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COMMUNICATIONS

Effect of blockade of 5-hydroxytryptamine re-uptake on drug-induced antinociception in the rat

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Central serotoninergic systems have been implicated in morphine-induced antinociception in the rat. For instance, a reduction in rat brain 5-hydroxytryptamine (5-HT) content either by the administration of the tryptophan hydroxylase inhibitor *p*-chlorophenylalanine (Tenen, 1968; Gorlitz & Frey, 1972) or by electrolytic lesion of the nucleus raphé medianus (Adler, Kostowski & others, 1975) or by the intraventricular injection of 5,6-dihydroxytryptamine (Genovese, Zonta & Mantegazza, 1973) attenuates the antinociceptive effect of the drug. Conversely, morphine antinociception is potentiated by the elevation of rat brain 5-HT either by the administration of 5-hydroxytryptophan (Contreras & Tamayo, 1967) or by the intraventricular injection of 5-HT (Sparkes & Spencer, 1971). Blockade of re-uptake is an alternative procedure for increasing 5-HT availability at the receptor and the objective of this study was to determine the effect of pretreatment with a specific inhibitor of 5-HT re-uptake on the antinociceptive effect of morphine, methadone and pethidine in the rat. Fluoxetine (Lilly 110140, 3-(*p*-trifluoromethylphenoxy)-*N*-methyl-3-phenyl-propylamine hydrochloride, generously supplied by Dr. R. W. Fuller, Eli Lilly and Co., Indianapolis, U.S.A.) was used since it has been shown to be a potent inhibitor of rat brain 5-HT re-uptake whilst being essentially devoid of effect on noradrenaline re-uptake (Wong, Horng & others, 1974; Fuller, Perry & Molloy, 1975). Furthermore, it has been reported that morphine-induced hypothermia in the rat is enhanced by pretreatment with fluoxetine (Fuller & Baker, 1974). The dose of fluoxetine used in this study (10 mg kg^{-1}) completely blocks rat brain 5-HT re-uptake (I. Goodlet, personal communication) as assessed by the effect of drug pretreatment on the ability of *p*-chloroamphetamine to lower rat brain 5-HT concentrations (Meek, Fuxe & Carlsson, 1971).

Male Wistar rats, 50–60 g were used. Antinociceptive activity was assessed by means of the hot plate (55°) test. Rats were placed on the hot plate 30 min after the subcutaneous injection of analgesic or vehicle (0·9% sodium chloride). The reaction time was the time between placement on the hot plate and the licking or flicking of hind paws. Fluoxetine (10 mg kg^{-1}) or saline was injected intraperitoneally 30 min before the analgesic or saline. Each result is the mean ± s.e.m. of ten observations. Statistical significance was determined by means of Student's *t*-test (two-tailed). Doses refer to the free base.

Morphine (3 mg kg^{-1}), methadone (1 mg kg^{-1}) and pethidine (10 mg kg^{-1}) significantly increased the reaction time of the rat 30 min after subcutaneous injection. The reaction time of the rat was unaltered 60 min after fluoxetine (10 mg kg^{-1}, i.p.). Pretreatment with fluoxetine significantly increased the reaction time of the morphine-treated rats. However, the reaction time of rats receiving either methadone or pethidine was unaltered by fluoxetine pretreatment (Table 1). Similar results were obtained when the time between injection of narcotic agonist and assessment of antinociceptive activity was increased from 30 min to 60 min. Hence, the results of this study reveal that fluoxetine, at a dose devoid of antinociceptive activity, potentiates the antinociceptive effect of morphine but not that of methadone and pethidine.

Table 1. *Effect of fluoxetine on the antinociceptive activity of morphine, methadone and pethidine.*

	Reaction time (s)	
Treatment	Saline-treated	Fluoxetine-treated
Saline	6·2 ± 0·5	6·8 ± 0·6
Morphine	15·0 ± 2·4**	26·4 ± 1·7†
Methadone	12·0 ± 2·3*	14·0 ± 2·0
Pethidine	11·1 ± 1·5*	13·7 ± 1·6

Reaction times (s) are the mean ± s.e.m. of ten observations. Morphine (3 mg kg^{-1}), methadone (1 mg kg^{-1}) and pethidine (10 mg kg^{-1}) were injected subcutaneously 30 min before rats were placed on the hot plate. Fluoxetine (10 mg kg^{-1}) was injected intraperitoneally 30 min before injection of analgesic.

* Differs from saline treated, $P < 0.05$; ** differs from saline-treated, $P < 0.01$; † differs from morphine-treated, $P < 0.01$.

* Correspondence.

In agreement with the findings of others (Yarbrough, Buxbaum & Sanders-Bush, 1971, 1973; Haubrich & Blake, 1973; Perez-Cruet, Thoa & Ng, 1975), it has been previously reported from this laboratory that acutely administered morphine increases rat brain 5-HT turnover, thus suggesting a stimulation of central serotoninergic neuronal activity. Furthermore, the morphine-induced increase in amine turnover would appear to be a specific effect since it is antagonized by pretreatment with the specific narcotic antagonist naloxone. Unlike morphine, acutely administered methadone and pethidine do not increase rat brain 5-HT turnover (Goodlet & Sugrue, 1972, 1974). This observation is in excellent agreement with the report that lesioning of the rat midbrain raphé decreased the antinociceptive effect of morphine but not that of methadone and pethidine (Samanin, Ghezzi & others, 1973). Sewell & Spencer (1975) have recently reported that the intraventricular injection of 5-HT to mice significantly increased the antinociceptive effect of a number of narcotic agonists, including morphine and pethidine. Hence, their findings for pethidine are at variance with those of Samanin & others (1973) and the results of this study. Perhaps, this discrepancy is best explained by the use of different species. Certainly, the effects of narcotic agonists on the turnover of 5-HT in rat and mouse brain differ. For example, methadone, whilst having no effect on rat brain 5-HT turnover (Sasame, Perez-Cruet & others, 1972; Goodlet & Sugrue, 1974), increases the turnover of the monoamine in the mouse brain (Bowers & Kleber, 1971).

Like fluoxetine, narcotic agonists such as pethidine (Carlsson & Lindqvist, 1969) and methadone (Ciofalo, 1974) also block 5-HT re-uptake. However, this property would appear to be unrelated to their antinociceptive effect since morphine is a very weak inhibitor (Ahtee & Saarnivaara, 1973; Ciofalo, 1974). Furthermore, (+)- and (−)-methadone are equieffective in blocking 5-HT re-uptake by rat hypothalamic slices (Moffat & Jhamandas, 1974). In addition, pretreatment with naloxone does not antagonize the pethidine-induced blockade of rat brain 5-HT re-uptake (Goodlet & Sugrue, 1974). The ability of fluoxetine to potentiate the antinociceptive effect of morphine, but not that of methadone and pethidine, is in all probability attributable to its ability to block 5-HT re-uptake. However, the precise mechanism whereby blockade of 5-HT re-uptake potentiates morphine-induced antinociception awaits clarification.

In summary, the results of this study lend further support to the concept that, of morphine, methadone and pethidine, only in the case of morphine would 5-HT appear to be involved in its antinociceptive effect in the rat (Goodlet & Sugrue, 1974).

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