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Original article

Effects of Sijunzi Dripping Pill on Gastrointestinal Motility of Mice

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ABSTRACT

Objective To study the effects of Sijunzi Dripping Pill (SDP) on gastrointestinal motility of mice. **Methods** The diarrhea and swimming model of mice was made by *Rhei Radix* et *Rhizoma*-induced spleen deficiency. The intestinal transit, gastric emptying test, serum motilin (MTL), vasoactive intestinal peptide (VIP), and substance P (SP) were chosen to observe the effects of high-, mid-, and low-dose SDP on stomach movements, and the water extractive of Sijunzi Decoction was used as positive control. **Results** Compared with the control group, the gastric emptying rate in the gastrointestinal motility group was significantly decreased, the intestinal propulsion rate was obviously increased, the levels of MTL, prostaglandin E_2 (PGE_2), and SP were increased ($P < 0.05$), while the level of VIP was decreased ($P < 0.05$). Compared with the model group, SDP could decrease the intestinal transit rate, whereas increase the gastric emptying rate and the level of MTL ($P < 0.05$); The high-dose SDP could decrease the level of PGE_2 ($P < 0.05$) and the low-dose SDP could decrease the level of VIP ($P < 0.05$); Each group had no significant effect on SP. **Conclusion** SDP has the good effect on increasing the gastrointestinal motility of mice, and its function may partly relate to the regulation of the levels of MTL and VIP as well as PGE_2.

Key words

amount of gastric emptying; gastrointestinal motility; serum motilin; Sijunzi Dripping Pill

1. Introduction

Sijunzi Decoction (SD) comes from a traditional Chinese medicine book called "*Taiping Benevolent Dispensary Bureau*", which is the basic recipe of tonifying *qi* and repairing the weakness of spleen. In modern recipe, *Codonopsis Radix* replaces *Ginseng Radix*, and in this study SD consists of *Codonopsis Radix*, *Poria*, roasted *Atractylodis Macrocephalae Rhizoma*, and *Glycyrrhizae Radix* et *Rhizoma Praeparata* cum *Melle*. The polysaccharids from SD have various functions and activities, such as aiding digestion, adjusting stomach movements, and affecting gastrointestinal hormone (Ye and Chen, 2005). Existing formulations made from above Chinese materia medica are water pill, mixture, and granules. Those formulations are produced by traditional methods which induce a lot of problems, such as complicated producing process, low absorbance, intake inconvenience, low efficiency, etc. Sijunzi Dripping Pill (SDP) mainly consisted of polysaccharids and lipid-soluble components in SD. This research focused on stomach movement function,

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examined the effects of SDP on intestinal propulsion, gastric emptying, serum motilin (MTL), vasoactive intestinal peptide (VIP), and substance P (SP) in mice with spleen deficiency.

2. Materials and methods

2.1 Experimental animals

Kunming mice (18–20 g), male and female in half, supplied by Experiment Center of Shanxi Medical University (SCXK-201101) were used in this study.

2.2 Drugs and reagents

Sijunzi Dripping Pill was supplied by Department of Pharmacology, Shanxi University of Traditional Chinese Medicine, and mainly contained 20% of polysaccharids and 0.02% of lipid-soluble components. *Rhei Radix* et *Rhizoma* (700 g, Jingwan Chinese Medicine Co., China, No. 110901) was immersed for 1 h in 2100 mL water, decocted for 15 min at 97 ^{o}C, filtered with a double layer gauze, then added with two fold amount of water to decoct the residues for 15 min, filtered and combined the decoction. The sample was concentrated to 1.5 g/mL, sealed, and kept in cool condition.

Codonopsis Radix (No. 100417101), *Poria* (No. 100301111), roasted *Atractylodis Macrocephalae Rhizoma* (No. 100428101), and *Glycyrrhizae Radix Rhizoma Praeparata* cum *Melle* (No. 105668192) were all purchased from Qiao Chinese Herbal Medicine Co., Ltd., China, at a ratio of 2:2:2:1, added with 1400 mL water to immerse 175 g medicine for 1 h, decocted for 2 h at 97 ^{o}C, and filtered with a double layer gauze after decocting. The sample was decocted for three times, the decoction was combined and concentrated to 1.75 g/mL, sealed, and kept in a cool condition. This decoction obtained was SD.

Polysaccharide sample (1 mg) was precisely weighed, together with dry KBr powder (200 mg) of chromatographic grade, finely grounded in agate mortar and mixed evenly, then shaped into tablet. MB104 FTIR (ABB Co., Canada) was used to characterize polysaccharide in SDP.

Xylitol (Shandong Futian Medicine Co., Ltd., No. 11031908), 1% methyl orange (Tianjin Guangfu Institute of Fine Chemicals), 5% sodium bicarbonate (Tianjin Shentai Chemicals Co., Ltd., No. 110801), and sodium chloride (Tianjin Kemimou Chemicals Co., Ltd., No. 20100427) were used. All reagents were of analytical grade. Motilin (MTL), vasoactive intestinal peptide (VIP), prostaglandin E_2 (PGE_2), and substance P (SP) were detected by ELISA kit (No. 201110, made by American R&D Co., imported, dispensed, and provided by Shanghai Chuangsai Co.).

2.3 Main instruments

721 Visible Spectrophotometer (Shanghai Qinghua Technology Co., Ltd.), FSH-II High-speed Electric Homogenizer (Jintan Danyangmen Quartz Glass Co.), LD5–10 Centrifuge (Beijing Medical Centrifuge Co.), JA4003 Analytical Balance (Shanghai Liangping Instrument Co., Ltd.), Spectra Max 190 Microplate Reader (Molecular Co., USA).

2.4 Groups and models

The Kunming mice (18–22 g) were housed in a room at (22 ± 1) ^{o}C, with free access to water and standard mouse chow. The mice were randomly divided into six groups (n = 8, half male and half female), such as control, model (spleen deficiency group), SD (35 g/kg fresh medicine), high-, mid-, and low-dose (2, 1, and 0.5 g/kg) SDP groups. In the morning, the mice in the control group were fed with water, the mice in the model group were ig administered with xylitol (3.5 g/kg) and the rest mice were ig given SDP with the relative doses. In the afternoon, all the model mice were given water extract from *Rhei Radix* et *Rhizoma* (37.5 g/kg) to establish the spleen deficiency model (Xu et al, 2004) except the mice in the control group. The mice in the control group were still given water, and after administration all the mice were put into a box full of water with the depth of 30 cm at (25 ± 1) ^{o}C. After swimming for 10 min, the mice were taken out and dried. The experiment lasted for two weeks.

2.5 Experimental protocol and measurement of intestinal propulsion and gastric emptying

Before the experiment, the mice were fasting for 18–20 h, but were supplied with tap water until 10 min before the experiment. On the day of the experiment, the animals were ig given 1% methyl orange (10 mL/kg) and absorbed blood 20 min later, then sacrificed. The stomach and attached small intestine of mice were immediately exposed by laparotomy. After ligation of esophagogastric, gastroduodenal, and ileocaecal junctions, the stomach and small intestine were carefully removed and placed on a wooden board to observe the leading edge of the methyl orange in the intestine (Wu et al, 2002). The length of small intestine from pylorus to ileocecal and the length of methyl orange promoting were measured. The formula of calculating intestinal propulsion was as follows: intestinal propulsion rate = length of methyl orange / whole length of small intestine.

The stomach was cut out, immersed into 1% sodium bicarbonate solution for 30 min, and centrifuged at 2000 r/min for 10 min. The supernatant was measured by determining the absorbance (A), and expressed as a percentage of gastric emptying. The formula for caculating gastric emptying rate was as follows:

gastric emptying rate = $(1 - A_n) / A_0$

where A_n was the A value of stomach, A_0 was the A value of 1% sodium bicarbonate solution.

2.6 Experimental protocol and measurement of serum MTL, small intestine homogenate, VIP, PGE_2, and SP

The blood samples were collected and centrifuged at 3500 r/min for 10 min, and ELISA kit was used to measure