/kg i.v.), BOL (1.0 - 1.5 mg/kg i.v.) and cinanserin (4.0 mg/kg i.v.) but not cyproheptadine (5.0 mg/kg i.v.) also blocked BSI (Clineschmidt and Anderson, 1970). The results of close-arterial injection of methysergide and LSD to the spinal cord and the brain stem suggested that they act at the spinal cord level. Thus, Clineschmidt and Anderson (1970) proposed that the bulbospinal inhibitory pathway contains a 5-HT interneurone in the spinal cord.

Supraspinal Effects on Renshaw Cells

Stimulation of the ventromedial mesencephalic reticular formation at the level of substantia nigra (A 6.5 - 1.0) or the ventrolateral bulbar reticular formation at the level of hypoglossal nucleus (P 9.5 - 11.0) was found to have an inhibitory effect on the number of Renshaw cell discharges evoked by antidromic volleys in the motoneurone axons (Koizumi et al., 1959; Haase and Meulen, 1961; Mac Lean and Leffman, 1967). This inhibitory effect can be obtained by stimulating either side of the reticular formation but is stronger when the side contralateral to the Ren-
shaw cell was stimulated (Haase and Meulen, 1961). The latency between stimulation of the mesencephalic reticular formation and the onset of its inhibitory effect on the Renshaw cell discharge is about 9 msec and this effect lasts at its maximum strength for about 20 to 25 msec. While stimulation of the mesencephalic reticular formation has a stronger inhibitory effect on Renshaw cell discharges, the stimulation of bulbar reticular formation has a longer duration of action (MacLean and Leffman, 1967).

Stimulation of ventral thalamus (Fields of Forel, Zona Incerta) or the pericruciate cortex inhibits the Renshaw cell discharges evoked by antidromic stimulation of the motor axons. Such an inhibition is rapid in onset and of short duration (MacLean and Leffman, 1967). The descending fibres from pericruciate cortex pass through the pyramids in the brain stem (MacLean and Leffman, 1967).

Stimulation of the anterior lobe of the cerebellum has a facilitatory effect on Renshaw cell discharges evoked by antidromic stimulation of the motoneurone axons. Stimulation in the brain stem reticular formation had no effect on these cells activated in the above manner (Haase and Meulen, 1961). However, when the Renshaw cell discharges were evoked by stimulation of a dorsal root, activation of the reticular formation with a conditioning interval of 12 msec, increased the discharge rate of the Renshaw cells (Haase and Meulen, 1961). In addition, stimulation of the ventral thalamus or the pericruciate cortex could activate Renshaw cells to discharge (MacLean and Leffman, 1967). The work of Haase and Meulen (1961) and MacLean and Leffman (1967) indicate that the Renshaw cell discharges are influenced by supraspinal inputs.

Von Tan and Henatsch (1968) have studied the effects of imipramine
(0.5 to 2.0 mg/kg i.v.) on recurrent inhibition of the MSR in decerebrate cats. They also showed that this drug blocks the inhibition at these doses. However, when the spinal cord of the animal was sectioned at the thoracic level the drug did not block recurrent inhibition (Von Tan and Henatsch, 1969). They suggested that a supraspinal monoaminergic pathway has a strong curbing effect on recurrent inhibition of the MSR.
EXPERIMENTAL

Adult cats of either sex (2.0 - 4.0 kg) were anaesthetized with ether. The trachea was cannulated and the animal was artificially respired using a respiratory pump (Type AC; HP 1/4; C.F. Palmer (Lond.) Ltd.). The left carotid artery was cannulated with No. 160 polyethylene tubing (Clay Adams, Div. of Becton, Dickinson and company) filled with diluted sodium heparin (Upjohn Company of Canada). This tubing was connected to a P-1000-A pressure transducer (Narco-Bio-Systems) which in turn was connected to a type DMP-4A desk model physiograph (Narco-Bio-Systems) for recording blood pressure. The other carotid artery was ligated. A cephalic vein was cannulated with No. 90 polyethylene tubing filled with normal saline. This cannula was used for intravenously injecting the test drugs.

The animal's head was mounted on a stereotaxic frame (Narishige Scientific Instrument Laboratory). The skull overlying the frontal and parietal lobes of the cerebral cortex was removed. The animal was decerebrated at the mid-collicular level (Fig. 1), the brain tissue above the transection was removed and the skull cavity was packed with gauze. The cut edges of the bone were covered with bone wax to control bleeding and prevent air embolism. Blood loss during decerebration was replaced by injecting dextran (6 w/v) immediately after decerebration. The occipital bone overlying the cerebellum was removed and the dura was cut to expose the cerebellum.

A laminectomy was performed in the lumbosacral region of the spinal
Fig. 1. A diagramatic representation of the experimental set up.
cord. In some preparations L6, L7 and S1 dorsal roots were cut bilaterally. In other preparations the dorsal roots on the left were left intact and the nerves on this side leading to the posterior biceps-semimembranosus (PBST) and quadriceps (QUAD) muscles were isolated and cut. The central ends of these nerves were attached to bipolar stimulating electrodes. The ventral roots L6, L7 and S1 on the left side were sectioned in all preparations. The skin flaps on the back of the animal were used to make a pool for holding mineral oil which prevented drying of the spinal cord and spread of the current. The temperature of the mineral oil pool and the body of the animal was maintained at $36 \pm 1^\circ C$ using automatic D.C. temperature regulators (Richardson et al., 1965) or a heating lamp.

In animals that were used in the 'cold block' experiments, the spinal cord was exposed at T10 - T12, the dura-matter was cut and warm oil was poured on the cord to maintain the temperature of the exposed spinal cord at about $37^0 C$.

Ether was discontinued following surgery and three hours were allowed for elimination of ether before recordings were taken. The animal was maintained on artificial respiration throughout the experiment.

The central end of the dorsal roots on the left side (in most of the cases L7) and the corresponding ventral root were placed on bipolar platinum hook electrodes, the monosynaptic reflex (MSR) was evoked every 5 sec by stimulation of the dorsal root (DR-MSR) with a square wave pulse (0.1 msec) from the S2 unit of a Grass stimulator and which passes through a SIU5 stimulation isolation unit. The stimulus strength used was supra-maximal for the MSR. In the animals with intact dorsal roots on the left side and cut nerves to QUAD and PBST muscles the central end of one of the muscle nerves was stimulated to evoke the QUAD-MSR or PBST-MSR using the above mentioned parameters. The compound action potential in the ventral
root was amplified using a Tektronix 2A61 differential amplifier and displayed on a Tektronix 560 model oscilloscope.

A bipolar coaxial stainless steel electrode (0.5 mm separation, 0.5 mm exposed tip) was directed stereotaxically to the vicinity of the caudal raphe nuclei (P 7.5 to 13.5; L 0.0 in most and 0.5 in a few preparations; V -6 to -10 in the Stereotaxic Atlas of Snider and Niemer, 1964) (Fig. 1). A locus in this area was stimulated by a train (300 msec duration) of square wave pulses of 0.5 msec duration and at 150 Hz using the S1 unit of a Grass S8 stimulator and a SIU5 stimulation isolation unit. This train was delivered so that the end of the train occurred 7.5 msec before a stimulus was delivered to evoke the MSR. The location of the electrode in the brain stem and the stimulus strength (usually less than 5.0 V) were adjusted so that the MSR was inhibited to about 40% of its original size. Furthermore, the electrode was considered to be placed only if there were no tonic contractures of the neck, back and the forelimbs or marked changes in the blood pressure during the stimulation.

Recurrent inhibition (RI) of the MSR was obtained by stimulation of the central ends of two ventral roots (usually L6 and S1) 7.5 msec before evoking the MSR. In some preparations, however, a single ventral root was stimulated. Square wave pulses of 0.5 msec duration using the S1 unit of a S8 Grass stimulator and a SIU5 stimulation isolation unit were delivered, the stimulus strength was adjusted so that the MSR was inhibited to approximately 40% of the unconditioned value.

In some animals, after obtaining stable recordings of bulbospinal inhibition (BSI) and RI of the MSR, Flaxedil (gallamine triethiodide) was injected to minimize the effect of movement of the animal on the recordings.

Bulbospinal and recurrent inhibition of the MSR were tested at 10
min intervals. The MSR was quantified by averaging 10 consecutive spikes. The average MSR before and after the drug administration was expressed as a percentage of the final control average MSR. The degree of either BSI or RI was quantified by averaging the amplitude of 10 consecutive MSR spikes as well as the following 5 spikes conditioned with either BSI or RI. The percentage difference between these values on the final control test was equal to 100% inhibition. Percent inhibition of previous and subsequent tests were calculated based on this figure.

Following control recordings, imipramine HCl* (5 mg/kg i.v.) or an equimolar amount of desipramine HCl* (4.8 mg/kg i.v.) was administered over a 10 min period. In other animals pargyline HCl** (30 mg/kg i.v.) was injected over a 30 min period. In two preparations methysergide bimaleate*** (0.5 mg/kg i.v.) was injected over a 5 min period. All injections were given using an infusion pump (Harvard Model 975).

To block supraspinal inputs to the spinal cord, 1 cm cubes of frozen mammalian Ringer solution were placed on the spinal cord which was exposed at the T10–T12 level (Wall, 1967). The absence of BSI of the MSR was taken as the criterion for a functional block of supraspinal inputs. In the subsequent discussion these animals will be referred to as the 'cold block' preparations. To reverse the 'cold block', Ringer cubes were removed and the cold Ringer solution was sucked out using an aspirator. Warm oil (about 37°C) was poured on the cord repeatedly until BSI of the MSR returned to its pre-'cold block' level. This reversible 'cold block' technique was used to test whether the effects of imipramine on the MSR and RI of the MSR were mediated through a supraspinal neuronal system.

Some animals were pretreated with DL-p-chlorophenylalanine*** (p-CPA) (300 mg/kg i.p. for 3 consecutive days) and prepared for record-
ings 24 hours after the last dose. Two doses of \( p \)-CPA 300 mg/kg i.p. given
two consecutive days reduced 5-hydroxytryptamine levels to 10 to 20 \% of
control values in the spinal cord (Taber, 1971).

Other animals were pretreated with DL-\( \alpha \)-methyl-\( p \)-tyrosine methyl
ester HCl **** (126 mg/kg i.p.) 16 and 4 hours before preparing the animal
for recordings. After a similar treatment noradrenaline was reported to
be depleted to an immeasurable quantity in the spinal cord (King and
Jewett, 1971).

Foot Note

* Geigy Limited.

** Abbott Pharmaceuticals

*** Sandoz Pharmaceuticals.

**** Sigma Chemical Company.
RESULTS

Bulbospinal Inhibition of the Monosynaptic Reflex

Imipramine HCl (5.0 mg/kg i.v.) completely blocked bulbospinal inhibition (BSI) of the DR-MSR. The effect was rapid in onset and was maximal after 20 - 30 min. The above and subsequent time refer to the start of the injection. The blocking action of imipramine started to decrease around 50 min and was completely absent after about 2 hours (Fig. 2A).

Bulbospinal inhibition of the QUAD-MSR and the PBST-MSR were blocked by the above dose of imipramine. However, the effect on the inhibition of the QUAD-MSR was more rapid in onset and greater in magnitude. At 10 min imipramine had converted the inhibition into facilitation in 5 of 6 QUAD experiments (% inhibition = -10.3 ± 6.3 S.E.M.). Conversion of the inhibition to facilitation at 10 min occurred in only 1 of 8 PBST experiments (% inhibition = 56.8 ± 15.2). The maximal effect of imipramine was -87.9 ± 19.4 % (50 min) in the QUAD experiments and -15.5 ± 26.5 % (30 min) in the PBST experiments (Fig. 3 and 4).

Pretreatment of the cats with DL-p-chlorophenylalanine (p-CPA) completely prevented the effect of imipramine in blocking BSI of the DR-MSR (Fig. 2B). However, pretreatment of the animals with DL-α-methyl-p-tyrosine methyl ester HCl (α-MPT) had no effect on the blocking action of imipramine (Fig. 2C).

The effects of desipramine HCl (4.8 mg/kg i.v.) on BSI of the DR-MSR were qualitatively similar to those of imipramine but, desipramine's
Fig. 2. The effect of imipramine (IMI) on bulbospinal inhibition of the DR-MSR in A. non-pretreated cats, n = 6; B. cats pretreated with p-CPA (300 mg/kg i.p. for 3 consecutive days), n = 7; and C. cats pretreated with α-MPT (126 mg/kg i.p. 16 and 4 hours prior to recording), n = 6. Imipramine HCl (5 mg/kg i.v.) was injected over 10 min as indicated on the abscissa. Each point in this and subsequent graphs represents the mean % ± S.E.M. of the conditioned (upper graph) and the unconditioned MSR (lower graph).
Fig. 3. The effect of imipramine HCl (IMI) (5 mg/kg i.v.) on bulbospinal inhibition of the QUAD-MSR, n = 6; and the PBST-MSR, n = 8.
Fig. 4. The blockade of bulbospinal inhibition by imipramine. A. The large spike represents the unconditioned PBST-MSR and the small spike represents inhibition of this reflex produced by a conditioning stimulus in the vicinity of the raphe magnus nucleus. B. A partial blockade of the inhibition at the end of a 10 min injection of imipramine HCl (5 mg/kg i.v.). C. Complete blockade of the inhibition 10 min after B. Each frame represents 3 unconditioned and 3 conditioned sweeps of the MSR.
action was quantitatively less and of longer duration compared to that of imipramine (Fig. 5).

Pargyline HCl (30 mg/kg i.v.) blocked BSI of the DR-MSR. The onset of this effect was gradual, and the effect was prolonged. The blocking action was maximal at about 90 min and there was no indication of recovery within 3 1/2 hours (Fig. 6).

In two p-CPA pretreated animals in which imipramine failed to block BSI of the MSR, methysergide (0.5 mg/kg i.v.) converted the inhibition to a 3 - 4 fold facilitation.

To test whether BSI and recurrent inhibition (RI) were stable, these inhibitions were recorded for 3 1/2 hours in two experiments. The maximum deviation from the control values was 13.7 % (BSI) and 17.4 % (RI).

Recurrent Inhibition of the Monosynaptic Reflex

Imipramine HCl (5.0 mg/kg i.v.) blocked RI of the DR-MSR. The effect was gradual and reached maximum in 50 min and persisted for longer than 90 min (Fig. 7).

Recurrent inhibition of the QUAD-MSR was blocked by imipramine, the effect was rapid in onset and reached maximum in about 10 min (Fig. 8A). However, the drug did not block RI of the PBST-MSR (Fig. 8B).

Application of a 'cold block' significantly enhanced RI of the QUAD-MSR (RI % of control = 115.42 ± 0.7, n = 6) but not RI of the PBST-MSR (RI % of control = 106.38 ± 10.52, n = 5). The effect of imipramine on RI of the QUAD-MSR was completely eliminated and RI was actually enhanced when a 'cold block' was applied 30 min after the injection of the drug (RI % of control = 116.02 ± 5.4, n = 6) (Fig. 8A).

Pretreatment of the cats with either p-CPA or α-MPT completely eliminated the blocking action of imipramine on RI of the DR-MSR (Fig. 7).
Fig. 5. The effect of desipramine HCl (DMI) (4.8 mg/kg i.v.) on bulbo-spinal inhibition of the MSR (upper graph) and the unconditioned MSR (lower graph), n = 6. Desipramine was injected over 10 min as indicated on the abscissa.
Fig. 6. The effect of pargyline HCl (PARG) (30 mg/kg i.v.) on bulbospinal inhibition of the MSR (upper graph) and the unconditioned MSR (lower graph), n = 6. Pargyline was injected over 30 min as indicated on the abscissa.
Fig. 7. The effect of imipramine (IMI) on recurrent inhibition of the DR-MSR in non-pretreated cats, n = 5, (A); cats pretreated with p-CPA (300 mg/kg i.p. for 3 consecutive days), n = 6, (B); and cats pretreated with α-MPT (126 mg/kg i.p. 16 and 4 hours prior to recording), n = 6, (C).
Fig. 8. The effect of imipramine HCl (IMI) (5 mg/kg i.v.) on recurrent inhibition of the QUAD-MSR, n = 6, (A); and the PBST-MSR, n = 6, (B). A 'cold block' (CB) was applied over 10 min as indicated on the abscissa.
Fig. 9. The effect of pargyline HCl (PARG) (30 mg/kg i.v.) on recurrent inhibition of the DR-MSR, n = 6.
Pargyline also antagonized RI of the DR-MSR in 4 of 6 animals. The effect was gradual and reached maximum in about 90 min. Although quantitatively the effect of pargyline on this inhibition was less than that produced by imipramine, the former's effect was longer lasting (Fig. 9).

The Unconditioned Monosynaptic Reflex

Imipramine depressed the DR-MSR to about 60% of its control value. The effect was maximal at about 30 min and the MSR started to return to control levels about 30 min later. Approximately 2 hours after the injection of the drug the MSR returned to its control value (Fig. 2A). The QUAD- and PBST-MSR were also depressed by imipramine (Fig. 8A, B). Application of a 'cold block' reduced the QUAD-MSR to about 60% of its control value (MSR % of control = 57.9 ± 6.9, n = 5) but did not have a significant effect on the PBST-MSR (MSR % of control = 96.5 ± 1.74, n = 5). After the injection of imipramine, when a 'cold block' was applied at 30 min, the QUAD-MSR was not depressed further (Fig. 8A).

Imipramine depressed the DR-MSR in the animals pretreated with either p-CPA or α-MPT and the depressant effect at 30 min was not significantly different from that of the DR-MSR in the non-pretreated animals (Fig. 2).

Desipramine also reduced the DR-MSR to about 65% of its control value. The drug effect reaches maximum in about 40 min and the spike did not return to control value within 2 hours (Fig. 5).

Pargyline exerted a biphasic effect on the DR-MSR. The drug initially depressed the MSR to about 88% of its control value. However, at about 90 min the spike was facilitated to about 106% of the control. This late facilitatory effect was variable from animal to animal and reached its max in about 150 min and persisted up to 210 min at which time the experiment was discontinued (Fig. 6).
**Blood Pressure Effects**

Imipramine depressed the mean pulse pressure to about 69% of its control value. In animals that were pretreated with p-CPA or α-MPT, imipramine depressed the mean pulse pressure to about 95% and 77% of the control values respectively. The time course of depression of blood pressure is similar to that of the blocking action of imipramine on BSI. However, the latter effect of the drug is not related to its effect on the blood pressure, because of the reasons narrated in the next section (Discussion).

Desipramine at the given dose did not have a significant effect on blood pressure of most of the animals, however, this drug significantly reduced blood pressure in two animals. Pargyline had no appreciable effect on blood pressure.
DISCUSSION

The finding that imipramine, desipramine and pargyline blocked bulbospinal inhibition (BSI) of the monosynaptic reflex (MSR) is not consistent with the proposal by Clineschmidt and Anderson (1970) that the bulbospinal inhibitory system contains a 5-hydroxytryptamine (5-HT) link.

Since imipramine failed to antagonize BSI of the MSR in cats pretreated with the tryptophan hydroxylase inhibitor, DL-\(\text{-}\)chlorophenylalanine (p-CPA) but still maintained its blocking action in animals pretreated with the tyrosine hydroxylase inhibitor, DL-\(\alpha\)-methyl-\(\text{-}\)tyrosine (\(\alpha\)-MPT), it is assumed that the action of imipramine is mediated through 5-HT. This assumption is strengthened by the observation that pargyline, which, at the given dose, significantly elevates the levels of 5-HT but not noradrenaline (NA) in the spinal cord of the cat (Anderson et al., 1967), blocked BSI. Furthermore, desipramine at an equimolar dose to that of imipramine was quantitatively less effective than imipramine in blocking BSI. It is known that imipramine preferentially blocks the uptake of 5-HT whereas desipramine preferentially blocks the uptake of NA (Carlsson et al., 1969; Ross and Renyi, 1969; Shaskan and Snyder, 1970). Thus, the findings in the present investigation strongly suggest that 5-HT is involved in antagonizing rather than producing BSI.

It is interesting to note that the experiments carried out by Anderson and coworkers (Anderson and Shibuya, 1966; Anderson et al., 1967; Shibuya and Anderson, 1968; Banna and Anderson, 1968) very strongly
indicate that the 5-HT axons descending in the spinal cord have an overall facilitatory effect on the MSR. It is tempting to speculate that at least a part of the facilitatory effect of 5-HT on the MSR is due to a tonic inhibitory action of a 5-HT system on the inhibitory pathways (disinhibition), Thus, the net effect on the MSR is facilitation. The following observations support the above suggestion: 1. Imipramine blocked BSI of the MSR. 2. This drug blocked recurrent inhibition (RI) (see subsequent discussion) and presynaptic inhibition (unpublished observations) of the QUAD-MSR. 3. Engberg et al. (1968) suggested that a descending 5-HT system has a tonic inhibitory action on Ib and flexor reflex afferents.

Although Clineschmidt and Anderson (1970) proposed that a 5-HT interneurone in the spinal cord is contained in the bulbospinal inhibitory pathway, there is no evidence to suggest that there are 5-HT interneurones in the spinal cord. Furthermore, d-lysergic acid diethylamide (LSD), which was used as a 5-HT antagonist by the above workers, is known to stimulate 5-HT receptors (Costa, 1956; Horita and Gogerty, 1958; Anden et al., 1968) and the effects may be dose dependent; stimulating at low doses and blocking at high doses (Costa, 1956). The effective dose of LSD in blocking BSI (0.25 mg/kg) produced an enhancement of the unconditioned MSR (Clineschmidt and Anderson, 1970). This is consistent with a 5-HT stimulant effect as seen following the injection of 5-HT precursors (Anderson and Shibuya, 1966). Moreover, the dose of LSD required to block the 5-HTTP induced enhancement of the MSR was higher than that which was effective in blocking BSI of the MSR (Banna and Anderson, 1968). The effect of 2-bromo-LSD (BOL) on BSI was transient; cyproheptadine, which blocked the 5-HTTP enhancement of the MSR at a lower dose, did not block BSI (Clineschmidt and Anderson, 1970). Also, the 5-HT antagonists
blocked the action of D-3,4-dihydroxyphenylalanine (l-dopa) on the MSR (Banna and Anderson, 1968). In the present study, when methysergide was tested on BSI in two animals that were pretreated with p-CPA and in which imipramine failed to block BSI, methysergide converted the inhibition to a 3 - 4 fold facilitation; this drug may be acting on a non-serotonergic system.

It is interesting that imipramine, pargyline and LSD all depressed the firing of the mesencephalic raphe neurones in rats (Sheard et al., 1972; Aghajanian et al., 1970; Aghajanian et al., 1968). Furthermore, this depressant effect of imipramine is absent in rats pretreated with p-CPA (Sheard et al., 1972). Hosli et al. (1971) reported that 5-HT had a general excitatory effect when iontophoretically applied on the bulbospinal neurones. Thus, if the 5-HT neurones in the raphe nuclei excite the bulbospinal inhibitory neurones, it may be possible that BSI is blocked by imipramine and pargyline since the 5-HT neurones in the raphe nuclei stop firing after these drugs. However, Clineschmidt and Anderson (1970) found that LSD was more effective in blocking BSI of the MSR when administered by close-arterial injection to the spinal cord than by close-arterial injection to the brain stem. Thus the site of action of LSD is most likely in the spinal cord. Also, the bulbospinal inhibitory pathway can not have a 5-HT neurone descending from the raphe nuclei to the spinal cord since, the conduction velocity of the inhibitory pathway was found to be high (Jankowska et al., 1968; Clineschmidt and Anderson, 1970) and the unmyelinated 5-HT fibres of 0.3 to 1.0 \( \mu \)m diameter (Dahlstrom and Fuxe, 1965) can not conduct impulses that fast. As mentioned earlier, there is no evidence for 5-HT interneurones to exist in the spinal cord.

Bulbospinal inhibition of the flexor and the extensor MSRs was found to be of the postsynaptic type (Llinas, 1964a; Llinas and Terzuolo, 1964,
1965; Jankowska et al., 1968). The inhibitory pathway most likely involves a disynaptic link and the conduction velocity of the pathway is high (Jankowska et al., 1968). While Jankowska et al. (1968) did not find any difference between the ionic mechanisms involved in BSI of the flexor and extensor MSRs, Llinas and Terzuolo (1965) noted differences between the two and suggested that the inhibitory synapses of the bulbospinal inhibitory pathway with the flexor motoneurones are on the dendrites, far from the soma. Huffman and McFadin (1972) found that bicuculline, a specific \textgamma-aminobutyric acid antagonist, blocked BSI of the flexor MSR but had no effect on BSI of the extensor MSR. It is interesting that in the present study imipramine's blocking action on BSI of the QUAD-MSR was quantitatively greater than that on BSI of the PBST-MSR. This difference in the blocking action of the drug may be due to the differences in the mechanism by which the bulbospinal inhibitory pathway exerts its action on the QUAD-MSR and the PBST-MSR or due to the difference in the 5-HT input to these two types of the MSRs.

Carpenter et al. (1966) found that stimulation in the medial caudal bulbar reticular formation (1 mm below the floor of the fourth ventricle) produced negative dorsal root potentials (DRPs) on the Ia afferents. These authors also reported that stimulation in the above reticular formation about 4 mm below the floor of the fourth ventricle did not produce negative DRPs on the Ia afferents. These observations suggest that the Magoun and Rhine's (1946) inhibitory area, the ventromedial caudal bulbar reticular formation, may not have a presynaptic type of bulbospinal inhibitory pathway. However, Chan and Barnes (1972) observed that stimulation in the ventral caudal bulbar reticular formation (2 mm lateral from the mid-sagittal line) produced negative DRPs on Ia afferents; these authors also noted a time correlation between the primary afferent
depolarization (PAD), the negative DRP and the inhibition of the MSR while stimulating in the above bulbar area. Since a negative DRP reflects PAD and presynaptic inhibition, these findings may suggest, contrary to those of Carpenter et al. (1966), that a presynaptic type of inhibitory component is present in BSI of the MSR. In the present study only the ventromedial caudal bulbar reticular formation was stimulated (V -6 to -10, L 0.0 in the Stereotaxic Atlas of Snider and Niemer, 1964). This area of stimulation is not exactly the same as that used in the study of Chan and Barnes' (1972) and it is not clear whether a presynaptic inhibitory component was involved in the present study of the MSR.

Assuming that in the present study only the postsynaptic type of the bulbospinal inhibitory pathway was stimulated and that the inhibitory pathway contains a disynaptic link (Jankowska et al., 1968) and that the interneurone is located in the spinal cord, it can be speculated that the blockade of BSI by 5-HT can be due to a 5-HT neurone that terminates either on the axon terminal or the soma of the bulbospinal neurone or the interneurone in the spinal cord. It seems unlikely that the inhibitory 5-HT neurone ends on the soma of the bulbospinal neurone since 5-HT was found to have a general excitatory effect on the latter neurones (Hosli et al., 1971). It is known that the 5-HT neurones in the raphe nuclei send axons that descend in the spinal cord and terminate in the dorsolateral and the ventrolateral motor nuclei of the ventral horn of the spinal cord (Fuxe, 1965). Thus it seems more likely that an inhibitory 5-HT neurone may end on the axon terminal of the bulbospinal neurone or on the interneurone in the spinal cord.

It is not known what other neuronal systems send inputs to the interneurones connected with the bulbospinal inhibitory pathway. Recently it has been reported that some interneurones in the spinal cord which
receive inputs from the primary afferents also receive supraspinal inputs (Koizumi et al., 1959; Engberg et al., 1968). For example, the Ia inhibitory interneurones receive supraspinal excitatory input (Hultborn and Udo, 1972). Llinas reported that strychnine, a specific glycine antagonist reduced the hyperpolarization of the motoneurone membrane produced by BSI. Strychnine blocks reciprocal inhibition by antagonizing the action of the putative neurotransmitter, glycine that is presumably released from the Ia inhibitory interneurone terminal at the interneurone-motoneurone synapses. It may be possible that these interneurones are connected with the bulbospinal inhibitory pathway. However, imipramine HCl (5 mg/kg i.v.) had no effect on reciprocal inhibition (unpublished observations) suggesting that there is no 5-HT input to the Ia inhibitory interneurones. Thus, if these interneurones are involved in BSI, a 5-HT inhibitory neurone must end on the axon terminal of the bulbospinal neurone. However, the idea that Ia inhibitory interneurones are connected with the bulbospinal inhibitory pathway is purely speculative.

Imipramine blocked RI of the QUAD-MSR and the effect of the drug is very likely mediated through a supraspinal monoaminergic system (see subsequent discussion). However, Renshaw cells do not seem to be connected with the bulbospinal inhibitory pathway because imipramine's effect on BSI was eliminated by pretreatment of the cats with p-CPA but not with α-MPT whereas imipramine's effect on RI was prevented by pretreatment of the cats with either p-CPA or α-MPT. Thus, the 5-HT input to block BSI and the monoaminergic input to block RI are probably two different systems.

Blockade of RI of the DR-MSR by imipramine and pargyline, and elimination of imipramine's blocking action by pretreatment of the cats with either p-CPA or α-MPT supports the finding of Von Tan and Henarsch
(1968) that a monoaminergic pathway antagonizes RI of the MSR.

Imipramine's antagonism of RI of the QUAD-MSR but not RI of the PBST-MSR indicates that the monoamine input is to the Renshaw cells involved in RI of the QUAD-MSR but not in RI of the PBST-MSR.

Potentiation of RI of the MSR by application of a 'cold block' and complete removal of imipramine's effect on RI of the QUAD-MSR by a 'cold block' indicate that a supraspinal monoaminergic system has a tonic inhibitory effect on RI of the QUAD-MSR.

Since pretreatment of the cats with either p-CPA or α-MPT completely eliminated the blocking action of imipramine on RI, there can not be two separate descending 5-HT and NA systems on RI. Instead, the descending system must have links involving both 5-HT and NA. It is interesting that the 5-HT and NA cells in the caudal brain stem have mutual synaptic contact and that there are both 5-HT and NA nerve terminals in the dorsolateral and ventrolateral motor nuclei of the ventral horn of the spinal cord (Fuxe, 1965).

Application of a 'cold block' significantly reduced the QUAD-MSR but had no effect on the PBST-MSR suggesting that the QUAD-MSR receives a supraspinal tonic facilitatory input. Imipramine decreased the DR-, QUAD- and PBST-MSRs. Application of a 'cold block' 30 min after the injection of imipramine did not reduce the QUAD-MSR any further. This may suggest that the effect of imipramine on the QUAD-MSR is due to the blockade of a supraspinal tonic facilitatory system. Since, a 'cold block' had no effect on the PBST-MSR and imipramine reduced this MSR, the effect of the drug may be due to its action on neuronal systems at the segmental level. Eccles and Lundberg (1959) found that the extensor motoneurones are under strong supraspinal influences and the flexor motoneurones are mostly independent of such influences. This agrees with
the present work.

Pretreatment of the cats with α-MPT did not have a significant effect on the depressant action of imipramine on the DR-MSR suggesting that this effect of imipramine on the DR-MSR is probably not mediated through a NA system. However, although pretreatment of the animals with p-CPA did not alter the action of imipramine on the DR-MSR upto 30 min of imipramine, the recovery of the MSR was faster than in nonpretreated animals. The reason for the faster recovery of the MSR in p-CPA pretreated animals is not understood. However, since imipramine did depress the MSR in these animals, this effect of the drug can not be entirely due to a 5-HT input.

The biphasic effect of pargyline on the MSR was previously reported by Anderson et al. (1967). Based on the monoamine levels in the spinal cord after pargyline and other pharmacological evidence they concluded that the enhancement phase of pargyline's action was mediated by increased endogenous levels of 5-HT. The enhancement of the MSR by pargyline in the present study is however smaller than that reported by Anderson et al. (1967). This may be due to the difference in the experimental preparations. Anderson et al. (1967) used cats with the spinal cord sectioned at the cervical level whereas in the present study cats decerebrated at the mid-collicular level were used. Thus the control MSR is considerably higher in most of the present experiments than the range (1.5 to 3.0 mV) used by Anderson et al. (1967). Therefore in the present study, possibly fewer motoneurones are available for recruitment into the discharge zone.

The blocking action of imipramine on BSI and RI and the drug's depressant action on the MSR do not seem to be related since pargyline, which blocked BSI and RI facilitated the MSR. However, a part of the
action of imipramine on the MSR may be mediated through 5-HT and it is surprising and not understood why this drug while blocking BSI, RI and presynaptic inhibition, reduces the MSR.

The time course of the depressant action of imipramine on blood pressure is found to be similar to that of the drug's action on BSI, RI and the MSR. However, the possibility that imipramine's effects on these are due to its action on the blood pressure is ruled out because of the following reasons: 1. Pargyline which does not have a significant effect on blood pressure blocked BSI and RI and had a biphasic action on the MSR. 2. Imipramine blocked RI of the QUAD-MSR but not RI of the PBST-MSR. 3. 'cold block' which had no effect on the blood pressure completely eliminated imipramine's effect on RI.

As outlined above, the findings in the present investigation strongly suggest that a 5-HT system antagonizes BSI of the MSR and that a supraspinal monoaminergic system having both 5-HT and NA links has a tonic inhibitory effect on RI of the QUAD-MSR. The 5-HT system that antagonizes BSI seems to be different from the monoaminergic system that blocks RI. Further investigation is necessary to establish the site of action of 5-HT involved in blocking these inhibitions.
REFERENCES


conductance and the ionic movements across the motoneuronal membrane that produce the inhibitory postsynaptic potential. J. Physiol. 130: 326-373.


HAASE, J. and VAN DER MEULEN, J.P. (1961). Effects of supraspinal stimul-


LLINAS, R. (1964b). Mechanisms of supraspinal action upon spinal cord
activities. Pharmacological studies on reticular inhibition of alpha-

LLINAS, R. and TERZUOLO, C.A. (1964). Mechanisms of supraspinal actions upon spinal cord activities. Reticular inhibitory mechanisms on alpha-


TABER, E., BRODAL, A. and WALBERG, F. (1960). The raphe nuclei of the brain stem in the cat I. Normal topography and cytoarchitecture and gene-


