maximum stimulation occurred over the range $1 \times 10^{-5} - 5 \times 10^{-5}$M. A 2.5 fold increase in cyclic AMP levels was routinely obtained. The pharmacological characteristics of agonists on this system are summarized in the table. The structures of active and inactive molecules demonstrates the strict requirements for a catechol grouping and a β-hydroxyl group with the correct stereochemistry. The blockade of the stimulation by 5×10^{-5}M noradrenaline in the presence of adrenolytics was stereoselective. The selective β_1 antagonist (±)-practolol (Dunlop & Shanks, 1968) was less active than (±)-propranolol, whilst the γ blocker H35/25 (1-(4'-methylphenyl)-2-isopropylamino-propranol HCl) (Levy & Wilkenfeld, 1969) was inactive; this is of interest as the β_2 agonist salbutamol (Farmer *et al.*, 1970) was also found to be inactive. The spectrum of activity for the neuroleptics was very different from that found for the blockade of the dopamine sensitive adenylate cyclase (Miller, Horn & Iversen, 1974). Promazine, clozapine, thioridazine and chlorpromazine were all quite active, whereas α-flupenthixol and trifluperazine, both potent blockers of the dopamine sensitive adenylate cyclase, were less active. Assuming the blockade by chlorpromazine is competitive it has an approximate Ki of 1.6×10^{-7}M whereas the value for the inhibition of the dopamine-adenylate cyclase is 4.8×10^{-8}M (Miller *et al.*, 1974). It is also of interest that no large differences in potency were found for the thioxanthene isomers; this contrasts strongly with results obtained from the dopaminergic cyclase (Miller *et al.*, 1974).

References

BLUMBERG, J.B. & SULSER, F. (1974). The effect of antipsychotic drugs on the 3', 5'-adenosine monophosphate (cAMP) system on rat limbic forebrain. *Fed. Proc.*, 33, 286.

BLUMBERG, J.B., TAYLOR, R.E. & SULSER, F. (1975). Blockade by pimozide of a noradrenaline sensitive adenylate cyclose in the limbic forebrain: possible role of limbic noradrenergic mechanisms in the mode of action of antipsychotics. *J. Pharm. Pharmac.*, 27, 125-128.

BROWN, B.L., EKINS, K.D. & ALBANO, J.D.M. (1972). Saturation assay for cyclic AMP using endogenous binding protein. *Adv. Cyclic Nucleotide Res.*, 2, 25-40.

CHASIN, M., MAMRAK, F. & SAMANIEGO, S.G. (1974). Preparation and properties of a cell-free hormonally responsive adenylate cyclase from guinea pig brain. *J. Neurochem.*, 22, 1031-1038.

DUNLOP, D. & SHANKS, R.G. (1968). Selective blockade of adrenoceptive beta receptors in the heart. *Br. J. Pharmac. Chemother.*, 32, 201-218.

FARMER, J.B., KENNEDY, I., LEVY, G.P. & MARSHALL, R.J. (1970). A comparison of the β-adrenoreceptor stimulant properties of isoprenaline, with those of orciprenaline, salbutamol, soterenol and trimetoquinol on isolated atria and trachea of the guinea-pig. *J. Pharm. Pharmac.*, 22, 61-63.

LEVY, B. & WILKENFELD, B.E. (1969). An analysis of selective beta receptor blockade. *Eur. J. Pharmac.*. 5, 227-234.

MILLER, R.J., HORN, A.S. & IVERSEN, L.L. (1974). The action of neuroleptic drugs on dopamine-stimulated adenosine cyclic 3', 5'-monophosphate production in rat neostriatum and limbic forebrain. *Mol. Pharmac.*, 10, 759-766.

Altered sensitivity of β-adrenoceptor-mediated cyclic AMP formation in brain

S.R. NAHORSKI* & K.J. ROGERS

Section of Pharmacology, Academic Division of Medicine, University of Sheffield

In several studies using homogenates or slices of brain tissue it has been demonstrated that the stimulation of catecholaminergic receptors causes an increased synthesis of cyclic AMP (Daly, 1975). In our studies of the effects of neurohormones on cerebral cyclic AMP metabolism we have found that the neonate chick with its immature blood-brain barrier is a useful experimental model for both *in vivo* and *in vitro* investigations (Edwards, Nahorski & Rogers, 1974), and we have provided evidence that catecholamines stimulate formation of the nucleotide in chick cerebral hemispheres via β-adrenoceptors (Nahorski, Rogers & Smith, 1974). In order to assess the potential importance of these responses we have, in the present study, used procedures that alter central adrenergic transmission, in an attempt to determine whether or not there are accompanying changes in the sensitivity of the cyclic AMP response.

Experiments were performed on 1-6 day old male Ranger chicks. Pretreatment with reserpine (2.5 mg/kg s.c.) daily for three days or 6-hydroxydopamine (60 μg in 10 μl intracerebroventricularly) on two successive days severely depleted brain catecholamines (> 70%) for at least six days. Five days after commencing the pretreatments, groups of chicks were injected intravenously with (−)-isoprenaline (5 μmol/kg) or histamine

(100 μmol/kg). The cerebral hemispheres were removed by freeze-blowing 2 min later and cerebral cyclic AMP measurements were performed by a protein binding assay. The increase in cyclic AMP in response to isoprenaline in reserpinized chicks (475%) or in chicks pretreated with 6-hydroxydopamine (441%) was significantly greater than that produced in control birds (266%), indicating an increased sensitivity to isoprenaline following the chronic depletion of cerebral catecholamines. Experiments were performed therefore to determine whether the chronic administration of isoprenaline would conversely reduce the sensitivity of the cyclic AMP response. (−)-Isoprenaline was suspended in glycerol trioleate and administered subcutaneously (150 μmol/kg) in two doses at 12 h intervals to effect a slow release of catecholamine. Six hours after the second injection the cerebral cyclic AMP response to intravenous isoprenaline was almost completely suppressed. The increase in cyclic AMP induced by histamine *in vivo* was not influenced by any of the drug treatments.

More detailed studies were performed *in vitro* using slices of chick cerebral hemispheres. Dose-response curves for isoprenaline indicated that the maximal increase in cyclic AMP produced by the catecholamines was significantly enhanced in slices prepared from reserpinized chicks but severely suppressed in the chronic isoprenaline group. However, although the supersensitive response was still observed in the presence of the potent phosphodiesterase inhibitor, Ro 20-1724 (4-(3-butoxy-4-methoxy benzyl)-2-imidazolidinone) (200 μM) the subsensitive response was almost restored to that of the controls.

The data suggest that the sensitivity of cerebral β-adrenoceptors mediating cyclic AMP formation may be regulated by the functional amount of transmitter at the receptor. The subsensitive condition may, in part, be due to a selective increase in phosphodiesterase activity but several alternative mechanisms need to be examined.

This work was supported by a grant from the Wellcome Trust.

References

DALY, J.W. (1975). Cyclic adenosine 3', 5'-monophosphate role in the physiology and pharmacology of the central nervous system. *Biochem. Pharmac.*, **24**, 159-164.

EDWARDS, C., NAHORSKI, S.R. & ROGERS, K.J. (1974). *In vivo* changes of cerebral cyclic adenosine 3', 5'-monophosphate induced by biogenic amines: association with phosphorylase activation. *J. Neurochem.*, **22**, 565-572.

NAHORSKI, S.R., ROGERS, K.J. & SMITH, B.M. (1974). Regulation of the cyclic AMP concentration in chick brain by β-adrenoceptors and histamine H_2 receptors. *Br. J. Pharmac.*, **52**, 121P.

Comparison of the effects of DOPA and noradrenaline on single cortical neurones

P. BEVAN, C.M. BRADSHAW* & E. SZABADI

Department of Psychiatry, University of Edinburgh, Morningside Park, Edinburgh EH10 5HF

L-3, 4-dihydroxyphenylalanine (DOPA) is widely used in the treatment of Parkinson's disease. DOPA is a metabolic precursor of dopamine, and it is generally believed that DOPA exerts its therapeutic effect via the release of dopamine in the caudate nucleus (Hornykiewicz, 1974). However, DOPA is also known to be a precursor of noradrenaline (NA), and it has been suggested that in structures receiving a NA innervation, exogenously administered DOPA may cause the release of NA from pre-synaptic terminals and thus mimic the actions of NA on post-synaptic cells (Andén, Carlsson & Häggendal, 1969). We have used the technique of microelectrophoresis to compare the action of DOPA and NA on single neurones in the cerebral cortex, a structure rich in NA-containing terminals (Fuxe, 1965).

Single spontaneously-active cortical neurones were studied in cats and rats anaesthetized with halothane. Drugs were applied from six-barrelled micropipettes by microelectrophoresis. Our techniques and methods of study have been described elsewhere (Bevan, Bradshaw, Roberts & Szabadi, 1974).

The effect of DOPA (released from 0.05 M DOPA methyl ester HCl solution) was examined on 51 neurones in the rat; 44 were excited and 7 depressed by the drug. Eleven cortical neurones were studied in the cat; of these 10 were excited and one depressed by DOPA. When the effects of DOPA were compared with those of NA on the same cells it was found that neurones invariably responded in the same direction to the two drugs (40 cells). In the case of both excitatory and depressant effects DOPA appeared to be less potent than NA, both in terms of the intensity of