molar excess of diethylstilboestrol (DES); unlabelled steroids were also present in specificity experiments.

Considerable amounts of DES suppressible binding of 17β-oestradiol (DSB) were found in cytosol from both brain areas of both sexes; no DSB was found in heart cytosol or plasma. Saturation binding capacities for 17β-oestradiol in both brain regions of either sex were of the order 1.5×10^8 sites/mg wet weight and equilibrium dissociation constants were all of the order 0.5×10^{-9} M; similar magnitudes and ranking orders of the affinities of other steroids for the oestradiol binding moiety were also found. Kds were of the order 10^{-6} for corticosterone, progesterone, testosterone and 5α-dihydrotestosterone, 4×10^{-8} M for 5α-androstan 3β 17β diol and 5α-androstan 3α-17β-diol, 10^{-9} M for 17α-oestradiol, oestriol, 16-epioestriol and DES. Thus the DSB we have measured and characterized from brain of 5-day-old rats is distinct from the high affinity receptors in adult brain in distribution, Kd, and specificity (Ginsburg *et al.*, 1973 and 1974) nor can it be identical with the oestradiol binding in perinatal rat brain cytosol described by Plapinger, McEwen & Clemens (1973) which, like oestradiol binding in blood, is not suppressed by DES and is of much lower oestradiol affinity. These oestradiol binding reactions do not necessarily represent the hormone receptor interactions involved in sexual differentiation—a protective function of the reaction should also be considered.

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Assessment of the agonist and antagonist activities of narcotic analgesic drugs by means of the mouse vas deferens

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It has been shown (Kosterlitz, Waterfield & Berthoud, 1974) that the correlation between the analgesic effect in man and the depression of the electrically induced contraction of the longitudinal muscle of the guinea-pig ileum is very high ($r = 0.930$), $n = 13$, potency spread >3 log units). Moreover, the correlation between the antagonist potencies determined in the morphine-dependent monkey and those measured in the guinea-pig ileum is also very high ($r = 0.974$, $n = 10$, potency spread >2 log units).

Recently it has been found that the responses of the longitudinal muscle of the mouse vas deferens to electrical stimulation (0.1 Hz, 1 ms, supramaximal voltage) are depressed by morphine-like drugs and that naloxone antagonizes this effect (Henderson, Hughes & Kosterlitz, 1972). For the assessment of the agonist and antagonist potencies of narcotic analgesic drugs by this preparation a method was used which is similar to that developed for the guinea-pig ileum (Kosterlitz & Watt, 1968). Normorphine, which in both preparations is equipotent with morphine, was the standard of reference because the onset of action and the recovery from it are very rapid. After two successive dose-response curves for normorphine had been constructed, the drug to be assayed was added to the organ bath (3 ml Mg-free Krebs solution) in a concentration to give a depression of the twitch by 20-40%. The degree of antagonist activity and the recovery from it were tested by suitable concentrations of normorphine at intervals of 7 minutes. The agonist and antagonist potencies were calculated as described for the guinea-pig ileum.

The values of the relative agonist activities obtained on the guinea-pig ileum and on the mouse vas deferens showed good agreement for compounds without antagonist component (codeine, pethidine, diamorphine and levorphanol) although the mean absolute sensitivity of the mouse vas deferens is only one-seventh of that of the guinea-pig ileum. For compounds with dual agonist and antagonist activities, the agreement between the relative agonist activities was not as

good, because in the mouse vas deferens the ID_{50} values were inversely related to concentration. On the other hand, the values of relative antagonist activities obtained by the two methods showed close correlation.

It would appear that the morphine receptors in the mouse vas deferens and the guinea-pig ileum are similar in principle but exhibit differences in detail.

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Modification of the antinociceptive activity of narcotic agonists and antagonists by intraventricular injection of biogenic amines in mice

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It has been reported that intracerebroventricular (i.c.v.) injections of 5-hydroxytryptamine (5-HT) potentiate, and those of noradrenaline (NA) attenuate, the antinociceptive activity of morphine in rats, although neither amine given alone acutely alters the nociceptive threshold (Sparkes & Spencer, 1969, 1971). Although further studies in mice confirmed these observations (Calcutt & Spencer, 1971), the results of studies with partial agonists were not clear since nalorphine was potentiated by 5-HT whilst pentazocine was not, and the antagonist naloxone abolished the prolongation in reaction time produced by 5-HT.

The present communication represents a further study of the interactions of i.c.v.-injected amines with narcotic agonist and partial agonist/antagonist drugs. Male mice of the ICI albino strain, weighing 18-22 g, were injected subcutaneously with agonist or partial agonist/antagonist agents, and the nociceptive threshold determined repeatedly during the next 3 h, using a tail-immersion technique similar to that described by Ben-Bassat, Peretz & Sulman (1959). In these experiments, mice were held in individual ventilated plastic containers at a room temperature of 21-22° C and nociceptive reaction times determined when the tail was immersed in a water bath maintained at 48° C.

Intraventricular 5-HT (10-100 μg) produced only brief increases in reaction time which were not dose-related. When injected 15 min before the peak effects of morphine (2.5 mg/kg), diacetylmorphine (0.75 mg/kg), pethidine (15 mg/kg) or etorphine (0.5 μg/kg), 5-HT (10 μg/mouse i.c.v.), produced a potentiation and prolongation of antinociceptive effect in each case. Conversely, when NA (10 μg/mouse i.c.v.) was injected 15 min prior to the peak effect of morphine (5 mg/kg), diacetylmorphine (1.5 mg/kg), pethidine (50 mg/kg) or etorphine (2 μg/kg), there was attenuation of the antinociceptive effects in each case.

In this tail immersion test, nalorphine, pentazocine and cyclazocine injected subcutaneously produced a dose-related antinociceptive effect which exhibited a low 'ceiling effect' when compared with predominantly agonist agents.

Concurrent injection of 5-HT (10 μg/mouse i.c.v.) potentiated and prolonged the antinociceptive effects of nalorphine (10 mg/kg), pentazocine (5 mg/kg) or cyclazocine (5 mg/kg), while concurrent injection of NA (10 μg/mouse i.c.v.) attenuated the effects of these agents administered in the same doses. Naloxone (5 mg/kg) was almost inactive in this test and its effects were not potentiated by 5-HT (10 μg/mouse i.c.v.).

It is suggested from these results that the possible dependence of the antinociceptive activity of morphine on the balance between concentrations of 5-HT and NA in the brain may be extended to the effects of other narcotic agonist and partial agonist agents.

The failure of other workers to show the effects of 5-HT and NA on narcotic partial agonists may