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Efficacy of combination therapy with probiotics and mosapride in patients with IBS without diarrhea: a randomized, double-blind, placebo-controlled, multicenter, phase II trial

C. H. CHOI,* J. G. KWON,† S. K. KIM,‡ S.-J. MYUNG,§ K. S. PARK,¶ C.-I. SOHN,** P.-L. RHEE,** K. J. LEE,†† O. Y. LEE,‡‡ H.-K. JUNG,§§ S. R. JEE,¶¶ Y. T. JEEN,*** M.-G. CHOI,††† S. C. CHOI,‡‡‡ K. C. HUH§§§ & H. PARK¶¶¶

*Department of Internal Medicine, Chung-Ang University College of Medicine, Seoul, Korea
†Catholic University of Daegu School of Medicine, Daegu, Korea
‡Kyungpook National University School of Medicine, Daegu, Korea
§University of Ulsan College of Medicine, Seoul, Korea
¶Keimyung University, School of Medicine, Daegu, Korea
**Sungkyunkwan University, School of Medicine, Seoul, Korea
††Ajou University School of Medicine, Suwon, Korea
‡‡Hanyang University College of Medicine, Seoul, Korea
§§Ewha Womans University School of Medicine, Seoul, Korea
¶¶Inje University College of Medicine, Busan, Korea
***Korea University College of Medicine, Seoul, Korea
†††The Catholic University of Korea, College of Medicine, Seoul, Korea
‡‡‡Wonkwang University School of Medicine, Iksan, Korea
§§§Konyang University College of Medicine, Daejeon, Korea
¶¶¶Yonsei University College of Medicine, Seoul, Korea

Key Messages

- Several probiotics are efficacious for the treatment of symptoms in patients with IBS. Mosapride stimulates gastrointestinal motility. A therapeutic intervention combining two therapeutic agents, such as probiotics and mosapride, has not been previously studied in patients with IBS.
- A randomized, double-blind, placebo-controlled, multicenter trial was conducted to determine the effect of such an intervention.
- The combination therapy with probiotics and mosapride is effective and safe for managing IBS symptoms and stool frequency and consistency in patients with non-diarrheal-type IBS.
- The highest dose of the study drug was most effective for producing improvement in abdominal pain/discomfort and spontaneous complete bowel movements.

Address for Correspondence
Hyojin Park, MD, PhD, Professor of Medicine, Department of Internal Medicine, Gangnam Severance Hospital, Yonsei University College of Medicine, 211, Eonju-ro, Gangnam-gu, Seoul, Korea.
Tel: +82-2-2019-3318; fax: +82-2-3463-3882;
e-mail: HJPARK21@yuhs.ac
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Abstract

Background *Probiotics can be beneficial in irritable bowel syndrome (IBS). Mosapride citrate, a selective 5-HT$_4$ receptor agonist, stimulates gastrointestinal motility. We investigated the efficacy of combination therapy with probiotics and mosapride for non-diarrheal-type IBS.* ***Methods*** *Two hundred and eighty-five IBS patients were randomly assigned to either a*

combination of probiotics (Bacillus subtilis and Streptococcus faecium) and mosapride at one of four different doses or a placebo for 4 weeks. The primary outcome was the proportion of patients experiencing adequate relief (AR) of global IBS symptoms at week 4. The secondary outcomes included subject's global assessment (SGA) of IBS symptom relief, individual symptoms, stool parameters, and IBS-quality of life. ***Key Results*** *The proportion of AR at week 4 was significantly higher in all treatment groups compared to the placebo group (53.7% in group 1, 55.0% in group 2, 55.2% in group 3, 53.6% in group 4 [the highest dose], and 35.1% in placebo group, respectively, $p < 0.05$). The proportion of patients reporting 'completely or considerably relieved' in the SGA was higher in the treatment groups than in the placebo group. The abdominal pain/discomfort score in the treatment group 4 was more prominently improved compared with that of the placebo group. In patients with constipation-predominant IBS, the improvements in stool frequency and consistency were significantly higher in the treatment groups 4 and 1, respectively, than those in the placebo group.* ***Conclusions & Inferences*** *Combination therapy with probiotics and mosapride is effective for relief of symptoms in patients with non-diarrheal-type IBS. The study has been registered in the US National Library of Medicine (http://www.clinicaltrials.gov, NCT01505777).*

Keywords *irritable bowel syndrome, mosapride, probiotics.*

INTRODUCTION

Irritable bowel syndrome (IBS) is the most common functional bowel disorder and is characterized by persistent or recurrent abdominal pain and discomfort with altered bowel habits. IBS may lead to impaired social and personal functions as well as a deterioration in the quality of life (QOL) of affected individuals.[1] IBS is considered a multifactorial disorder associated with visceral hypersensitivity, altered gut motility, and dysfunction of the brain–gut axis and immune system.[2–5] In the past, therapy has been symptom-based, but recent advances in the understanding of the pathophysiology have led to the development of therapies directed at specific phenotypes of IBS, such as serotonergic agents and prosecretory agents.[6,7] However, the treatments are still unsatisfactory in some patients. Recently, the involvement of microbial factors, such as alterations in the gut microbiota, has been suggested as a possible etiological mechanism.[8–11] Several microbiology studies have demonstrated quantitative and qualitative alterations in the gut microbiota in patients with IBS compared with healthy controls (HC).[9,12,13] Interventional clinical studies targeting the gut microbiota with antibiotics or probiotics suggested beneficial effects in some patients with IBS.[14,15]

Probiotics are characterized very broadly as 'live microorganisms which, when administered in adequate amounts, confer a health benefit on the host'.[16] Meta-analyses have suggested that probiotics have positive effects on patients with IBS.[17–19] However, not all probiotic strains are effective for the relief of IBS symptoms, and the mechanisms of action remain unclear. Suggested mechanisms of action of probiotics in IBS include modulation of the microbiota composition and brain–gut axis, regulation of local and systemic immune responses, and reduction of visceral hypersensitivity.[20–24]

5-Hydroxytryptamine (5-HT; serotonin) is a neurotransmitter and mucosal signaling molecule mostly produced by enterochromaffin cells. It has diverse functions in regulating gastrointestinal (GI) motility and visceral sensitivity, emotion, appetite, pain and sensory perception, cognition, sexual activity, and sleep.[25,26] Dysfunction of serotonergic signaling is considered important in the pathogenesis of IBS and has been a therapeutic target.[25,26] Studies have shown that serotonergic agents, acting primarily through 5-HT_3 and 5-HT_4 receptors, provide clinical benefit to patients with functional GI disorders including IBS.[6,25,27] The 5-HT_4 receptor agonist prucalopride accelerates colonic transit and has been effective in patients with chronic constipation.[28,29] Mosapride citrate (mosapride) is a selective 5-HT_4 receptor agonist, and its metabolite has a weak 5-HT_3 receptor antagonistic effect.[30] Mosapride has been reported to stimulate gastric motor activity *in vivo* and *in vitro*,[31] and its clinical use includes functional dyspepsia and diabetic gastroparesis.[30,32] In addition, mosapride stimulates colonic motility[33,34] and has been shown to increase bowel frequency in patients with constipation related to Parkinsonism[35] and diabetic neuropathy.[36]

We hypothesized that combining mosapride with probiotics will exert more beneficial effects than placebo in the treatment of patients with IBS without diarrhea. The study aimed to evaluate the efficacy and safety of combination therapy with probiotics and mosapride in a randomized, double-blind, placebo-controlled, phase II trial in patients with non-diarrheal-type IBS and to determine the optimal dosage

of combined probiotics and mosapride for a following phase III study.

MATERIALS AND METHODS

Subjects

This multicenter study was carried out at 16 academic teaching hospitals in South Korea from June 2011 until June 2012. Eligible patients satisfied Rome III criteria for diagnosis of IBS[37] and complained of abdominal pain or discomfort for at least 2 days during the 1-week run-in period.

The inclusion criteria were age between 18 and 75 years, organic abnormality excluded by physical examination with a complete blood cell count and blood chemistry performed during the screening period, and written informed consent. Colonoscopy was performed during the screening period in subjects who had not undergone a colonoscopy in the preceding 5 years. Exclusion criteria included intolerance to probiotics or lactose, pregnancy, lactation, being of childbearing age without using contraception, severe systemic illness (liver cirrhosis, congestive heart failure, chronic renal failure, angina, uncontrolled hypertension, endocrine disorder, metabolic disorder, or malignant tumors), history of inflammatory bowel disease or psychiatric disorder, history of alcohol or drug addiction, previous abdominal surgery other than appendectomy, participation in another clinical trial within 2 months before the onset of this trial, use of drugs influencing the evaluation of efficacy during the study period, and being judged ineligible by clinicians.

Signed informed consent was obtained from each patient prior to entering the study. The study was conducted according to the Declaration of Helsinki and was approved by the Institutional Review Boards of all participating hospitals. The study was registered at www.clinicaltrials.gov (NCT01505777).

Study protocol

This was a multicenter, randomized, double-blind, placebo-controlled, parallel-group phase II trial. During the 1-month screening period, each potentially eligible patient was evaluated by a full review of his or her clinical history, physical examination, and full blood count and serum chemistry. Clinically significant abnormalities in any of the latter test results led to exclusion from randomization. Eligible patients who fulfilled the inclusion criteria received daily questionnaires to be answered in a symptom diary during the 1-week run-in period (the final week of the screening period) to assess IBS symptoms as well as stool frequency and consistency, and the patients also recorded their responses to the IBS-QOL questionnaire (Fig. 1).

According to the recommendation of the Design of Treatment Trials for Functional Gastrointestinal Disorders,[38] only patients who had a pain/discomfort frequency of at least 2 days during the 1-week run-in period were included in the study. After the screening period, patients returned for a second visit and were entered into the treatment period only if they fulfilled the randomization criteria and still met all inclusion and exclusion criteria.

Patients were then randomized to receive either a combination of probiotics (Medilac®; Hanmi Pharma Korea Inc., Seoul, South Korea)[39] and mosapride (Mosasal®; Hanmi Pharma Korea Inc.) at one of four different doses or the placebo, each to be taken three times daily for 4 weeks (see below). Treatment assignments were carried out according to a computer-generated randomization schedule that was designed to allocate patients to one of the four treatment arms or the placebo. Randomization was balanced using permuted blocks and stratified by the investigator and run-in status of the treatment groups. The subjects were assigned to sequential allocation numbers at each site, and the medications were presented as one capsule (probiotic or placebo) plus one tablet (mosapride or placebo) of identical appearance in all five groups to maintain the double-blind condition. The documents for the study groups correspond to the allocation numbers have been kept sealed, and all investigators and subjects remained blinded to the treatment allocation until completion of the trial.

The treatment period lasted 4 weeks, after which the patients were followed up for another 2 weeks. During the treatment and posttreatment periods, IBS symptoms and stool parameters were recorded daily using self-administered questionnaires. Follow-up visits during the study period were scheduled at weeks 4 and 6 to assess IBS symptoms, IBS-QOL, drug compliance, and adverse events. Each week during the study period, patients responded to questions pertaining to adequate relief (AR) and subject global assessment (SGA).[40,41] Adequate relief and SGA were assessed weekly using a patient interview at each follow-up visit (weeks 4 and 6) or via an interactive voice response survey by telephone at weeks 1, 3, and 5 (Fig. 1).

Compliance was calculated as the percentage of planned ingestion of the study product, and a compliance rate greater than 80% was set as the minimum. Dulcolax suppository (bisacodyl, Boehringer Ingelheim GmbH, Ingelheim Rhein, Germany) 10 mg was allowed as a rescue medication if the patient had not passed a bowel movement for at least 3 days and could not stand the constipation symptoms.

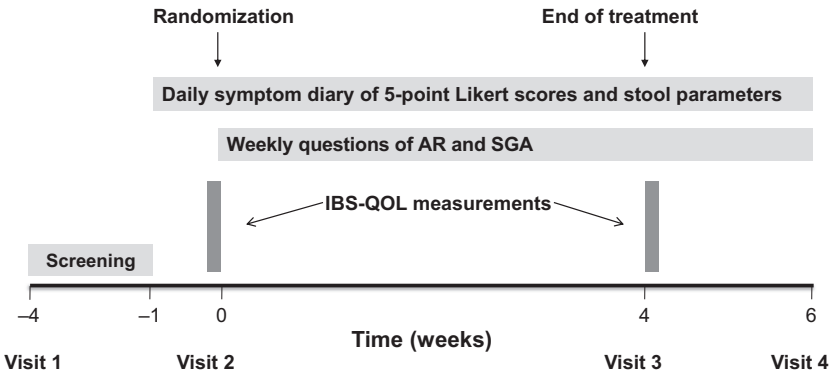

Figure 1 Schematic presentation of the study design. IBS-QOL, irritable bowel syndrome quality of life; AR, adequate relief; SGA, subject's global assessment.

Study medications

Medilac® (Hanmi Pharma Korea Inc.) contains two species of lactic acid bacteria (*Bacillus subtilis* and *Streptococcus faecium*),[39] and Mosasal® (Hanmi Pharma Korea Inc.) contains 5 mg of mosapride per tablet.

Treatment group 4 was given the highest dose (probiotics 3×10^{10} CFU and mosapride 15 mg/day), followed by group 3 (probiotics 1.5×10^{10} CFU and mosapride 15 mg/day), group 2 (probiotics 1.5×10^{10} CFU and mosapride 10 mg/day), and group 1 (probiotics 1.0×10^{10} CFU and mosapride 10 mg/day).

Clinical outcome assessments

During the treatment and posttreatment periods, patients responded weekly to the question: 'In the past 7 days, have you had AR of overall IBS symptoms?' (Yes/No)[42] and SGA (range 1–5; 1, completely relieved; 2, considerably relieved; 3, somewhat relieved; 4, unchanged; or 5, worse) of global IBS symptoms. The following five symptoms were self-assessed daily in a diary: abdominal pain/discomfort, bloating, passage of gas, straining, and sense of incomplete evacuation. Each symptom was evaluated using a five-point Likert scale (range 0–4) and was recorded based on the worst intensity in the previous 24 h.[43] Stool frequency and spontaneous complete bowel movements (SCBMs) were recorded as numbers per day, and stool consistency was evaluated using the Bristol Stool Scale (range 1–7).[44] Quality of life was assessed during the screening period and at the end of treatment by administration of an IBS-QOL questionnaire developed and validated by Drossman *et al.*[45] and translated into Korean.[46]

The primary end point was the proportion of patients who had AR of global IBS symptoms at week 4. The secondary end points included the SGA of IBS symptom relief, individual symptom scores, IBS-QOL, and stool parameters (consistency, frequency, and SCBM).

Safety assessments

Treatment-emergent adverse events (TEAEs) were monitored throughout the study. Relationship between the adverse events and the study drugs were classified by the investigators as (i) definitely related, (ii) probably related, (iii) possibly related, (iv) unknown or unable to determine, (v) probably not related, and (vi) definitely not related, and the first four categories were considered to be study drug-related adverse events. Physical examination, eletrocardiography (ECG), and routine laboratory tests (hematology/biochemistry/urinalysis) were performed at visits 1 and 4 or at early withdrawal. Vital signs were measured at each visit. Safety assessments included adverse events, abnormalities in laboratory findings, and ECG and vital signs from the screening phase until study completion. The baseline was the final evaluation performed before study drug administration.

Statistical analyses

All data were collected and analyzed independent of the investigators, none of whom had access to the data or data analysis until the analyses had been completed. All of the efficacy analyses were based on the intention-to-treat paradigm and were conducted in the full analysis set (FAS) population, which consisted of all randomized patients who received at least one dose of study medication and had at least the primary efficacy measurement. To address missing values, the last observation carried forward imputation method was implemented in variables associated with primary endpoint. Imputation was not applied for missing values of secondary variables or safety measures, and only raw data were used in the analyses.

The sample size calculated for this study intended to detect a 21% difference in the proportion of responders between the treatment and the placebo groups based on previous data.[47] This study was a phase 2 exploratory study for determination of clinically appropriate dose of the investigational product, and thus a more lenient 10% significance level was used instead of the traditional 5%. Using this 10% significance level and a statistical power of 80% with a one-sided test, a sample size of 56 patients was required for each treatment and placebo group, assuming a dropout rate of 10%.

SAS version 9.2 (SAS Institute, Cary, NC, USA) was used to perform all data analyses. Unless otherwise stated, data are presented as mean ± standard deviation (SD). Categorical variables were compared using Pearson's chi-squared test or Fisher's exact test, whereas continuous variables were compared using an independent two sample *t*-test or one-way ANOVA. Baseline characteristics were evaluated by one-way ANOVA, Pearson's chi-squared test, or Fisher's exact test. Rates of symptom improvement (AR and SGA) were described as percentages and were compared between groups using Pearson's chi-squared test or Fisher's exact test. The changes from baseline in Likert scores for individual symptoms, IBS-QOL, and stool parameters were evaluated by independent two sample *t*-test. For the safety variables, Pearson's chi-squared test or Fisher's exact test was used to evaluate the differences in incidence between the groups.

RESULTS

Baseline characteristics

A total of 350 patients who were eligible for inclusion were screened at 16 sites. After the exclusion of 62 patients during the screening period, 286 patients were randomized and entered the treatment phase. One patient was excluded from the placebo group after randomization, and the remaining 285 patients were available for the FAS analysis. A flowchart of patient progression through the study with reasons for premature discontinuation is presented in Fig. 2. Among the 285 evaluable subjects, 144 (50.5%) were female, and the average patient age was 47.0 years (range 20–73). The proportions of male patients were somewhat higher in treatment groups 2 and 3 than in treatment group 1, but the proportions were not significantly different from that of the placebo. There were no significant differences in age, body mass index, or IBS subtype (Table 1).

Adequate relief and subject's global assessment of overall IBS symptoms

In the primary outcome analysis, the proportion of AR for overall IBS symptoms at week 4 was 53.7% (29/54) in treatment group 1, 55.0% (33/60) in treatment group 2, 55.2% (32/58) in treatment group 3, 53.6% (30/56) in

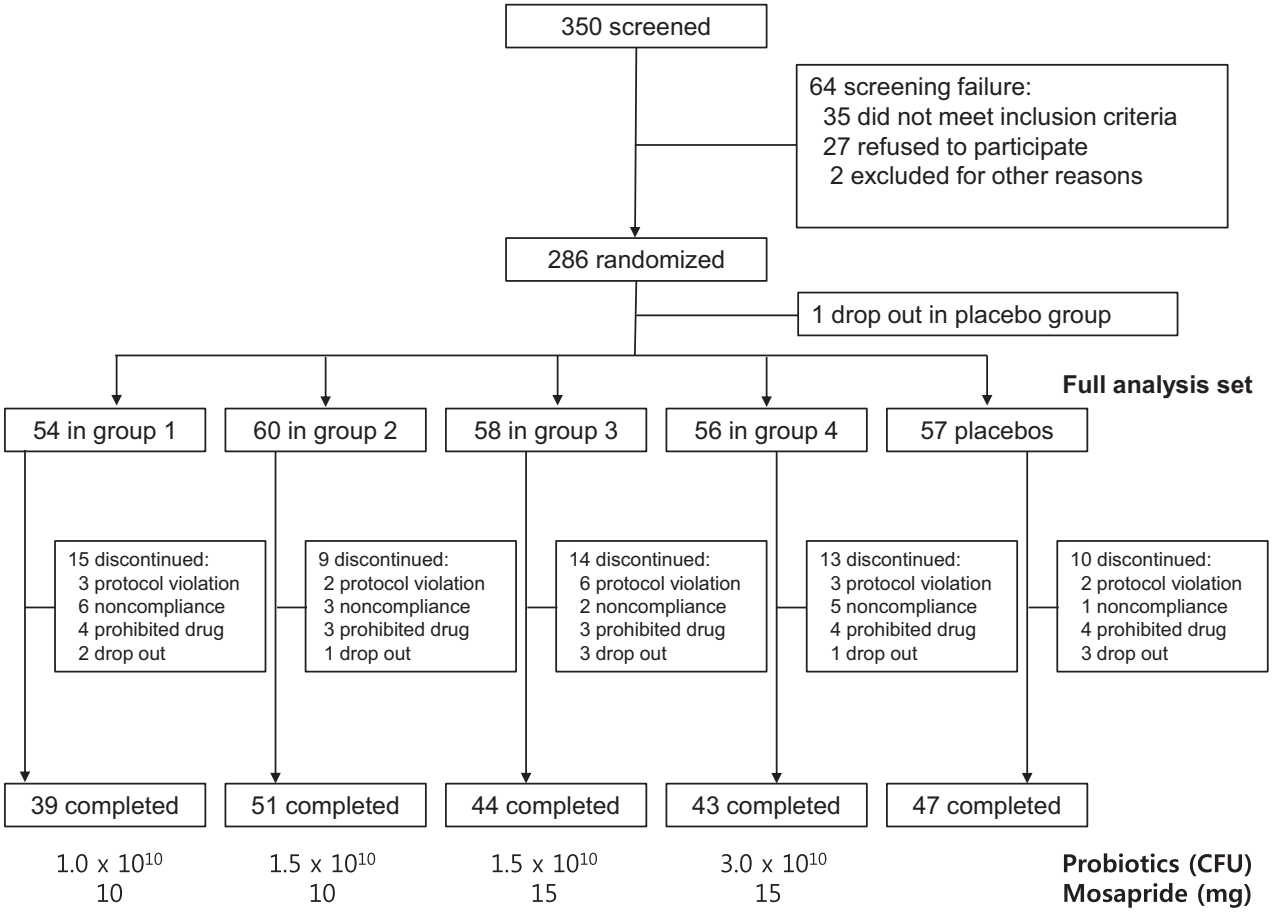

Figure 2 Flowchart of study participants.

Table 1 Baseline characteristics of the patients in the full analysis set

Characteristic	Treatment group 1 (n = 54)	Treatment group 2 (n = 60)	Treatment group 3 (n = 58)	Treatment group 4 (n = 56)	Placebo (n = 57)	p-value*
Medication dose						
Probiotics, CFU	1.0×10^{10}	1.5×10^{10}	1.5×10^{10}	3.0×10^{10}		
Mosapride, mg	10	10	15	15		
Age, mean ± SD	44.8 ± 13.4	48.9 ± 14.2	46.2 ± 13.8	45.9 ± 12.8	48.5 ± 13.2	0.42
Sex, n (%)						
Male	20 (37.0%)	35 (58.3%)	35 (60.3%)	25 (44.6%)	26 (45.6%)	0.07
Female	34 (63.0%)	25 (41.7%)	23 (39.7%)	31 (55.4%)	31 (54.4%)	
BMI (kg/m^2), mean ± sd	22.7 ± 2.7	22.9 ± 3.5	22.8 ± 2.7	22.6 ± 2.7	22.9 ± 2.5	0.96
IBS subtypes, n (%)						
IBS-C	22	25	24	21	21	0.83
IBS-M	31	32	33	33	36	
IBS-U	1	3	1	2	0	

*p-value for analysis of variance. Data are shown as mean ± SD and frequency. BMI, body mass index; IBS, irritable bowel syndrome; IBS-C, constipation-predominant IBS; IBS-M, mixed-type IBS-U; Unspecified-type IBS.

treatment group 4, and 35.1% (20/57) in the placebo group, with statistically significant differences between all of the treatment groups and the placebo ($p < 0.05$, Fig. 3). The proportion of AR tended to be higher in all treatment groups than in the placebo group throughout the whole treatment period and was significantly higher at weeks 3 and 4 ($p < 0.05$, Fig. 4).

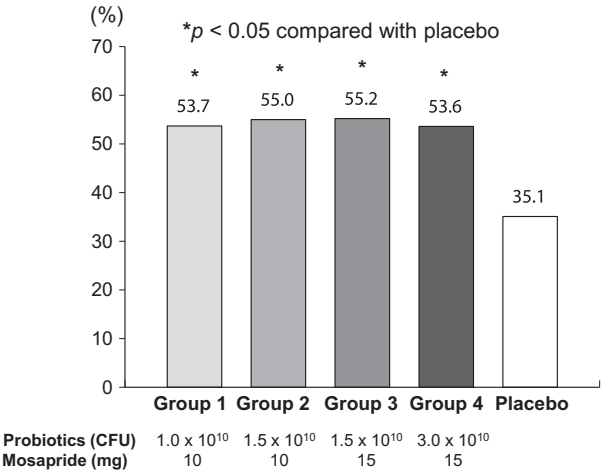

Figure 3 Primary efficacy variable: adequate relief (AR) at week 4. The proportion of AR at week 4 was significantly lower in patients receiving the placebo compared to those receiving treatment with probiotics and mosapride.

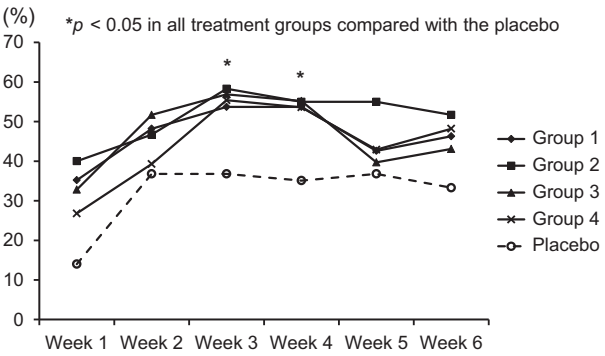

Figure 4 Adequate relief (AR) by week. The proportion of AR was significantly higher in patients receiving treatment with probiotics and mosapride compared to that of the placebo at weeks 3 and 4.

In the secondary outcome analyses, the proportion of patients answering that their symptoms were 'completely or considerably relieved' in the SGA-related question tended to be higher in the treatment groups than in the placebo group. The proportion was significantly higher in all treatment groups than in the placebo group at week 4 ($p < 0.1$, Fig. 5).

IBS symptoms, IBS-QOL, and stool parameters

All individual symptoms, except for straining and the sense of incomplete evacuation in treatment group 4, and the IBS-QOL were significantly improved during the treatment period in all treatment groups. Among the differences in the individual symptom scores at baseline

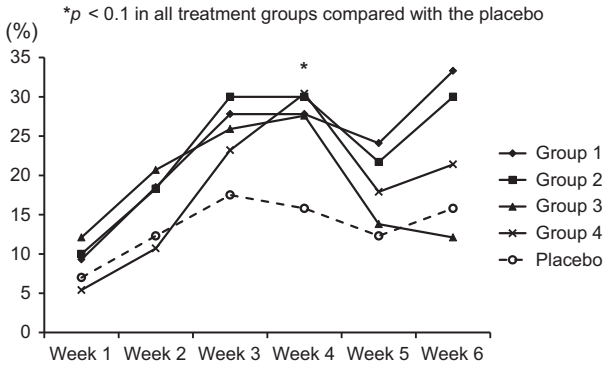

Figure 5 Subject's global assessment (SGA) by week. The proportion of SGA scores of 1–2 (completely or considerably relieved) was significantly higher in patients receiving treatment with probiotics and mosapride compared with that of the placebo at week 4.

and at week 4, only the abdominal pain/discomfort score in treatment group 4 was more prominently improved compared with that of the placebo group (-0.59 ± 0.79 *vs* -0.32 ± 0.76, $p = 0.08$, Table 2). There were more favorable tendencies of effects on bloating in all treatment groups than in the placebo group.

Spontaneous complete bowel movement and BSS scores were analyzed separately according to the IBS subtype. In the analyses for constipation-predominant IBS (IBS-C), overall SCBM and BSS scores were improved only in the treatment groups, and the differences in SCBM and BSS scores at baseline and at week 4 were significantly higher in treatment groups 4 and 1, respectively, than those in the placebo group ($p < 0.05$, Table 3). In mixed-type IBS (IBS-M), the SCBM and BSS scores did not significantly change after treatment in any study group.

Safety and tolerability

Adverse events reported during the study are shown in Table 4. The overall safety profile of the treatment groups was similar to that of the placebo. The number of patients who reported at least one TEAE was not different between the treatment groups and the placebo group: 4 (7.4%), 8 (13.3%), 8 (13.8%), 8 (14.3%), and 6 (10.3%) in treatment groups 1, 2, 3, 4, and the placebo group, respectively. Serious adverse events developed in two patients, with one in treatment group 3 and the other in treatment group 4. In one patient (female, 55 years), uterine myoma was diagnosed during the study period but was not related to the study drugs and was improved by operation without any sequela. In the other patient (male, 23 years), blood alanine aminotransferase, aspartate aminotrans-

Table 2 Changes in Likert scores for individual symptoms and IBS-QOL from baseline to week 4 in all patients with IBS

Symptom	Treatment group 1 (n = 54)	Treatment group 2 (n = 60)	Treatment group 3 (n = 58)	Treatment group 4 (n = 56)	Placebo (n = 57)
Abdominal pain/discomfort					
Baseline	1.49 ± 0.70	1.50 ± 0.69	1.60 ± 0.83	1.73 ± 0.79	1.48 ± 0.71
At week 4	1.00 ± 0.73	1.04 ± 0.79	1.15 ± 0.82	1.19 ± 0.77	1.17 ± 0.77
Difference	−0.45 ± 0.85	−0.46 ± 0.81	−0.43 ± 0.61	−0.59 ± 0.79*	−0.32 ± 0.76
In-group p-value	<0.01	<0.01	<0.01	<0.01	<0.01
Bloating					
Baseline	1.48 ± 0.64	1.57 ± 0.67	1.61 ± 0.86	1.77 ± 0.85	1.55 ± 0.66
At week 4	1.01 ± 0.77	1.13 ± 0.76	1.13 ± 0.89	1.31 ± 0.84	1.21 ± 0.79
Difference	−0.43 ± 0.85	−0.44 ± 0.77	−0.46 ± 0.70	−0.52 ± 0.91	−0.33 ± 0.77
In-group p-value	<0.01	<0.01	<0.01	<0.01	<0.01
Passage of gas					
Baseline	1.52 ± 0.62	1.51 ± 0.64	1.74 ± 0.87	1.59 ± 0.79	1.61 ± 0.65
At week 4	1.20 ± 0.77	1.29 ± 0.64	1.28 ± 0.63	1.39 ± 0.73	1.26 ± 0.61
Difference	−0.29 ± 0.84	−0.22 ± 0.66	−0.39 ± 0.72	−0.21 ± 0.63	−0.36 ± 0.61
In-group p-value	0.02	0.01	<0.01	0.02	<0.01
Straining					
Baseline	0.86 ± 0.54	0.95 ± 0.59	1.11 ± 0.84	1.07 ± 0.64	0.99 ± 0.73
At week 4	0.57 ± 0.53	0.73 ± 0.73	0.69 ± 0.69	0.62 ± 0.62	0.74 ± 0.74
Difference	−0.30 ± 0.62	−0.18 ± 0.70	−0.29 ± 0.60	−0.12 ± 0.65	−0.15 ± 0.66
In-group p-value	<0.01	0.06	<0.01	0.19	0.11
Sense of incomplete evacuation					
Baseline	0.96 ± 0.66	1.10 ± 0.69	1.12 ± 0.86	0.96 ± 0.66	1.09 ± 0.76
At week 4	0.65 ± 0.64	0.78 ± 0.68	0.73 ± 0.69	0.84 ± 0.59	0.88 ± 0.66
Difference	−0.28 ± 0.67	−0.31 ± 0.74	−0.35 ± 0.65	−0.13 ± 0.63	−0.22 ± 0.71
In-group p-value	0.01	<0.01	<0.01	0.13	0.03
IBS-QOL					
Baseline	66.19 ± 19.08	62.30 ± 18.64	60.10 ± 22.90	65.15 ± 20.75	61.31 ± 18.49
At week 4	78.51 ± 16.42	75.74 ± 17.28	75.25 ± 17.07	76.23 ± 18.65	75.32 ± 16.45
Difference	12.20 ± 18.70	13.30 ± 18.19	14.37 ± 14.14	10.68 ± 14.15	14.10 ± 16.52
In-group p-value	<0.01	<0.01	<0.01	<0.01	<0.01

*p = 0.08 compared with the placebo. Data are shown as mean ± SD. Difference = Week 4 score – Baseline score. IBS, irritable bowel syndrome; IBS-QOL, IBS quality of life.

Table 3 Changes in stool parameters from baseline to week 4 in constipation-predominant IBS

Symptom	Treatment group 1 (n = 22)	Treatment group 2 (n = 25)	Treatment group 3 (n = 24)	Treatment group 4 (n = 21)	Placebo (n = 21)
SCBM					
Baseline	0.51 ± 0.42	0.41 ± 0.41	0.45 ± 0.48	0.33 ± 0.33	0.66 ± 0.80
At week 4	0.57 ± 0.37	0.56 ± 0.41	0.63 ± 0.48	0.57 ± 0.47	0.53 ± 0.51
Difference	0.07 ± 0.47	0.15 ± 0.50	0.14 ± 0.38	0.22 ± 0.35*	0.00 ± 0.31
In-group p-value	0.50	0.16	0.12	0.01	0.95
Bristol Stool Scale					
Baseline	3.28 ± 1.12	3.35 ± 1.08	3.27 ± 1.34	2.85 ± 0.98	3.55 ± 1.23
At week 4	3.99 ± 0.98	3.53 ± 1.28	3.28 ± 0.94	3.56 ± 1.34	3.45 ± 1.26
Difference	0.71 ± 0.95†	0.13 ± 1.07	0.26 ± 1.14	0.65 ± 1.70	−0.02 ± 0.70
In-group p-value	<0.01	0.58	0.30	0.10	0.91

*p = 0.04, †p = 0.01 compared with the placebo. Data are shown as mean ± SD. Difference = Week 4 score – Baseline score. IBS, irritable bowel syndrome; SCBM, spontaneous complete bowel movements.

Table 4 Summary of TEAE in all treated patients*

Event, *n* (%)†	Treatment group 1 (*n* = 54)	Treatment group 2 (*n* = 60)	Treatment group 3 (*n* = 58)	Treatment group 4 (*n* = 56)	Placebo (*n* = 58)
Any adverse event	4 (7.4%)	8 (13.3%)	8 (13.8%)	8 (14.3%)	6 (10.3%)
Serious adverse event	0	0	1 (1.7%)	1 (1.8%)	0
Discontinuation due to adverse event	0	1 (1.7%)	2 (3.4%)	1 (1.8%)	0
Study drug-related adverse event	1 (1.9%)	2 (3.3%)	2 (3.4%)	1 (1.8%)	0
Abdominal pain	0	0	1 (1.7%)	0	0
Thirst	0	1 (1.7%)	0	0	0
Dizziness	1 (1.9%)	0	0	0	0
Headache	1 (1.9%)	0	0	0	0
Dysgeusia	0	1 (1.7%)	0	0	0
Atopic dermatitis	0	0	0	1 (1.8%)	0
Increased γ-GT	0	1 (1.7%)	0	0	0
Prolonged PT	0	1 (1.7%)	0	0	0
Study drug-related serious adverse event	0	0	1 (1.7%)	0	0
Increased ALT	0	0	1 (1.7%)	0	0
Increased AST	0	0	1 (1.7%)	0	0
Increased blood CPK	0	0	1 (1.7%)	0	0

*Includes relationships that were 'possible,' 'probable,' and 'definite,' as well as those unable to be assessed. †Incidence is based on the number of patients experiencing at least one adverse event, not the number of events. ALT, Alanine aminotransferase; AST, Aspartate aminotransferase; CPK, Creatine phosphokinase; γ-GT, Gamma-glutamyltransferase; PT, Prothrombin time; TEAE, treatment-emergent adverse event.

ferase, and creatine phosphokinase were increased to 590 IU/L, 149 IU/L, and 8690 IU/L, respectively, at visit 3 after the treatment phase. However, anti-HAV IgM, HBs Ag, and HCV antibody were all negative, and the abnormal chemistry findings normalized at visit 4 (11 days later) with conservative treatment. The patient reported that he had exercised too hard the day prior to visit 3, and so this adverse event was considered to be associated with excessive exercise. However, this event could not be eliminated as unrelated to the study drugs and was classified as a study drug-related serious adverse event.

The treatments were well-tolerated, and the mean drug compliance rate was >90% in all study groups (mean total compliance rate; 93.3 ± 11.6). The number of patients who needed rescue medication (bisacodyl suppository 10 mg) was four (7.1%) in treatment group 1, six (10.0%) in treatment group 2, five (8.6%) in treatment group 3, two (3.6%) in treatment group 4, and six (10.5%) in the placebo group, with no significant difference among the study groups ($p = 0.66$). The mean frequency of recue medication was 1.7 ± 0.5.

DISCUSSION

The present phase II study was designed to assess the efficacy, safety, and dose effect of combination therapy with probiotics and mosapride compared to placebo in patients with non-diarrheal-type IBS. This randomized, double-blind, placebo-controlled, multicenter study evaluated the effects of four different doses of combination treatment of probiotics and mosapride. The therapeutic benefits of these combination therapies were demonstrated through the primary and some secondary measures of efficacy. A significantly higher percentage of patients receiving the combination therapies achieved AR at week 4 in all treatment groups compared with the placebo. The percentage of patients answering that their symptoms were 'completely or considerably relieved' for the SGA question at week 4 was significantly higher in all treatment groups than in the placebo group. Although there were no dose–response effects on AR and SGA, the Likert scores for abdominal pain/discomfort in treatment group 4 decreased more prominently than those of the placebo group. In patients with IBS-C, the SCBM increased significantly in treatment group 4, and the BSS score increased in treatment groups 1 and 4 however, no such improvements were observed in the placebo group. Stool parameters composed of SCBM and BSS scores were analyzed separately based on IBS subtype because the measures usually have meaning only in constipation- or diarrhea-predominant IBS. Collectively, the clinical efficacy of the combination therapies was greater than the effects noted in the placebo group, and these effects were the most prominent and consistent in treatment group 4 considering the effects on abdominal pain and IBS-C subtype.

The gut microbiota has been suggested to be an important factor that may contribute to the pathophysiology of IBS. The proposed beneficial effects of the gut microbiota include the maintenance of intestinal

homoeostasis and mucosal integrity, control over intestinal sensation and motility, and protection against pathogens through bacterial antagonism.[8–11] Probiotics may improve these functions of the gut microbiota and might have beneficial effects via controlling visceral hypersensitivity, abnormal motility, and abnormal brain–gut interactions.[21] Currently, several probiotics are known to be efficacious for the treatment of patients with IBS. Meta-analyses have suggested that the overall efficacy of probiotics is modest and varies by strain and IBS symptoms evaluated.[17–19] In some reports, probiotics were shown to have greater effects on all IBS symptoms,[48] whereas in other studies, they did not improve all IBS symptoms compared with placebo.[22,49–51] Accordantly, in our study, probiotics combined with mosapride were not effective on all IBS symptoms evaluated. The treatments showed significant superior effects on the relief of overall symptoms (all treatment groups), abdominal pain/discomfort (group 4), and stool frequency (group 4) and consistency (group 1) and more favorable tendencies of effects on bloating. However, no superior effects were reported for passage of gas, straining, sense of incomplete evacuation, or IBS-QOL compared with the placebo. As IBS is characterized by the symptoms of abdominal pain and disturbed bowel habits, an effective trial is one that affects these major symptoms.[51] The overall better tendencies for improvements in abdominal pain, bloating, and stool frequency and consistency might have led to the superior effects on AR and SGA in all treatment groups.

Mosapride citrate (mosapride) is a selective 5-HT_4 receptor agonist known to have prokinetic properties on the stomach[31] and is widely used in patients with dyspeptic symptoms in Japan, South Korea, and China.[30] Mosapride has also been reported to stimulate colonic motility[33,34] and increase bowel frequency in patients with constipation related to Parkinsonism[35] and diabetic neuropathy.[36] This drug may exert an additive effect on the effects of probiotics in IBS patients without diarrhea. In addition, mosapride has been well-tolerated in clinical trials with no associated life-threatening adverse events in human or animal studies, which is not the case for other 5-HT_4 receptor agonists such as cisapride and tegaserod.[30] Moreover, many primary care physicians and gastroenterologists agree that specific probiotics can be used as adjuncts to conventional treatment and alone for the management of some IBS symptoms.[21] Thus, we evaluated the effects of combining a therapeutic agent (mosapride) with probiotics for potential benefits for managing IBS symptoms. As expected, the combination therapy with probiotics and mosapride improved not only overall symptoms in patients with non-diarrheal-type IBS but also stool frequency and consistency, especially in patients with IBS-C. However, we did not investigate how the combination therapies exerted their effects in patients with IBS. The stabilization of the composition of gut microbiota by probiotics and the improvement in visceral hypersensitivity and altered intestinal motility by both probiotics and mosapride might be the action mechanisms responsible for the effects of this mixture in our study.[21,52,53] Further studies are needed to evaluate the exact mechanisms of the effects of this combined treatment in patients with IBS.

Mosapride is, as aforementioned, a prokinetic agent that one may use as a symptomatic treatment of constipation and related symptoms on an on-demand basis. Probiotics, on the other hand, act via modulation of intestinal microbiota, influencing gut immune functions and brain–gut interactions, and is rather considered a 'regimen for a certain time frame' with suspected persisting long-term effects. However, probiotics and prokinetics have been shown in many clinical trials to be generally effective only during the treatment periods, with the efficacy decreasing after stopping the treatment. A recent systematic review suggested that regular consumption for a reasonable period of time is important in probiotic therapy for a chronic GI problem, because probiotic strains are transient and will generally be washed out within days.[18] In our study, the effect of combination therapy of probiotics and mosapride increased gradually with time and became evident at treatment week 3, thus this combination therapy seem to be used for at least 2 or 3 weeks regardless of whether the treatment is on an on-demand or continuous basis for relief of symptoms in IBS without diarrhea.

Previous studies on IBS have generally included all IBS subtypes, and only a few studies have focused on IBS-C or IBS-D. However, the US Food and Drug Administration (FDA) recommended that these two IBS subtypes (IBS-C and IBS-D) are optimally studied in separate clinical trials to adequately assess the efficacy of drug products for the treatment of each condition, because the clinical signs and symptoms associated with IBS-C and IBS-D can be significantly different.[54] Clinical studies need to be randomized and placebo-controlled to adequately assess treatment benefit in the setting of IBS. Following these recommendations, we conducted our study with a randomized, double-blind, placebo-controlled design in patients with non-diarrheal-type IBS. However, our study did not fulfill all of the FDA guidance, such as primary endpoints and responder definition. Future

phase III trial will follow the FDA or European Medicines Agency guidelines.

There are several limitations of our study. First, probiotics or mosapride alone groups were not included in the study. As a result, we could not determine if only one of the two drugs was effective, if both were effective, or if they had a synergistic effect. Additional randomized controlled trial may be needed, including mosapride or probiotics alone groups, to identify the active component in this mixture and to determine if the two treatments have a synergistic effect. Second, our study included relatively short treatment (4 weeks) and posttreatment (2 weeks) periods. The treatment effects decreased at the end of treatment (week 5) but appeared to resume in week 6, especially with regard to the SGA results, although the changes were not statistically significant. These variable effects seemed to be due to the characteristics of IBS, with symptom severity varying over time. Thus, additional long-term clinical trials, including both longer treatment and posttreatment periods, are needed to confirm the favorable effects of this acute phase intervention and to determine the sustainability of the effects after treatment completion. Third, the dropout rates were high in general and particularly in group 1. Fourth, we did not evaluate the mechanisms of action by which the two study drugs exerted their effects.

In conclusion, our study demonstrated that combination therapy of mosapride and probiotics is effective and safe for managing IBS symptoms and stool frequency and consistency in patients with non-diarrheal-type IBS. Even if this study has some limitations, it is a useful reference for future studies that evaluate the efficacy of various combination treatments of pre-existing effective drugs, especially based on probiotics, in patients with IBS. Further large-scale, phase III studies are warranted to confirm the effects of combination therapy with probiotics and mosapride, and to elucidate their mechanism of action.

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CONFLICTS OF INTEREST

All authors have no competing interests.

AUTHOR CONTRIBUTION

All authors (CC, JK, SK, SM, KP, CS, PR, KL, OL, HC, SJ, YJ, MC, SC, KH, and HP) were involved with study concept and design, and performed data collection. CC, SM, and HP analyzed and interpreted the data. CC drafted and edited the manuscript. SM and HP reviewed and advised editing the manuscript. All authors approved the final version of the manuscript.

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