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Interactions of 5-Hydroxytryptamine and Narcotic Analgesics on Dog Intestine *in vivo*

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Abstract. The effects of intra-arterially (i.a.) injected morphine and dextromoramide on 5-hydroxytryptamine (5-HT)-induced contraction of the jejunum of dog were tested. Dextromoramide increased the intestinal contractile action of 5-HT from 44.7 ± 8.8 cm H_2O to 154.6 ± 17.3 cm H_2O. This effect was reversible in about 15 min. The tryptamine antagonists LSD, BOL, dibenamine, and cyproheptadine failed in blocking 5-HT and the narcotic analgesics. Atropine (50 μg/kg i.a.), however, was active in this respect. We propose that 5-HT released by morphine acts on tryptamine receptors located on the ganglionar cells and is not blocked by LSD. Possibly morphine inhibits also the uptake of 5-HT by the myenteric plexus which would explain the potentiation of exogenous 5-HT.

Key Words
Dextromoramide
5-HT
Morphine

Morphine and other narcotic analgesics release 5-hydroxytryptamine (5-HT) from the intestinal wall of dogs (2–4). Both the 5-HT release and the 5-HT content of the intestinal wall have been related to the contractile response induced by narcotic analgesics on dog intestine *in vivo* (3, 4). In isolated but vascularly perfused dog duodenum, intra-arterial morphine induces a contractile response and at the same time increases the concentration of 5-HT in the venous effluent (2, 3). Treatment with reserpine as well as tachyphylaxis to 5-HT reduced the contractile effect evoked by morphine (2, 3). We previously reported (4, 5) similar results in the dog duodenum *in vivo* with dextromoramide, a potent analgesic of the methadone class.

These results suggest that 5-HT may interact on the narcotic-analgesic-induced intestinal contraction in dogs. To investigate this hypothesis, we studied on dog intestine *in vivo* the influence of narcotic analgesics on 5-HT effect and that of some tryptamine antagonists (LSD, BOL, dibenamine, cyproheptadine and methysergide) on the actions of 5-HT and some narcotic analgesics.

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Materials and Methods

38 mongrel dogs weighing from 6 to 12 kg were anesthetized with pentobarbital sodium (30 mg/kg i.v.), intubated and mechanically ventilated.

Arterial blood pressure was recorded continuously from a femoral artery by a pressure transducer, and a polyethylene catheter was placed in a femoral vein.

The superior mesenteric artery was isolated near the aorta. A 23-gauge needle connected to a polyethylene catheter filled with heparinized saline (10 IU/ml) was then inserted to allow intra-arterial (i.a.) injections.

Next, a thin-walled water-filled balloon was inserted into the lumen of a jejunum loop through a small longitudinal incision on the intestine, and advanced 5–10 cm. The incision was then sutured and the balloon connected to a pressure transducer adjusting the pressure to 10 cm H_2O. An hour was allowed to pass before starting the experiments. All recordings were made with a Grass polygraph model 7.

Controls. The average standard response to the i.a. injection of 5-HT (5 μg/kg) and morphine (10 μg/kg) or dextromoramide (10 μg/kg) was determined for each animal. After each injection of morphine or dextromoramide (as soon as the intestinal motility returned to control levels), repeated i.a. injections of 5-HT were made, until recovery of 5-HT standard responses. 5-HT injections were spaced at least 5 min from each other to avoid tachyphylaxis.

For the study of the tryptamine antagonists the animals were divided into one group with 5 animals and four groups with 4 animals. In each group we studied one of the following drugs: LSD (lysergic acid diethylamide, 50 μg/kg i.a.), BOL (bromolysergic acid diethylamide, 50 μg/kg i.a.), dibenamine (100 μg/kg i.a.), methysergide (50 μg/kg i.a.) and cyproheptadine (100 μg/kg i.a.). The blocking agent was injected i.a. 30–60 sec prior to injection of the narcotic analgesics or 5-HT.

The following agents were used: 5-hydroxytryptamine creatine phosphate (5-HT; Sigma Chem. Co.), morphine chlorhydrate (Merck Co.), dextromoramide tartarate (SARSA), diethylamide of lysergic acid (LSD; Sandoz), diethylamide of bromolysergic acid (BOL; Sandoz), methysergide maleate (Sandoz), cyproheptadine chlorhydrate (Periatin®, Merck Sharp & Dohme), atropine sulfate (Merck Co.), dibenamine chlorhydrate, acetylcholine chlorhydrate (Roche), hexamethonium chlorhydrate.

Weights cited refer to the salts. All compounds were dissolved in saline solution. Solutions for i.a. injections were buffered with phosphate buffer (Sorensen) to pH 7. Volumes administered i.a. did not exceed 0.5 ml and were usually less than 0.2 ml.

All results are expressed as the mean ± standard error ($\bar{x} \pm s_{\bar{x}}$). Student's paired t-test was used to test significance.

Results

5-HT and Morphine

In 5 dogs, morphine (10 μg/kg i.a.) constantly and significantly increased the contractile effect induced by 5-HT. The potentiation was greater and highly significant at the first injection of 5-HT (by 4–6 min after injection of mor-

phine) (fig. 1, table I). On the average, potentiation disappeared by 15 min after morphine injection. Morphine did not significantly change the contractile effect induced by acetylcholine (10 μg/kg i.a.).

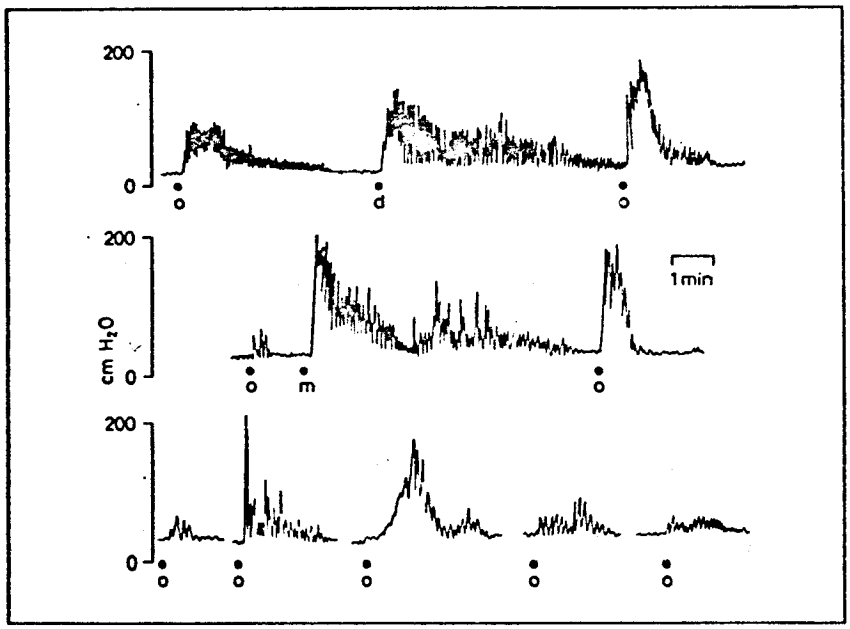

Fig. 1. Potentiation by morphine (m – 10 μg/kg i.a.) and dextromoramide (d – 10 μg/kg i.a.) of the intestinal response to 5-HT (o – 5 μg/kg i.a.) (dogs 16 and 4). Below (dog 5) 5-HT (o) before and at 5, 10, 15 and 20 min after injection of dextromoramide.

Table I. Potentiation by dextromoramide (10 μg/kg i.a.) and morphine (10 μg/kg i.a.) of the effect of 5 μg/kg i.a. of 5-HT, in the intraluminal pressure of the jejunum of the dog

n[1]	Controls $\bar{x} \pm \rho_{\bar{x}}$	5 min after		P[2]
	cm H_2O	morphine $\bar{x} \pm \rho_{\bar{x}}$ cm H_2O	dextromoramide $\bar{x} \pm \rho_{\bar{x}}$ cm H_2O	
5	+ 48.50 ± 9.01	+ 150.80 ± 20.50		<0.001
12	+ 44.75 ± 8.80		+ 154.66 ± 17.34	<0.001

1 n = Number of animals in the sample.
2 = Paired student's t-test.

5-HT and Dextromoramide

Results were the same as those obtained with morphine (fig. 1). Dextromoramide also induced a highly significant potentiation of the contractile effect induced by the first 5-HT injection (table I). This effect disappeared by the fifteenth minute after dextromoramide injection. The intestinal response to acetylcholine (10 μg/kg i.a.) did not change.

Hexamethonium

In 5 dogs the effect of intravenous administration of hexamethonium (1 mg/kg) was studied. Though hexamethonium induced a considerable drop in blood pressure (usually falling to 60 – 50 % of control level) and a decrease in spontaneous motility of intestine, it did not affect the intestinal response to 5-HT or dextromoramide (i.a.).

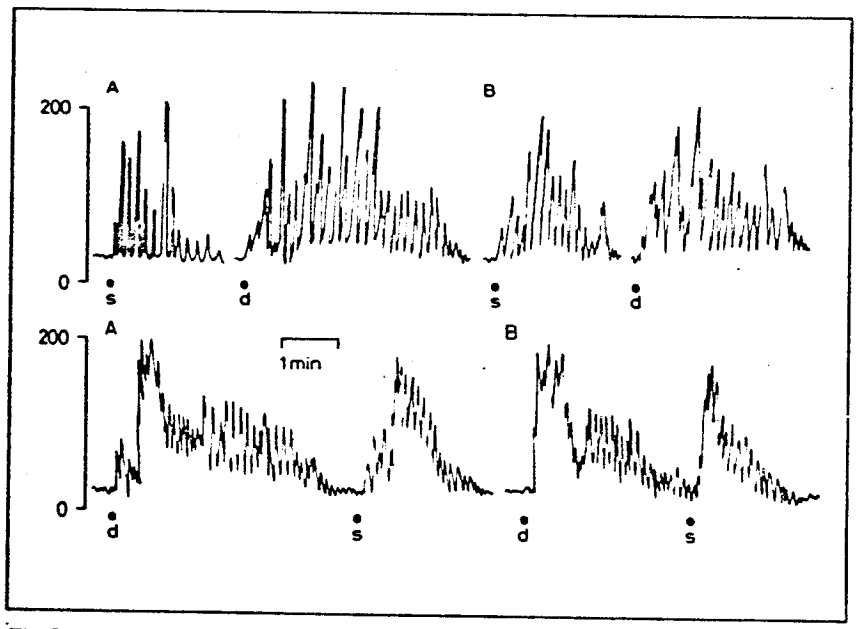

Fig. 2. Effects of LSD-25 and BOL upon the intestinal contractile response induced by 5 μg/kg i.a. of 5-HT (s) and 10 μg/kg i.a. of dextromoramide (d). In the y axis, intraluminal pressure in cm H_2O. Above (at B) 50 μg/kg i.a. of LSD-25 was injected 1 min before s and d (dog 19). Below (at B) 50 μg/kg i.a. of BOL was injected 1 min before d (dog 23).

Tryptamine Blockers

The tryptamine blockers tested had no significant effect on intestinal contractile response elicited either by 5-HT or the narcotic analgesics. They also did not change the potentiation of 5-HT by the narcotic analgesics (fig. 2). Even high doses of LSD-25 (100 μg/kg i.a. – 2 dogs) had no significant effect on the intestinal contractile response to 5-HT or narcotic analgesics. In this dose LSD-25 alone had a discrete direct contractile effect.

Atropine

In 4 dogs, 50 μg/kg i.a. of atropine blocked entirely the effect of Ach (5 μg/kg i.a.), almost entirely morphine (89.5 ± 5.2 %) but only 60–70 % of the 5-HT effect. Administration of LSD-25 (50 μg/kg i.a.) after atropine did not increase significantly the block of 5-HT, morphine, and dextromoramide, respectively (fig. 3).

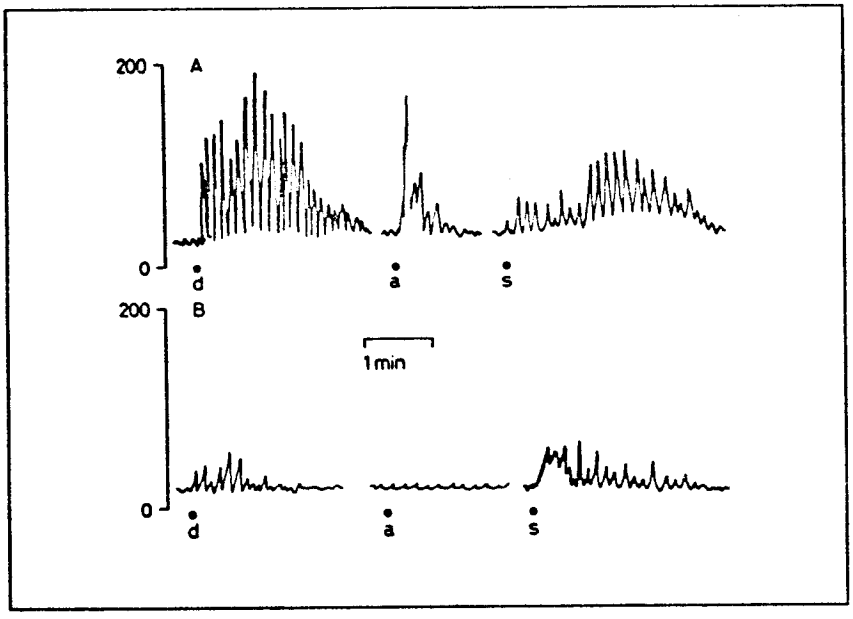

Fig. 3. Effects of atropine on acetylcholine (a – 5 μg/kg i.a.), dextromoramide (d – 10 μg/kg i.a.) and 5-HT (s – 5 μg/kg i.a.) induced contraction of the intestine (dog 11). In B, 50 μg/kg of atropine was injected i.a. immediately before d, a and s. Between each injection 15 min were allowed to pass.

Discussion

Our results show that morphine and dextromoramide have a marked potentiating effect on the 5-HT-induced contraction of dog intestine *in vivo*. The reason for the sensitization is not clear at present but could be related to a blockade by morphine of the 5-HT uptake by the tryptaminergic neurons of the intestine. Our results contrast with the work of *Gaddum and Picarelli* (8) who showed that morphine is a 5-HT antagonist in guinea-pig intestine *in vitro* ('M' receptors). However, in 1967, *Burks and Long* (2, 3), have already shown that morphine did not block the effect of 5-HT on isolated and arterially perfused (with oxygenated Krebs) dog duodenum. Possibly the different preparations are responsible for the different drug responses, because in isolated dog duodenum strips, morphine partially blocks 5-HT (6). As in all nerve tissue it is possible that the multisynaptic myenteric plexus could be affected by isolation procedures (probably hypoxia) thus changing its normal vital characteristics. On the other hand, as the tryptamine blockers blocked neither morphine nor 5-HT, we cannot discard the hypothesis that 5-HT mediates the effect of narcotic analgesics on intestine. As *Gyermeck* (10) showed, BOL, LSD and dibenamine block chiefly, on the intestine, the tryptamine receptors of the smooth muscle ('D' receptors) having only a very slight action upon receptors on the ganglion cells (neural receptors or 'M' receptors). The same has been shown by *Drakontides and Gershon* (7) in the duodenum of the mouse. It seems that 5-HT has chiefly a neural action on *in vivo* dog intestine with only a slight direct action on the smooth muscle. *Gershon et al.* (9) and *Brownlee and Johnson* (1) showed that 5-HT seems to act as a neurotransmitter between the sensorial terminals (preganglionic fibers) and the motor neurons of the myenteric plexus involved in the peristaltic reflex. Thus, if released 5-HT acts chiefly upon neural structures (probably upon the ganglion cells of the myenteric plexus) and not upon smooth muscle cells, the tryptamine blocking agents used would be rather ineffective. We propose that 5-HT released by morphine and other narcotic analgesics from the preganglionic stores in the myenteric plexus could act upon the tryptamine receptors on the postganglionic neurons. This will evoke release of acetylcholine by the postganglionic terminals and contraction of the intestine. This hypothesis would explain why atropine in low doses blocks to a large extent the effect of 5-HT and narcotic analgesics on the intestine. It also explains why LSD and hexamethonium fail to block the effects of 5-HT and narcotic analgesics on the intestine.

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