Morphological studies of the developing lymphoid tissues have been completed in the mouse, rabbit, guinea pig and dog. These support the foregoing conclusions and will be reported elsewhere.

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EXCITATION AND INHIBITION OF BRAIN-STEM NEURONES BY *NOR*ADRENALINE AND ACETYLCHOLINE

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THE technique of iontophoretic application of drugs by micro-pipettes introduced by Nastuk[1] and developed by del Castillo and Katz[2] was successfully adapted for an examination of the responses of spinal neurones by Curtis and Eccles[3]. The precise analysis of the pharmacological properties of single neurones which can be achieved with this technique gives great promise for the study of drug action and chemical transmission in the central nervous system.

It is natural that the known peripheral transmitters, acetylcholine and *nor*adrenaline, should be tested for actions in the central nervous system and neurones which respond to iontophoretic application of acetylcholine have been discovered in the spinal cord[3], inferior colliculus[4] and cerebral cortex[5,6]. More recently Salmoiraghi and Steiner[7] have found that acetylcholine has both excitatory and inhibitory effects on neurones in the brain-stem. *Nor*adrenaline, so far as it has been tried[4,8,9], has been found to be ineffective or to have a non-specific inhibitory action. Previous investigations with other techniques (see review[10]) have suggested that adrenaline and *nor*adrenaline can act on neurones in the brain-stem and Bradley and Mollica[11] found that intracarotid injections of these substances excited some neurones and inhibited others.

In the experiments recorded here a survey has been made of the responses of neurones in a limited region of the brain-stem of decerebrate cats to the local application of acetylcholine and *nor*adrenaline. Four barrelled electrodes of overall tip diameter 2–12μ were used. One barrel was filled with 4 M sodium chloride for extracellular recording of unit activity; current could be passed through the other three barrels, one of which was routinely filled with 0·9 per cent sodium chloride acidified to *p*H 3 with hydrochloric acid, while the other two contained 10 per cent acetylcholine hydrochloride and 3 per cent l-*nor*adrenaline bitartrate, both at *p*H 4. A small negative backing current of 3 n.amp or less was used to prevent diffusion of the positive ions from the electrode tip. To expel the ions the current was reversed and increased to not more than 100 n.amp. The electrode was inserted under visual observation at positions in the rostral third of the medulla and

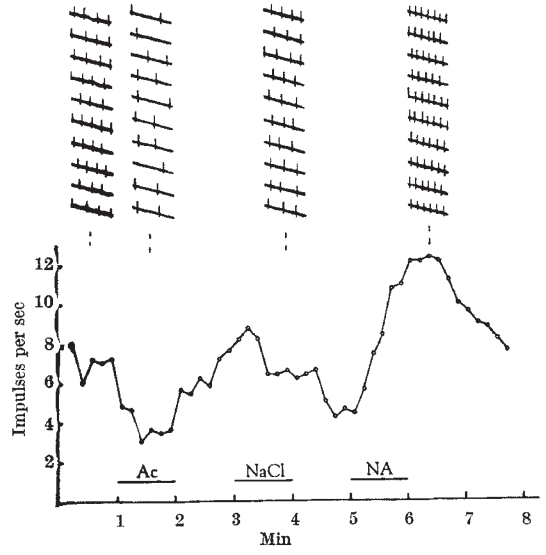

Fig. 1. Above, oscillograph records of neurone activity at times indicated; below, graph of impulse frequency against time. The black bars indicate the period of application of drugs using positive currents of 50 n.amp (electrode tip 10μ)

caudal half of the pons within 3 mm from the midline. Each neurone encountered as the electrode was moved inwards was tested for its response to current passed through each barrel. The neurone impulse frequency was sampled every second and the mean frequency over a 10-sec period was plotted against time (Fig. 1). A positive current applied to the control barrel (acidified saline) usually depressed neuronal activity but occasionally had no effect or produced a slight excitation. By carefully adjusting the current, these effects could be distinguished from drug effects by their magnitude and time course.

More neurones were found to be affected by l-*nor*adrenaline than by acetylcholine. Of 95 neurones tested, 44 (46 per cent) showed a response to this substance and of these 30 (31 per cent) were excited and 14 (15 per cent) inhibited (Figs. 1 and 2). The

(*Continued on page 873*)

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action of *nor*adrenaline persisted for some time (up to 3 min) after the passage of ions had been stopped (Fig. 1); but this effect was less marked when smaller currents were passed through finer electrodes (Fig. 2).

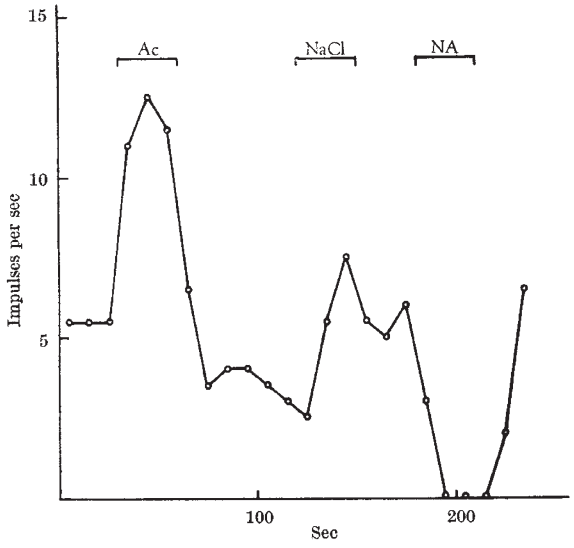

Fig. 2. Acetylcholine and sodium chloride applied with currents of 25 n.amp, *nor*adrenaline with 12 n.amp (electrode tip 2μ)

Acetylcholine was tested on 111 neurones, of which 30 (27 per cent) showed a response, 25 (22 per cent) being excited and 5 (5 per cent) being inhibited. Examples of these effects are shown in Figs. 1 and 2. The action of acetylcholine was usually more rapid in onset than that of *nor*adrenaline and rarely outlasted the period of application by more than 10 sec. These results with acetylcholine thus confirm the findings of Salmoiraghi and Steiner[7].

Interesting results were obtained when both drugs were tested on the same neurone. Ninety-three neurones were tested in this way and 22 (24 per cent) were affected by both *nor*adrenaline and acetylcholine. The majority of these, thirteen, were excited by both, but nine were excited by one drug and inhibited by the other (Figs. 1 and 2). These results are summarized in Table 1. All the results described here could be obtained consistently, that is, repeated applications of the drugs to the same neurone produced the same effects. They thus support the findings of Bradley and Mollica[11] using intracarotid injections.

Table 1. SUMMARY OF THE RESPONSES OF NEURONES TESTED WITH BOTH ACETYLCHOLINE AND *nor*ADRENALINE

		*nor*adrenaline		
		+	−	0
acetylcholine	+	13	5	5
	−	4	0	1
	0	12	9	44

+, Excitation; −, inhibition; 0, no effect or doubtful effect.

In these experiments no attempt has yet been made to identify the type of neurone physiologically but to judge from the electrode positions and the fact that most of the neurones responded to somatic stimulation over a large area of the body, it is likely that many of them were reticular. It is interesting to note that some of the neurones inhibited by one drug and excited by the other were of the type that are inhibited by some somatic stimuli and excited by others[12].

The relevance of these results to chemical transmission in this region of the brain remains to be examined, although they suggest that cholinergic and adrenergic receptors may be present on the same neurones.

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MUTAGENIC ACTIVITY OF CELLULAR MACROMOLECULES IN *DROSOPHILA MELANOGASTER*

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THE mutagenic activity of deoxyribonucleic acid (DNA) in *Drosophila* has now been conclusively demonstrated[1], especially as regards the induction of small chromosome deletions resulting in the *Minute* phenotype. On the other hand, it was not possible to induce transformation at specific gene loci by DNA extracted from organisms with the wild-type alleles. Clearly, therefore, the extracted DNA lacks the specific organization of metazoan genes and is accordingly unacceptable for incorporation into the functional genome. Mutagenic activity was not restricted to any specific DNA organization: heterologous samples from rat liver as well as from bacteriophage *T4* were found to be active. This raised the question as to whether the DNA type of mutagenesis could also occur with other biological macromolecules. Genetic tests were accordingly undertaken with deoxyribonucleoproteins (DNP), ribonucleic acid (RNA) and histones, and the results are presented in this communication.

Drosophila DNA has been successfully extracted from developing eggs (pre-larval embryos) by a method[2] devised by Dr. K. S. Kirby involving the use of diethyldithiocarbamate and phenolphthalein diphosphate. Heterologous DNA, from rat liver and bacteriophage *T4*, were prepared by the usual *p*-aminosalicylate/phenol technique[3]. DNP was prepared from embryonic cells (developing eggs) or larval