NA. Electrolytic lesions in the ventral tegmental area resulted in a significant depletion of DA from both the medial prefrontal and cingulate cortex but produced no change in cortical NA. The depletion of DA from the cingulate cortex was not complete and indicated the possible presence of a separate group of DA terminals whose cell bodies of origin were outside the ventral tegmentum. Unilateral injections of 6OHDA (4 μg/1 μl) into the substantia nigra produced a small but significant depletion of DA from the superficial layers of the cingulate cortex. These results are consistent with previous fluorescent histochemical findings (Lindvall *et al.*, 1974).

References

CUELLO, A.C., HILEY, C.R. & IVERSEN, L.L. (1973). Use of catechol-*o*-methyltransferase for the enzyme radiochemical assay of dopamine. *J. Neurochem.*, **21**, 1337–1340.

LINDVALL, O., BJÖRKLUND, A., MOORE, R.Y. & STENEVI, U. (1974). Mesencephalic dopamine neurons projecting to neocortex. *Brain Res.*, **81**, 325–331.

MASON, S.T. & IVERSEN, S.D. (1975). Learning in the absence of forebrain noradrenaline. *Nature, Lond.*, **258**, 422–424.

THIERRY, A.M., STINUS, L., BLANC, G. & GLOWINSKI, J. (1973). Some evidence for the existence of dopaminergic neurons in rat cortex. *Brain Res.*, **50**, 230–234.

The actions of cholinomimetics and catecholamines on rat substantia nigra neurones

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Acetylcholine and choline acetylase occur in the substantia nigra (Cheney, Le Fevre & Racagni, 1975) and acetylcholine excites nigra cells (Crossman, Walker & Woodruff, 1974a; Dray & Straughan, 1976). Recent evidence suggests that dopamine is released from dopamine-containing zona compacta neurone dendrites (Geffen, Jessell, Cuello & Iversen, 1976) and iontophoretically applied dopamine produces inhibition or excitation (Dray, Gonye, Oakley & Tanner, 1976). Although there is little noradrenaline in the substantia nigra, histochemical studies reveal a small population of noradrenaline-containing terminals in the zona reticulata. In this study we have examined the characteristics of the acetylcholine and catecholamine receptors of nigra neurones.

Experiments were performed on 150 g female Wistar rats, anaesthetized with urethane (1.5–2 g/kg i.p.). Extracellular recordings were made from single substantia nigra neurones using multibarrel glass microelectrodes (Crossman *et al.*, 1974b). Drugs were applied iontophoretically. The iontophoretic barrels contained acetylcholine, nicotine, carbachol, furtrethonium, atropine, physostigmine, dopamine, adrenaline and noradrenaline, all 0.2 M, pH 4–5.

Acetylcholine has predominantly excitatory effects applied iontophoretically to nigra cells. Of 190 cells tested, acetylcholine (20–60 nA) excited 105 and inhibited 20. Nicotine (30–60 nA) excited 41 and inhibited 2 of 70 cells tested. Furtrethonium (20–60 nA) excited 11 and inhibited 3 of 19 cells tested. Carbachol (20–60 nA) excited 15 and inhibited one of 19 cells tested. All four carbachol responses tested with atropine (40 nA for 45 s) were reversibly inhibited. Atropine blocked two of three actylcholine excitations and both nicotine excitations on which it was tested. Physostigmine (50 nA) applied prior and concurrently with acetylcholine potentiated the acetylcholine response on all five occasions.

Close correlation occurred between the effects following application of noradrenaline and adrenaline to nigra cells. Of 14 cells tested both adrenaline (30–60 nA) and noradrenaline (30–60 nA) excited three cells, inhibited five cells and three gave biphasic responses to both compounds; three cells were unaffected by adrenaline and two of these were similarly unaffected by noradrenaline while the third gave a biphasic response. No such correlation occurs between dopamine and noradrenaline responses. Of 13 cells excited by dopamine (30–60 nA), noradrenaline (30–60 nA) excited four, inhibited three, one was biphasic and five were unaffected. Of eight cells inhibited by dopamine, noradrenaline inhibited five, excited one and had no effect on two. Of 12 cells unaffected by dopamine, noradrenaline inhibited three, excited two, two were biphasic and five were unaffected.

It is suggested that cholinergic excitations are produced by stimulation of a mixed nicotinic/muscarinic receptor while inhibitions may be muscarinic. The catecholamine results are consistent with two populations of adrenergic receptors, one stimulated by dopamine and one by noradrenaline and adrenaline.

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References

CHENEY, D.L., LE FEVRE, H.F. & RACAGNI, G. (1975). Choline acetyltransferase activity and mass fragmentographic measurement of acetylcholine in specific nuclei and tracts of rat brain. *Neuropharmac.*, **14**, 801–809.

CROSSMAN, A.R., WALKER, R.J. & WOODRUFF, G.N. (1974a). Pharmacological studies on single neurones in the substantia nigra of the rat. *Br. J. Pharmac.*, **51**, 137–138P.

CROSSMAN, A.R., WALKER, R.J. & WOODRUFF, G.N. (1974b). Problems associated with iontophoretic studies in the caudate nucleus and substantia nigra. *Neuropharmac.*, **13**, 547–552.

DRAY, A., GONYE, T.J., OAKLEY, N.R. & TANNER, T. (1976). Evidence for the existence of a raphe projection to the substantia nigra in the rat. *Brain Res.*, **113**, 45–57.

DRAY, A. & STRAUGHAN, D.W. (1976). Synaptic mechanisms in the substantia nigra. *J. Pharm. Pharmac.*, **28**, 400–405.

GEFFEN, L.B., JESSEL, T.M., CUELLO, A.C. & IVERSEN, L.L. (1976). Release of dopamine from dendrites in the rat substantia nigra. *Nature*, **260**, 258–260.

Effects of C-fragment on brain stem neurones in the cat

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The C-fragment of β-lipotropin is a potent analgesic when injected into the cerebral ventricles of the cat (Feldberg & Smythe, 1976, 1977).

We have compared the actions of C-fragment with those of morphine and methionine-enkephalin by iontophoretic application from micropipettes to neurones in the brain stem of decerebrate cats. The neurones tested were in the periaqueductal gray and other medial areas of the mid-brain, in nucleus raphe magnus, and in the medial bulbar reticular formation. Most cells were strongly inhibited by C-fragment while the remainder (6 out of 82) were unaffected, and the time course of the inhibition was similar to that of the iontophoretic application. Morphine and met-enkephalin had depressant effects on the same neurones, except that morphine produced a biphasic response (inhibition followed by excitation) in two neurones in the mid-brain on which C-fragment exerted only a depressant action. Naloxone (Endo Laboratories) applied by iontophoresis did not block the effects of C-fragment, morphine or met-enkephalin in the areas tested and occasionally exhibited agonist activity.

Although it is difficult to compare potencies with the iontophoretic method, the results suggest that C-fragment is more potent than met-enkephalin since comparable effects were obtained with smaller currents. As C-fragment has more charged side groups than met-enkephalin fewer ions will be ejected by the same current, and it is also likely to be less mobile than the smaller peptide. These considerations suggest that C-fragment is more potent than met-enkephalin in depressing the activity of neurones in the brain stem.

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References

FELDBERG, W. & SMYTH, D.G. (1976). The C-fragment of lipotropin—a potent analgesic. *J. Physiol.*, **260**, 30–31P.

FELDBERG, W. & SMYTH, D.G. (1977). Analgesia produced in cats by the C-fragment of lipotropin and by a synthetic pentapeptide. *J. Physiol.* (in press).