the morphine-induced contractions but when administered alone, caused contractions similar to those produced by morphine. In contrast, cyproheptadine (3×10^{-5} M) not only abolished morphine-induced contractions but also inhibited responses to carbachol (10^{-7} M) and potassium (30 mM).

Morphine-induced contractions were potentiated by phentolamine (10^{-5} M) and inhibited by noradrenaline (5×10^{-7} M) or tyramine (3×10^{-5} M). The dose of morphine required to produce contractions was lower in tissue from rats pretreated with reserpine (0.3 mg/kg daily for three days). In these tissues in which the frequency of spontaneous contraction was increased, tyramine (10^{-5} M) did not inhibit morphine-induced contractions.

Morphine-induced contractions were inhibited by adenosine (5×10^{-6} M), adenosinetriphosphate (ATP) (5×10^{-6} M), papaverine (5×10^{-5} M) and by removing calcium from the bathing medium, but were unaffected by tetrodotoxin (3×10^{-7} g/ml), which caused contractions similar to those produced by morphine.

These results suggest the mechanism underlying the actions of morphine in rat colon is unlike that responsible for the actions of morphine in dog intestine (Burks, 1973). They suggest that morphine may cause contractions in rat colon by removing a tonic inhibitory nervous influence, which normally suppresses myogenic activity involving a pacemaker (Connor, Prosser & Weems, 1974) within smooth muscle (Wood, 1972; Tonini, Leccinini & Crema, 1974).

M.G.C.G. is an M.R.C. Scholar.

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Increased cerebral cyclic GMP concentration induced by muscarinic cholinergic agonists and prostaglandin $F_{2\alpha}$

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An important role for cyclic nucleotides in the functioning of the central nervous system has been indicated by a number of investigations (see Daly, 1975). In previous studies in this laboratory the use of the neonate chick with its immature blood-brain barrier has allowed the investigation of the effects of several neurohormones on cerebral cyclic AMP formation *in vivo* (Edwards, Nahorski & Rogers, 1974). In view of the growing evidence that cyclic GMP may also be involved in the processes of neurotransmission (Goldberg, O'Dea & Maddox, 1973), we have made appropriate studies on the nucleotide using the above experimental model.

All experiments were performed on 3-day old male Ranger chicks. Drugs were injected into the right jugular vein and at appropriate intervals the cerebral hemispheres were removed by a freeze-blowing apparatus (Veech, Harris, Veloso & Veech, 1973). Cyclic GMP was assayed, after chromatographic purification with alumina and Dowex 50 resin, by a modification of the protein binding method of Dinnendahl (1974).

Unlike cyclic AMP (Nahorski, Rees & Rogers, 1975) the concentration of cyclic GMP (50–70 pmol/g) in the cerebral hemispheres was not influenced by decapitation or by age over the perinatal period. The administration of oxotremorine, arecoline or methacholine (0.1–0.5 μmol/kg) induced a rapid increase in cyclic GMP concentration. Maximum increases (100–200%) were observed 3 min after injection. Of a number of other substances studied prostaglandin $F_{2\alpha}$ (0.5 μmol/kg was also effective in increasing (80–100%) the nucleotide concentration *in vivo*. However, noradrenaline, dopamine, 5-hydroxytryptamine, adrenaline, isoprenaline, clonidine, histamine, γ-aminobutyric acid and prostaglandin E_1 were found to be ineffective, in doses up to those producing marked behavioural effects.

Further *in vitro* studies of the effects of muscarinic agonists were made. Slices of chick cerebral hemispheres were pre-incubated for 30 min in Krebs-bicarbonate buffer. In the presence of the potent phosphodiesterase inhibitor papaverine (0.05 mM) the concentration of cyclic GMP in the slices (12–14 pmol/mg protein) was 2-fold higher than in its absence. Moreover exposure to oxotremorine (10 μM) produced a further increase of 100% in cyclic GMP only in the presence of papaverine.

From these results it is clear that the factors regulating cerebral cyclic GMP concentration differ from those controlling cyclic AMP. These initial studies show that in chick cerebral tissue, muscarinic agonists and prostaglandin $F_{2\alpha}$ stimulate cyclic GMP formation, but whether or not these effects result from the direct stimulation of cell surface receptors remains to be established.

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The effect of chronic lithium administration on stimulation-induced changes in forebrain 5-hydroxyindoles: modification by chlorimipramine

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Collard & Roberts (1974) showed that chlorimipramine reduced the production of 5-hydroxyindoleacetic acid (5-HIAA) by stimulation of the medial raphe nucleus, probably by inhibiting reuptake of neuronally released 5-hydroxytryptamine (5-HT). Using this method of examining 5-HT release this study reports the effect of lithium (Li^+).

Male Albino Wistar rats weighing 150–250 g were divided into two groups of 36 animals. One group received a daily injection of isotonic (0.15 M) LiCl (0.75 mEq/kg i.p.) for 10 days while the control group received saline. Twenty-four hours after the last dose of Li^+ or saline, half the animals in each group received chlorimipramine (5 mg/kg i.p.), while the others received saline. From each of the four

Table 1 The effect of chlorimipramine (5 mg/kg i.p.) on the stimulation-induced changes in forebrain 5-hydroxyindole concentrations in control animals and in animals which had received 10 day Li^+ treatment. Results are expressed as the mean ± s.e. mean of 9 pairs of animals and analysed by the paired t test. (+ and − indicate stimulation-induced increases and decreases, respectively in 5-hydroxyindole concentration).

Stimulation-induced change in 5-HT concentration (ng/g)

	Saline	*Chlorimipramine*	*Chlorimipramine minus saline*	*P*
Control	+44 ± 53	+116 ± 59	+72 ± 99	n.s.
Lithium	+71 ± 50	−42 ± 51	−113 ± 48	0.05

Stimulation-induced change in 5-HIAA concentration (ng/g)

	Saline	*Chlorimipramine*	*Chlorimipramine minus saline*	*P*
Control	+93 ± 12	−16 ± 15	−109 ± 20	0.001
Lithium	+98 ± 21	+78 ± 14	−20 ± 22	n.s.