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Investigation of 5-HT_3 receptor-mediated contraction in guinea-pig distal colon

Mayumi Yamano *, Keiji Miyata

Neuroscience and Gastrointestinal Research Laboratories, Institute for Drug Discovery Research, Yamanouchi Pharmaceutical Co. Ltd., 21 Miyukigaoka, Tsukuba, Ibaraki 305, Japan

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Abstract

We investigated the participation of cholinergic and tachykininergic mechanisms in 5-hydroxytryptamine (5-HT)-induced contraction via 5-HT_3 receptors in longitudinal and circular muscle of guinea-pig isolated distal colon. 5-HT produced concentration-dependent contractile responses in longitudinal and circular muscle. The 5-HT_3 receptor antagonists ramosetron (YM060) ((*R*)-5-[(1-methyl-3-indolyl)carbonyl]-4,5,6,7-tetrahydro-1*H*-benzimidazole hydrochloride), YM114 (KAE-393) ((*R*)-5-[(2,3-dihydro-1-indolyl)carbonyl]-4,5,6,7-tetrahydro-1*H*-benzimidazole hydrochloride), ondansetron and granisetron produced a concentration-dependent shift to the right of the 5-HT concentration-response curves in both muscle. However, methysergide and GR113808 had no effect on 5-HT-induced contraction. In the longitudinal muscle, atropine concentration-dependently inhibited 5-HT-induced contraction, and tetrodotoxin abolished it. (±)-CP96,345 attenuated the contractile response to 5-HT, but (±)-SR48,968 had no effect on it. In the presence of atropine, (±)-CP96,345 completely blocked 5-HT-induced contraction. In the circular muscle, atropine had no effect on the contractile response to 5-HT, whereas tetrodotoxin completely suppressed it. The contractile response elicited by 5-HT in the circular muscle was not inhibited by either (±)-CP96,345, (±)-SR48,968, devazepide, L-365,260 or indomethacin. It is suggested that 5-HT acts via 5-HT_3 receptors to release acetylcholine and substance P, which in turn are responsible for contraction of the longitudinal muscle. In the circular muscle, as in the longitudinal muscle, 5-HT-induced contraction is mediated by the 5-HT_3 receptor. Unlike the case in longitudinal muscle, however, this contraction involves neither cholinergic nor tachykininergic transmission. It is also suggested that neither cholecystokinin (CCK) nor prostaglandins participate in 5-HT_3 receptor-mediated contraction in circular muscle.

Keywords: 5-HT_3 receptor; Colon, guinea pig; Longitudinal muscle; Circular muscle; Tachykinin

1. Introduction

Gastrointestinal motility is regulated by stimulatory and inhibitory neurotransmitters from the myenteric plexus. Among these, 5-hydroxytryptamine (5-HT) is present in the mucosa and myenteric plexus throughout the gastrointestinal tract and contributes to induction of the peristaltic reflex (Bulbring and Crema, 1958; Costa et al., 1982; Furness and Costa, 1982; Griffith and Burnstock, 1983; Yuan et al., 1994). 5-HT stimulates intestinal smooth muscle both directly and also through the release of acetylcholine and other neurotransmitters from intramural neurons (Gaddum and Picarelli, 1957; Costa and Furness, 1979; Buchheit et al., 1985; Ramírez et al., 1994).

The contractile effect of 5-HT in the longitudinal muscle of guinea-pig isolated ileum and colon has been closely investigated. Woollard et al. (1994) reported that 5-HT-induced contraction in the longitudinal muscle of guinea-pig isolated distal colon was mediated by activation of 5-HT_3 and 5-HT_4 receptors on myenteric neurons and 5-HT_1-like receptors on the muscle. This 5-HT_3 receptor-mediated contraction involved the release of acetylcholine and substance P. In the circular muscle of guinea-pig isolated ileum, Grbović and Radmanović (1987) reported that 5-HT-induced contraction was inhibited by both methysergide and atropine. Until now, however, the mechanism of 5-HT-induced contraction in the circular muscle of guinea-pig isolated colon has not been clear. In the present study, we sought evidence for the involvement of 5-HT_3 receptors in 5-HT-induced contraction in the circular muscle of guinea-pig isolated distal colon. The participation of cholinergic and tachykininergic mechanisms in 5-HT-in-

* Corresponding author. Tel.: (81-298) 525-111; Fax: (81-298) 522-965.

duced contraction through 5-HT_3 receptors was investigated in both the longitudinal and circular muscles by the use of new specific tachykinin NK_1 and NK_2 receptor antagonists. In the circular muscle preparation, we also studied the involvement of cholecystokinin (CCK) and prostaglandins in the contractile response elicited by activation of 5-HT_3 receptors.

2. Materials and methods

Male Hartley guinea pigs weighing 600–1000 g were killed by cervical dislocation and exsanguinated. The distal portion of the colon (discarding the terminal 4–5 cm) was removed.

2.1. Contractility of longitudinal muscle layer

The colon was divided longitudinally into segments approximately 2 cm in length. The longitudinal muscle with myenteric plexus was removed by gentle stroking with a cotton swab at an angle to the mesenteric attachment. The tissues were vertically suspended in a 10-ml organ bath containing Krebs-bicarbonate solution (composition in mM: NaCl 118.4, $CaCl_2$ 2.5, KH_2PO_4 1.2, KCl 4.7, $MgSO_4$ 1.2, $NaHCO_3$ 25.0, glucose 10.0) maintained at 37°C and gassed with a mixture of 95% O_2 and 5% CO_2. Tissues were attached to isometric force-displacement transducers (SB-1T, Nihon Kohden, Tokyo, Japan) connected to a recorder (MC 6621, Graphtec, Tokyo, Japan) through a carrier amplifier (AP-621G, Nihon Kohden). Equilibration was undertaken for approximately 30 min before exposure to test compounds. Isometric contractions under a loading tension of 1 g were recorded. Cumulative concentration-response curves for 5-HT (0.1–30 μM) were constructed by increasing bath concentrations of the agonist approximately 3-fold. As the cumulative concentration-response curve for 5-HT under these conditions could be constructed 4 times at 30-min intervals in the same preparation without significant change in E_{max} or EC_{50} values (data not shown), each antagonist was examined at up to three different concentrations in the same preparation. (±)-SR48,968 was allowed to pre-equilibrate for 60 min and other antagonists were allowed to pre-equilibrate for 30 min prior to construction of the second, third and fourth concentration-response curves for 5-HT, because the preliminary experiment showed that sufficient equilibration times of (±)-SR48,968, indomethacin and other antagonists in the muscle preparation were 60, 30 and 15 min, respectively. We used GR113808 (100 nM), since GR113808 (1–100 nM) produced a parallel and concentration-dependent shift to the right of the 5-HT_4 receptor agonist (5-methoxytryptamine and renzapride) concentration-response curves in the longitudinal muscle of guinea-pig isolated distal ileum in the preliminary experiment.

2.2. Contractility of circular muscle strip

The colon was divided longitudinally into segments approximately 1 cm in length. A ring of colon was opened. A circularly oriented strip (1 cm in length) was suspended in a 10-ml organ bath containing Krebs-bicarbonate solution maintained at 37°C and gassed with a mixture of 95% O_2 and 5% CO_2. Isometric contractions under a loading tension of 1 g were recorded as above. Concentration-response curves for 5-HT (0.1–30 μM) were constructed in a non-cumulative fashion by adding increasing concentrations of agonist at 15-min intervals. The non-cumulative concentration-response curve for 5-HT under these conditions could be constructed twice in the same preparation without significant change in E_{max} or EC_{50} values. In studies with antagonists, each strip was used to record two concentration-response curves, the first for the agonist alone and the second for the agonist in the presence of an antagonist. (±)-SR48,968 or indomethacin was allowed to pre-equilibrate for 60 or 30 min prior to the addition of 5-HT, respectively. Other antagonists were allowed to pre-equilibrate for 15 min. We used atropine (0.1–1 μM) in the present study, since the supramaximal concentration of acetylcholine (10 μM)-induced contraction was abolished by atropine (1 μM) in the circular muscle of guinea-pig isolated distal colon (data not shown).

2.3. Characterization of concentration-response curves

Responses were measured as an increase in isometric tension. In the preliminary experiment, 5-HT (0.1–100 μM) produced concentration-dependent contractile responses in the longitudinal and circular muscle preparations. The maximum contraction was elicited by 5-HT at 30 μM in both preparations. Responses were expressed as a percentage of the contraction produced by 5-HT (30 μM). In the presence of an antagonist, results were expressed as a percentage of the response obtained with 5-HT (30 μM) alone. Concentration-response curves for 5-HT were characterised by their maxima (E_{max}), and by the 5-HT concentration yielding a half-maximal effect (EC_{50}).

2.4. Data analysis

All values are expressed as the mean ± S.E.M. or as the mean with 95% confidence limits. Probit analysis was used to obtain EC_{50} values. The dose ratio was obtained from the ratio of EC_{50} values for 5-HT in the presence and absence of an antagonist. The pA_2 value and slope of the curves were calculated (Arunlakshana and Schild, 1959). Significance between groups was assessed by the paired *t*-test. Probabilities of $< 5\%$ ($P < 0.05$) were considered significant.

2.5. Drugs

Ramosetron hydrochloride (YM060) ((*R*)-5-[(1-methyl-3-indolyl)carbonyl]-4,5,6,7-tetrahydro-1*H*-benzimidazole hydrochloride), YM114 (KAE-393) ((*R*)-5-[(2,3-dihydro-1-indolyl)carbonyl]-4,5,6,7-tetrahydro-1*H*-benzimidazole hydrochloride), ondansetron hydrochloride, granisetron hydrochloride, GR113808 ([1-[2-[(methylsulfonyl)amino]ethyl]-4-piperidyl]methyl 1-methyl-1*H*-indole-3-carboxylate), (±)-CP96,345 ((±)-*cis*-2-(diphenylmethyl)-*N*-(2-methoxyphenyl)-methyl]-1-azabicyclo[2.2.2]octan-3-amine), (±)-SR48,968 ((±)-*N*-methyl-*N*[4-(4-acethylamino-4-phenylpiperidino)-2-(3,4-dichlorophenyl)butyl]benzamide hemifumarate), devazepide and L-365,260 (3*R*(+)-*N*-(2,3-dihydro-1-methyl-2-oxo-5-phenyl-1*H*-1,4-benzodiazepin-3-yl)-*N*′-3-methylphenyl urea) were prepared by Yamanouchi Pharmaceutical (Tsukuba, Japan). Methysergide hydrogen maleate was kindly donated by Sandoz (Basle, Switzerland). 5-HT creatinine sulfate and acetylcholine chloride (Ovisot) were purchased from E. Merck (Darmstadt, Germany) and Daiichi Pharmaceutical (Tokyo, Japan), respectively. Atropine sulfate, tetrodotoxin and indomethacin were purchased from Wako (Osaka, Japan), Sigma (St. Louis, MO, USA) and Nakalai Tesque (Kyoto, Japan), respectively. Ramosetron, GR113808, methysergide and (±)-CP96,345 were dissolved in a minimal amount of 0.1 M HCl and diluted with Krebs-bicarbonate solution. (±)-SR48,968, devazepide, L-365,260 and indomethacin were dissolved in a minimal amount of dimethyl sulfoxide and diluted with Krebs-bicarbonate solution. Other drugs were dissolved in Krebs-bicarbonate solution. The reported concentrations are the final bath concentrations.

3. Results

3.1. Contractility studies

5-HT (0.1–30 μM) produced concentration-dependent contractile responses in the longitudinal and circular muscles of guinea-pig isolated distal colon, with EC_{50} values of 1.10 (0.65–1.86) and 1.73 (1.34–2.24) μM, and with E_{max} values of 4.64 ± 0.33 g ($n = 6$) and 2.37 ± 0.47 g ($n = 11$), respectively. Fig. 1 shows representative records of these contractile responses to 5-HT in the longitudinal and circular muscle preparations.

3.2. Effect of 5-HT receptor antagonists on 5-HT-induced contraction

The selective 5-HT_3 receptor antagonists ramosetron, YM114, ondansetron and granisetron produced a parallel and concentration-dependent shift to the right of the 5-HT concentration-response curves without a decrease in maximal response in both longitudinal and circular muscle (Figs. 2 and 3). pA_2 values of these 5-HT_3 receptor antagonists are listed in Table 1. Methysergide (10 μM), which was sufficient to block 5-HT_1 and 5-HT_2 receptors, and GR113808 (100 nM), a selective 5-HT_4 receptor antagonist, had no effect on 5-HT-induced contraction in

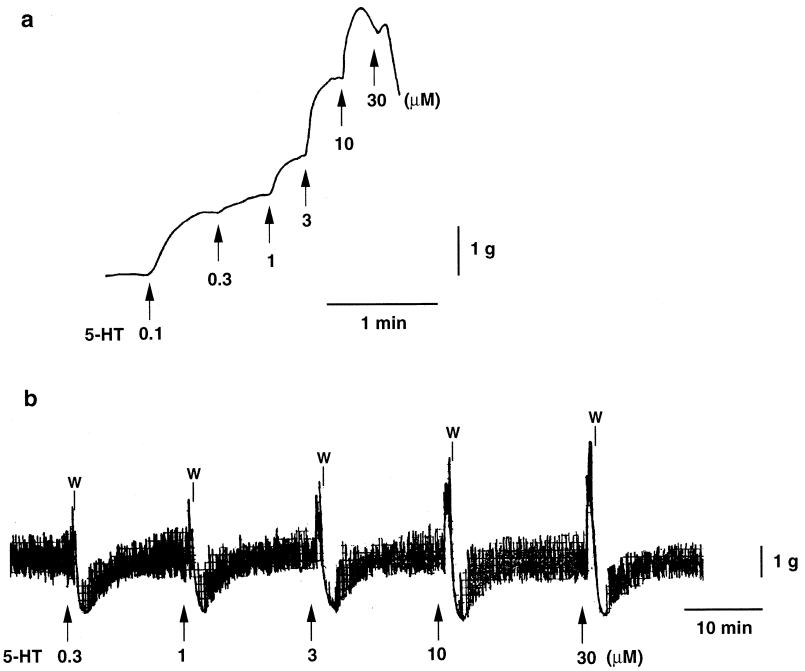

Fig. 1. Representative records showing the contractile responses evoked by 5-HT in longitudinal (a) and circular muscle (b) of guinea-pig isolated distal colon. 5-HT (0.1–30 μM) was applied cumulatively to the longitudinal muscle preparation, and non-cumulatively (0.3–30 μM) to the circular muscle preparation at 15-min intervals. Arrows indicate the addition of 5-HT.

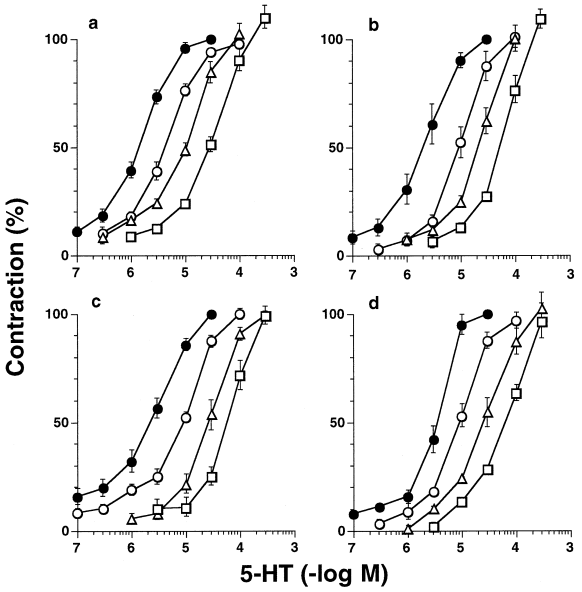

Fig. 2. Antagonism by ramosetron, YM114, ondansetron and granisetron of 5-HT-induced contraction in the longitudinal muscle of guinea-pig isolated distal colon. (a) Control (●), ramosetron 3 (○), 10 (△) and 30 nM (□). (b) Control (●), YM114 3 (○), 10 (△) and 30 nM (□). (c) Control (●), ondansetron 300 (○), 1000 (△) and 3000 nM (□). (d) Control (●), granisetron 30 (○), 100 (△) and 300 nM (□). Antagonists were incubated with the colon for 30 min prior to the construction of the 5-HT concentration-response curve. Values are the mean ± S.E.M. from 3–6 preparations and are expressed as a percentage of 5-HT (30 μM) response.

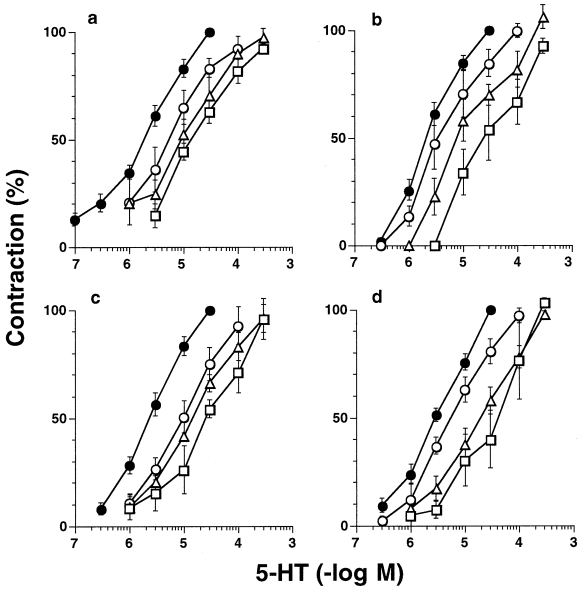

Fig. 3. Antagonism by ramosetron, YM114, ondansetron and granisetron of 5-HT-induced contraction in the circular muscle of guinea-pig isolated distal colon. (a) Control (●), ramosetron 1 (○), 3 (△) and 10 nM (□). (b) Control (●), YM114 0.3 (○), 1 (△) and 3 nM (□). (c) Control (●), ondansetron 100 (○), 300 (△) and 1000 nM (□). (d) Control (●), granisetron 10 (○), 30 (△) and 100 nM (□). Antagonists were incubated with the colon for 15 min prior to the addition of each concentration of 5-HT. Values are the mean ± S.E.M. from 3–4 preparations and are expressed as a percentage of 5-HT (30 μM) response.

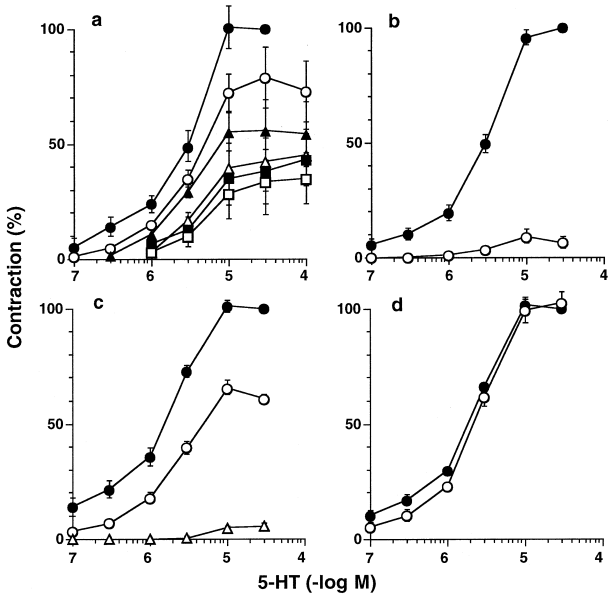

Fig. 4. Inhibitory effects of atropine, tetrodotoxin, (±)-CP96,345 and (±)-SR48,968 on 5-HT-induced contraction in the longitudinal muscle of guinea-pig isolated distal colon. (a) Control (●), atropine 10 (○), 30 (▲), 100 (△), 300 (■) and 1000 nM (□). (b) Control (●) and tetrodotoxin 1 μM (○). (c) Control (●), (±)-CP96,345 300 nM (○) and (±)-CP96,345 300 nM plus atropine 1000 nM (△). (d) Control (●) and (±)-SR48,968 100 nM (○). Antagonists were incubated with the colon for 30 (a–c) and 60 min (d) prior to the construction of the 5-HT concentration-response curve. Values are the mean ± S.E.M. from 4–6 preparations and are expressed as a percentage of 5-HT (30 μM) response.

either muscle preparation (data not shown, $n = 3–4$). None of the antagonists examined had any significant influence on baseline tension of the preparations at the concentrations used.

3.3. Effect of atropine, tetrodotoxin and tachykinin receptor antagonists on the 5-HT-induced contraction in longitudinal muscle

In the longitudinal muscle, atropine (0.01–1 μM) concentration-dependently inhibited, but did not abolish, 5-HT-induced contraction (Fig. 4a). In contrast, tetrodotoxin (1 μM) abolished 5-HT-induced contraction (Fig. 4b). Further, (±)-CP96,345 (300 nM), a selective tachykinin NK_1 receptor antagonist, attenuated this contraction, while (±)-SR48,968 (100 nM), which was sufficient to block tachykinin NK_2 receptors, had no effect on it (Fig. 4c, d). When atropine (1 μM) and (±)-CP96,345 (300 nM) were combined, however, 5-HT-induced contraction was completely blocked (Fig. 4c).

3.4. Effect of atropine, tetrodotoxin, tachykinin receptor antagonists, CCK receptor antagonists and indomethacin on the 5-HT-induced contraction in circular muscle

In the circular muscle, atropine (0.1–1 μM) had no effect on the contractile response to 5-HT (Fig. 5a), whereas

Table 1
Estimates of pA_2 values of antagonists on the 5-HT-induced contraction in the longitudinal and circular muscle of guinea-pig isolated distal colon

	Ramosetron	YM114	Ondansetron	Granisetron
Longitudinal muscle	8.76 ± 0.05 (18)	9.03 ± 0.09 (12)	6.97 ± 0.09 (11)	7.84 ± 0.04 (12)
	1.11 [0.82–1.39]	0.83 [0.36–1.30]	1.15 [0.81–1.50]	1.00 [0.76–1.24]
Circular muscle	9.01 ± 0.08 (11)	9.50 ± 0.10 (12)	7.23 ± 0.12 (12)	8.09 ± 0.06 (10)
	0.97 [0.49–1.45]	1.40 [0.88–1.91]	0.43 [0.10–0.95]	0.98 [0.62–1.33]

Data are the mean ± S.E.M. (pA_2) and the 95% slope of Schild plot with the number of experiments (n).

tetrodotoxin (1 μM) completely suppressed it (Fig. 5b). The contractile response elicited by 5-HT in the circular muscle was not affected by either (±)-CP96,345 (300 nM), (±)-SR48,968 (100 nM), devazepide (10 nM), a CCK_A receptor antagonist, or L-365,260 (300 nM), a CCK_B receptor antagonist (data not shown, $n = 3–5$). In contrast, indomethacin (3 μM), a cyclooxygenase inhibitor, significantly enhanced the magnitude of contractile response evoked by 5-HT (Fig. 6) and the amplitude of spontaneous contraction (data not shown, $n = 5$). The magnitude of acetylcholine (10 μM)-induced contractile response, however, was also significantly enhanced by indomethacin (data not shown, $n = 6$).

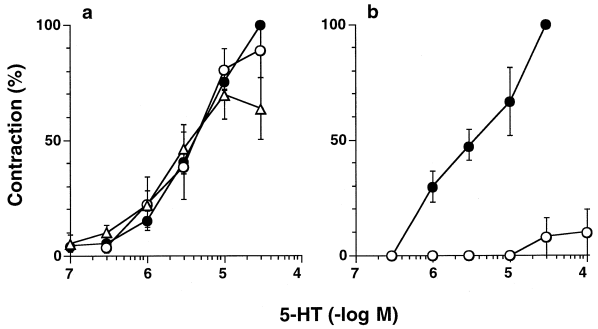

Fig. 5. Inhibitory effects of atropine and tetrodotoxin on 5-HT-induced contraction in the circular muscle of guinea-pig isolated distal colon. (a) Control (●), atropine 100 (○) and 1000 nM (△). (b) Control (●) and tetrodotoxin 1 μM (○). Antagonists were incubated with the colon for 15 min prior to the addition of each concentration of 5-HT. Values are the mean ± S.E.M. from 3–5 preparations and are expressed as a percentage of 5-HT (30 μM) response.

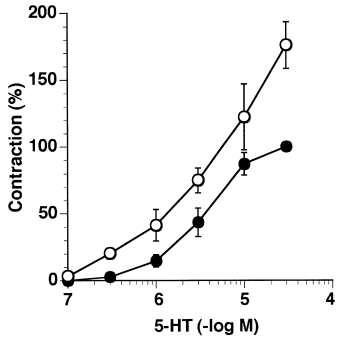

Fig. 6. Concentration-response curves to 5-HT in the circular muscle of guinea-pig isolated distal colon in the absence (●) or presence (○) of indomethacin 3 μM. Indomethacin was incubated with the colon for 30 min prior to the addition of each concentration of 5-HT. Values are the mean ± S.E.M. from 5 preparations and are expressed as a percentage of 5-HT (30 μM) response.

4. Discussion

The results of the present study suggest that the contractile response induced by 5-HT is mediated through the activation of 5-HT_3 receptors on neurons innervating both longitudinal and circular muscle of guinea-pig isolated distal colon. 5-HT produced concentration-dependent contractile responses in both the longitudinal and circular muscle of guinea-pig isolated distal colon. On the basis of EC_{50} values, there was no marked difference in the potency of 5-HT between longitudinal and circular muscle. The selective 5-HT_3 receptor antagonists ramosetron, YM114, ondansetron and granisetron competitively antagonized the 5-HT concentration-response curves in both muscle preparation. On the basis of pA_2 values, their rank order of 5-HT_3 receptor blocking activity in both the longitudinal and circular muscle was YM114 > ramosetron > granisetron > ondansetron, with no marked difference in potency between muscle preparation. In contrast, neither the 5-HT_1 and 5-HT_2 receptor antagonist methysergide nor the selective 5-HT_4 receptor antagonist GR113808 had any effect on 5-HT-induced contraction in either preparation. Woollard et al. (1994) reported that 5-HT-induced contraction was mediated by the activation of 5-HT_3 and 5-HT_4 receptors on myenteric neurons and 5-HT_1-like receptors on the longitudinal muscle of young guinea pig (150–300 g) isolated distal colon. This discrepancy with our present findings may have been due to the difference in the age of the guinea pigs. In support of this explanation, Bayol et al. (1985) indicated that there were significant alterations during ontogenesis in the methysergide-sensitive contraction elicited by 5-HT in airway muscle preparations from guinea pigs.

It is known that the contractile response mediated by the 5-HT_3 receptor is sensitive to muscarinic and tachykinin NK_1 receptor antagonists in the longitudinal muscle of guinea-pig ileum and colon (Ramírez et al., 1994; Woollard et al., 1994). In the present study, 5-HT_3 receptor-mediated contraction in the longitudinal muscle of guinea-pig isolated distal colon was partially suppressed by atropine and (±)-CP96,345, a selective tachykinin NK_1 receptor antagonist, and abolished when both atropine and

(±)-CP96,345 were present. These results are consistent with previous reports, and indicate that 5-HT-induced contraction in the longitudinal muscle involves the stimulation of both acetylcholine and substance P release in the enteric nervous system. In contrast, unlike the situation in the longitudinal muscle, atropine and (±)-CP96,345 had no effect on 5-HT-induced contraction via 5-HT_3 receptors in the circular muscle of guinea-pig isolated distal colon. However, 5-HT-induced contraction in the circular muscle was completely inhibited by tetrodotoxin, indicating that it was mediated through the stimulation of release of neurotransmitters other than acetylcholine and substance P via 5-HT_3 receptors. Wardell et al. (1994) reported that 5-HT-immunoreactive nerves innervated both 5-HT and non-5-HT neurons in enteric neurons of the guinea-pig distal colon.

A number of candidates may be proposed as neurotransmitter released in the enteric nervous system which, in turn, is responsible for contraction in the circular muscle of guinea-pig intestine. Maggi et al. (1994a,b) reported that atropine-resistant electrical stimulation-induced contraction was inhibited not only by a tachykinin NK_1 but also by an NK_2 receptor antagonist in the circular muscle of guinea-pig ileum and colon. In the present study, however, (±)-SR48,968, a tachykinin NK_2 receptor antagonist, had no effect on 5-HT-induced contraction in the circular muscle. CCK is reported to produce contraction in the circular muscle of guinea-pig isolated ileum (Bartho et al., 1987; Botella et al., 1992, 1994), but neither devazepide, a CCK_A receptor antagonist, nor L-365,260, a CCK_B receptor antagonist, had any effect on 5-HT-induced contraction in the present study. Prostaglandins (PGE_2 and $PGF_{2\alpha}$) also induce contraction in the circular muscle of guinea-pig isolated ileum and colon (Ishizawa, 1988, 1991; Botella et al., 1993). Further, Beubler et al. (1989) showed that 5-HT stimulated the release of PGE_2 in the intestine of rats in vivo. Although these results may indicate that prostaglandins released in the enteric nervous system are involved in the 5-HT_3 receptor-mediated contraction, indomethacin significantly enhanced the magnitude of contractile response evoked by 5-HT and the amplitude of spontaneous contraction in the circular muscle preparation in the present study. It is therefore considered that neither prostaglandins nor CCK participate in 5-HT_3 receptor-mediated contraction in the circular muscle of guinea-pig isolated distal colon. However, our results suggest that a cyclooxygenase-derived eicosanoid mediates relaxation and limits 5-HT-induced contraction in the circular muscle preparation. It was reported that PGE_2 caused not only contraction but also relaxation in the circular muscle of isolated guinea-pig ileum and colon (Ishizawa, 1988; Calixto and Medeiros, 1991; Botella et al., 1993). Taken together, it might be suggested that 5-HT increases the release of prostaglandin, which in turn is responsible for relaxation in the circular muscle of guinea-pig isolated distal colon. In the present report, however, indomethacin enhanced the magnitude of the contractile response induced not only by 5-HT but also by acetylcholine in the circular muscle preparation. Therefore, the mechanism of the indomethacin-induced effect on the circular muscle has not been clarified, and further studies are needed to make it clear.

A number of other candidates as neurotransmitter involved in 5-HT_3 receptor-mediated contraction in the circular muscle preparation may be considered. It is reported that galanin (Botella et al., 1992, 1995) and calcitonin gene-related peptide (Holzer et al., 1989) evoke contraction in the circular muscle of guinea-pig ileum. Furthermore, contractile effects of glutamate (Wiley et al., 1991; Koyuncuoglu et al., 1992), ATP (Wiklund and Gustafsson, 1988), γ-aminobutyric acid (Ong and Kerr, 1984), bombesin (Garzon et al., 1985; Takeda et al., 1990), somatostatin (Takeda et al., 1989) and pituitary adenylyl cyclase activating peptide (Katsoulis et al., 1993) have been reported in the longitudinal muscle of guinea-pig ileum, while those in the circular muscle have not been investigated. These findings suggest that these neurotransmitters may participate in 5-HT_3 receptor-mediated contraction in the circular muscle of guinea-pig isolated distal colon. However, further studies are needed to confirm which neurotransmitter is involved.

In conclusion, these results suggest that 5-HT acts via 5-HT_3 receptors to release acetylcholine and substance P which, in turn, are responsible for contraction in the longitudinal muscle of guinea-pig isolated distal colon. In the circular muscle, 5-HT-induced contraction is mediated by the 5-HT_3 receptor, as in longitudinal muscle, but, unlike the longitudinal muscle, this contraction involves neither cholinergic nor tachykininergic transmission. It is also suggested that neither CCK nor prostaglandins participate in 5-HT_3 receptor-mediated contraction in the circular muscle.

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