The effect of phosphodiesterase inhibitors on the stimulation of cerebral cyclic AMP formation by biogenic amines *in vitro* and *in vivo*

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In contrast to the situation in several extracerebral tissues, the classical methylxanthine phosphodiesterase inhibitors theophylline and caffeine do not potentiate neurohormone stimulation of cyclic AMP formation in brain slices (Forn & Krishna, 1971; Schultz & Daly, 1973). Recently a number of other compounds have been shown to be more potent inhibitors of phosphodiesterase than theophylline in cell free preparations of brain and we have examined some of these drugs on biogenic amine-stimulated cyclic AMP formation both *in vitro* and *in vivo*.

In vitro experiments were performed on 0.37 mm thick slices of mouse forebrain preincubated for 60 min in Krebs-bicarbonate buffer containing 10 mM glucose. Cyclic AMP was assayed by the method of Brown, Albano, Ekins, Sgherzi & Tampion (1971). Of a number of compounds examined at a concentration of 200 μM, 4-(3-butoxy-4-methoxy)-2-imidazolidinone (Ro 20-1724) and 2-amino-6-methyl-5-oxo-4-*n*-propyl-4,5-dihydro-*s*-triazolo(1,5-a) pyrimidine (ICI 63 197) markedly potentiated the cyclic AMP response to a submaximal concentration (100 μM) of noradrenaline. Theophylline, papaverine and medazepam did not significantly enhance the effect of noradrenaline.

Histamine and dopamine were completely inactive in stimulating cyclic AMP formation in mouse forebrain slices, but in the presence of 200 μM Ro 20-1724 or ICI 63.197, significant (2-3 fold) accumulations of the nucleotide were produced by both of these amines. Prostaglandin E_1 and 5-hydroxytryptamine at concentrations of up to 100 μM did not enhance cyclic AMP formation in the presence or absence of these phosphodiesterase inhibitors.

In this laboratory we have shown that systemically administered biogenic amines increase cerebral cyclic AMP in the neonate chick (Edwards, Nahorski & Rogers, 1974). Using this model an attempt was made to demonstrate inhibition of cerebral phosphodiesterase *in vivo*. Phosphodiesterase inhibitors were injected s.c. or in the case of RO 20-1724 i.v., 20 min before killing. The cerebral hemispheres were removed using a freeze-blowing technique to eliminate post-mortem changes in cerebral cyclic AMP (Nahorski & Rogers, 1973). None of the phosphodiesterase inhibitors alone increased cerebral cyclic AMP *in vivo*. However, the intravenous injection of isoprenaline (5 μmoles/kg) raised brain levels of the nucleotide to 228% of control values in two minutes. In chicks pretreated with RO 1724 (1 mg/kg), isoprenaline increased cerebral cyclic AMP to 342% of levels observed with the phosphodiesterase inhibitor alone. Similarly pretreatment with ICI 63 197 (5 mg/kg) markedly enhanced the response to isoprenaline in that nucleotide levels of 951% above those of controls were observed. Theophylline (50 mg/kg), papaverine (20 mg/kg) and medazepam (20 mg/kg) did not modify the isoprenaline response *in vivo*.

The present results demonstrate that dialkoxyimidazolidinone and triazolopyrimidine phosphodiesterase inhibitors potentiate the increase in cyclic AMP induced by biogenic amines in mouse cerebral slices. Moreover, since these compounds appear to be effective phosphodiesterase inhibitors *in vivo*, they may prove to be valuable tools in assessing the functional role of cyclic nucleotides in the central nervous system.

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