visual searching. In rodents, the nose and mouth are importantly involved in investigating the immediate environment; all these activities may, therefore, be expected to decrease in frequency of occurence if lithium does in fact suppress the performance of behaviour associated with awareness of the environment.

The increase in frequency of scratching amongst the lithium-treated female rats cannot readily be explained in terms of reduced environmental awareness. It may be that this behaviour compensates for an observed slight, but statistically non-significant, decrease in grooming with the teeth, but it is also possible that it represents a response to a local irritation caused by the drug injection: its non-occurrence in males, however, remains puzzling.

Lithium salts are toxic and are known to produce a condition in rats which has been referred to as 'lithium sickness'[6]; it is not easy to see how this could have produced the pattern of findings noted in the present study, though the possibility must still be borne in mind that some of the effects may represent toxic reactions.

Our understanding of the actions of lithium in the treatment of recurrent mood disorders[7], and of the mechanisms underlying those disorders themselves, will be enhanced by the closer investigation of the drug's action upon animal behaviour, but such studies must involve the use of appropriate test situations which are likely to prove sensitive to the slight behaviour modifications which lithium can produce. Such subtle effects may eventually be found to be crucial in defining the psychopharmacological profile of lithium.

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The Action of Substance P on Mesencephalic Reticular and Substantia Nigral Neurones of the Rat

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Summary. Extracellular recordings were made from neurones in the mesencephalic reticular formation and substantia nigra of the rat which was anaesthetized with urethane 1.5–2 g/kg i.p. Out of 44 cells tested 42 were excited by Substance P applied iontophoretically and in some cases this excitation was rapid. Evidence is presented for Substance P as a putative excitatory transmitter onto reticular and nigral neurones possibly released from primary sensory afferents.

Substance P is found distributed widely in the central nervous system[2] and there are particularly high levels in the substantia nigra[3]. Recently NILSSON, HÖKFELT and PERNOW[4] have investigated the distribution of Substance P in the rat central nervous system using immunohistochemistry. They found evidence for its location in nerve endings but not in cell bodies. Particularly high levels occurred in the substantia gelatinosa, nucleus parasolitarius, dorsal and ventral lateral geniculate body, medial and central amygdaloid nuclei and the substantia nigra. They suggested these nerve endings could arise from primary sensory afferents. Substance P has been shown to have an effect on cells in several areas of the brain and cord[5–7]. For example, KRNJEVIC and MORRIS[6] found that it was excitatory on cuneate neurones but while the excitation was strong it was slow in onset, the onset occurring after a delay of 10–30 sec and reaching a peak after 30 sec. Following termination of ejection the response decreased gradually over a 60 sec or longer period. These authors concluded that Substance P was unlikely to be the quick acting transmitter released from primary afferent terminals. In the present study we have investigated the effect of Substance P on reticular and nigral cells and compared its action with that of acetylcholine.

Materials and methods. Experiments were performed on 18 female Wistar Albino rats weighing 150 g and anaesthetized with urethane 1.5–2 g/kg i.p. Extracellular recordings were made from single neurones in the mesencephalic reticular formation and substantia nigra using parallel multibarrel glass microelectrodes[8]. Cell position was located by ejection of Pontamine Sky Blue from the recording barrel[9] and then preparing frozen sections. Substance P was synthesized by a method fully described by YAJIMA, KITAGAWA and SEGAWA[10], and ejected iontophoretically from a 0.003 *M* solution of dilute tartaric acid, pH 5.5–6.0. Both cathodal and anodal current were used to release Substance P. Positive results were obtained only with an anodal current. As previous workers[6] reported release of Substance P by a cathodal current, the release of Substance P was measured from electrodes which had produced results in vivo. Release of Substance P was measured by placing the tip of the electrode in 0.2 ml Krebs solution and applying 50 nA of either a cathodal or anodal current for 20 min. The amount of Substance P in the solutions was measured using a bio-assay technique[2]. Acetylcholine and γ-aminobutyric acid (GABA) were ejected iontophoretically as cations from 0.2 *M* solution in distilled water, pH 4.5. Current balancing was used during the ejection of all drugs[11].

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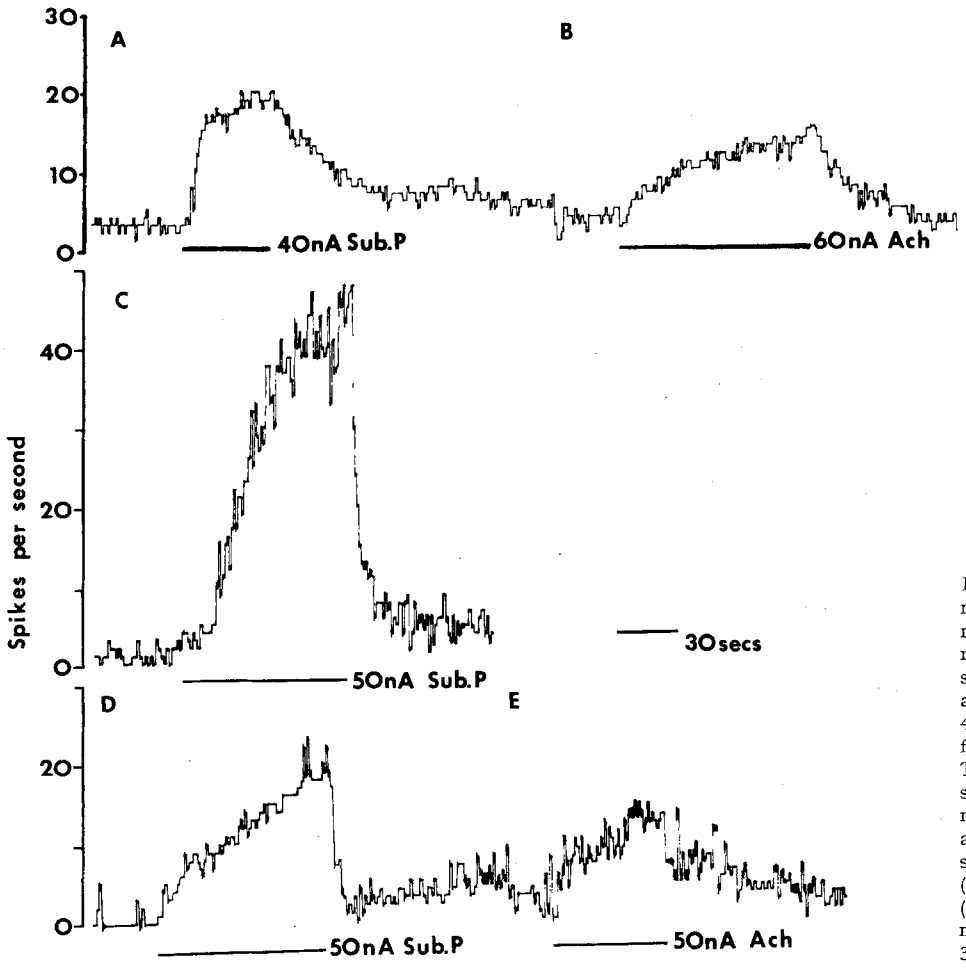

Rate meter recordings from 3 neurones in the mesencephalic reticular formation and substantia nigra of the rat. Traces A and B show the effect of iontophoretically applied Substance P (40 nA for 45 sec) and acetylcholine (60 nA for 100 sec) on a single neurone. Trace C show the effects of Substance P (50 nA for 85 sec) on a neurone which was unaffected by acetylcholine. Traces D and E show the effect of Substance P (50 nA for 85 sec) and acetylcholine (50 nA for 60 sec) on another neurone. The time bar represents 30 sec.

Results and discussion. Substance P (8–50 nA) was tested on 44 neurones of which 42 responded by excitation (Figure A, C, D) and 2 had no effect, none were inhibited. Of the 44 cells, 27 were also examined for their response to acetylcholine (30–60 nA) and of these 11 were excited (Figure B and E), and 7 were inhibited by acetylcholine while the remainder failed to respond, for example, the cell shown in Figure C. GABA (10–40 nA) inhibited all cells tested. In all cases where a cell responded to both Substance P and acetylcholine, Substance P was more potent. There did not appear to be any correlation between the action of Substance P and acetylcholine. Certain of the cells responded rapidly to Substance P, the response being within 4 sec (Figure A), while other cells responded after a latency of 16 sec (Figure C). The delay before reaching peak firing after the application of Substance P also varied from 8–72 sec (see Figure); following cessation of the pulse the response either rapidly terminated within 8 sec (Figure D), or recovered slowly over a period of 140 sec (Figure A). All the cells illustrated in the Figure have a slow firing rate of 2 to 4 per sec resting discharge without activation with homocysteic acid. This firing rate is characteristic of the dopamine cell bodies of the zona compacta[12]. Histological evidence from the present study indicates that some of the recordings were made from this area.

As previous workers[6,7] had expelled Substance P using cathodal current, release of Substance P was determined from electrodes using both cathodal and anodal current (see methods section). Substance P, approximately 3 p g/min, was found to be released from the anodal current but none was released with cathodal current. Controls were also used where a barrel of the multibarrel microelectrode was filled with tartaric acid and this had no effect on cells which were excited by Substance P.

In contrast to cells in the cuneate nucleus[6] and the cerebral cortex[7], certain of the cells in this study responded very rapidly to Substance P, reaching a peak after only 8 sec. This would be in accordance with a role for Substance P as the rapidly acting transmitter released from primary afferent nerve endings[13]. Sensory information passes to the substantia nigra area[14] and from the immunohistochemical studies of Nilsson, Hökfelt and Pernow[4] Substance P terminals end in the substantia nigra where there are high level of Substance P[3] and so there is good histochemical and neurochemical evidence for it as a putative transmitter in this area. And the present neuropharmacological study presents evidence for a direct action of Substance P on cells in this area[15].

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