Short communications

Lethality of the morphinan isomers levorphanol and dextrorphan

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Significantly different ($P<0.05$) LD_{50} values were found in Swiss-Webster mice for levorphanol (73 mg/kg, i.p.) and dextrorphan (120 mg/kg, i.p.). A subcutaneous injection of naloxone 15 min before challenge prevented the lethal effect of an LD_{98} of levorphanol, with ED_{50} value of 1·36 mg/kg. Naloxone, in doses from 2 to 100 mg/kg, did not prevent death caused by 150 mg/kg of either dextrorphan or levorphanol. Levorphanol was lethal for mice pretreated with 10 mg/kg of naloxone, a dose sufficient to block opiate-specific lethal effects, but the LD_{50} value was 109 mg/kg, in contrast to 73 mg/kg in the absence of naloxone. By the criteria of stereospecificity and naloxone blockade, levorphanol-induced mortality in mice is a typical opiate effect in the lower of the two dose ranges studied. At higher doses of levorphanol a non-specific effect supervenes, with an LD_{50} value virtually the same as that of dextrorphan.

The lethal effect of narcotic analgesics is widely recognized, but the underlying mechanisms are not completely understood. For example, death due to central respiratory depression (Jaffe, 1970) and pulmonary oedema following an anaphylactic reaction (Helpern & Rho, 1966) have both been described. Stereospecificity is one of the criteria for a specific opiate effect; e.g., levorphanol, the (−) isomer of 3-hydroxy-*N*-methylmorphinan, is an opiate in all respects, while dextrorphan, the (+)-isomer of levorphanol, is completely inactive as a narcotic agonist. A previous report (Goldstein & Sheehan, 1969) showed that both levorphanol and dextrorphan have lethal effects in mice at about the same high dose range and suggested that this lethality is due to some nonspecific mechanism. Nonspecific inhibitory effects of morphine at high concentrations have already been demonstrated on transmission in the superior cervical ganglion of the cat and rabbit, and on the contraction of the myenteric plexus-longitudinal muscle preparation of the guinea-pig ileum (Kosterlitz, Lees & Watt, 1969). In this paper we show that levorphanol can exert a lethal effect on mice in two ways, one of which is a typical opiate effect.

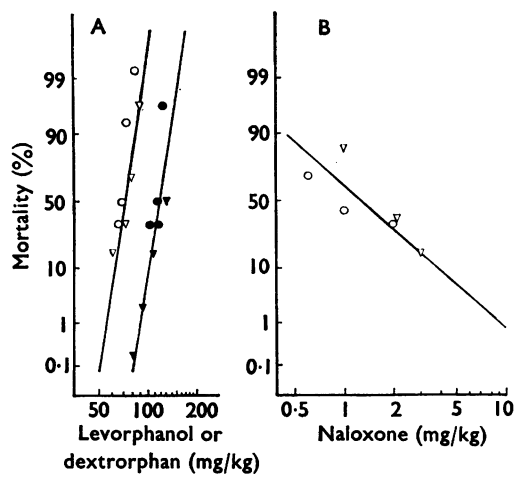

FIG. 1. Log dose–mortality curves. (A) Mice were challenged i.p. with different doses of levorphanol (open symbols) or dextrorphan (closed symbols). (B) Mice were injected s.c. with different doses of naloxone, and 15 min later were challenged i.p. with 85 (○) or 100 (▽) mg/kg of levorphanol, doses in the LD_{98} range. Each point represents 6 mice. Mortality (%) within all groups was determined 90 min after challenge and the data were plotted on logarithm-probability paper according to Litchfield & Wilcoxon (1949). Circles and triangles represent separate experiments.

Methods.—Male Swiss-Webster mice, 25–30 g, were supplied with food and water *ad libitum,* and maintained on a 12 h light-dark cycle. Levorphanol and dextrorphan were administered intraperitoneally as the tartrates (0·01 ml solution/g body weight); naloxone and 0·9% NaCl solutions were injected subcutaneously 15 min prior to challenge with levorphanol or dextrorphan. Stated doses of all drugs refer to the free bases. Mice were randomly divided into groups of 6 before each experiment and used only once. The mortality (%) within groups was determined 90 min after challenge and the data were analysed by the method of Litchfield & Wilcoxon (1949). Levorphanol tartrate and dextrorphan tartrate were generously furnished by Hoffmann-La Roche, Inc.; naloxone hydrochloride was a gift from Endo Laboratories, Inc.

Results.—Figure 1A shows log dose–% mortality curves for levorphanol and dextrorphan. The curves are parallel and yield different ($P<0·05$) LD_{50} values of 73 mg/kg (95% confidence interval, 70–76 mg/kg) for levorphanol and 120 mg/kg (113–127 mg/kg) for dextrorphan. Pretreatment with the specific narcotic antagonist naloxone prevented the lethal effects of an LD_{98} dose of levorphanol (Fig. 1B), with an ED_{50} of 1·36 mg/kg (0·94–1·97 mg/kg). However, pretreatment with naloxone, in eight doses ranging from 2·0 to 100 mg/kg, did not protect mice from the lethal effects of an LD_{98} dose of dextrorphan (not shown). Mice challenged with 150 mg/kg of levorphanol (approximately the LD_{98} dose of dextrorphan) could not be protected by pretreatment with any dose of naloxone from 5·0 to 100 mg/kg.

Four groups of mice were pretreated with 10 mg/kg of naloxone and 15 min later were challenged with different doses of levorphanol. This dose of naloxone should completely protect the mice from opiate-specific lethal effects (see Fig. 1B). Levorphanol was lethal in these naloxone-pretreated mice, but with an LD_{50} of 109 mg/kg (105–114 mg/kg), in contrast to an LD_{50} of 73 mg/kg in the absence of naloxone (see above). By the Litchfield–Wilcoxon test (1949) this value is not significantly different from the LD_{50} of dextrorphan, but it is different ($P<0·05$) from the levorphanol LD_{50} determined in the absence of naloxone (Fig. 1A). Sodium tartrate (Baker), injected intraperitoneally in amounts equimolar with the highest dose of dextrorphan tartrate tested, did not have a lethal effect.

The behaviour of mice following injection of levorphanol or dextrorphan was similar, except that only levorphanol caused the Straub tail reaction and stereotyped running activity. A few minutes after injection of either drug, the mice became ataxic and fell onto their sides. Breathing was slow and irregular, anoxia was evident, and preterminal convulsions occurred. The whole sequence of events, from injection to cessation of movement, took 10–20 minutes.

Discussion.—By the criteria of stereospecificity and naloxone blockade, it was shown that levorphanol-induced mortality in mice is a typical opiate effect in the lower of the two dose ranges studied. At higher doses of levorphanol a nonspecific lethal effect supervenes, with an LD_{50} value virtually the same as that found for dextrorphan. Neither this lethal effect nor that of dextrorphan was blocked by naloxone. It has been shown (Goldstein & Sheehan, 1969) that the slope of the log dose–mortality curve for levorphanol is different from that for the analgesic and running effects in mice, suggesting that the lethal effects of levorphanol may be mediated by receptors that are different from those mediating analgesia and the stereotyped running behaviour. Two classes of putative opiate receptors have recently been described (Goldstein, Lowney, & Pal, 1971; Pal, Lowney & Goldstein, 1972; Goldstein, 1972), based on the stereospecific binding of levorphanol in mouse brain. The opiate receptor mediating lethal effects in mice may represent the 'low affinity, high capacity' class of opiate binding sites found in mouse brain.

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