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Dual therapy for third-line *Helicobacter pylori* eradication and urea breath test prediction

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Abstract

We evaluated the efficacy and tolerability of a dual therapy with rabeprazole and amoxicillin (AMX) as an empiric third-line rescue therapy. In patients with failure of first-line treatment with a proton pump inhibitor (PPI)-AMX-clarithromycin regimen and second-line treatment with the PPI-AMX-metronidazole regimen, a third-line eradication regimen with rabeprazole (10 mg q.i.d.) and AMX (500 mg q.i.d.) was prescribed for 2 wk. Eradication was confirmed by the results of the ^{13}C-urea breath test (UBT) at 12 wk after the therapy. A total of 46 patients were included; however, two were lost to follow-up. The eradication rates as determined by per-protocol and intention-to-treat analyses were 65.9% and 63.0%,

respectively. The pretreatment UBT results in the subjects showing eradication failure; those patients showing successful eradication comprised 32.9 ± 28.8 permil and 14.8 ± 12.8 permil, respectively. The pretreatment UBT results in the subjects with eradication failure were significantly higher than those in the patients with successful eradication (P = 0.019). A low pretreatment UBT result (≤ 28.5 permil) predicted the success of the eradication therapy with a positive predictive value of 81.3% and a sensitivity of 89.7%. Adverse effects were reported in 18.2% of the patients, mainly diarrhea and stomatitis. Dual therapy with rabeprazole and AMX appears to serve as a potential empirical third-line strategy for patients with low values on pretreatment UBT.

Key words: *Helicobacter pylori*; Amoxicillin; Dual therapy; Eradication; Urea breath test

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TO THE EDITOR

Eradication of *Helicobacter pylori* (*H. pylori*) has been reported as an effective strategy in the treatment of peptic ulcers and gastric mucosa-associated lymphoid tissue lymphomas and also prevents the recurrence of gastric cancer after endoscopic resection[1-7]. The first-line regimen for the treatment of *H. pylori* infection in Japan is triple therapy with a proton pump inhibitor (PPI), amoxicillin (AMX) and clarithromycin (CLR) administered for 7 d. Failure of this first-line therapy against *H. pylori* infection has been reported in approximately 20% of infected patients[8,9]. With the increase in the frequency of CLR-resistant *H. pylori*, there is rising concern about the potential decline in the eradication rate of this infection[10]. Although therapy with PPI-AMX-metronidazole (MNZ) administered for 1 wk has been found to be effective as a second-line regimen in patients failing the first-line regimen, approximately 10% of patients fail to respond to even second-line treatment, necessitating the establishment of an alternative third-line strategy for the effective eradication of *H. pylori*[3,11].

Although *H. pylori* bacteria easily develop resistance to CLR and MNZ, *H. pylori* has been considered to seldom become resistant to AMX. AMX is the preferred antibiotic because it is bactericidal and resistance is rare; therefore, it can be used again after treatment failure[8]. A number of studies have suggested that good success rates for *H. pylori* eradication could be obtained with AMX and PPI dual therapy if the effective PPI dose and frequency of administration were increased[12]. The majority of patients who experience two eradication failures have the rapid metabolizer genotype of CYP2C19. Because omeprazole and lansoprazole are extensively metabolized by CYP2C19 in this genotype, their plasma concentrations will not attain levels sufficient to inhibit acid secretion, and therefore, antibiotics such as AMX will be less stable in the stomach, resulting in a lower eradication rate[13]. The PPI rabeprazole is a substitute of benzimidazole. CYP2C19 is less involved in the metabolism of rabeprazole than in that of omeprazole and lansoprazole[14]. Moreover, rabeprazole has a greater and more rapid acid-inhibitory effect than does omeprazole. Several reports on the pharmacokinetics and pharmacodynamic characteristics of PPIs have indicated that a sufficient plasma concentration of PPIs can be achieved in patients with the rapid metabolizer genotype of CYP2C19 by frequent PPI dosing[12,15]. Furuta *et al*[16] recently reported an excellent eradication rate of 87.8% following dual therapy with rabeprazole 4 times/day and AMX as a third-line rescue. However, their study was completed at only one or two centers. Our study was designed as a prospective, multicenter trial with the participation of 16 Japanese hospitals affiliated with the National Hospital Organization to investigate the efficacy of dual therapy with 4 times daily dosing of rabeprazole and AMX as empiric third-line rescue therapy.

A total of 46 patients (26 males, 20 females; age 60.7 ± 12.9 years, mean ± SD) referred to us between January 2009 and January 2012 were enrolled. Endoscopic examinations were conducted before treatment in all patients, and *H. pylori* positivity was confirmed by histology, stool antigen test, *H. pylori*-specific IgG antibodies or the ^{13}C-urea breath test. All patients had a history of two treatment failures (first-line treatment used: triple therapy with PPI- AMX-CLR for 7 d; second-line treatment used; triple therapy with PPI-AMX-MNZ for 7 d). The exclusion criteria in this study were (1) age < 18 years; (2) presence of clinically significant underlying disease (hepatic or renal disease, diabetes mellitus); (3) history of gastric surgery; and (4) allergy to any of the drugs used in the study. *H. pylori* eradication failure was defined as a positive ^{13}C-urea breath test (UBT) at the end of 12 wk after completion of treatment. The ^{13}C-urea used was 100 mg ^{13}C-labelled urea, produced by Otsuka pharmaceutical Co., LTD, Japan. The procedure was modified from the European standard protocol for the detection of *H. pylori*[17]. We

Table 1 Demographic characteristics of the patients and the results of eradication therapy

Characteristics	Total (n = 46)	Eradication success (n = 29)	Eradication failure (n = 15)	P value
Age (mean ± SD, yr)	60.7 ± 12.9	59.8 ± 13.4	60.8 ± 12.1	0.813
Sex (male/female)	26/20	15/14	10/5	0.530
Diagnosis (GU/DU/CG)	23/15/8	15/10/4	8/3/4	0.450
Pretreatment UBT	20.4 ± 21.2	14.8 ± 12.8	32.9 ± 28.8	0.019
Eradication rate (ITT) %	63.0			
Eradication rate (PP) %	65.9			

GU: Gastric ulcer; DU: Duodenal ulcer; CG: Chronic gastritis; UBT: Urea breath test; ITT: Intention-to-treat; PP: Per protocol.

chose 2.5 permil for cut-off level of the rise in the delta value of $^{13}CO_2$ at 15 min after the ingestion of ^{13}C-urea.

The treatment regimen was rabeprazole 10 mg q.i.d. and AMX 500 mg q.i.d. administered for 2 wk. Participants were requested to return at the conclusion of the therapy for an interview regarding any adverse events. Successful *H. pylori* eradication was defined as a negative UBT at the end of 12 wk after completion of treatment. Statistical analyses were performed using the chi-square, Fisher's exact and Student's t tests, as appropriate. P values of less than 0.05 were accepted as representing statistical significance. The study was conducted with the approval of the Ethics Committee of the National Hospital Organization Tokyo Medical Center, and informed consent was obtained from all patients prior to the examinations. The clinical trial registration number of the University Hospital Medical Information Network was R000003204.

Of the 46 patients enrolled, 2 dropped out of the study, leaving 44 patients in the per protocol (PP) set. *H. pylori* eradication was confirmed in 29 patients, representing an eradication rate of 63.0% [95% confidence intervals (CI): 47.6%-76.8%] by intention-to-treat (ITT) analysis and 65.9% (95% CI: 50.1%-79.5%) by PP analysis (Table 1). Patient compliance with the prescribed treatment was excellent. Adverse events were recorded in 8 patients (18.2%; 95% CI: 8.2%-32.7%). Six patients had mild diarrhea or soft stools but went on to complete the study. Two patients developed stomatitis.

Because the numerical results of the UBT are a function of the total urease activity within the stomach, they represent a quantitative index of the density of gastric *H. pylori* colonization[18]. As a low pretreatment UBT value could be one of the predictive factors for eradication success, the pretreatment UBT value was analyzed. The pretreatment UBT results in the subjects with eradication failure and in those with successful eradication were 32.9 ± 28.8 and 14.8 ± 12.8 (permil, mean ± SD), respectively. The results of the statistical analysis showed that the pretreatment UBT results in the subjects with eradication failure were significantly higher than those in the patients with successful eradication (P = 0.019, effect size 0.81). We plotted original receiver operator characteristic (ROC) curves for the pretreatment UBT results to establish the appropriate cutoff value. According to the ROC curves, the optimal cutoff value in our population was 28.5. When patients were assigned to two groups (UBT results ≤ 28.5 permil and > 28.5 permil), the eradication rates were 81.3% (26/32) and 25.0% (3/12), respectively (P = 0.001). A low pre-treatment UBT value (≤ 28.5 permil) predicted the success of the eradication therapy with a sensitivity of 89.7 %, specificity of 60.0 %, positive predictive value of 81.3%, negative predictive value of 75.0% and accuracy of 79.5%.

Currently, a standard third-line therapy still remains to be established. *H. pylori* isolates after two eradication failures are often resistant to both MNZ and CLR. The alternative candidates for third-line therapy are fluoroquinolones-AMX-PPI, rifabutin-AMX-PPI, and high-dose PPI/AMX therapy[2,19-21]. Gisbert *et al*[22] conducted a prospective multicenter study to evaluate the outcomes of treatment with a third-line levofloxacin-based regimen. The patients were treated for 10 d with a regimen consisting of omeprazole, levofloxacin and AMX. The eradication rates as determined by PP and ITT analyses were 66% and 60%, respectively. However, resistance to fluoroquinolones has been shown to be easily acquired, and in countries with a high rate of use of these drugs, the resistance rates are relatively high. González Carro *et al*[23] evaluated the efficacy of a third-line rifabutin-based triple therapy. The patients were treated with PPI, rifabutin and AMX for 10 d. The eradication rates as determined by PP and ITT analyses were 62.2% and 60.8%, respectively. However, it has been suggested that the use of rifabutin be reserved for the treatment of multidrug-resistant *Mycobacterium tuberculosis* strains[24].

Our results for the dual therapy with 4 times daily dosing of rabeprazole and AMX for 14 d, which yielded eradication rates in the PP and ITT analyses of 65.9% and 63.0%, were as successful as other empiric third-line therapy regimens. In particular, a low pretreatment UBT result (≤ 28.5 permil) predicted the success of the eradication therapy with a positive predictive value of 81.3%, sensitivity of 89.7% and specificity of 60.0%, so the dual therapy appeared to serve as a promising option for empiric third-line rescue therapy in patients with a low pretreatment UBT value.

We recently reported the resistant rates of *H. pylori* to AMX. The resistance rates to AMX (MIC ≥ 0.06 μg/mL) in the groups with no history of eradication treatment, a history of one treatment failure, and a history of two treatment failures were 13.6%, 26.5% and 49.5%, respectively. The MIC_{90} of AMX increased by 2-fold after each eradication failure[25]. Resistance to AMX in *H. pylori* was gradually induced after unsuccessful eradication. Because the AMX resistance rate after two treatment failures was relatively high, the eradication rate of the present study was lower than that of previous report by Furuta *et al*[16]. Therefore, antimicrobial susceptibility testing of *H. pylori* is desirable before the selection of a suitable third-line therapy, although the culture-based antibiotic susceptibil-

ity testing for *H. pylori* is expensive, time-consuming, and not always available on a routine basis[26]. There are several limitations to our study. First, our eradication study was single armed using the dual therapy, and different doses or superiority over quinolone-based therapy was not evaluated. Second, we did not examine the *in vitro* susceptibility in patients treated with the dual therapy. Thus, *in vitro* resistance to AMX was not elucidated. These issues should be re-evaluated in future studies.

Finally, although we did not achieve excellent eradication success, the dual therapy appeared to serve as a promising option for empiric third-line rescue therapy in patients with low pretreatment UBT values. The antimicrobial susceptibility testing of *H. pylori* is desirable before the selection of a suitable third-line therapy in patients with high pretreatment UBT values.

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Original Article

Efficacy of Solifenacin on Irritable Bowel Syndrome With Diarrhea: Open-label Prospective Pilot Trial

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Background/Aims
Solifenacin, a muscarinic type 3 receptor antagonist, is used to treat overactive bladder in adults. The aim of this study is to examine the efficacy of solifenacin on the symptomatic relief of diarrhea predominant irritable bowel syndrome (IBS-D).

Methods
A total of 20 patients with IBS-D were enrolled. After a 2-week observation period, all participants received solifenacin for 6 weeks. Subsequently, the administration of solifenacin was discontinued and ramosetron, a serotonin 3 receptor antagonist, was administered for 4 weeks. Overall improvement, the IBS-symptom severity scale (IBS-SSS), and frequency of defecation were assessed.

Results
Six weeks after initiation of solifenacin treatment and 4 weeks after initiation of ramosetron treatment, overall improvement was observed in 19 out of 20 (95%) and 17 out of 20 (85%) participants, respectively. At 2 weeks after initiation of solifenacin, overall improvement was observed in 16 out of 20 participants (80%). Total IBS-SSS scores at 2 and 6 weeks after the administration of solifenacin, and at 4 weeks after administration of ramosetron, were significantly lower than those at week 0. Compared to before administration, the participants' quality of life and frequency of defecation were significantly lower in all participants at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after administration of ramosetron.

Conclusions
The efficacy of solifenacin in the treatment of IBS with diarrhea was not inferior to that of ramosetron. Further placebo-controlled parallel studies are needed.
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Key Words
Diarrhea; Overactive; Ramosetron; Solifenacin succinate; Urinary bladder

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Conflicts of interest: None.

Introduction

Irritable bowel syndrome (IBS) is a condition characterized by the presence of chronic abdominal pain or abdominal discomfort accompanied by abnormal bowel movements, such as diarrhea or constipation.[1] In IBS, symptoms are improved by defecation, and there appears to be no organic substance or biochemical abnormality that can explain the symptoms. In population-based Japanese surveys, the prevalence of IBS has been estimated as 10%-15% and the annual incidence as 1%-2%.[2,3] Because gastroenterologists frequently focus mainly on inflammatory or malignant disorders, functional disorders such as IBS, that are associated with subjective symptoms, are less likely to be the target of aggressive treatment. The complaints of patients with IBS consist of general gastrointestinal symptoms, and differential diagnosis of complications such as infectious enteritis is necessary. Therefore, it is important to obtain a detailed history of the disease. The treatment for IBS tends to consist of merely the prescription of common gastrointestinal medications. For healthcare providers, IBS can be difficult to detect, and patients are often dissatisfied with the outcome even when they consult a physician, resulting in a low consultation rate at medical institutions. Currently, the majority of patients remain undiagnosed, including those who are themselves unaware of their disease. Although the disease is not life-threatening, the symptoms of IBS clearly cause deterioration in patients' quality of life, and it affects a large number of patients. The societal losses due to IBS are immeasurable. Depending on the type of stool, IBS can be classified into 4 categories: constipation predominant IBS (IBS-C), diarrhea predominant IBS (IBS-D), mixed IBS (IBS-M), and unsubtyped IBS.[1] Among those categories, IBS-D is a particularly serious problem for patients who commute to work or to school by public transportation. Anticholinergic drugs, the serotonin 3 (5-HT_3) receptor antagonist, ramosetron,[4] high molecular weight polymers (polycarbophil calcium),[5] gastrointestinal motility regulators, Probiotics preparations (such as *Bifidobacterium infantis* 35624)[6] and laxatives are used in the treatment of IBS. However, no medication for the treatment of IBS has been able to provide the same levels of efficacy as proton pump inhibitors that are used for the treatment of peptic ulcers or gastroesophageal reflux disease.

Muscarinic type 3 (M_3) receptors are believed to be the key molecule for the pathogenesis of IBS,[7] and the efficacy of M_3 receptor antagonists in the treatment of IBS has been the focus of several studies.[8,9] Although a M_3 receptor antagonist such as mepenzolate bromide has been used as a modulating agent of gastrointestinal motility since 1967 in Japan, no clinical trials had been conducted to reveal the efficacy for IBS defined under the modern Rome criteria. Until now, even though mepenzolate bromide has been used empirically to IBS, no significant effect on IBS has been reported even in the non-randomized clinical study or in animal study. Recently, solifenacin [(+)-(1S,3′R)-quinuclidin-3′-yl 1-phenyl-1,2,3,4-tetrahydroisoquinoline-2-carboxylate monosuccinate], a M_3 receptor antagonist, has been used in the treatment of overactive bladder (OAB) in Japan, and its usage is covered by national insurance. Our recent epidemiological study also demonstrated a high rate of comorbidity between IBS and OAB.[10] In addition, the mode of solifenacin action on bowel dysfunction in vivo using experimental models that reproduced the symptoms present in IBS was similar to that of darifenacin, a selective M_3 receptor antagonist, with equivalent potencies. By contrast, propantheline, an anti-muscarinic drug that has been used for IBS, was much less potent.[9]

Because of the pathogenetic similarities between IBS-D and OAB with respect to the presence of hyperactive smooth muscles,[9] the present study was designed to examine the efficacy of solifenacin for the treatment of IBS-D.

Materials and Methods

Study Population

The present study is a single-cohort prospective trial. The protocol for this study was approved by the ethics committee of Tokyo-Eki Center-Building Clinic (TEC-C C0005, Nov. 7, 2010, UMIN000005577). This study included IBS-D patients, age 20 years or older, who were treated as outpatients in Tokyo-Eki Center-Building Clinic. The required sample size for testing the equality of proportions was 16 patients based on a minimum expected difference of 10% and standard deviation of 10% in the overall improvement between solifenacin and ramosetron, with an alpha error of 5% and 80% power.[11] Thus, after considering the number of patients who dropped out, a total of 20 patients were recruited for the present study.

The IBS was diagnosed according to the Rome III criteria.[1] Namely, participants were defined as having IBS if they had suffered recurrent abdominal pain or discomfort for more than 2 days in a week and also had 2 or more of the following: improvement with defecation, onset associated with change in (increased

or decreased) frequency of stool production, and onset associated with change in stool consistency (hard or soft). IBS patients were subcategorized as having IBS-C, IBS-D and IBS-M. In IBS-C, onset was associated with decreased frequency of stool production or hard stool, while in IBS-D onset was associated with increased frequency of stool production or soft stool, including diarrhea; patients with IBS-M experienced both decreased and increased frequency of stool production or presence of both hard and soft stool at different times. Among them, only patients with IBS-D were recruited to the present study.

The following participants were excluded from the study: subjects with a history of laparotomy for upper or lower digestive tract surgery, narrow-angle glaucoma, severe diseases (such as urinary retention) or disabilities that could have affected the participants' condition or the test results; and whose physical examination, laboratory tests, vital signs (blood pressure and pulse rate) and electrocardiogram had shown clinically problematic abnormalities.

Interventions

After a 2-week run-in period, the administration of solifenacin 5 mg tablets was initiated. In participants who showed overall improvement 2 weeks later, solifenacin 5 mg was continued for another 4-week period. In participants who showed no overall improvement, the dose of solifenacin was increased to 10 mg and was continued for 4 weeks. However, in participants who had difficulties taking the 5 mg dose after 2 weeks, the treatment was either discontinued or the dose was decreased to 2.5 mg. Starting at 6 weeks, ramosetron 5 μg was administered continuously for 4 weeks if the attending physician determined that no problems had appeared during the preceding 4 week treatment. A flowchart of the tests is shown in Figure 1.

During the study period, parallel administration of therapeutic agents targeting the digestive system was prohibited, except for medications for purposes other than the treatment of IBS that were administered chronically or taken as needed. In addi-

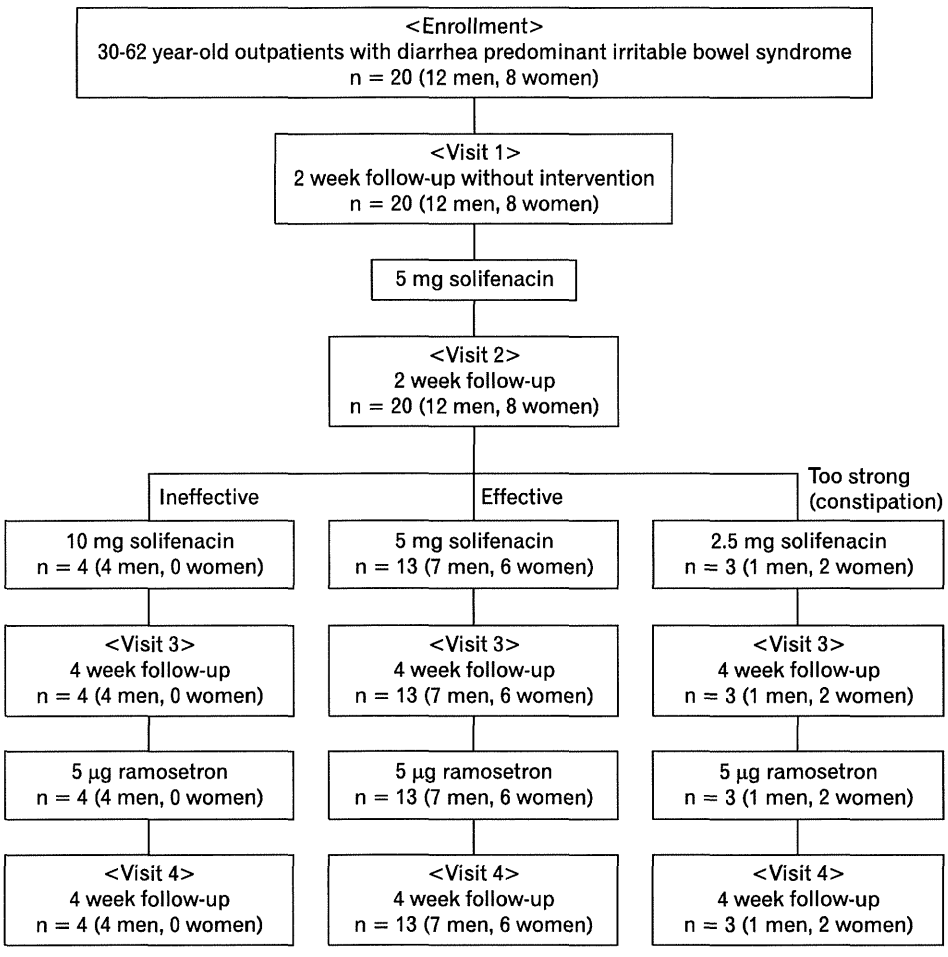

Figure 1. The flowchart of the tests.

tion, medications that were likely to affect gastrointestinal motility were also prohibited.

Assessments

The presence or absence of overall improvement was used as the primary efficacy endpoint according to the method of previous randomized double-blind, placebo-controlled clinical trial on the effectiveness of ramosetron.[12] In this study, the subjective symptoms of IBS in the 1 week before initiation of solifenacin treatment were compared with those during a 1-week period just before the day of the assessement, and when improvements in the subjective symptoms were found, we considered this to be evidence of an "overall improvement." When no improvement was found, patients were said to have "no overall improvement." The IBS-symptom severity scale scores (IBS-SSS; < 75 = no IBS, 75-175 = mild IBS, 175-300 = moderate IBS, and 300 = severe IBS) and the number of stools per day were used as secondary endpoints. Safety was assessed on the basis of adverse events and vital signs. The assessments were performed 4 times: before initiation of solifenacin (visit 1), 2 weeks after initiation of solifenacin (visit 2), 6 weeks after initiation of solifenacin (visit 3) and 4 weeks after initiation of ramosetron (visit 4) (Fig. 1).

Statistical Methods

The differences in background characteristics between participants showing improvement in symptoms and those showing no improvement were analyzed using the Chi-squared test and unpaired *t* test. IBS-SSS scores on the day of each hospital visit and the differences in the average numbers of stools per day were analyzed using a paired *t* test. All statistical analyses were conducted using the SPSS statistics version 18.0 for Windows software (SPSS Japan, Tokyo, Japan; SPSS Inc., IL, USA). The data in the tables were expressed as mean ± standard deviation. Two-sided *P*-values were considered as statistically significant at a level of 0.05.

Results

Prescribed Dose of Solifenacin

Twenty subjects (12 men and 8 women; mean age, 44.8 ± 8.3 years) agreed to participate in the study. None of the 20 participants had been taking medication that needed to be discontinued. The participants' backgrounds are shown in Table. Before administration of the test drugs, 8 of the participants had IBS-SSS scores indicating mild symptoms, 10 had IBS-SSS scores indicating moderate symptoms, and 2 had IBS-SSS scores indicating severe symptoms. All 20 participants started taking solifenacin 5 mg tablets 2 weeks later. For 4 participants who showed no overall improvement 2 weeks after administration of solifenacin (visit 2), the dose was increased to 10 mg. Among the 16 participants who showed overall improvement, the dose of solifenacin was decreased to 2.5 mg in 3 subjects who were constipated and was continued at 5 mg in the other 13 subjects. Administration of solifenacin was then continued for further 4 weeks. At week 6 (visit 3), solifenacin treatment was switched to 5 μg dose of ramosetron, 5-HT_3 receptor antagonist, for all participants, and ramosetron treatment was continued for further 4 weeks. We confirmed that the medication was taken by all participants in accordance with the protocol. The flowchart of the study is shown in Figure 1.

Effectiveness Measures

Overall improvement was observed in 16 out of 20 participants at 2 weeks after initiation of solifenacin treatment. At 6 weeks after initiation of solifenacin treatment (visit 3) and 4 weeks after initiation of ramosetron treatment (visit 4), overall improvement was observed in 19 (95%) and 17 (85%) out of 20 participants, respectively. No statistically significant differences in background characteristics were found between the patients who showed improvement and the patients who did not show improvement.

Table. Participant Characteristics (n = 20)

Age (mean ± SD, yr)	44.8 ± 1.6
Gender (n [%])	
Men	12 (60)
Women	8 (40)
Smoking habit (n [%])	
Non smoker	10 (50)
Former smoker	2 (10)
Smoker	8 (40)
Alcohol habit (n [%])	
None	2 (10)
Sometimes	11 (55)
Everyday	7 (35)
BMI (mean ± SD, kg/m^2)	24.1 ± 3.2
Duration of illness (mean ± SD, yr)	13.0 ± 12.1
Total IBS-SSS (mean ± SD)	212 ± 58
Frequency of defecation (mean ± SD/day)	3.3 ± 1.6

BMI, body mass index; IBS-SSS, irritable bowel syndrome-symptom severity score.

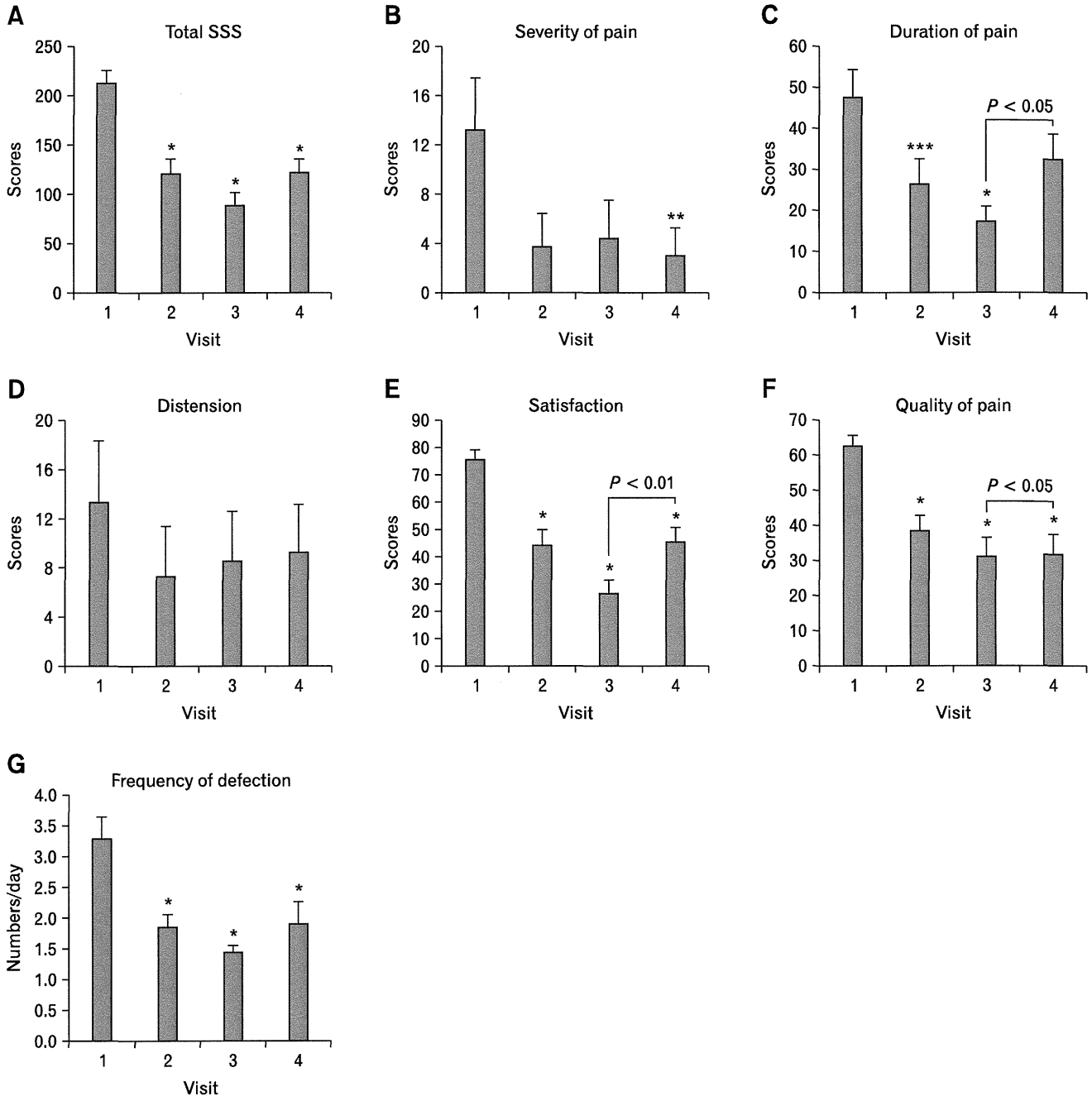

Figure 2. The irritable bowel syndrome-symptom severity scale (IBS-SSS) scores at 0, 2 and 6 weeks after the administration of solifenacin (visit 1, 2 and 3) and at 4 weeks after the administration of ramosetron (visit 4). (A) Total IBS-SSS scores at 0, 2 and 6 weeks after the administration of solifenacin (visit 1, 2 and 3) and at 4 weeks after the administration of ramosetron, $^{*}P < 0.001$ as compared with the values at week 0. (B) The score for the severity of pain at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after the administration of ramosetron, $^{**}P < 0.05$ as compared with the values at week 0. (C) The scores for the duration of pain at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after the administration of ramosetron, $^{***}P < 0.01$ as compared with the values at week 0. (D) The scores for the distension at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after the administration of ramosetron, (E) The scores for the satisfaction at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after the administration of ramosetron, $^{*}P < 0.001$ as compared with the values at week 0. (F) The scores for the quality of life at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after the administration of ramosetron, $^{*}P < 0.001$ as compared with the values at week 0. (G) The scores for the frequency of defecation at 2 and 6 weeks after the administration of solifenacin and at 4 weeks after the administration of ramosetron, $^{*}P < 0.001$ as compared with the values at week 0.

Total IBS-SSS scores at 2 and 6 weeks after the administration of solifenacin (visit 2 and 3) and at 4 weeks after the administration of ramosetron (visit 4) were significantly lower than those at week 0 (visit 1) (Fig. 2). IBS-SSS scores lower than 75 were viewed as remission of IBS symptoms. Of the 20 participants, 6 subjects (30%) had IBS-SSS scores lower than 75 at 2 weeks after administration of solifenacin, while 10 subjects (50%) had IBS-SSS scores lower than 75 at 6 weeks after administration. Further, the IBS-SSS scores of 4 out of 20 participants (20%) were lower than 75 at 4 weeks after administration of ramosetron. No statistically significant differences were found between the IBS-SSS scores at 6 weeks after administration of solifenacin and those at 4 weeks after administration of ramosetron.

The differences between each outcome measure of the IBS-SSS scores at each hospital visit day were evaluated (Fig. 2). Compared to pain intensity at week 0, no improvement was found after solifenacin treatment, but significant improvement was observed 4 weeks after administration of ramosetron. However, the number of days of pain and the degree of satisfaction with defecation habits were more significantly improved at 6 weeks after administration of solifenacin than at 4 weeks after administration of ramosetron. The patients' quality of life and the number of stools per day were significantly lower at 2 and 6 weeks after administration of solifenacin and at 4 weeks after administration of ramosetron than before the administration of treatment.

There were 3 solifenacin medication groups: dose increment group (10 mg, n = 4), no dose change group (5 mg, n = 13), and dose-decreased group (2.5 mg, n = 3). At the end of the solifenacin treatment period (visit 3), no significant difference was observed in overall improvement, total SSS and number of stools. At the end of the ramosetron medication, no significant difference was observed in these 3 groups in terms of overall improvement ($P = 0.481$). Moreover, although stool number tended to decrease in the dose-decreased group, no statistically significant difference was observed with respect to total SSS and stool number per day between these 3 groups: dose increment group (123 ± 69 and 2.3 ± 1.0), no dose change group (119 ± 56 and 2.0 ± 1.9), and dose-decreased group (140 ± 49 and 1.1 ± 0.8; $P = 0.853$ and $P = 0.640$). Furthermore, the difference in these 3 groups could not be a cofounding factor that affected the results of the ramosetron treatment.

Safety Profile

The use of solifenacin caused dry mouth in 1 participant and constipation in 3 other participants. The use of ramosetron caused constipation in 4 participants and loose stools in 2 other participants. No other problematic adverse events were observed.

Discussion

Since this study was an open-label study and since the evaluation was based on symptoms, the possible involvement of the placebo effect in the results cannot be excluded. However, even when this is taken into consideration, the present study verified the efficacy of solifenacin on the symptomatic amelioration of IBS-D. Solifenacin at doses from 2.5 to 10 mg resulted in overall improvement at 6 weeks in 19 participants (95%), 10 of whom (50%) showed remission of symptoms. Since pooled estimate of placebo response was reported to be 42.6% (95% confidence interval, 38.0%-46.5%) in 19 complementary and alternative medicine trials in IBS,[13] solifenacin may be more effective than placebo. In addition, the effects of solifenacin on the symptoms of IBS were comparable to the effects of ramosetron, a medication that is used in the treatment of IBS-D in men and is covered by national insurance in Japan.

Currently, no therapeutic drugs have been shown to be definitively effective in the treatment of IBS. In addition, IBS has very high prevalence; according to a recent questionnaire survey of 10,000 Japanese citizens, the prevalence of IBS was 13.1%, indicating that approximately 12 million (12.5%) adult Japanese citizens (20-79 years old) have IBS.[3] Meanwhile, in a 10-year follow-up study conducted on 3,873 patients, the incidence of new cases of IBS over the 10-year period was 15%.[14] Thus, while IBS is a disease with high incidence, patient awareness of the disease is low, and since the majority of IBS patients treat themselves by self-medicating with over the counter drugs instead of consulting medical professionals, there are significant economic and social losses. Accordingly, therapeutic drugs effective against IBS are highly anticipated.

The sequential use of therapeutic agents in functional gastrointestinal disorders has been criticized because symptom severity may fail to return to baseline after the first treatment period. In the present study, since the first treatment was fixed to solifenacin and the second to ramosetron, there is a possibility for the overestimation of therapeutic effect of ramosetron in comparison with that of solifenacin. Therefore, we cannot conclude which medication is better for the treatment of IBS-D based on the results obtained in the present study. However, the present results suggest at least in the management of IBS-D that solifenacin is not

inferior to ramosetron, with a possible superiority of solifenacin in terms of the days of pain and the degree of satisfaction with defecation habit.

This study was an open-label trial, and the data are neither those of parallel group trial nor crossover study; however, the results showed the potential therapeutic application of solifenacin in the treatment of IBS-D, and the data from this study are significant in that they indicate at least the possibility of a new therapeutic drug for IBS-D. On the other hand, with the present non-parallel study, the possible placebo effect could not be excluded. On the basis of these results, further placebo-controlled parallel group studies remain awaited to confirm the efficacy of solifenacin.

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Author Contributions: YF, HS designed the research study. YF and AK conducted the clinical study. YF, HS & JM analyzed and interpreted the data. HS drafted the article. TH supervised and approved to be published.

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High frequency of overlap between functional dyspepsia and overactive bladder

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Abstract

Background *Overactive bladder syndrome (OAB) is defined as a symptom complex comprising urgency, with or without urge incontinence, and usually frequency and nocturia. The association between irritable bowel syndrome (IBS) and bladder symptoms has been reported. This study is designed to investigate whether functional dyspepsia (FD), like IBS, is associated with OAB.* ***Methods*** *A web surveys containing questions about OAB, FD, IBS, and demographics were completed by 5494 public individuals (2302 men and 3192 women) who have no history of severe illness. The prevalence and overlap of OAB, FD, and IBS were examined.* ***Key Results*** *Among participants with FD, 20.5% could also be diagnosed with OAB (odds ratio [OR]: 2.85; 95% confidence interval [CI]: 2.21–3.67). Although concomitant FD and IBS were more strongly associated with OAB (OR: 4.34; 95% CI: 2.81–6.73), OAB was also highly prevalent among participants with FD but without IBS (OR: 3.09; 95% CI: 2.29–4.18). Among participants with FD, an overlapping OAB condition was more prevalent in those with both postprandial distress syndrome (PDS) and epigastric pain syndrome (EPS) (OR: 3.75; 95% CI: 2.48–5.67) than in those with PDS or EPS alone. Among participants with OAB, the severity of bladder symptoms was greater in participants with dyspeptic symptoms than without them.* ***Conclusions & Inferences*** *Overactive bladder syndrome is common among FD patients, even if they do not have IBS. To improve FD patients' quality of life, it will be important to provide management for OAB.*

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INTRODUCTION

Overactive bladder syndrome (OAB) is defined as a symptom complex comprising urgency, with or without urge incontinence, and usually frequency and nocturia, in the absence of other local factors that would account for the symptoms. OAB is a complex of one or more of the following symptoms, which occur in the absence of other local factors that would account for their presence: urgency, urge incontinence, frequent urination, and nocturia.[1] Functional gastrointestinal disorders (FGIDs), including irritable bowel syndrome (IBS) and functional dyspepsia (FD), are defined as chronic disorders of the digestive system in which symptoms cannot be explained by the presence of structural or tissue abnormality. Both OAB and FGIDs are significant health issues, as they are highly prevalent and have negative effects on quality of life.[2] The association between IBS and bladder symptoms was documented as early as 1986,[3,4] when Whorwell *et al.* reported that IBS patients frequently experienced symptoms of irritable bladder, including frequency, urgency, hesitancy, nocturia, and incomplete bladder emptying. Coyne *et al.* recently reported that chronic constipation occurs more frequently in patients with OAB.[5] However, studies on the potential association between OAB and FD are generally lacking.

Functional dyspepsia impairs the health-related quality of life in patients, and the impact seems to be on all major variables of quality of life, namely mental, social, and physical functioning.[6] A recent population-based study showed that postprandial distress syndrome (PDS) seems to impair the quality of life more than epigastric pain syndrome (EPS), while FD-IBS overlap has a significant impact on bodily pain.[6] In

addition, a large employee-based study showed that employees with FD had greater average annual medical and prescription drug costs than those without FD.[7] The employees with FD were absent for an additional 0.83 days per year and produced 12% fewer units per hour than those without FD.[7] On the other hand, patients with OAB tend to limit their fluid intake, avoid sexual intimacy, wear pads, and be more anxious about knowing the location of toilets. In particular, older OAB patients not only have an increased risk of injury and fractures,[8] but also have a higher incidence of sleep disturbance, depression, and visits to physicians.[9,10] Therefore, it is important to evaluate the frequency of overlap between FD and OAB to improve the patients' quality of life and reduce the economic losses incurred.

The standard treatment for OAB is anti-muscarinic drugs, which have gastrointestinal side effects. Although the most well-known side-effect is constipation, patients may also experience dyspepsia and abdominal pain during treatment of OAB. In three phase III, randomized, placebo-controlled, 12-week trials that evaluated the efficacy, tolerability, and safety of once-daily controlled-release darifenacin for OAB, dry mouth (20.2%–35.5%), constipation (14.8%–21.3%), dyspepsia (2.7%–8.4%), abdominal pain (2.4%–3.9%), nausea (1.5%–2.7%), and diarrhea (0.9%–2.1%) were reported as adverse events.[11] The high prevalence of gastrointestinal (GI) side effects also indicates that it is important to better understand the relationship not only between OAB and IBS, but also OAB and FD.

The aim of the present study was to investigate the frequency of overlap between OAB and FD. As the presence of IBS is a potential confounding factor, we also investigated overlap between OAB and IBS.

MATERIALS AND METHODS

Study participants

The protocol for this study was approved by the ethics committee of Tokyo-Eki Center Building Clinic (TEC-C E-002, July 14, 2010). We conducted a web-based cross-sectional study including participants from a list of 177 615 individuals (age range, 20–75 years) who had previously provided informed consent and enrolled for unspecified clinical research trials conducted by the Tokyo-Eki Center-Building Clinic. No participants in the list have severe chronic or life-threatening illnesses, such as progressive malignant diseases or systemic autoimmune diseases, or serious mental illnesses, such as major depression or schizophrenia. Individuals with a history of prescription drug use were initially excluded. The questionnaires collected sufficient data for us to use the OABSS[12] to evaluate OAB, and the Rome III criteria to evaluate FD[13] and IBS.[14] Using the questionnaires, the presence/absence of structural disease in the urinary tract was also determined. In the questionnaires, we also asked whether dyspeptic symptoms were relieved with defecation. In addition, prior receipt of an upper GI screening examination was elicited. If it was identified, the presence/absence of structural disease in the upper GI was abstracted. We also collected the following demographic information: age, gender, smoking and alcohol-drinking habits, height, and weight. Participants could select one of three smoking habit categories based on the number of cigarettes consumed per day (0 = 'none', 1–4 = 'light', ≥15 = 'heavy') and one of three alcohol intake categories based on the number of days per week on which alcohol was consumed (0 = 'none', 1–3 = 'light', 4–7 = 'heavy'). We calculated body mass index (BMI) (weight height^{-2}) using the morphometric data provided. Participants with urethral calculus, bladder cancer, or prostate cancer were excluded from the study.

Definitions of OAB, FD, and IBS

As OAB is a collection of symptoms, symptom assessment tools are used for quantitative assessment of the syndrome. The OABSS is a validated self-assessment questionnaire that provides a simple sum of 4 symptom scores that address daytime frequency, nighttime frequency, urgency, and urgency incontinence. The maximum scores for each component are defined as 2, 3, 5, and 5, respectively.[12,15] Here, OAB was defined as a urinary urgency score (third question of OABSS) of 2 or more, and a total OABSS of 3 or more, based on the clinical guidelines for OAB prepared by the Neurogenic Bladder Society.[16]

Based on the Rome III criteria, participants were defined as having dyspepsia if they had experienced one or more symptoms, such as postprandial fullness, early satiation, or epigastric pain or burning, for at least 6 months prior to the survey. Participants with only epigastric pain that was relieved by defecation were not included into those with dyspepsia, as their symptoms would be caused by unrecognized IBS. Participants with dyspepsia who had undergone upper gastrointestinal examination and had no evidence of structural disease to explain their symptoms were defined as having 'FD'. Functional dyspepsia participants with postprandial fullness or early satiation were defined as having PDS, while those with epigastric pain or burning were defined as having EPS; some participants had both PDS and EPS.

Based on the Rome III criteria, participants were defined as having IBS if they had suffered recurrent abdominal pain or discomfort for more than 2 days in a week and also had two or more of the following: improvement with defecation, onset associated with a change (increased or decreased) in frequency of stool production, and onset associated with a change in stool consistency (hard or soft). Irritable bowel syndrome participants were subcategorized as having constipation-predominant IBS (IBS-C), diarrhea-predominant IBS (IBS-D), or mixed IBS (IBS-M). In IBS-C, onset was associated with decreased frequency of stool production or hard stool, while in IBS-D onset was associated with increased frequency of stool production or soft stool, including diarrhea; participants with IBS-M experienced both decreased and increased frequency of stool production or presence of both hard and soft stool at different times.

Statistical analysis

Differences between non-OAB and OAB, non-FD and FD, and non-IBS and IBS participants were examined with unpaired Student's *t*-tests (for age and BMI) and Pearson's chi-squared tests (for gender, smoking habits, and alcohol-drinking habits). Associations between OAB and FGIDs or other clinical factors were evaluated using univariate and multivariate logistic regression. Associations between the OABSS and dyspeptic symptoms were examined with unpaired Student's *t*-tests. All statistical analyses

were conducted using SPSS version 18.0 for Windows (SPSS Japan Inc., Tokyo, Japan). The data in the tables are expressed as mean ± standard deviation. Two-sided *P*-values were considered to be statistically significant at a level of less than 0.05.

RESULTS

Participant characteristics

A total of 5494 individuals completed the web-based surveys (Fig. 1). After we excluded 164 participants who had an organic urinary tract disease, our final sample size was 5330 participants (2187 men and 3143 women). OAB, FD, and IBS were diagnosed in 497 (9.3%), 438 (8.2%), and 728 (13.7%) participants, respectively. Among the 438 participants with FD, 267 (61.0%) were categorized as having PDS alone, 45 (10.3%) were classified as having EPS alone, and 126 (28.8%) were found to have both PDS and EPS. Among the 728 participants with IBS, 147 (20.2%) were categorized as having IBS-C, 456 (62.6%) were classified as having IBS-D, and 125 (17.2%) were found to have IBS-M.

Participant characteristics are shown in Table 1. Both mean age and alcohol consumption levels were higher in OAB participants than in non-OAB participants. Alcohol consumption was more prevalent in FD participants than in non-FD participants. Mean age was lower in participants with IBS than in those without this condition.

Overlap of OAB, FD, and IBS

The numbers of participants with OAB, FD, or IBS are shown in Fig. 2A. Among participants with either FD or OAB, 10.7% (90/844) had both FD and OAB. On the other hand, among participants with either IBS or OAB, 12.3% (134/1091) had both IBS and OAB. Overlap between FD and OAB was almost as often as overlap between FD and IBS (11.2%; 117/1049) (Fig. 2B–D).

Logistic regression analyses showed that OAB was associated with FD (odds ratio [OR]: 2.85; 95% confidence interval [CI]: 2.21–3.67) almost at the same level as it was associated with IBS (OR: 2.63; 95% CI: 2.12–3.27) (Table 2). OAB was also significantly associ-

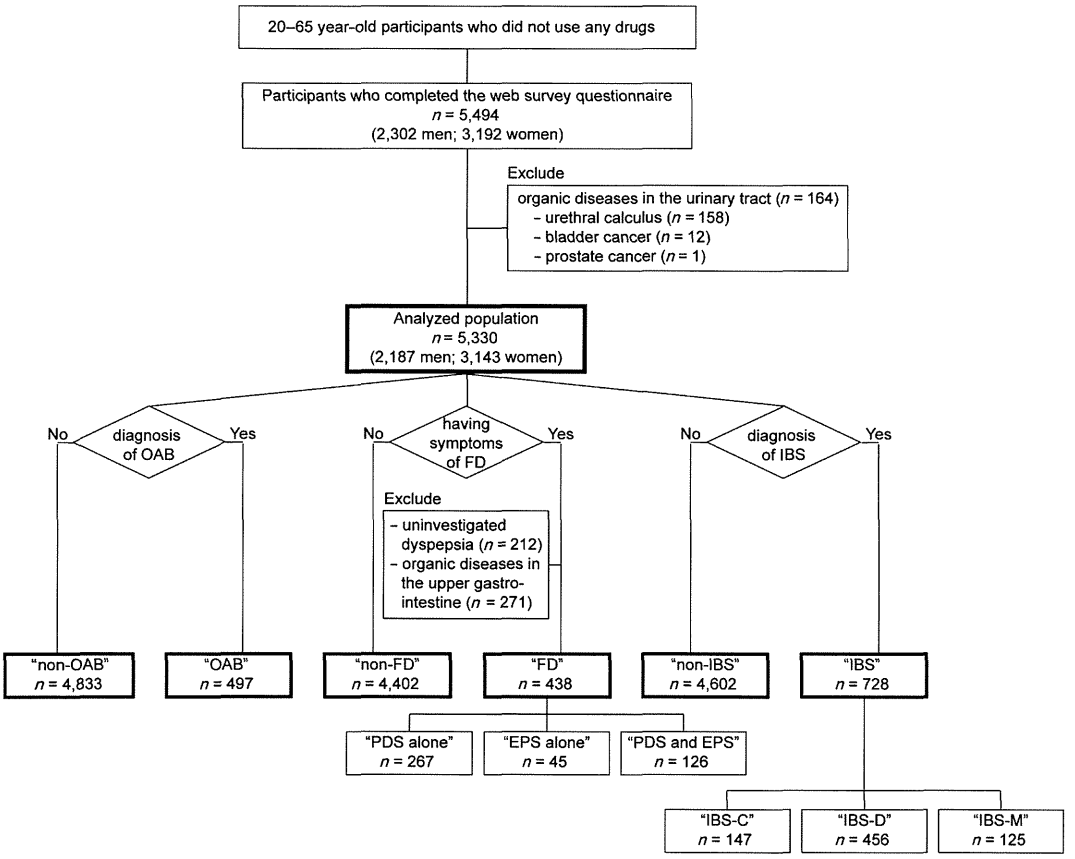

Figure 1 The study population. OAB, overactive bladder; FD, functional dyspepsia; IBS, irritable bowel syndrome; IBS-C, constipation-predominant irritable bowel syndrome; IBS-D, diarrhea-predominant irritable bowel syndrome; IBS-M, mixed irritable bowel syndrome; PDS, postprandial distress syndrome; EPS, epigastric pain syndrome.

Table 1 Participant characteristics

	Non-OAB (n = 4833)	OAB (n = 497)	Non-FD (n = 4402)	FD (n = 438)	Non-IBS (n = 4602)	IBS (n = 728)
Age, years (mean ± SD)	42.5 ± 9.7	**45.9 ± 10.2**	42.9 ± 9.9	42.9 ± 8.4	43.2 ± 9.8	**40.3 ± 9.7**
Gender, n (%)						
Men	1985 (41.1%)	202 (40.6%)	1820 (41.3%)	188 (42.9%)	1897 (41.2%)	290 (39.8%)
Women	2848 (58.9%)	295 (59.4%)	2582 (58.7%)	250 (57.1%)	2705 (58.8%)	438 (60.2%)
Smoking habit, n (%) (number of consumptions/day)						
None (0)	3712 (76.8%)	385 (77.5%)	3412 (77.5%)	328 (74.9%)	3555 (77.2%)	542 (74.5%)
Light (1–14)	453 (9.4%)	51 (10.3%)	395 (9.0%)	45 (10.3%)	433 (9.4%)	71 (9.8%)
Heavy (15 ≤)	668 (13.8%)	61 (12.3%)	595 (13.5%)	65 (14.8%)	614 (13.3%)	115 (15.8%)
Alcohol habit, n (%) (number of days of consumption/week)						
None (0)	1837 (38.0%)	**176 (35.4%)**	1709 (38.8%)	**133 (30.4%)**	1758 (38.2%)	255 (35.0%)
Light (1–3)	1616 (33.4%)	**145 (29.2%)**	1449 (32.9%)	**158 (36.1%)**	1510 (32.8%)	251 (34.5%)
Heavy (4–7)	1380 (28.6%)	**176 (35.4%)**	1244 (28.3%)	**147 (33.6%)**	1334 (29.0%)	222 (30.5%)
BMI, kg m^{-2} (mean ± SD)	22.2 ± 3.5	22.4 ± 3.9	22.2 ± 3.5	22.2 ± 3.7	22.2 ± 3.6	22.1 ± 3.6

Bold values indicate significant differences between non-OAB and OAB, non-dyspepsia and FD, or non-IBS and IBS. Differences of age and BMI were analyzed by unpaired Student's t-tests. Differences of gender, smoking habit, and alcohol habit were analyzed by Pearson's chi-squared tests. OAB, overactive bladder; IBS, irritable bowel syndrome; BMI, body mass index; FD, functional dyspepsia.

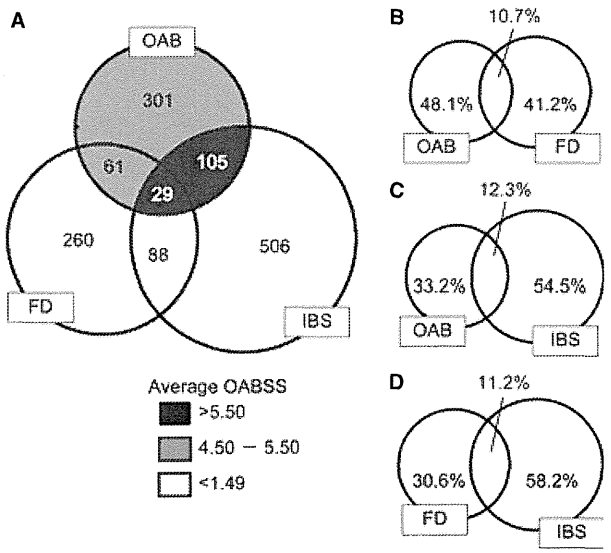

Figure 2 Overlap between OAB, FD, and IBS. (A) The number of participants in each partition. Each partition is painted in different colors to be classified using Overactive Bladder Symptom Score (OABSS). The symptoms of OAB were the most severe in OAB participants with both FD and IBS. The prevalence of overlap (B) between OAB and FD, (C) between OAB and IBS, and (D) between FD and IBS are shown. OAB; overactive bladder; FD, functional dyspepsia; IBS, irritable bowel syndrome.

Table 2 Symptom overlap of OAB with other conditions

	Non-OAB (n = 4833)	OAB (n = 497)	Odds ratio (95% CI)†
Non-FD	4485 (92.8%)	407 (81.9%)	ref.
FD	348 (7.2%)	90 (18.1%)	2.85 (2.21–3.67)
PDS alone	218 (4.5%)	49 (9.9%)	2.48 (1.79–3.43)
EPS alone	36 (0.7%)	9 (1.8%)	2.76 (1.32–5.76)
PDS and EPS	94 (1.9%)	32 (6.4%)	3.75 (2.48–5.67)
Non-IBS	4239 (87.7%)	363 (73.0%)	ref.
IBS	594 (12.3%)	134 (27.0%)	2.63 (2.12–3.27)
Neither FD nor IBS	3979 (82.3%)	302 (60.8%)	ref.
FD without IBS	260 (5.4%)	61 (12.3%)	3.09 (2.29–4.18)
IBS without FD	506 (10.4%)	105 (21.1%)	2.73 (2.15–3.48)
Both FD and IBS	88 (1.9%)	29 (5.8%)	4.34 (2.81–6.73)

†Analyzed by univariate logistic regression model. OAB, overactive bladder; CI, confidence interval; FD, functional dyspepsia; IBS, irritable bowel syndrome; PDS, postprandial distress syndrome; EPS, epigastric pain syndrome.

ated with all subcategories of FD (e.g., PDS alone, EPS alone, or concomitant PDS and EPS). In particular, OAB was more common in participants with both PDS and EPS (OR: 3.75; 95% CI: 2.48–5.67).

OAB was strongly associated with the presence of both FD and IBS (OR: 4.34; 95% CI: 2.81–6.73). In addition, OAB was also commonly found even in participants with FD but without IBS (OR: 3.09; 95% CI: 2.29–4.18). This result shows that FD and IBS are independently associated with the presence of OAB.

Differences in FD participants with and without OAB

We compared demographic and symptomatic characteristics between participants with both FD and OAB and those with FD but without OAB (Table 3). The multivariate logistic regression analyses revealed that older age (OR: 1.04; 95% CI: 1.01–1.07) and the presence of IBS-C (OR: 3.08; 95% CI: 1.24–7.63) were independently associated with overlap of FD and OAB. However, IBS-D, IBS-M, gender, smoking, alcohol use,

Table 3 Difference between FD with and without OAB

FD (*n* = 438)	FD without OAB (*n* = 348)	FD with OAB (*n* = 90)	Univariate analysis, Odds Ratio (95% CI)	Multivariate analysis† Odds Ratio (95% CI)
IBS-C, *n* (%)	12 (3.4%)	9 (10.0%)	**3.11 (1.27–7.63)**	**3.08 (1.24–7.63)**
IBS-D, *n* (%)	55 (15.8%)	17 (18.9%)	1.24 (0.68–2.26)	
IBS-M, *n* (%)	21 (6.0%)	3 (3.3%)	0.54 (0.16–1.84)	
Age, years (mean ± SD)	42.4 ± 8.4	45.1 ± 8.2	**1.04 (1.01–1.07)**	**1.04 (1.01–1.07)**
Gender, men, *n* (%)	147 (42.2%)	41 (45.6%)	1.14 (0.72–1.82)	
Smoking habit, *n* (%)				
None	257 (73.9%)	71 (78.9%)	ref.	
Light	36 (10.3%)	9 (10.0%)	0.29 (0.42–1.97)	
Heavy	55 (15.8%)	10 (11.1%)	0.66 (0.32–1.37)	
Alcohol habit, *n* (%)				
None	106 (30.5%)	27 (30.0%)	ref.	
Light	132 (37.9%)	26 (28.9%)	0.91 (0.42–1.40)	
Heavy	110 (31.6%)	37 (41.1%)	1.31 (0.75–2.30)	
BMI, kg m^{-2} (mean ± SD)	22.3 ± 3.7	22.1 ± 3.7	0.99 (0.93–1.06)	

†Analyzed by multivariable logistic regression model with adjustment for IBS-C and age. Bold values indicate significant associations. FD, functional dyspepsia; OAB, overactive bladder; IBS, irritable bowel syndrome; CI, confidence interval; BMI, body mass index.

and BMI were not associated with overlap of these two conditions.

Influences of dyspeptic symptoms on the severity of OAB symptoms

Among participants with OAB, the severity of OAB symptoms in participants with and without dyspeptic symptoms (postprandial fullness, early satiation, epigastric pain, and epigastric burning) is shown in Table 4. The average score of total OABSS in OAB participants with postprandial fullness was significantly higher than that in OAB participants without postprandial fullness. The presence of early satiation, epigastric pain, and epigastric burning also enhanced the scores of total OABSS, although these differences were not significant.

Table 4 Associations between the severities of OAB and dyspeptic symptoms

	Total OABSS	
OAB (*n* = 497)	Score (mean ± SD)	*P*-value
Postprandial fullness		
Absence (*n* = 283)	**5.24 ± 1.93**	**0.03**
Presence (*n* = 214)	**5.65 ± 2.20**	
Early satiation		
Absence (*n* = 345)	5.32 ± 1.95	0.14
Presence (*n* = 152)	5.64 ± 2.29	
Epigastric pain		
Absence (*n* = 405)	5.35 ± 1.98	0.12
Presence (*n* = 92)	5.72 ± 2.37	
Epigastric burning		
Absence (*n* = 421)	5.36 ± 2.02	0.13
Presence (*n* = 76)	5.75 ± 2.26	

Bold values indicate significant associations. OAB, overactive bladder.

These results suggest that concomitant dyspeptic symptoms influence the symptom severity of OAB.

DISCUSSION

This large-scale cross-sectional study of a Japanese population revealed a high frequency of overlap between FD and OAB. To our knowledge, this is the first evidence for an independent association between FD and OAB. In IBS patients, many extra-intestinal co-morbidities, such as migraine, fibromyalgia, chronic fatigue syndrome, chronic pelvic pain, and depression, are more common.[4,17,18] According to the guidance on IBS from the National Institute for Health and Clinical Excellence (NICE), bladder symptoms are common in people with IBS, and may be used to support the diagnosis of IBS.[19] However, the present study revealed that OAB is a significant co-morbidity not only in IBS, but also in FD.

Overlap between OAB and IBS, diagnosed using the modern criteria, has not been reported previously, although Nickel *et al.*[20] recently documented the overlap of interstitial cystitis/painful bladder syndrome (IC/PBS) with IBS, diagnosed using the Rome III. Although IC/PBS and OAB have similar symptoms, such as urgency, frequency, and nocturia, IC/PBS is differentiated from OAB by the presence of pain. The present study also provides the first evidence for the high prevalence of overlap between IBS and OAB. In addition, our work has revealed that OAB is more prevalent in participants with both IBS and FD than in those with IBS but without FD. Usually, different specialists (e.g., gastroenterologists and urologists) independently treat FD and OAB; this often occurs in the setting of a tertiary care hospital. Both

specialists must recognize the impact of overlap between FD and OAB.

We recently reported that FD patients with concomitant constipation or diarrhea show different characteristics from those without bowel symptoms.[21] We reported that FD patients with bowel symptoms have greater symptom severity than those without bowel symptoms. There were a greater proportion of women, especially with low BMI, in constipation-predominant FD. Alcohol consumption was associated with diarrhea-predominant FD in both genders.[21] The present study revealed that FD participants with IBS-C are likely to have concomitant OAB. Namely, high prevalence of concomitant OAB is thought to be one of the characteristics among constipation predominant FD patients. Previous studies also showed that there is a significant overlap between lower urinary tract symptoms and constipation.[5,22,23] Charach *et al.* reported that medical relief of constipation significantly improves lower urinary tract symptoms in the elderly, and also improves the patient's mood, sexual activity, and quality of life,[24] suggesting that treatment for FD would also improve symptoms of OAB, or vice versa.

We were unable to use the web survey to investigate the mechanism driving the strong association among OAB, FD, and IBS. However, our findings suggest that these conditions may result from common pathogenic mechanisms such as visceral (gastrointestinal and bladder) hypersensitivity, dysfunction of the central or autonomic nervous system, genetic susceptibility, alterations in serotonin (5-HT) signalling and metabolism, and psychological factors including somatization and anxiety. In the present study, participants with early satiation have to urinate more frequently (Table 4), suggesting that impaired gastric accommodation and bladder accommodation might be caused by common mechanisms. Recent studies demonstrated that elevated levels of neurotrophins, namely nerve growth factor (NGF) and brain-derived neurotrophic factor (BDNF), contribute to bladder overactivity in OAB patients.[25] These factors might also contribute to visceral hypersensitivity in FGIDs.[26] In addition, as gut inflammation is known to cause functional and structural changes in the central nervous system as a result of abnormal afferent input from the gut,[27] it may alter urinary bladder smooth muscle function. Noronha *et al.* reported that a transient colonic inflammatory insult attenuates the amplitude of bladder detrusor muscle contractions in rats.[28] Thus, investigating common mechanisms of OAB and FGIDs might be able to reveal new pathophysiology in these disorders. Further studies are warranted to define underlying mechanisms.

It is possible that our population was not representative of the general population. As patients who were prescribed any drugs were excluded, some of individuals who have more severe symptoms of FD or OAB may have been filtered out. A potential bias also exists if some individuals do not participate in a web-based panel because of concerns about the technology. However, recent Japanese study to clarify the difference between of the survey methods (electronic survey and postal survey) on the epidemiology of FGID symptoms showed that the proportions of symptom subtypes and the patterns of the overlaps were similar in the two methods, despite the difference in the prevalence.[29] In the same way, it is believed that population biases would not have a large impact on the associations between diseases (odds ratios) shown in the present study.[30]

In conclusion, clinical overlap between OAB and FD is very common. This association was not confounded by coexisting IBS, although concomitant IBS-C increased the risk of OAB among participants with FD. Among participants with OAB, the severity of bladder symptoms was greater in participants with dyspeptic symptoms than without them. These results suggest that a subgroup of patients may show development of OAB and FD through the same pathophysiology, which may lead to the discovery of a novel mechanism in FD. Furthermore, potential OAB symptoms should be considered when evaluating and treating patients with FD.

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DISCLOSURE

The authors have no competing interests.

AUTHOR CONTRIBUTIONS

HS and YF designed the research study, conducted the web survey, and collected the data; JM, HS, KH, SF, and SO analyzed and interpreted the data; JM and HS drafted the article; TH supervised and approved final publication.

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ORIGINAL ARTICLE

Transmural Pressure Loading Enhances Gastric Mucosal Cell Proliferation

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Abstract
Aim Although increased intraluminal pressure in the stomach due to gastric outlet obstruction or functional gastric motor dysfunction, including gastroparesis, may affect gastric mucosal integrity, the direct effect of mechanical pressure on gastric mucosal cells has not yet been fully investigated. The aims of this study were to determine whether exposure to transmural pressure would affect the proliferation of gastric mucosal cells and to elucidate the intracellular signaling pathways involved.
Methods Cellular proliferation and DNA synthesis were evaluated in rat gastric epithelial cells exposed to high transmural pressures. The levels of activation of 3 MAP kinases, ERK, JNK, and p38, were assessed, and the induction of immediate early gene expression was examined. The activation of nuclear factor activator protein-1 (AP-1) was evaluated by an electrophoretic mobility shift assay.
Results Exposure to high transmural pressure significantly increased DNA synthesis within 24 h, with the most marked increase observed after exposure to a pressure of 80 mmHg, and this increase was inhibited by the MEK1 inhibitor PD98059. Early activation of ERK kinase, but not of JNK or p38 kinase, was detected after pressure loading. Early induction of the *c-fos* and *c-myc* genes and activation of the AP-1 transcription factor were also demonstrated within 3 h of exposure to 80 mmHg of pressure.
Conclusion Gastric mucosal cell proliferation induced by exposure to high transmural pressure may be related to early activation of ERK, the induction of *c-fos* and *c-myc*, and the activation of AP-1.

Keywords Transmural pressure · Cell proliferation · Immediate early response gene (IERG) · Mitogen-activated protein kinase (MAPK) · Gastric motor dysfunction

Introduction

The gastric mucosa serves as an important barrier to the contents of the gastric lumen, including intraluminal acid, and the epithelium of the gastric mucosa is therefore a site of continual proliferation and differentiation. These processes are regulated by the division of undifferentiated mucous neck cells, and most of the newly produced cells differentiate into surface mucous cells as they migrate rapidly to the surface [1–3]. After injury or ingestion and digestion of food, the functional capacity of the gut can be increased adaptively by increasing the rate at which the gastric epithelium is renewed [4, 5]. Although the precise mechanisms regulating the normal and adaptive growth of the gastric epithelium remain unknown, not only neural mechanisms [6], hormones [7, 8], and growth factors, but also mechanical stimuli such as intragastric pressure or volume overload, are thought to be involved.

Specifically, the pyloric canal is characterized by hypertrophic muscle up to 6-mm thick in patients with infantile hypertrophic pyloric stenosis (IHPS) [9]. In these

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cases, the canal lumens are filled with compressed and redundant mucosa, which protrudes into the gastric antrum because the rigid antropyloric canal is unable to accommodate it [10]. The decreased diameter of the canal lumen in IHPS (3 mm or less) makes the intraluminal pressure extraordinarily high; this could account for the accompanying mucosal hypertrophy, which typically equals or far exceeds the muscle thickness. We recently reported that surgically induced rat gastric outlet obstruction resulted in an increased wet weight of the stomach as well as remarkable hyperplasia of the gastric muscle layers with enhanced levels of fasting plasma ghrelin and expression of the c-kit, a marker for interstitial cells of Cajal, choline acetyltransferase and stem cell factor, a c-kit ligand [6], suggesting that the mechanism by which narrowing of the pyloric canal promotes cell proliferation may involve elevated intragastric pressure [8]. Even without the persistently high pressure loading possibly encountered in IHPS or gastric outlet obstruction, in functional dyspepsia (FD), especially postprandial distress syndrome (PDS), based on the Rome III classification [11, 12], the deregulated antral peristalsis or disturbed fundic accommodation may lead to the development of intragastric pressure after food intake is intermittently enhanced, possibly to supraphysiological levels, leading to possible high transmural pressure overloading at the gastric wall.

The expression of genes encoding regulators of gastric function is modulated over a period of several hours by the gastric contents and also changes rapidly with changes in the digestive state. For example, antral gastrin mRNA levels in rats are significantly elevated relative to fasting levels 1–2 h after refeeding, while the somatostatin mRNA levels are depressed at this time point [13]. The factors mediating these feeding-induced changes in gene expression are not yet clear, but the rapidity of the onset of these changes suggests the involvement of immediate early gene expression and mitogen-activated protein kinase (MAPK) activation. Two aspects of feeding that might trigger the expression of immediate early genes are physical distension of the stomach and chemical stimulation of the mucosa, and Dimaline et al. [14] have shown that distension of gastric mucosa can itself induce the expression of *c-fos and c-myc*.

The mean intragastric pressure produced by fundic waves in healthy dogs has been shown to be >53 cm H_2O (39 mmHg) [15]. Thomas and Kelly classified changes in the intraluminal pressure recorded from the proximal gastric pouch into several types of waves and showed that type III waves (fundic waves) were contractions with amplitudes in excess of 60 cmH_2O (44 mmHg) [16]. The characteristic contractions of the distal stomach represent peristaltic waves. Human antral contractions during fasting produce intraluminal pressure changes ranging from a few to over 100 cmH_2O (74 mmHg) in amplitude and from 1 to 4 s in duration [17], and such intraluminal pressures influence the biological activity of the gastric mucosa. According to ambulatory gastrojejunal manometry data [18], antral motor activity in humans has amplitude of about 50 mmHg with a duration of 4.7–4.9 s. We have recently demonstrated that exposure to transmural pressure induces cell proliferation and DNA synthesis in cultured intestinal epithelial cells, suggesting that mechanical pressure is involved in the regulation of mucosal remodeling in the intestine [19]. In the present study, the term "remodeling" was used to describe the enhancement of cell proliferation in response to transmural pressure, resulting in hyperplastic changes of the entire gastric wall, a totally different term from restitution. Intraluminal pressure, may also be involved in mucosal remodeling of the gastric mucosa, but little is known about the mechanisms underlying pressure-induced growth of gastric mucosal epithelial cells.

In the model of shear stress loading to vascular endothelial cells, the molecular mechanisms that transduce physical stress into gene transcriptional changes have been well investigated. Some of the endothelial second messenger activities associated with this transduction is ion channel activation, stimulation of protein kinase C (PKC) activity, and activation of MAPK pathways [20–22]. Downstream targets of activated MAPK pathways include immediate early response genes (IERG), such as *c-myc* [23], *c-fos* [24] and *c-jun* [25]. These IERGs have been shown to respond to changes in shear stress in endothelial cells [26]. c-Fos/c-Jun heterodimers (assembled by leucine zipper formation) bind primarily to the nuclear factor activator protein-1 (AP-1) site, where they activate transcription [27]. The phosphorylation and activation of these transcription factors induce immediate early gene expression with subsequent entry of the cells into the cell cycle.

In the present study, we first investigated whether exposure of gastric mucosal cells to increased transmural pressure affected their proliferation. Having established that it did, we attempted to investigate the mechanisms by which mechanical pressure might affect the intracellular signaling pathways leading to cell growth. Our results revealed that in cultured gastric epithelial cell lines, mechanical stimulation activates MAPK pathways, induces nuclear proto-oncogenes, and stimulates the formation of AP-1-DNA complexes.

Materials and Methods

Pressure Loading and Evaluation of Cell Proliferation

A gastric mucosal epithelial cell line (RGM-1) established from the gastric mucosa of adult Wistar rats [28] was