Effects of *Hange-shashin-to* (TJ-14) and *Keishi-ka-shakuyaku-to* (TJ-60) on contractile activity of circular smooth muscle of the rat distal colon

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Submitted 25 May 2012; accepted in final form 16 August 2012

**Kito Y, Teramoto N. Effects of *Hange-shashin-to* (TJ-14) and *Keishi-ka-shakuyaku-to* (TJ-60) on contractile activity of circular smooth muscle of the rat distal colon. Am J Physiol Gastrointest Liver Physiol 303: G1059–G1066, 2012. First published August 23, 2012; doi:10.1152/ajpgi.00219.2012.—The Japanese Kampo medicines *Hange-shashin-to* (TJ-14) and *Keishi-ka-shakuyaku-to* (TJ-60) have been used to treat symptoms of human diarrhea on an empirical basis as Japanese traditional medicines. However, it remains unclear how these drugs affect smooth muscle tissues in the distal colon. The aim of the present study was to investigate the effects of TJ-14 and TJ-60 on the contractile activity of circular smooth muscle from the rat distal colon. TJ-14 and TJ-60 (both 1 mg/ml) inhibited spontaneous contractions of circumferentially cut preparations with the mucosa intact. Blockade of nitric oxide (NO) synthase or soluble guanylate cyclase activity abolished the inhibitory effects of TJ-60 but only attenuated the inhibitory effects of TJ-14. Apatin (1 μM), a blocker of small-conductance Ca²⁺-activated K⁺ channels (SK channels), attenuated the inhibitory effects of 5 mg/ml TJ-60 but not those of 5 mg/ml TJ-14. TJ-14 suppressed contractile responses (phasic contractions and off-contractions) evoked by transmural nerve stimulation and increased basal tone, whereas TJ-60 had little effect on these parameters. These results suggest that 1 mg/ml TJ-14 or TJ-60 likely inhibits spontaneous contractions of the rat distal colon through the production of NO. Activation of SK channels seems to be involved in the inhibitory effects of 5 mg/ml TJ-60. Since TJ-14 has potent inhibitory effects on myogenic and neurogenic contractile activity, TJ-14 may be useful in suppressing gastrointestinal motility.**

**DIARRHEA, CHARACTERIZED BY** frequent intestinal evacuation with abnormally high fluid content in the stool, is an important worldwide public health challenge. It is often caused by a gastrointestinal infection, which can be spread through contaminated food or drinking water, or from person to person as a result of poor personal hygiene. According to the World Health Organization (WHO), diarrhea was the second leading cause of death in low-income countries in 2008. However, diarrhea is also common in developed countries. It has been reported that ~5% of the general population of developed countries suffer from chronic diarrhea, defined as a decrease in fecal consistency lasting for ≥4 wk (7, 11, 27). The common causes of chronic diarrhea are irritable bowel syndrome (IBS), inflammatory bowel diseases (IBDs) such as ulcerative colitis and Crohn’s disease, malabsorption syndromes such as lactose intolerance and celiac disease, and chronic infections (particularly in patients who are immune-compromised) (3, 5, 27). Thus it is important to develop new treatments to ameliorate chronic diarrhea that occurs in these conditions.

*Hange-shashin-to* (TJ-14) and *Keishi-ka-shakuyaku-to* (TJ-60) are traditional Japanese Kampo medicines, both of which have been used to treat symptoms of diarrhea. TJ-14 consists of seven herbs, namely *Pinelliae tuber, Scutellariae radix, Glycyr rhizae radix, Zizyphi fructus, Ginseng radix, Coptidis rhizoma,* and *Zingiberis siccatum rhizoma* (12, 13, 14, TJ). TJ-60 consists of five herbs: *Cinnamomum cortex, Paeoniae radix, Zingiberis rhizoma, Zizyphi fructus,* and *Glycyr rhizae radix* (24). It has been reported that TJ-14 is effective in the treatment of acute or chronic gastrointestinal catarrh, fermentative diarrhea, and acute enterogastritis (14). TJ-14 is also useful for preventing and controlling diarrhea induced by irinotecan (CPT-11), an anticancer drug (21). It has been reported that TJ-14 inhibits prostaglandin E₂ (PGE₂) production directly by decreasing cyclooxygenase-2 (COX2) activity and indirectly by suppressing the secretion of corticosterone (12, 13); it also enhances water reabsorption in the rat colon (12). TJ-14 also ameliorates colonic inflammation and body weight loss induced by 2,4,6-trinitrobenzene sulfonic acid (TNBS) (16). Compared with TJ-14, less is known about the effects of TJ-60 in the treatment of diarrhea (24). Surprisingly, there have been no reports into the mechanisms by which these Kampo medicines affect colonic motility.

We have investigated the effects of TJ-14 and TJ-60 on the contractile activity of circular smooth muscle (CSM) from the rat distal colon. Kampo medicines were found to inhibit spontaneous CSM activity through different mechanisms: TJ-14 inhibited transmural nerve stimulation (TNS)-induced phasic contractions, off-contractions, and increases in basal tone, whereas TJ-60 had little effect on these parameters.

A preliminary account of the present results was communicated to the 85th Annual Meeting of the Japanese Pharmacological Society (18).

**MATERIALS AND METHODS**

Male Wistar rats (aged 6 wk) were obtained from a commercial breeder (Charles River, Shizuoka, Japan). Rats were weighed, anesthetized with fluoromethyl 2,2,2-trifluoro-1-(trifluoromethyl) ethyl ether (sevoflurane; Maruishi Pharmaceuticals, Osaka, Japan), and then exsanguinated by bleeding from the femoral arteries. All animals were treated according to the Guidelines for the Care and Use of Laboratory Animals of Nagoya City University Medical School, accredited by The Physiological Society of Japan, and were approved by Nagoya City University Animal Center (approval no. H19-26). The distal colon was excised and placed in a dish containing Krebs solution (see below). The colon was opened up longitudinally by a cut along the mesenteric border. For tension measurements, circumferentially cut strips (circa 2- to 3-mm wide and 10- to 12-mm long) of CSM were prepared.
CSM preparations were transferred to 2-ml organ baths and superfused with warmed (36°C) Krebs solution at a constant flow rate (4 ml/min) using a peristaltic pump (PO-1; Tokyo Rikakikai, Tokyo, Japan). Silk threads were tied around both ends of each strip; one was fixed to the bottom of the organ bath, and the other was connected to an isometric force transducer (FD-pick up TB-612T; Nihon Kohden, Tokyo, Japan) that was coupled to a bridge amplifier. An initial tension of 1 g was applied to each muscle strip preparation. After 90–120 min, strips displayed spontaneous phasic contractions. Isometric tension changes were digitized using a Digidata 1200 Interface (Axon Instruments) and stored on a personal computer for subsequent analysis. Intramural nerves were selectively stimulated by passing brief pulses of constant current (pulse duration of 0.3 ms) between two parallel silver electrodes placed in the organ bath. The amplitude and frequency of spontaneous contractions, the area-under-contraction, and the increase of basal tone evoked by TNS were determined using PClamp 9.0 software (Axon Instruments). Each of these parameters was expressed as a percentage of the control value for statistical analysis. The neural selectivity of TNS-evoked responses was confirmed by their sensitivity to tetrodotoxin (TTX) (1 μM).

Data are expressed as means ± SD (n = 5). Significant difference from control: *P < 0.05; **P < 0.01.

The ionic composition of the Krebs solution was as follows (in mM): 137.4 Na⁺, 5.9 K⁺, 2.5 Ca²⁺, 1.2 Mg²⁺, 15.5 HCO₃⁻, 1.2 H₂PO₄⁻, 134 Cl⁻, and 11.5 glucose. The solutions were gassed with 95% O₂ containing 5% CO₂, and the pH was maintained at 7.2–7.3.

Drugs used were apamin (from Peptide Institute, Osaka, Japan), atropine sulfate, N-nitro-L-arginine (L-NNA), verapamil (all from Sigma-Aldrich Japan, Tokyo, Japan), 1H-[1,2,4]oxadiazolo[4,3-a]quinoxalin-1-one (ODQ) (from Tocris Cookson, Bristol, UK), and TTX (from Wako Pure Chemical Industries, Osaka, Japan). Verapamil and ODQ were dissolved in dimethyl sulphoxide (DMSO) to make stock solutions and were added to Krebs solution to produce the required concentrations.

Fig. 1. Effects of N-nitro-L-arginine (L-NNA) and atropine (Atr) on spontaneous contractions of circular muscle tissue isolated from the rat distal colon. A: data obtained from the same tissue. Spontaneous contractions were recorded in both the absence (Aa) and presence of 100 μM L-NNA (Ab), and after co-application of 100 μM L-NNA and 3 μM Atr (Ac). B: summary of the effects of L-NNA, L-NNA + Atr, or ODQ on the amplitude of spontaneous contractions. C: summary of the effects of L-NNA, L-NNA + Atr, or ODQ on the frequency of spontaneous contractions. Data are expressed as means ± SD (n = 5). Significant difference from control: *P < 0.05; **P < 0.01.

Fig. 2. Inhibitory effects of TJ-14 on spontaneous contractions in the presence of L-NNA or ODQ. Spontaneous contractions were recorded before (A) and during application of 100 μM L-NNA (B) or 10 μM ODQ (C). In the absence or presence of these drugs, 1 mg/ml TJ-14 (a) or 5 mg/ml TJ-14 (b) were applied at the bar under each record. A–C were recorded from different tissues.
final concentrations just before their use. TJ-14 and TJ-60 (obtained from Tsumura and Company, Tokyo, Japan) were dissolved in Krebs solution directly. All other drugs were dissolved initially in distilled water. The final concentration of the solvent in Krebs solution did not exceed 1/1,000. Addition of these chemicals to the Krebs solution did not significantly alter the pH.

RESULTS

General observations. CSM preparations with an intact mucosa were spontaneously active, displaying periodical phasic contractions (spontaneous contractions) at a frequency of 0.5 ± 0.2 min⁻¹ (n = 5; Fig. 1A). The mean amplitude of the spontaneous contractions was 489 ± 200 mg (n = 5). Higher-frequency, phasic contractions of smaller amplitude were also observed between the larger low-frequency contractions, as previously reported (22). However, the nature of these small amplitude contractions was not further examined in the present study. Application of 100 μM L-NNA, an inhibitor of NO synthase, significantly increased both the amplitude and frequency of spontaneous contractions (Fig. 1, Ab, B, and C). The L-NNA-induced enhancement of spontaneous contractions was inhibited by 3 μM atropine (Fig. 1, Ac, B, and C). Application of 10 μM ODQ, an inhibitor of soluble guanylate cyclase, also increased the amplitude of spontaneous contractions (Fig. 1, B and C). Enhancement of spontaneous contractions by L-NNA was also observed in the presence of 1 μM TTX (amplitude: 167 ± 19%, n = 5, P < 0.01; frequency: 135 ± 6%, n = 5, P < 0.01). Even in quiescent tissues, spontaneous contractions were generated by the application of L-NNA (n = 3) or TTX (n = 2). These results suggest that NO released from nitrergic nerves may produce a sustained inhibition of spontaneous contractions of CSMs in the rat distal colon.

Effects of TJ-14 and TJ-60. Experiments were carried out to investigate the effects of TJ-14 and TJ-60 on the spontaneous contractions of rat colon CSM. When 0.1 mg/ml TJ-14 was applied, TJ-14 caused little effect on spontaneous contractile activity. TJ-14, at concentrations of >1 mg/ml, abolished all spontaneous contractions (Figs. 2A and 3). In the presence of L-NNA (100 μM) or ODQ (10 μM), the inhibitory effects of 1 mg/ml TJ-14 on spontaneous contractions was partially attenuated, whereas that of 5 mg/ml TJ-14 was unchanged (Figs. 2, B and C, and 3). TJ-14 (5 mg/ml) abolished spontaneous contractions in the presence of 1 μM TTX (n = 4, data not shown). TJ-14 (5 mg/ml) also decreased the basal tone (−19 ± 10 mg, n = 7, P < 0.01). The inhibition of spontaneous contractions produced by TJ-14 (5 mg/ml) was reversible over a 40- to 50-min period (n = 7, data not shown).

Fig. 3. Summary of the effects of TJ-14 on spontaneous contractions in the presence of L-NNA or ODQ. A: inhibitory effects of TJ-14 (0.1, 1, and 5 mg/ml) on the amplitude of spontaneous contractions in the absence or presence of L-NNA (100 μM) or ODQ (10 μM). B: inhibitory effects of TJ-14 (0.1, 1, and 5 mg/ml) on the frequency of spontaneous contractions in the absence or presence of L-NNA (100 μM) or ODQ (10 μM). Data are expressed as means ± SD (n = 4–7). Significant difference from control (in the absence of TJ-14): *P < 0.05; **P < 0.01. *Significant difference from control (in the absence of L-NNA or ODQ) (P < 0.05).

Fig. 4. Inhibitory effects of TJ-60 on spontaneous contractions in the presence of L-NNA or ODQ. Spontaneous contractions were recorded before (A) and during the application of 100 μM L-NNA (B) and 10 μM ODQ (C). In the absence or presence of these drugs, 1 mg/ml TJ-60 (a) or 5 mg/ml TJ-60 (b) was applied during the bar under each tension record. Note that A, B, and C were recorded from different tissues.
When 0.1 mg/ml TJ-60 was applied, TJ-60 caused little effect on spontaneous contraction. However, spontaneous contractions were also abolished by higher concentrations of TJ-60 (≥1 mg/ml) (Figs. 4A and 5). In the presence of l-NNA or ODQ, the additional application of TJ-60 (1 mg/ml) had little effect on spontaneous contractions, although a higher concentration of TJ-60 (5 mg/ml) suppressed these contractions (Figs. 4, B and C, and 5). Similar results were observed in the presence of 1 μM TTX (n = 3, data not shown). TJ-60 (5 mg/ml) had little effect on the basal tone (~2 ± 15 mg, n = 7, P > 0.05). On removal of TJ-60 (5 mg/ml), suppression of spontaneous contractions was gradually reversed over ~40–50 min (n = 7).

In the presence of 1 μM TTX, apamin (1 μM) gradually elicited an increase in the amplitude and frequency of spontaneous contractions (Fig. 6, A–C). As shown in Fig. 6, A–C, TJ-60 (5 mg/ml)-induced inhibition of spontaneous contractions was attenuated by apamin. In contrast, TJ-14 (5 mg/ml) significantly inhibited spontaneous contractions, even in the presence of apamin (Fig. 6, B and C).

Taken together, these results indicate that the inhibitory effects of TJ-60 (1 mg/ml) on spontaneous contractions of CSMs may well be mediated by the production of NO, whereas those produced by TJ-60 (5 mg/ml) are likely mediated by an apamin-sensitive pathway. TJ-14 (1 mg/ml)-induced inhibitory actions on spontaneous contractions of CSM were partly sensitive to l-NNA. However, TJ-14 (5 mg/ml)-induced inhibitory effects were unaffected by l-NNA or apamin. The effects of TJ-14 and TJ-60 on the spontaneous contractions are summarized in Table 1.

Properties of TNS-induced contractile activity. To examine the properties of the motor innervations of the rat distal colon, TNS was used. Application of TNS (3 min at 4 Hz with 0.3-ms pulse duration) evoked several phasic contractions, which were quantified as area-under-contraction data (58.6 ± 27.3 g × s, n = 8); this was followed by a transient contraction upon cessation of TNS (off-contraction) (1,874 ± 419 mg, n = 8; Fig. 7Aa). Although the off-contraction was a single phasic contraction, repetitive generation of a group of phasic contractions was observed on cessation of TNS in the presence of l-NNA and atropine (Fig. 7Ac). The basal tone was increased by TNS (62 ± 60 mg, n = 8; Fig. 7Ab). The TNS-induced contractile responses (area-under-contraction, off-contraction, and increase in the basal tone) were enhanced by 100 μM l-NNA (Fig. 7, Ab and B–D). The additional application of 3 μM atropine abolished the TNS-induced contractile responses.
but not the off-contraction (Fig. 7, Ac, B–D). In the presence of L-NNA plus atropine, TNS generated a transient relaxation \((-45 \pm 19 \text{ mg}, n = 8; P < 0.01\) followed by a sustained relaxation as shown in Fig. 7Ac. Both the transient relaxation and off-contraction were abolished by 1 \(\mu\text{M}\) apamin \((n = 3)\). These results suggest that the phasic contractions and increase in basal tone evoked by TNS may be cholinergic in origin. This would indicate that the CSM receives cholinergic excitatory, nitrergic inhibitory, and non-nitrergic (apamin-sensitive) inhibitory innervation, as previously reported \((9, 10)\).

**Effects of TJ-14 and TJ-60 on TNS-induced contractile activity.** Further experiments were performed to investigate the effects of TJ-14 and TJ-60 on the TNS-induced contractile responses of CSM. To eliminate NO-induced inhibitory influences, experiments were carried out in the presence of 100 \(\mu\text{M}\) L-NNA. Application of TJ-14 \((5 \text{ mg/ml})\) caused inhibitory effects on several phasic contractions (Fig. 8, Ab and B) as well as on the off-contraction (Fig. 8, Ab and C) and, interestingly, increased basal tone (Fig. 8, Ab and D). In contrast, TJ-60 \((5 \text{ mg/ml})\) had no significant inhibitory actions on the three parameters of the TNS-induced contractile responses (Fig. 8, Ac, B, and D). Since several components of the phasic contractions and the increase in basal tone were atropine-sensitive responses evoked by TNS, these results imply that TJ-14 may possess inhibitory effects on the cholinergic contraction, whereas TJ-60 is unlikely to exert such effects (Table 1).

### Table 1. Comparison of the effects of TJ-14 and TJ-60 on spontaneous contractions and TNS-induced contractile activities

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TNS, transmural nerve stimulation; L-NNA, \(N^\text{N}\)-nitro-L-arginine; ODQ, 1H-[1,2,4]oxadiazolo[4,3-a]quinoxalin-1-one. *TNS was applied in the presence of L-NNA. †On contraction means contractile activity generated during application of TNS.

### Effects of verapamil on TNS-induced contractile activity.
To examine the roles of voltage-dependent L-type \(\text{Ca}^{2+}\) channels (VDLCCs), the effects of verapamil, a selective blocker of VDLCCs, on TNS-evoked responses were investigated. Verapamil \((3 \mu\text{M})\) abolished spontaneous contractions in the presence of 100 \(\mu\text{M}\) L-NNA (Fig. 9A), suggesting that spontaneous contractions are associated with \(\text{Ca}^{2+}\)-influx through VDLCCs, as previously reported \((15, 22)\). Verapamil decreased basal tone \((-60 \pm 11 \text{ mg}, n = 5, P < 0.01\) (Fig. 9A). Verapamil greatly attenuated the phasic contractions (Fig. 9, A and B) and the increase in basal tone (Fig. 9, A and D) evoked by TNS. Interestingly, verapamil slightly decreased the off-contraction (Fig. 9, A and C). The off-contraction seems to be generated by rebound excitation after inhibitory junction potentials (IJPs) induced by purinergic neurotransmitters, since it was abolished by apamin. It has been reported that active repolarization of IJPs may result from activation of a nicardipine-insensitive, voltage-dependent, nonselective cation conductance (NSCC) in murine colon \((1)\). Thus, the off-contraction observed in our experiments might be evoked by activation of NSCC in rat colonic CSM. These results indicate that cholinergic responses cause phasic contractions and that the increment of basal tone is related to the activation of VDLCCs in the CSM of rat distal colon.

**Fig. 7.** Effects of L-NNA and atropine (Atr) on TNS-evoked contractions in CSM isolated from the rat distal colon. **A:** response to TNS under control conditions (Aa), in the presence of 100 \(\mu\text{M}\) L-NNA (Ab) and in the presence of 100 \(\mu\text{M}\) L-NNA and 3 \(\mu\text{M}\) Atr (Ac). The basal tone is indicated by the dotted line. **B:** summary of the effects of L-NNA and L-NNA + Atr on the area of contraction induced by TNS. **C:** summary of the effects of L-NNA and L-NNA + Atr on the off-contraction induced by TNS. **D:** summary of the effects of L-NNA and L-NNA + Atr on the increase of the basal tone induced by TNS. Data are presented as means with SD \((n = 8)\). Significant difference from control: *\(P < 0.05\); **\(P < 0.01\).
DISCUSSION

The aim of the present study was to characterize the inhibitory effects of two Japanese Kampo medicines (TJ-14 and TJ-60), which have been used to treat diarrhea, on the contractile activity of CSM from the rat distal colon. The results demonstrate unequivocally that TJ-14 inhibited spontaneous contractions, partly through the production of NO, and that TJ-60 inhibited these contractions through both the production of NO and via an apamin-sensitive inhibitory pathway. These data indicate that TJ-14 caused an inhibition of TNS-evoked cholinergic responses, whereas TJ-60 had no significant effect.

The inhibitory effects of TJ-14 (1 mg/ml) were partially sensitive to l-NNA. However, the inhibitory effects induced by the higher concentration of TJ-14 (5 mg/ml) were insensitive to l-NNA or apamin; hence the inhibitory mechanisms underlying the actions of 5 mg/ml TJ-14 remain unclear. We suggest that TJ-14 may inhibit spontaneous contractions in the rat distal colon by directly affecting the CSM but not through the release of non-nitrergic inhibitory neurotransmitters, since the TJ-14-induced inhibition was observed in the presence of TTX. Moreover, TJ-14 inhibited TNS-induced cholinergic responses to the level found in the presence of verapamil (Figs. 8 and 9). Additional studies will be required to shed further light on the mechanisms that underlie the effects of TJ-14 on VDLCCs in rat colon CSM.

TJ-60 (1 mg/ml) inhibited spontaneous contractions of rat colon CSM by activating l-NNA- or ODQ-sensitive pathways, suggesting the involvement of NO (25). In contrast, a higher concentration of TJ-60 (5 mg/ml) inhibited spontaneous con-
tractions through an apamin-sensitive pathway, indicating that TJ-60 may activate SK channels, an effect that has been shown for purinergic neurotransmission (9, 19, 25). A previous study demonstrated that TJ-43, another Japanese Kampo medicine, relaxed CSM in the rat fundus by activating SK channels (17). Interestingly, three herbs (Zingiberis rhizoma, Zizyphi fructus, and Glycyrrhizae radix) are ingredients common to both TJ-43 and TJ-60 (14, 17), so it is plausible that compounds that activate SK channels may be contained in one or all of these herbs. In contrast to TJ-14, TJ-60 had little effect on the TNS-induced cholinergic responses. It is generally known that cholinergic nerves are potent excitatory nerves in the colon and that their stimulation is associated with the generation of a rhythmic migrating motor pattern, called the colonic migrating motor complex (CMMC), that underlies movement of feces or stool in the colon (4, 28–30, 32). Inhibition of cholinergic responses may suppress the generation of CMMC and cause constipation (6). Thus the lack of an inhibitory effect of TJ-60 on TNS-evoked cholinergic responses does not simply lead to the conclusion that TJ-60 is not effective in the treatment of diarrhea symptoms. It seems that TJ-60 is indeed effective in the treatment of IBS (personal communication from Tsumura & Company), although, to date, there are no studies published in international journals describing the effectiveness of TJ-60 on IBS. There are three major symptom subtypes of IBS: diarrhea predominant, constipation predominant, and alternating (31). Taken together, these results imply that the weak effects of TJ-60 on the TNS-evoked cholinergic responses might be better suited to ameliorate IBS with alternating bouts of diarrhea and constipation.

Chronic diarrhea, defined as diarrhea lasting for >4 wk and with more than three loose stools per day, is associated with IBD, IBS, and infections and also presents as an adverse effect of many drug treatments (7, 11, 27). The impact of chronic diarrhea on a patient’s quality of life (QOL) and on health-care costs is likely to be considerable. Thus successful treatment of chronic diarrhea will be paramount to improve patients’ QOL and reduce health-care costs. Loperamide, a μ-opioid receptor agonist, is widely utilized to treat various diarrhea syndromes. However, loperamide does not decrease the abdominal pain associated with IBS (5, 23). In Japan, TJ-14 has recently been used to treat several forms of diarrhea, such as acute diarrhea, fermentative diarrhea, and diarrhea induced by anticancer drugs (14). Several studies have shown that TJ-14 inhibits the production of PG$E_2$, enhances water absorption, and reduces inflammation in the rat colon (12, 13). In addition, the present study has unequivocally demonstrated that TJ-14 has potent inhibitory effects on both myogenic and neurogenic colonic motility. These results strongly suggest that TJ-14 could be used not only as an antidiarrheal drug but also as an antispasmodic. It is known that antispasmodics are effective in IBS and chronic constipation (14). Thus the lack of an inhibitory effect of TJ-60 on TNS-evoked cholinergic responses does not simply lead to the conclusion that TJ-60 is not effective in the treatment of the abdominal pain associated with IBD.

In conclusion, the present study has shown that both TJ-14 and TJ-60 inhibit contractile activity in the rat colon CSM by different mechanisms. TJ-14 inhibits both spontaneous contractions (myogenic contractile activity) and TNS-evoked cholinergic responses (neurogenic contractile activity), whereas TJ-60 inhibits spontaneous contractions. Since TJ-14 has inhibitory effects on both myogenic and neurogenic contractile activity in the colon, TJ-14 may well be effective in the treatment of the abdominal pain associated with gastrointestinal motor dysfunction.

ACKNOWLEDGMENTS

The authors are grateful to Dr. R. J. Lang for critical reading of the manuscript. Hange-shashin-to (TJ-14) and Keishi-ka-shakuyaku-to (TJ-60) were kindly provided by Tsumura & Company.

GRANTS

This project was supported by a Grant-in-Aid for Young Scientists (B) from the Japanese Society for the Promotion of Science to Y. Kito (grant no. 21790629). This work was also supported by a Funding Program for Next Generation World-Leading Researcher (Noriyoshi Teramoto, grant no. LS906) from the Japanese Society for the Promotion of Science.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS

Author contributions: Y.K. conception and design of research; Y.K. performed experiments; Y.K. analyzed data; Y.K. interpreted results of experiments; Y.K. prepared figures; Y.K. drafted manuscript; Y.K. and N.T. edited and revised manuscript; Y.K. and N.T. approved final version of manuscript.

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