Phase III Comparative Clinical Study of AJG533

- Examination of Efficacy and Safety of AJG533 in Patients with Chronic Constipation -

Protocol

Sponsor: AJINOMOTO PHARMACEUTICALS CO., LTD.

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Summary

(1) Purposes of the study

To verify the superiority of AJG533 to placebo and examine its safety in the double-blind comparative study design in which 10 mg of AJG533 or placebo is orally administered once daily for 14 days in patients with chronic constipation using the change in the frequency of spontaneous bowel movements (SBMs) in treatment period Week 1 from screening period Week 2 as the primary endpoint

(2) Subject

1) Target disease

Chronic constipation

2) Inclusion Criteria

Patients who meet all of the following criteria (i) to (ix)

<<At provisional enrollment>>

(i) Patients with chronic constipation

(ii) Patients with the mean SBM* frequency < 3 per week from at least 6 months before the informed consent

(iii) Patients with at least one of the following symptoms related to SBM from at least 6 months before the informed consent:

  (a) Straining during at least 25% of defecations;

  (b) Lumpy or hard stools in at least 25% of defecations; and/or

  (c) Sensation of incomplete evacuation for at least 25% of defecations.

(iv) Patients confirmed to have no organic lesion in the large intestine by colonoscopy or radiographic contrast enema in 5 years

(v) Age: 20 years or older (at the time of the informed consent)

(vi) Gender: Male and female

(vii) Inpatient/outpatient status: Outpatient

(viii) Patients who are capable of providing written consent

<<At enrollment>>

(ix) Patients with SBM* frequency < 6 during the 2-week screening period

*: Defecation without laxatives/enema or digital evacuation. Defecation within 24 hours after the use of a laxative or rescue medication will not be deemed as SBM in this study.

3) Exclusion Criteria

Patients who meet any of the following criteria will not be included in the study:

<<At provisional enrollment and enrollment>>

(i) Patients who have or are suspected to have organic constipation

(ii) Patients who have or are suspected to have symptomatic or drug-induced constipation

(iii) Patients who have or are suspected to have slow colon transit type constipation

(iv) Patients who have or are suspected to have excretory disorder constipation

(v) Patients with a current or past history of gastrointestinal obstruction

(vi) Patients with a current or past history of abdominal hernia

(vii) Patients with a history of laparotomy except simple appendectomy

(viii) Patients with a history of surgical or endoscopic procedures for cholecystectomy and papillotomy

(ix) Patients in whom the dosage regimens of medications, of which changing the dosage regimens is prohibited, were changed after the day of informed consent

(x) Patients who cannot use the rescue medication (bisacodyl suppositories 10 mg)

(xi) Pregnant, lactating or potentially pregnant women, women who wish to become pregnant from the time of the informed consent to the last screening/test point, or women who do not agree to use appropriate birth control methods (oral contraceptives, intrauterine devices, diaphragm, or compliance with the use of condoms by the partner) (women of childbearing potential will receive pregnancy test to check pregnancy status)

(xii) Patients with concurrent serious renal disease (creatinine ≥ 2.00 mg/dL) or liver disease (total bilirubin ≥ 3.0 mg/dL, or AST or ALT ≥ 100 U/L)

(xiii) Patients with concurrent serious heart disease

(xiv) Patients with malignancies

(xv) Patients with a history of serious drug allergy

(xvi) Patients who have participated in a clinical study of AJG533 (who have received AJG533)

(xvii) Patients who are taking part in another clinical study or patients who took part in another clinical study within 12 weeks before enrollment (providing informed consent) in this study

(xviii) Patients who are determined by the investigator or subinvestigator to be not suitable for the conduct of the study for any other reasons.

<<At enrollment>>

(xix) Patients who used the rescue medication (bisacodyl suppositories 10 mg) at least 6 times during the 2-week screening period or patients who used the rescue medication at least 3 times in screening period Week 2

(xx) Patients who used the rescue medication for less than 72 hours after defecation during the 2-week screening period

(xxi) Patients with mushy stool or watery stool (Bristol Stool Form Scale type 6 or 7) in SBM during the 2-week screening period

(xxii) Patients who used prohibited medications/therapies during the 2-week screening period

(3) Study Drug

1) Name of study drug

AJG533

2) Content and dosage form

(i) Study drug: AJG533 5 mg tablet

A light-yellow, round, film-coated tablet containing 5 mg of elobixibat

(ii) Comparator: AJG533 placebo tablet

A film-coated tablet not containing elobixibat that is indistinguishable from an AJG533 5 mg tablet in appearance, odor or weight

3) Package of study drug

Two tablets × 7 lines/sheet of AJG533 5 mg tablets or AJG533 placebo tablets will be packed in a PTP sheet (for 1 week), and two PTP sheets (for 2 weeks) will be packed in an aluminum pouch, and the pouches will be packed in a small box.

(4) Study design

1) Study design

(i) Type of study

A multicenter, randomized, double-blind, placebo-controlled, parallel-group study

(ii) Group structure and target sample size

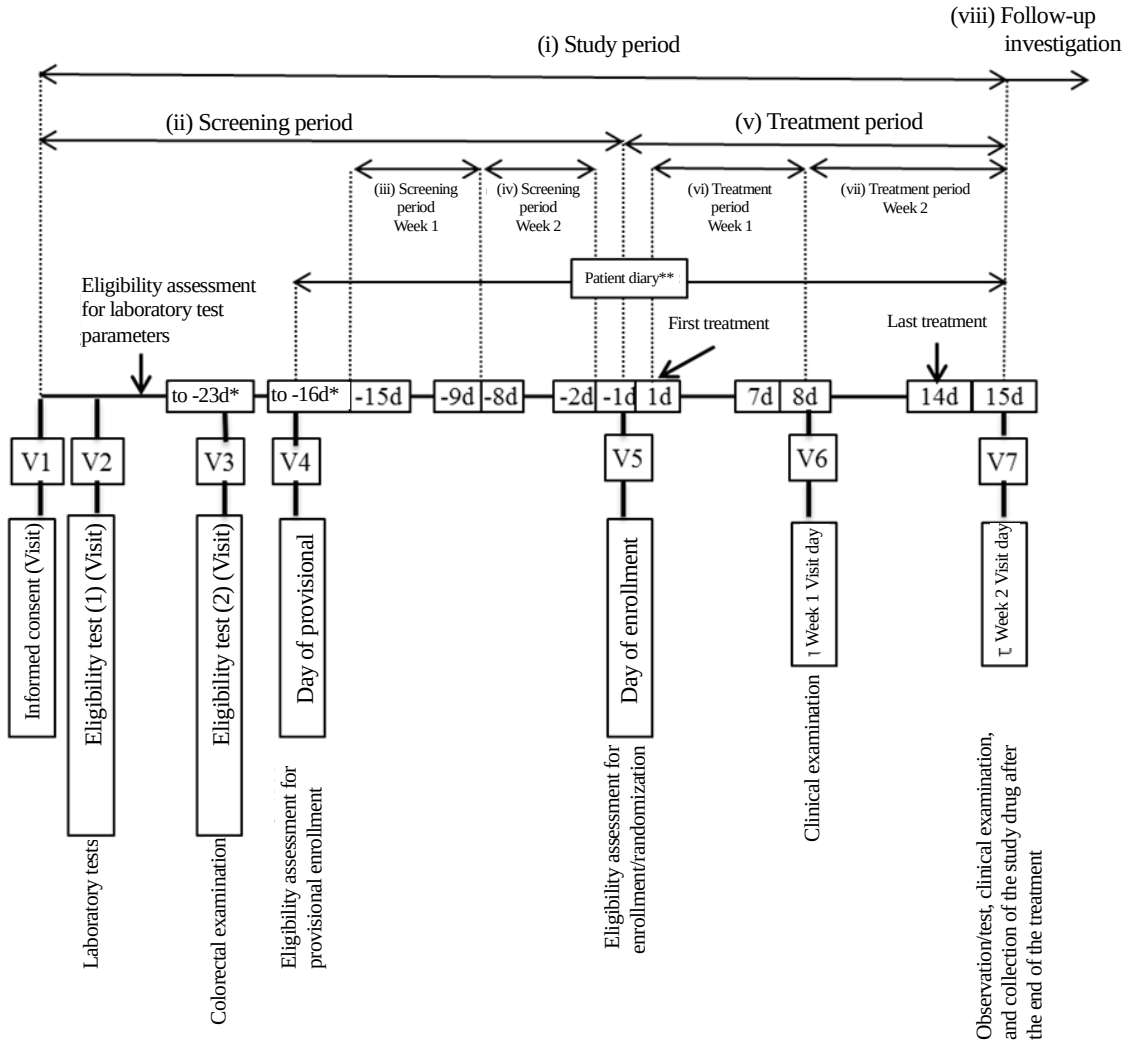
AJG533 10 mg group	: 60 subjects
AJG533 Placebo group	: 60 subjects
Total	: 120 subjects

(iii) Doses, treatment method and duration of treatment

After completing the 2-week screening period, the study drug will be started on the day after the day of enrollment.

AJG533 at 10 mg or placebo will be orally administered once daily approximately 30 minutes before breakfast for 14 days.

(iv) Study schedule



*: Colonoscopy or radiographic contrast enema for reviewing eligibility will be performed by 8 days (Day -23) before the start of the screening period after obtaining the informed consent and reviewing eligibility for the other items. After confirming eligibility, the patient will be provisionally enrolled by Day -16.

**: The use of laxatives and rescue medication and bowel movement status will be recorded from the day of the provisional enrollment. Eligibility will be assessed based on the records from Day -15 (0:00) to Day -2 (24:00) and the patient enrolled based on the results.

2) Endpoints

(i) Efficacy Endpoints

i) Primary endpoint

Change in SBM* frequency from screening period Week 2 to treatment period Week 1

*: Defecation without laxatives/enema or digital evacuation. Defecation within 24 hours after the use of a laxative or rescue medication will not be deemed as SBM in this study.

ii) Secondary endpoints

- Change in SBM frequency from screening period Week 2 to treatment period Week 2
- Change in SBM frequency from the screening period to the treatment period
- Changes in complete spontaneous bowel movement (CSBM)** frequency from screening period Week 2 to treatment period Weeks 1 and 2

  **: SBM without sensation of incomplete evacuation
- Change in CSBM frequency from the screening period to the treatment period
- Proportion of patients who experienced SBM within 24 hours/48 hours after the start of the study treatment
- SBM and CSBM responder *** rates for treatment period Week 1 and Week 2

  *** : A responder will be defined as a patient with weekly SBM or CSBM frequency of ≥ 3 and weekly SBM or CSBM frequency improved by ≥ 1 from Week 2 of the screening period.
- Time to the first SBM
- Use of rescue medication
- Stool consistency as measured by Bristol Stool Form Scale
- Evaluation of weekly-based severity of constipation for treatment period Week 1 and Week 2

(ii) Safety Endpoints

- TEAEs
- Laboratory test parameters
- Vital signs

3) Concomitant Therapies

(i) Prohibited medications and therapies

The use of the following medications and therapies that may affect the study will be prohibited during the period from the start of the screening period to the last observation/test. However, bisacodyl suppositories 10 mg may be used based on the rules specified in (ii) (a) Rescue medication.

- Different types of laxatives (e.g. magnesium oxide preparations, sodium picosulfate, and sennoside)
- Chinese herbal medicines indicated for constipation (e.g., daio-kanzo-to, choi-joki-to, and dai-saiko-to)
- Drugs for the treatment of irritable bowel syndrome (IBS) (e.g., ramosetron hydrochloride, polycarbophil calcium, and trimebutine maleate)
- 5-HT_3 antiemetics
- Enteric movement accelerating agents (e.g., mosapride citrate, metoclopramide, and domperidone)
- Macrolide antibiotics (e.g., erythromycin, roxithromycin, and azithromycin)
- Antidepressants, antipsychotics, antianxiety agents, and tranquilizers (excluding those used for the treatment of insomnia)
- Anticholinergic drugs (excluding topical agents)
- Nonsteroidal anti-inflammatory drugs (excluding topical agents)
- Supplements, etc. for improving constipation
- Enema and intestinal lavage
- Intestinal tract cleaning agents
- Drugs affecting the amount of bile acid (e.g., colestimide and ursodeoxycholic acid)
- Other study drugs
- Constipation therapies such as biofeedback
- Digital evacuation

(ii) Restricted concomitant medications (therapies)

Restricted medications are described below.

(a) Rescue medication

Bisacodyl suppositories 10 mg prescribed for this study may be used under the following conditions. Bisacodyl suppositories 10 mg may be utilized as a rescue medication only when there is no bowel movement for at least 72 consecutive hours during the period from the start of the screening period to the last observation/test. When there is no bowel movement even after using one dose of bisacodyl suppositories 10 mg, the investigator will determine whether or not the subject can continue the study.

When there is no bowel movement for at least 72 consecutive hours after having a bowel movement following the use of the rescue medication, one dose of the rescue medication may be used again. However, the use of the rescue medication within 24 hours before and 48 hours after the start of the study treatment is prohibited.

(b) Drugs of which changes in dosage regimens are prohibited

When any of the following drugs is used at the time of receiving the informed consent and needs to be continued during the study period, the change in its dosage regimen is prohibited during the study period from the time of the informed consent to the last observation/test.

For the drugs of which changing the dosage regimen is prohibited, the name of the drug used from the day of the informed consent to the last observation/test, period of use, and dosage regimen will be investigated and recorded.

- Hypnotics for the treatment of insomnia
- Calcium antagonists
- Iron preparations

4) Guidance for subjects

Subjects will be instructed not to greatly change their dietary life and exercise patterns from the start of the screening period to the end of the study.

(5) Observation, test and investigation items

1) Observation, test and investigation items

Table a Observation, test and investigation items for eligibility check

	Contents
Subject background	Date of birth, inpatient/outpatient status, medical history (including surgical history)[*1] and complications [*2]
Observation/investigation items	Subjective symptoms, objective findings, and use of concomitant medications
Vital signs	Blood pressure (systolic and diastolic blood pressures) and pulse rate
Hematology	White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count
Blood biochemistry	Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin, urea nitrogen, creatinine, uric acid, LDL-cholesterol, HDL-cholesterol, total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl, and Ca
Urinalysis (qualitative)	Glucose, protein, urobilinogen, occult blood, pregnancy test (women only) [*3]
Items related to bowel movements	Bowel movement status (date and time, stool consistency as measured by Bristol Stool Form Scale) and use of laxatives[*4] and rescue medication
Colorectal examination (colonoscopy or radiographic contrast enema)	Colorectal examination will be performed by 8 days before the start of the screening period for subjects for whom the absence of organic disease has not been confirmed with the examination in 5 years before the provisional enrolment.

*1: Medical history: Medically important symptoms or diseases (e.g., those related to the exclusion criteria) that developed within 5 years and cured before the start of the study treatment All laparotomy histories will be recorded.

*2: Complications: Diseases present or symptoms or signs observed at the start of the study treatment and considered as clinically important

*3: Pregnancy test will be performed at Visit 2 only in women of childbearing potential.

*4: Laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and intestinal tract cleaning agents (including commercially available drugs) used on the day before the start of the screening period will also be investigated.

Table b Observation, test and investigation items

	Contents
Subject background	Gender, birth date, race, medical history (including surgical history)[*1], complications[*2], and presence or absence of constipation-predominant IBS
Observation/investigation items	Study drug compliance, use of concomitant medications, subjective symptoms, objective findings, and study completion
Vital signs	Height, body weight, blood pressure (systolic and diastolic blood pressures), and pulse rate
Hematology	White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count
Blood biochemistry	Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin, urea nitrogen, creatinine, uric acid, LDL-cholesterol[*3], HDL-cholesterol[*3], total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl, and Ca
Urinalysis (qualitative)	Glucose, protein, urobilinogen and occult blood
Items related to bowel movements	Bowel movement status (date and time, stool consistency as measured by Bristol Stool Form Scale, and sensation of complete evacuation), constipation severity assessment (date of assessment and severity assessment), and use of the rescue medication

*1: Medical history: Medically important symptoms or diseases (e.g., those related to the exclusion criteria) that developed within 5 years and cured before the start of the study treatment All laparotomy histories will be recorded.

*2: Complications: Diseases present or symptoms or signs observed at the start of the study treatment and considered as clinically important

*3: LDL-cholesterol and HDL-cholesterol at Visit 7 and at discontinuation will be investigated in a blinded manner.

2) Investigation items and schedule

The investigation items and schedule in this study are shown in Table c. The acceptable ranges at each visit are presented in Table d.

Table c Investigation items and schedule

Study period

Screening period

	Informed consent	Eligibility test (1)	Confirmation of laboratory test eligibility assessment results *1	Eligibility test (2) *2	Day of provisional enrollment		Day of enrollment	Starting day of study treatment		Week 1 Visit day		Week 2 Visit day /at discontinuation
visit	1	2		3	4		5			6		7
Test/observation time points (day)				to -23	to -16		-1	1		8	14	15
Informed consent	○											
Enrollment and confirmation of eligibility assessment results *1			○		○		○					
Medical history *3, complications *4, presence or absence of constipation-predominant IBS							○					
Clinical examination		○		○	○		○			○		○
TEAEs								←				→
Subject background (gender, birth date, inpatient/outpatient status, and race)		○										
Vital signs (height)							○					
Vital signs (body weight)							○					○
Vital signs (blood pressure, pulse rate)		○					○					○
Colonoscopy or radiographic contrast enema *2				○								
Study drug compliance								←				→
Use of rescue medication (patient diary)						←						→
Use of laxatives *5 (patient diary)					↔							
Use of concomitant medications (patient diary)	←											→
Bowel movement status *6 (patient diary)					←							→
Constipation severity evaluation (patient diary)							○			○		○
Hematology		○					○					○
Blood biochemistry (i) (excluding LDL-C and HDL-C)		○					○					○
Blood biochemistry (ii) (LDL-C and HDL-C)		○					○					○
Pregnancy test *7		○										
Urinalysis (qualitative)		○					○					○
Blood collection volume (mL)		7mL					7mL					7mL
Urine collection volume (mL)		10mL					10mL					10mL

○: To be performed

*1: Confirmation of laboratory test eligibility assessment results: Eligibility assessment results will be confirmed for renal and hepatic functions in the eligibility test (1). If eligible, the eligibility test (2) or provisional enrollment will be implemented. If ineligible, the patient will be registered as ineligible.

*2: Colonoscopy or radiographic contrast enema will be performed by 8 days before the start of the screening period (Day -23) for patients for whom the absence of organic disease has not been confirmed with the examination in 5 years before the provisional enrolment.

*3: Medical history: Medically important symptoms or diseases that developed within 5 years and cured before the start of the study treatment. However, all laparotomy histories will be recorded.

*4: Complications: Diseases present or symptoms or signs observed at the start of the study treatment and considered as clinically important

*5: The use of laxatives after the end of provisional enrollment will be recorded. The use of laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and intestinal tract cleaning agents (including commerically available drugs) will be recorded.

*6: The bowel movement status from the day of provisional enrollment will be recorded.

*7: To be performed only in women of childbearing potential.

Table d Acceptable range at each visit

Visit	5	6	7 *1
No. of days from the start of study treatment	- 1 day	8 days	15 days
Acceptable range (day)	-1	8 to 10	15 to 17

*1: In principle, the tests at discontinuation will be performed within 2 weeks after the date of the last dose.

Note: Any deviation from these acceptable ranges is considered as a protocol deviation. The acceptable ranges for data adoption are presented in Section "13.3 Data Handling."

(6) Statistical analysis

1) Primary endpoint

(i) Primary analysis

The primary endpoint will be the change in the frequency of SBM in treatment period Week 1 fromscreening period Week 2 in the AJG533 10 mg and placebo groups in the efficacy analysis set, FAS. It will be calculated by determining the change in the frequency of SBM for 7 days in each subject (SBM frequency in treatment period Week 1 - SBM frequency in screening period Week 2). Summary statistics will be calculated for each treatment group. The AJG533 10 mg and placebo groups will be compared by the analysis of covariance (ANCOVA) using the SBM frequency in screening period Week 2 as a covariate.

(ii) Secondary analyses

- The same analysis as the primary analysis will be performed in the efficacy analysis set, PPS for the sensitivity analysis of the primary analysis.
- Adjusted analyses using subject demographic parameters as a covariate will be performed. The items and categories based on the blinded review will be presented in the Statistical Analysis Plan.
- Subgroup analyses with appropriately categorized subject demographic parameters will be performed. The items and categories based on the blinded review will be presented in the Statistical Analysis Plan.

2) Secondary endpoints

- Change in SBM frequency from screening period Week 2 to o treatment period Week 2
- Change in SBM frequency from the screening period to the treatment period
- Changes in CSBM frequency from screening period Week 2 to treatment period Week 1 and Week 2
- Change in CSBM frequency from the screening period to the study drug treatment period
- Proportion of patients who experienced SBM within 24 hours/48 hours after the start of the study treatment

- SBM and CSBM responder rates for treatment period Week 1 and Week 2
- Time to the first SBM
- Use of rescue medication
- Stool consistency as measured by Bristol Stool Form Scale
- Evaluation of weekly-based severity of constipation for treatment period Week 1 and Week 2

3) Safety analysis

- TEAEs
- Laboratory test parameters
- Vital signs

(7) Discontinuation criteria by subjects

The investigator or subinvestigator will withdraw subjects from the study in the following cases. The investigator or subinvestigator will promptly notify the sponsor of the date of discontinuation judgment * and reason for discontinuation when any enrolled subject discontinues the study.

(1) [SCREEN FAILURE]: A subject is evaluated as ineligible and therefore not enrolled in the study.
(2) [WITHDRAWAL BY SUBJECT]: A subject requests the withdrawal from the study for his/her personal reasons including moving and hospital transfer (discontinuation due to "TEAEs" or "lack of efficacy" will not be classified as "withdrawal by subject").
(3) [ADVERSE EVENT]: The investigator or subinvestigator judges that a subject can no longer continue the study drug due to a TEAE or a subject requests the withdrawal from the study due to a TEAE.
(4) [PROTOCOL DEVIATION]: A subject is found ineligible after he/she is enrolled in the study.
(5) [LACK OF EFFICACY]: The target disease is not worsened, but the continued participation in the study will pose an unacceptable risk to the subject due to inadequate clinical efficacy.
(6) [STUDY TERMINATED BY SPONSOR]: The entire study is discontinued.
(7) [LOST TO FOLLOW-UP]: A subject is lost to follow-up and cannot continue the study.
(8) [OTHER]: A subject cannot continue the study in compliance with the protocol or the investigator or subinvestigator judges that a subject has to be withdrawn from the study for other reasons.

*: The day of discontinuation judgment is defined as the day when the investigator or subinvestigator judges that the subject meets any of the discontinuation criteria. If the investigator or subinvestigator recognizes that the subject meets any of the discontinuation criteria ex post facto due to emergency or for other reasons, the day the situation arises is considered as the day of discontinuation judgment.

(8) Study period

October 30, 2015 to June 30, 2016 (scheduled date of the conclusion of the first study agreement to the date of the last visit of the last subject)

List of Abbreviations and Definition of Terms

Abbreviations and Terms	Non-abbreviated terms
AJG533	Development code of elobixibat, A3309 (foreign development code)
ALP	Alkaline phosphatase
ALT(GPT)	Alanine aminotransferase(Glutamic-pyruvic transaminase)
ASP	Application service provider
AST(GOT)	Aspartate aminotransferase(Glutamic-oxaloacetic transaminase)
AUEC	Area under the effect curve
AUC	Area under the plasma concentration-time curve
$AUC_{0\text{-last(all)}}$	AUC from time 0 after administration to not detectable (N.D.)
$AUC_{0\text{-t}}$	AUC from time 0 after administration to the last quantitation time
$AUC_{0\text{-}\infty\text{(inf)}}$	AUC from time 0 after administration to infinity
BMI	Body mass index
BUN	Blood urea nitrogen
C4	7α-hydroxy-4-cholesten-3-one
C_{max}	Maximum concentration
CD-R	Compact disc recordable
CK(CPK)	Creatine kinase (Creatine phosphokinase)
CRP	C-reactive protein
DVD-R	Digital versatile disc recordable
eCRF	electronic Case report form
EDC	Electronic data capture
EDTA-2K	Dipotassium dihydrogen -ethylenediaminetetraacetate
Emax	Maximum effect
FAS	Full analysis set
GCP	Good clinical practice
GMP	Good manufacturing practice
γ-GTP	γ-glutamyltranspeptidase
HDL	High density lipoprotein
HDL-C	High density lipoprotein cholesterol
5-HT_4	5-Hydroxytryptamine type4 receptor
hERG	human Ether-a-go-go related gene
IBAT	Ileal bile acid transporter
IBS	Irritable bowel syndrome
INN	International nonproprietary name

Abbreviations and Terms	Non-abbreviated terms
in vitro	*In vitro* test to detect drug reaction in an artificially created body environment from human or animal tissues
in vivo	*In vivo* test to detect drug reaction in the body or cells by directly administering the test substance in an experimental animal
ITT	Intention-to-treat
LAP	Leucine aminopeptidase
LDH	Lactate dehydrogenase
LDL	Low density lipoprotein
LDL-C	Low density lipoprotein cholesterol
MBq(mBq)	Milli-becquerel (unit of radioactivity)
MCHC	Mean corpuscular hemoglobin concentration
MedDRA	Medical dictionary for regulatory activities
mITT	Modified intention-to-treat
nmol	nano mole
PPS(PP)	Per protocol set
PT	Preferred term
PTP	Press through package
QOL	Quality of life
QT	Time from cardiac depolarization to repolarization
SE	Standard error
SOC	System organ class
SRL	Abbreviation of SRL, Inc.
TK	Toxicokinetics
TG	triglyceride
t_{max}	maximum drug concentration time
Last observations/tests	Observations/tests to be performed at Visit 7 or at discontinuation; those for investigating the outcome of TEAEs are not included.

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1. Background

Constipation is a condition in which patients complain of unpleasant symptoms such as difficulty in defecation, sensation of incomplete evacuation, and abdominal pain due to decreased bowel movement frequency, decreased amount of defecation, or hardening of stool. It is managed by improving dietary and lifestyle habits, self-treatment using over-the-counter (OTC) drugs, and treatment mainly with drugs at medical institutions. Pharmacotherapy given by Japanese medical institutions uses bulk-forming laxatives, such as Bulkose and Vencoll, irritant laxatives, such as Pursennid and Laxoberon, and saline laxatives, such as magnesium oxide and magnesium sulfate, with saline and irritant laxatives being more frequently used. However, they have failed to bring a feeling of satisfaction because saline laxatives cause mushy stools by excessive water secretion and hypermagnesemia and because irritant laxatives are associated with habituation and secondary failure due to long-term use. Therefore, effective drugs that can be used safely for a long time have been awaited.

In recent years, drugs with new mechanisms of actions, such as lubiprostone, a type-2 chloride channel agonist, linaclotide, a guanylate cyclase type-C agonist, and prucalopride, a selective, high affinity 5-HT_4 receptor agonist, have been developed and launched in Western countries. In Japan, lubiprostone (brand name: Amitiza Capsules 24 μg) has been approved for marketing with an indication for "chronic constipation (excluding constipation caused by organic disease)," in June 2012 and marketed. In addition to this, several compounds with different mechanisms of actions are under development.

Elobixibat (INN) is a new drug for the treatment of chronic constipation that has been developed by Albireo AB in Sweden (development code: A3309) (it was previously developed as a drug for hyperlipidemia by AstraZeneca, but the development was discontinued). Elobixibat is poorly absorbed and specifically inhibits the ileal bile acid transporter (IBAT), a transporter associated with bile acid reabsorption in the terminal ileum, to prevent bile acid reabsorption and stimulate bile acids to move into the colon, thereby exerting its effect to promote bowel movements. The foreign Phase I study of elobixibat in patients with chronic idiopathic constipation (A3309-001) showed that the drug was well tolerated up to 10 mg once daily for 14 days and promoted gastrointestinal transit, improved stool consistency, and increased bowel movement frequency after administration at 10 mg. In the foreign late Phase II study (A3309-002) in which 5, 10, or 15 mg of elobixibat or placebo was administered once daily in patients with chronic idiopathic constipation, the change in weekly spontaneous bowel movement (SBM) frequency from the screening period, which was the primary endpoint of the study, was significantly increased in the 10 and 15 mg groups as compared to the placebo group, and no safety concerns were noted.

Ajinomoto Pharmaceuticals obtained the right to develop elobixibat in Japan from Albireo and started to develop it as a drug for chronic constipation (development code: AJG533) in April 2012.

In the Japanese Phase I clinical study (AJG533/CP1) in which elobixibat was administered repeatedly once daily for 14 days in Japanese patients with chronic constipation, elobixibat was well tolerated up to 20 mg and dose-dependently increased the SBM frequency: the difference from the placebo was significant at 2.5 mg or higher.

In the Japanese Phase II study (AJG533/ET1), 5, 10, or 15 mg of elobixibat or placebo was orally administered once daily for 14 days in patients with chronic constipation to examine efficacy and safety. The results showed that the 10 mg or 15 mg groups had significantly superior efficacy to the placebo group for the efficacy primary endpoint (change in SBM frequency in treatment period Week 1 from the screening period) and many of the secondary endpoints, and that the drug was well tolerated up to 15 mg. It was therefore decided that the clinical recommended dose of the drug would be 10 mg when it was orally administered once daily.

Based on the above results, it was decided to perform the Japanese Phase III clinical study (AJG533/CT1) designed to verify the efficacy of elobixibat as orally administered once daily at 10 mg and the Japanese long-term administration study (AJG533/LT1) designed to examine the long-term safety of the drug.

Non-clinical safety results

For the effect on the central nervous system, the single-dose oral administration of up to 350 mg/kg of elobixibat in rats had no neurotoxic, neurobehavioral, autonomic, or psychotropic effect or effect on body temperature or motor coordination. For the effect on motor activity, the drug reduced irritability at 35 mg/kg and motor activity at 350 mg/kg, although these effects were slight.

For the effect on the cardiovascular system, the intravenous administration of 0.0035 to 3.5 mg/kg of the drug in anesthetized beagle dogs did not prolong QT period and cause any ECG or T wave form abnormality. Although mild reduction of coronary blood flow by vascular constriction was observed at 0.0035 to 3.5 mg/kg, the no-observed effect level (NOEL) was set at 0.35 mg/kg, considering the intensity of the effect. The drug had no effect on blood pressure or heart rate when it was orally administered in rats up to 350 mg/kg. An *in vitro* experiment showed that elobixibat at 0.65 μM had a slight inhibitory effect on hERG current (6.0%).

For the effect on the respiratory system, the single oral administration of elobixibat up to 350 mg/kg had no effect on respiratory functions.

For the effect on the digestive system, the single-dose oral administration of elobixibat in rats reduced carbon powder transportation rate in the test system in which a carbon powder suspension was administered at 70 minutes after the administration of the drug (reduction by 30% as compared to the control group) at 350 mg/kg, but caused no effect at 35 mg/kg.

The effect on water and electrolyte metabolism was examined by the single-dose oral administration of elobixibat in rats to evaluate urinary volume, urinary electrolytes, and creatinine clearance. The results showed that 350 mg/kg of the drug significantly reduced the urinary excretion of Na^+ (by 26% as compared to the control group) and increased urinary urea concentration.

The minimum lethal dose of elobixibat was estimated to be above 2000 mg/kg in mice or rats in the single-dose oral toxicity study because 2000 mg/kg of the drug caused no death. The maximum tolerated dose of elobixibat was estimated to be 70 mg/kg because the drug caused vomiting at 140 mg/kg when the dose was escalated from 8.4 mg/kg in the single-dose maximum tolerated dose study in dogs.

In repeated-dose toxicity studies, the lethal dose of elobixibat was estimated to be 500 mg/kg/day or higher because urgent autopsy was required in animals treated with 500 mg/kg/day or higher in the 13-week administration study in mice. Cecal inflammation was observed in rats treated with 200 or 1000 mg/kg/day of elobixibat in the 5-day administration study, while no such finding was observed in the 4-week, 13-week, or 26-week administration study, in which the no observed adverse effect level (NOAEL) was estimated to be the maximum dose used, or 350 or 348 mg/kg/day. NOAEL was estimated to be 17.4 mg/kg/day because vomiting was observed at 139.2 mg/kg/day in the 4-week administration study in dogs. NOAEL was estimated to be 140 mg/kg/day in males and 17.5 mg/kg/day in females because vomiting was observed in females treated with 140 mg/kg/day in the 13-week administration study in dogs. NOAEL was estimated to be 140 mg/kg/day for males and 17.5 mg/kg/day for females because the skin lesions by demodicosis was considered related to elobixibat in

females treated with 140 mg/kg/day in the 52-week administration study in dogs. Additionally, loose or liquid stool observed in dogs treated with elobixibat is considered to be the pharmacological effect of the drug.

For genotoxicity, elobixibat caused negative results in the reverse mutation test using bacteria, *in vitro* mouse lymphoma TK study, and *in vivo* rat bone marrow micronucleus assay.

For reproduction/development toxicity, NOAEL for fertility and embryofetal development was estimated to be 1000 mg/kg/day because no toxic change was observed up to the highest dose of 1000 mg/kg/day in the preliminary embryofetal development study and fertility and embryofetal development study in rats. Similarly, NOAEL was estimated to be the highest dose examined, or 150 mg/kg/day, in the embryofetal development study in rabbits because no toxic change in embryofetal development was observed at the level.

Japanese clinical study results

Japanese clinical studies are listed in Table 1-1.

Table 1-1 List of Japanese clinical studies of AJG533

Study category (study number)	Study design	Purposes	Subject	Study drug, comparator, dose and mode of administration	Sample Size	Treatment period
Japanese Phase I (AJG533/CP1)	Randomized, double-blind, placebo-controlled, single-dose and repeated-dose study	Safety Pharmacokinetics Pharmacodynamics Efficacy	Patients with chronic constipation	(Single-dose) 2.5, 5, 10, 15, or 20 mg of AJG533 or placebo (Repeated-dose) 2.5, 5, 10, 15, or 20 mg of AJG533 or placebo	120 subjects (Single-dose) 10 subjects per group 60 subjects in total (Repeated-dose) 10 subjects per group 60 subjects in total	(Single-dose) Single dose (Repeated-dose) 14 days
Japanese Phase II (AJG533/ET1)	A multicenter, randomized, double-blind, placebo-controlled, parallel-group study	Efficacy Safety	Patients with chronic constipation	5, 10, or 15 mg of AJG533 or placebo	163 subjects	14 days

(1) Japanese Phase I study (AJG533/CP1)

The Japanese Phase I study was performed in Japanese patients with chronic constipation by Ajinomoto Pharmaceuticals.

AJG533 at 2.5, 5, 10, 15 or 20 mg or placebo was orally administered once daily in 60 patients in a double-blind, cross-over manner to examine safety, pharmacokinetics, and food effect. After single-dose safety was confirmed in each group, AJG533 at 2.5, 5, 10, 15 or 20 mg or placebo was orally administered once daily for 14 days in 59 patients to examine the safety by the repeated-dose administration. Additionally, the pharmacokinetics and pharmacodynamics of the drug was examined and the efficacy of the drug was exploratorily examined.

Pharmacokinetic results showed that the exposure to the drug was dose-dependently increased in the dose range of 2.5 to 20 mg, and that the intake of breakfast reduced the exposure to about 20 to 30%. No accumulation was observed when the drug was repeatedly administered at 2.5, 10, or 20 mg, while accumulation was observed at 5 or 15 mg: it seemed that steady state was reached on Day 8 after the start of treatment at 15 mg. Pharmacodynamic results showed reduced serum LDL-cholesterol concentration and increased plasma C4 concentration. No specific tendency was noted for serum HDL-cholesterol concentration. Exploratory efficacy examination showed that the change in SBM frequency was increased at 2.5 mg or higher, and that most of the subjects experienced SBM within 24 hours after treatment. The incidence of TEAEs did not increase dose dependently. Although the incidence of diarrhoea was high after the repeated administration at 20 mg, it was determined that AJG533 was tolerated up to 20 mg because all the TEAEs were mild in severity.

(2) Japanese Phase II study (AJG533/ET1)

The Japanese Phase II study was performed in patients with chronic constipation by Ajinomoto Pharmaceuticals.

AJG533 at 5, 10 or 15 mg or placebo was orally administered once daily in 163 patients to examine efficacy and safety.

For efficacy, the primary endpoint, or the change in SBM frequency from screening period Week 2 to treatment period Week 1 (FAS), was significantly increased in the 10 and 15 mg groups as compared to the placebo group. Significant improvement was also observed in the 10 mg and 15 mg groups as compared to the placebo group for many of the other efficacy endpoints set as secondary endpoint. These results suggest the efficacy of AJG533 in patients with chronic constipation. In the 5 mg group, significant improvement as compared to the placebo group was demonstrated for only a few endpoints, although many of the endpoints tended to improve. Based on these efficacy results, AJG533 at 10 and 15 mg once daily was anticipated to show adequate efficacy. The effect of the drug at 5 mg was considered to be inferior to that at 10 or 15 mg.

For safety, the incidence of TEAEs and TRAEs in the AJG533 groups was higher than that in the placebo group for all dose levels, although the incidence did not increase dose proportionally. Although AJG533 caused gastrointestinal disorders including abdominal pain and diarrhoea, there was no death, serious adverse events other than death, or severe TEAEs. TEAEs led to treatment discontinuation in 4 subjects in the 5 mg group, 1 subject in the 10 mg group, and 2 subjects in the 15 mg group. All the events were mild to moderate and resolved after treatment discontinuation. These safety results indicate that AJG533 was well tolerated up to 15 mg when it was orally administered once daily for 14 days.

Based on the above results, the recommended clinical dose of AJG533 as orally administered once daily was determined to be 10 mg.

Foreign clinical study results

Foreign clinical studies are listed in Table 1-2.

Table 1-2 List of foreign clinical studies of AJG533

Study category (study number)	Study design	Purposes	Subject	Study drug, comparator, dose and mode of administration	Sample Size	Treatment period
Foreign Phase I (D1240C00001)	Single-blind, randomized, placebo-controlled, single-dose and repeated-dose study	Safety Pharmacokinetics	Healthy male volunteers	0.1, 0.5, 2.5, or 5 mg of AJG533 (single dose) 0.25 mg/day (repeated-dose) Placebo	38 subjects (Single-dose) 30 subjects (Repeated-dose) 8 subjects	(Single-dose) Single dose (Repeated-dose) 7 days
Foreign late Phase I(A3309-001)	Double-blind, randomized, placebo-controlled, single-center, prospective, dose-escalating study	Safety Pharmacokinetics	Patients with chronic idiopathic constipation	0.1, 0.3, 1.0, 3.0, or 10 mg/day of AJG533 Placebo	30 subjects	14 days

Study category (study number)	Study design	Purposes	Subject	Study drug, comparator, dose and mode of administration	Sample Size	Treatment period
Foreign late Phase II(A3309-002)	Multicenter, double-blind, randomized, placebo-controlled, parallel-group, dose-finding study	Efficacy Safety	Patients with chronic idiopathic constipation	5, 10, or 15 mg/day of AJG533 Placebo	190 subjects	56 days
Foreign Phase II(A3309-003)	Single-center, double-blind, randomized, placebo-controlled, parallel-group, dose-response study	Efficacy Pharmacokinetics	Patients with functional constipation	15 or 20 mg/day of AJG533 Placebo	36 subjects	14 days
Foreign Phase I(A3309-004)	Open-label, non-randomized study	Pharmacokinetics	Healthy male volunteers	[^{14}C]-AJG533(5 mg, 2.75 MBq)	6 subjects	Single dose
Foreign Phase I (000132)	Open-label, fixed sequence study	Drug-drug interaction (pharmacokinetics)	Healthy male and female volunteers	Elobixibat 10 mg/day Drugs examined for interaction: midazolam, dabigatran, and etexilate	25 subjects	5 days

*: A3309 is the foreign development code of elobixibat.

**: Additionally, two Phase III studies in patients with chronic idiopathic constipation were started (but terminated due to a distribution issue with the trial medication). A long-term administration study was performed as an extension trial of the Phase III study. An early Phase II study was performed in patients with dyslipidemia during the early development phase of the drug.

Foreign clinical studies performed by Albireo (or AstraZeneca) or Ferring Pharmaceutical provided the following results.

(1) Foreign Phase I study (D1240C00001)

The Phase I single-dose and repeated-dose study was performed in healthy male subjects in Sweden by AstraZeneca. A3309 (0.1, 0.5, 2.5, or 5 mg) or placebo was administered in a single dose in a single-blind manner in 30 subjects. A3309 (0.25 mg) or placebo was repeatedly administered in 8 subjects for 7 days to examine the tolerability, safety, and pharmacokinetics of A3309.

Because the single-dose administration study started at 2.5 mg often caused gastrointestinal disorders at 2.5 or 5 mg (including 1 serious adverse event of proctitis at 5 mg), the protocol was revised to administer the drug at 0.1 or 0.5 mg in a single dose and 0.25 mg in repeated doses without dose escalation.

The single-dose administration at 0.1 or 0.5 mg made no difference in the incidence in TEAEs from the placebo group. The repeated-dose administration at 0.25 mg caused gastrointestinal disorders, most of which were mild and not clinically relevant. It caused almost no other TEAEs.

The plasma concentration of A3309 was below the detection limit for almost all the plasma samples analyzed.

(2) Foreign late Phase I study (A3309-001)

The late Phase I study was performed in patients with chronic idiopathic constipation in Sweden by Albireo. A3309 at 0.1, 0.3, 1.0, 3.0, or 10 mg or placebo was administered once daily for 14 days in 30 patients in a double-blind manner to examine the safety, tolerability of, and systemic exposure to, A3309.

None of the treated groups showed any difference in the incidence of TEAEs from the placebo group. Most of observed TEAEs were mild and no death, serious adverse event, adverse event leading to treatment discontinuation, or severe adverse event was observed.

The plasma concentration of A3309 detected in the 1 to 10 mg groups was in the order of picomole and increased dose proportionally. The plasma concentration reached C_{max} within 4 hours after administration and then lowered to a level below the detection limit at 24 hours after administration in the 10-mg group. It was suggested that SBM and complete SBM frequencies were increased and stool consistency improved in the 10-mg group, and that the transit time in the large intestine was shortened (gastrointestinal propulsion was improved) in the 3-mg and 10-mg groups. Total cholesterol and LDL-cholesterol significantly and dose-dependently decreased and C4 dose dependently increased.

(3) Foreign late Phase II study (A3309-002)

The late Phase II study was performed in patients with chronic idiopathic constipation in the United States by Albireo. A3309 at 5, 10, or 15 mg or placebo was administered once daily for 56 days in 190 patients in a double-blind manner to examine the efficacy and safety of A3309.

The change in the weekly SBM frequency from the screening period to Week 1 or to Weeks 1 to 8 was significantly improved in the 10 or 15 mg group as compared to the placebo group. That in the weekly CSBM frequency was significantly improved in the 5, 10, or 15 mg group as compared to the placebo group. The time until the first SBM was significantly reduced and the proportion of subjects with SBM within 24 hours and overall therapeutic effect on stool hardness (improvement) were significantly increased in the 10 or 15 mg group as compared to the placebo group.

The C4 value was significantly increased in the 5, 10, or 15 mg group as compared to the placebo group. Cholesterol and LDL-cholesterol were significantly reduced in the 10 or 15 mg group as compared to the placebo group.

Although TEAEs were generally observed in the active drug and placebo groups, many of them were mild and no serious adverse drug reaction occurred. Many of TEAEs were classified into mild to moderate gastrointestinal disorders. Although they tended to increase dose dependently, they were resolved promptly.

(4) Foreign Phase II study (A3309-003)

The Phase II study was performed in patients with functional constipation in the United States by Albireo. A3309 at 15 or 20 mg or placebo was administered once daily for 14 days in 36 patients in a double-blind manner to examine the effect on gastrointestinal and colonic motor functions.

The results of the primary endpoints including the colonic geometric center at 24 hours, ascending colon half-dose transit time, and colonic geometric center at 48 hours showed that A3309 had an effect to accelerate colon transit. In contrast, no difference was noted between the placebo and active drug groups for the colonic transfer rate/small intestine transit rate at 6 hours or solid gastric half-quantity transit time, indicating that there was no sign of the effect of A3309 on small intestine transit. These results indicated that A3309 would exert its effect in the colon.

Although 2 of the 11 subjects in the 20 mg group discontinued the study due to an adverse drug reaction of diarrhoea, A3309 was generally well tolerated at 15 or 20 mg.

(5) Foreign Phase I study (A3309-004)

The Phase I study of [^{14}C]-A3309 was performed in healthy male volunteers in the United Kingdom by Albireo. [^{14}C]-A3309 was orally administered in a single dose in 6 subjects to examine the absorption, distribution, metabolism, and excretion of A3309.

After the oral administration of [^{14}C]-A3309 (4.16 mg; 2.26 MBq), total radioactivity was mainly excreted in feces: 103.1% of the dose was excreted by 144 hours after administration. The rate of the dose excreted in urine was 0.00% to 0.02%. After the administration of [^{14}C]-A3309, the concentration of total radioactivity in whole blood or plasma was close to the background value and slightly above the detection limit.A3309 had a C_{max} of 0.522 ± 0.339 nmol/L, median t_{max} of 0.75 hours after administration, and $AUC_{0\text{-}t}$ of 0.664 ± 0.504 nmol･h/L. The unchanged form of A3309 (49.84% of the radioactivity administered and equivalent to 96.06% of the radioactivity in the sample) and 4 mono-hydroxylated isomers of A3309 (1.64% of the radioactivity administered and equivalent to 3.16% of the radioactivity in the sample) were identified in the analysis of the extract of pooled feces. No metabolite was detected in the plasma extract.

All TEAEs were mild and no serious TEAEs or those resulting in treatment discontinuation were observed.

(6) Foreign Phase I study (000132)

An open-label fixed-sequence study was performed in healthy male and female subjects in the United States by Ferring Pharmaceuticals to examine the interaction between oral elobixibat and midazolam, a substrate of CYP3A4, or dabigatran etexilate, a substrate of P-gp. Elobixibat at 10 mg/day was administered once daily for 5 days in 25 subjects.

The 90% confidence interval of the ratio of midazolam + elobixibat to midazolam alone was within the predetermined range of 0.80 to 1.25 for C_{max} on Days 1 and 5 and AUC_t on Day 1 of midazolam when midazolam was administered in a single dose in combination with elobixibat on Days 1 and 5.

The 90% confidence interval of the ratio of dabigatran + elobixibat to dabigatran alone was within the predetermined range of 0.40 to 2.5 for C_{max} and AUC_t of dabigatran etexilate when dabigatran etexilate was administered in a single dose in combination with elobixibat on Day 1.

Although the clinical trial report prepared by Ferring Pharmaceuticals concluded that there was no clinically relevant interaction between elobixibat and intestinal CYP3A4 or P-gp, Ajinomoto Pharmaceuticals determined that the possible mild inhibitory effect of elobixibat on P-gp could not be ruled out because the upper limit of the 90% confidence interval was above the standard range of bioequivalence of 0.80 to 1.25.

Positioning of the present clinical study

The efficacy and safety of elobixibat has been confirmed in foreign dose-finding studies in patients with chronic idiopathic constipation.

In the Japanese Phase I clinical study (AJG533/CP1) in which elobixibat was administered once daily for 14 days in Japanese patients with chronic constipation, elobixibat was well tolerated up to 20 mg and dose-dependently increased SBM frequency, with a significant difference from placebo being observed at 2.5 mg or higher.

In the Japanese Phase II study (AJG533/ET1) in which 5, 10, or 15 mg of elobixibat or placebo was orally administered once daily for 14 days in patients with chronic constipation, the 10 and 15 mg groups had significantly superior efficacy to the placebo group for the efficacy primary endpoint (change in SBM frequency in the treatment period Week 1from the screening period) and many of the secondary endpoints, and the drug was well tolerated up to 15 mg. It was therefore decided that the clinical recommended dose of the drug would be 10 mg when it was orally administered once daily.

Based on these results, the present Japanese Phase III clinical study (AJG533/CT1) is planned to verify the efficacy of the drug as orally administered at 10 mg once daily. Furthermore, because the drug is expected to be used for a long period, the Japanese long-term administration study (AJG533/LT1) is also planned to examine the long-term safety of the drug. The long-term efficacy of the drug is also examined in the long-term administration by allowing investigators to adjust the daily dose of the drug among the dose levels of 5, 10, and 15 mg because constipation symptoms are expected to vary in the long term.

2. GCP Compliance

This study will be conducted in compliance with the ethical principles based on the Declaration of Helsinki, standards stipulated in Article 14, Paragraph 3 and Article 80-2 of the Act on Securing Quality, Efficacy and Safety of Pharmaceuticals, Medical Devices, Regenerative and Cellular Therapy Products, Gene Therapy Products, and Cosmetics, and Good Clinical Practice (GCP).

3. Purposes of the study

3.1 Purposes of the study

To verify the superiority of AJG533 to placebo and examine its safety in the double-blind comparative study design in which 10 mg of AJG533 or placebo is orally administered once daily for 14 days in patients with chronic constipation using the change in the frequency of spontaneous bowel movements (SBMs) in treatment period Week 1 from screening period Week 2 as the primary endpoint

3.2 Type of Study

Confirmatory study (Phase III)

4. Subject

4.1 Target disease

Chronic constipation

4.2 Inclusion Criteria

Patients who meet all of the following criteria (i) to (ix)

<<At provisional enrollment>>

(i) Patients with chronic constipation

(ii) Patients with the mean SBM frequency < 3 per week from at least 6 months before informed consent

(iii) Patients with at least one of the following symptoms related to SBM from at least 6 months before the informed consent:

  (a) Straining during at least 25% of defecations;

  (b) Lumpy or hard stools in at least 25% of defecations; and/or

  (c) Sensation of incomplete evacuation for at least 25% of defecations.

(iv) Patients confirmed to have no organic lesion in the large intestine by colonoscopy or radiographic contrast enema in 5 years

(v) Age: 20 years or older (at the time of informed consent)

(vi) Gender: Male and female

(vii) Inpatient/outpatient status: Outpatient

(viii) Patients who are capable of providing written consent

<<At enrollment>>

(ix) Patients with SBM frequency* < 6 during the 2-week screening period

*: Defecation without laxatives/enema or digital evacuation. Defecation within 24 hours after the use of a laxative or rescue medication will not be deemed as SBM in this study.

[Rationales for the selection]

(ii) and (iii): These criteria were set to exclude rectoanal abnormalities, which are excretory disorder symptoms, based on the Rome III Diagnostic Criteria for Functional Constipation[1)].

(iv): The criterion was set to definitely exclude organic constipation in consideration of safety.

(v): The age of 20 years or older, at which the consent of individuals is legally effective, was chosen for patients to voluntarily participate in a clinical study.

(vii): The criterion was set because constipation is mainly treated on an outpatient basis.

(viii): The criterion was set in compliance with the spirit of the Declaration of Helsinki.

(ix): The criterion was set to include patients who have mean SBM frequency of less than 3/week immediately before the start of the study drug.

4.3 Exclusion Criteria

Patients who meet any of the following criteria will not be included in the study:

<<At provisional enrollment and enrollment>>

(i) Patients who have or are suspected to have organic constipation

(ii) Patients who have or are suspected to have symptomatic or drug-induced constipation

(iii) Patients who have or are suspected to have slow colon transit type constipation

(iv) Patients who have or are suspected to have excretory disorder constipation

(v) Patients with a current or past history of gastrointestinal obstruction

(vi) Patients with a current or past history of abdominal hernia

(vii) Patients with a history of laparotomy except simple appendectomy

(viii) Patients with a history of surgical or endoscopic procedures for cholecystectomy and papillotomy

(ix) Patients in whom the dosage regimens of medications, of which changing the dosage regimens is prohibited, were changed after the day of informed consent

(x) Patients who cannot use the rescue medication (bisacodyl suppositories 10 mg)

(xi) Pregnant, lactating or potentially pregnant women, women who wish to become pregnant from the time of the informed consent to the last observation/test point, or women who do not agree to use appropriate birth control methods (oral contraceptives, intrauterine devices, diaphragm, or compliance with the use of condoms by the partner) (women of childbearing potential will receive pregnancy test to check pregnancy status)

(xii) Patients with concurrent serious renal disease (creatinine ≥ 2.00 mg/dL) or liver disease (total bilirubin ≥ 3.0 mg/dL, or AST or ALT ≥ 100 U/L)

(xiii) Patients with concurrent serious heart disease

(xiv) Patients with malignancies

(xv) Patients with a history of serious drug allergy

(xvi) Patients who have participated in a clinical study of AJG533 (who have received AJG533)

(xvii) Patients who are taking part in another clinical study or patients who took part in another clinical study within 12 weeks before enrollment (providing informed consent) in this study

(xviii) Patients who are determined by the investigator or subinvestigator to be not suitable for the conduct of the study for any other reasons.

<<At enrollment>>

(xix) Patients who used the rescue medication (bisacodyl suppositories 10 mg) at least 6 times during the 2-week screening period or patients who used the rescue medication at least 3 times in Week 2 of the screening period

(xx) Patients who used the rescue medication for less than 72 hours after defecation during the 2-week screening period

(xxi) Patients with mushy stool or watery stool (Bristol Stool Form Scale type 6 or 7) in SBM during the 2-week screening period

(xxii) Patients who used prohibited medications/therapies during the 2-week screening period

[Rationales for the selection]

(i) to (ix) and (xix) to (xxii): The criteria were set because they might affect the evaluation of the pharmacological efficacy of AJG533.

(x) to (xv): The criteria were set to secure patient safety.

(xvi): The criterion was set to eliminate the influence of the investigator's or subinvestigator's and patient's preconception caused by the fact that the patient had been treated with AJG533.

(xvii): The criterion was set from the ethical perspective and for eliminating the effect on evaluation of AJG533.

(xviii): The criterion was set assuming cases for which the investigator or subinvestigator might determine that the patient was not suitable for the study for reasons other than the above criteria from scientific and ethical viewpoints.

5. Patient Informed Consent and Information Provision

5.1 Preparation of informed consent form and patient information leaflet

The investigator will prepare the informed consent form and patient information leaflet based on the reference examples of the informed consent form and patient information leaflet provided by the sponsor and other related materials and information.

The investigator will obtain the prior approval of the IRB for the informed consent form and patient information leaflet.

5.2 Contents to be Included in Informed Consent form and Patient Information Leaflet

(1) That the study involves research

(2) Purposes of the study

(3) Name, job title, and contact information of the investigator or subinvestigator

(4) Method of the study (including the experimental aspect of the study, inclusion/exclusion criteria for subjects, and probability of the random assignment to each treatment at randomization, if any)

(5) Anticipated clinical benefits and risks or inconveniences

(6) Availability of other therapeutic methods for the patient (disease) and anticipated important benefits and risks of the therapeutic methods

(7) Expected duration of participation of subjects in the study

(8) That the patient's participation in the study is voluntary and that the patient may refuse to participate or withdraw from the study, at any time, without penalty or loss of benefits to which the patient is otherwise entitled

(9) That the monitor(s), the auditor(s), the IRB, and the regulatory authority(ies) will be granted direct access to original medical records without violating the confidentiality of the patient, and that, by attaching his/her name/seal or signing the informed consent form, the patient is authorizing such access

(10) In case the results of the study are published, the patient's identity will remain confidential.

(11) The person(s) to contact at the study site for further information regarding the study and the rights of subjects, and whom to contact in the event of study-related health injury.

(12) The compensation and/or treatment available to the patient in the event of study-related health injury.

(13) The approximate number of patients involved in the study.

(14) That subjects will be informed in a timely manner in case information becomes available that may be relevant to their willingness to continue participation in the study.

(15) The foreseeable circumstances and/or reasons under which the patient's participation in the study may be discontinued.

(16) The anticipated expenses, if any, to the patient for participating in the study.

(17) The anticipated payment, if any, to the patient for participating in the study (agreement on the calculation method of amount of payment, etc.).

(18) Type of the IRB investigating and reviewing the appropriateness of the study, matters to be investigated and reviewed by each IRB, and other study-related matters concerning the IRB (including the name and address of the founder of each IRB, accessible information, etc.) and matters concerning written procedures for access by the IRB.

(19) Mattes to be complied with by subjects

(20) That data to be checked at enrollment may include data before the informed consent.

(21) That women of childbearing potential will receive pregnant test before provisional enrollment (eligibility test (1)) to confirm that they are not pregnant

(22) That women of childbearing potential are expected to practice contraception using appropriate methods (compliance with the use of oral contraceptives, intrauterine devices, diaphragm or condom by the partner, etc.) under the instruction of the investigator or subinvestigator during the period from the informed consent to the last observation/test.

5.3 Timing and Method for Obtaining Informed Consent

(1) The investigator or subinvestigator will give adequate explanation to prospective subjects using the informed consent form and patient information leaflet approved by IRB, and the study collaborator will, as necessary, provide supplementary explanation as a person giving a supplementary explanation before performing study-related activities specified in this protocol (such as tests/observations, and administration of the study drug).

(2) The investigator, subinvestigator, or study collaborator will provide prospective subjects with an opportunity to ask questions and adequately answer all the questions prior to obtaining consent. In addition, they will provide prospective subjects with a sufficient amount of time for deciding whether or not to participate in the study.

(3) The investigator or subinvestigator will record the date of giving the explanation for consent in and affix his/her name/seal or signature to the informed consent form. The study collaborator who gave the supplementary explanation will also affix his/her name/seal or signature to and record the date of explanation in the informed consent form.

(4) In case an impartial witness is required at the time of giving the explanation (in case the subject is incapable of reading the written information, etc. but is able to understand the contents thereof using oral or other means of communication), not only the prospective subject, but also the witness will affix his/her name/seal or signature to and record the date on the informed consent form. The impartial witness shall be a person who is independent from the conduct of the study and shall not be unduly

influenced by persons involved in the study. The investigator, subinvestigator or study collaborator shall not serve as the witness.

(5) The investigator or subinvestigator will record the date of explanation in the subject screening list/register for patients to whom the explanation was given (prospective subjects).

(6) The investigator or subinvestigator will obtain the written voluntary consent of each prospective subject to participate in the study. The prospective subject will affix his/her name/seal or signature to and record the date of consent in the form.

(7) When the prospective subject is unable to personally sign the informed consent form due to a compelling reason, his/her consent will be obtained by his/her name/seal whenever possible. If this is also impossible, the impartial witness shall attend and name/seal or sign and date the informed consent form to prove that the subject has received adequate explanation and provided his/her voluntary consent. In such a case, the columns for signature and the date of consent for the subject shall be left blank. Furthermore, the witness should additionally record the name of the subject, background reason why the subject is incapable of personally providing his/her signature, and the relationship between the subject and witness in the margin of the informed consent form.

(8) The investigator or subinvestigator shall give the subject this named/sealed or signed and dated informed consent form (copy) and patient information leaflet before the subject is enrolled in the study.

(9) The investigator, subinvestigator or study collaborator will record the subject identification (ID) code to be assigned at the time of obtaining consent, the date of informed consent obtained, and the date of handing the informed consent form to the subject in the subject screening list/register.

(10) Informed consent shall always be obtained before implementing tests and observation for the purpose of this study, provided, however, that test/observation data within the acceptable time window before receiving consent may be used if the subject has agreed to this.

(11) The informed consent form will be attached to medical records and retained unless otherwise stipulated at each study site.

(12) The investigator or subinvestigator shall check whether or not the subject is treated by other physicians and provide the applicable physicians with information on his/her participation in the study based on his/her consent.

5.4 Considerations in obtaining informed consent

(1) The investigator, subinvestigator or study collaborator must not force the prospective subject to participate in or continue to participate in the clinical study or exert undue influence on the prospective subject.

(2) The informed consent form and patient information leaflet and supplementary oral information to be given during explanation must not contain any expression that makes subjects or prospective subjects

give up or doubt their rights or any expression that releases, reduces, or doubts the legal responsibilities of the study site, investigator, subinvestigators, study collaborators, or sponsor.

(3) The oral and written explanation to be given to subjects and informed consent form and patient information leaflet shall use terms understandable to subjects and avoid technical terms as much as possible.

(4) The investigator, subinvestigator, or study collaborator must not participate in the study as a subject.

(5) In principle, the personnel of the study sites (including students) must not participate in the study as a subject. However, this does not apply to the case where the voluntary consent of subjects can be assured by records or other measures, considering whether subjects depend on (are subservient to) the investigator or subinvestigator.

5.5 Actions to be taken when information that may influence the willingness of subjects becomes available and revision of informed consent and patient information leaflet

(1) The investigator or subinvestigator will inform subjects in a timely manner if any information that may affect their willingness to continue participation in the study (e.g. additional information on adverse drug reactions) becomes available, confirm their willingness to continue to participate in the study, and document the result and its date.

(2) When the investigator decides that the informed consent form and patient information leaflet should be revised due to the information, the investigator will immediately revise the informed consent form and patient information leaflet in cooperation with the sponsor and obtain the approval of IRB.

(3) When the informed consent form and patient information leaflet is revised, the investigator or subinvestigator will explain their contents to subjects and obtain their voluntary consent to continue to participate in the study. The procedures and considerations in obtaining the written consent are as specified in Sections “5.3 Timing and method for receiving informed consent” and “5.4 Points to consider for obtaining informed consent.”

6. Study Drug

The study drug manufactured and packed in compliance with the Good Manufacturing Practice (GMP) for Study Drugs will be used.

6.1 Name of study drug

(1) Study drug: AJG533 5-mg tablet

1) Study drug code: AJG533

2) Nonproprietary name: Elobixibat Hydrate

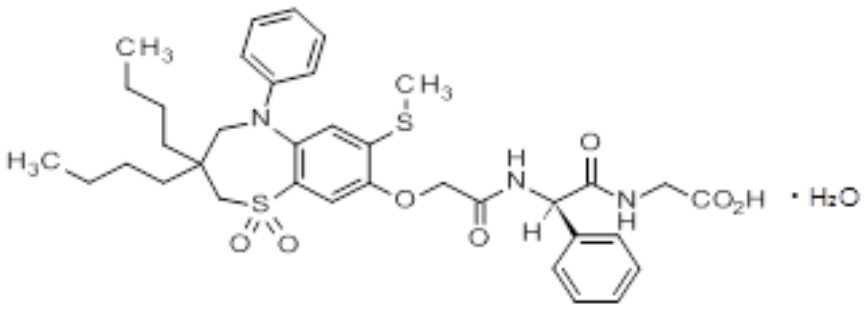
3) Chemical name:

[(2R)-2-(2-{[3,3-Dibutyl-7-(methylsulfanyl)-1,1-dioxo-5-phenyl-2,3,4,5-tetrahydro-1H-1λ^6,
5-benzothiazepin-8-yl]oxy}acetamido)-2-phenylacetamido]acetic acid monohydrate

4) Molecular formula: $C_{36}H_{45}N_3O_7S_2 \cdot H_2O$

5) Molecular weight: 713.90

6) Chemical structure:



7) Content and dosage form

A light-yellow, round, film-coated tablet containing 5 mg of elobixibat

(2) Comparator: AJG533 placebo tablet

1) Study drug code: AJG533

2) Content and dosage form

A film-coated tablet not containing elobixibat that is indistinguishable from an AJG533 5 mg tablet in appearance, odor or weight

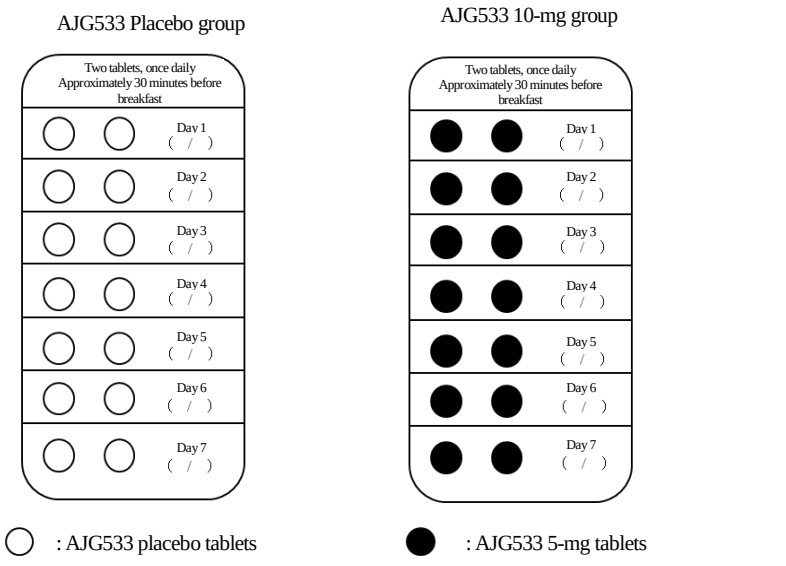
[Rationales for selecting comparator]

The placebo was selected as a comparator to verify the superiority of AJG533 to the placebo.

6.2 Package and Labelling

Two tablets × 7 lines/sheet of AJG533 5-mg tablets or AJG533 placebo tablets will be packed in a PTP sheet (for 1 week), and two PTP sheets (for 2 weeks) will be packed in an aluminum pouch, and pouches will be packed in a small box.

[PTP package form]



* The actual AJG533 5-mg tablets (●) and AJG533 placebo tablets (○) are indistinguishable from each other.

[Label on the small box]

(i) Top side

FOR CLINICAL STUDY USE ONLY

Group XX No. X

AJG533

Protocol No. AJG533/CT1

Contents:	14 tablets × 2 sheets
Manufacturing No.:	AJG533CT1
Storage:	Store at room temperature.
Expiration:	Expiration specified in the “document stipulating storage conditions of the study drug”

Sponsor: AJINOMOTO PHARMACEUTICALS CO., LTD.
2-1-1 Irifune, Chuo-ku, Tokyo

(ii) Long side

FOR CLINICAL STUDY USE ONLY	AJG533/CT1	Group XX No. X

6.3 Storage and expiration

(1) Storage: Store at room temperature.

(2) Expiration: Date specified in the separately specified “document stipulating storage conditions of the study drug”

6.4 Control of study drugs

(1) The sponsor will conclude the study agreement with each study site and then deliver the study drugs to the study drug controller. The sponsor will give the study drug delivery note to the study drug controller in delivering the study drugs.

(2) The study drug controller will check the manufacturing number, quantity, expiration date, and storage of the study drugs at receipt and send the receipt of the study drugs to the sponsor. The study drug controller will appropriately store and control the study drugs according to the study drug handling procedures prepared by the sponsor.

(3) The study drug controller will prescribe the study drugs to each subject from the small box with the drug number assigned to each subject. The study drug controller will keep the small box dedicated for each subject and will not use the drugs in it for other subjects or patients.

(4) The study drug controller will collect all unused drugs from subjects and record the date and quantity of collection in the study drug control table.

(5) The sponsor will check the study drug inventory retained by the study drug controller against the study drug control table at appropriate intervals.

(6) The study drug controller will record the quantity of the study drugs delivered from the sponsor, quantity of the study drugs prescribed, quantity of the study drugs collected from subjects, if any, and quantity of the unused study drugs at the end of the study in the study drug control table and appropriately store and control the table with the study drug delivery note.

(7) Unused study drugs, study drugs collected from the patients and empty boxes after prescription shall be, in principle, properly stored by the study drug controller at each study site until key code breaking, and the sponsor shall collect them after key code breaking. In addition, when the study drugs and other relevant items are collected before key code breaking upon request of the study site, etc., the study drug controller shall seal the unused tudy drugs and thestudy drugs collected from the patients.

(8) The sponsor will collect and store the unused study drugs, study drugs collected from subjects, and empty drug boxes after prescription from each study site.

(9) The study drug controller at each study site and sponsor will check the quantity of the study drugs delivered, quantity of the study drugs prescribed, quantity of the study drugs collected from subjects, and quantity of unused study drugs against the study drug control table when the sponsor collects the study drugs. The study drug controller will check the results and then affix his name/seal or signature to

the study drug control table. The study drug controller will return the study drug and personally deliver a copy of the study drug control table and study drug return note to the sponsor. The sponsor will personally deliver the study drug collection note to the study drug controller. The information that identifies subjects on the copy of the study drug control table or small boxes, if any, will be made unreadable. The study drug controller will investigate the cause and record the details in the study drug control table if any discrepancy is found between the study drug control table and quantity of the unused study drugs. The study drug controller will record the name of the drug, quantity, and cause of the discrepancy in the study drug return note.

7. Study design

7.1 Study design

7.1.1 Type of study

A multicenter, randomized, double-blind, placebo-controlled, parallel-group study

[Rationales for the selection]

A multicenter, randomized, double-blind, parallel-group study to compare 2 groups using placebo as the comparator was planned to verify the superiority of AJG533 to placebo.

7.1.2 Group structure

AJG533 10 mg group	: 60 subjects
AJG533 placebo group	: 60 subjects
Total	: 120 subjects

[Rationales for the selection]

The rationale for the sample size is described in “13.1 Sample Size.”

7.1.3 Doses, treatment method and duration of treatment

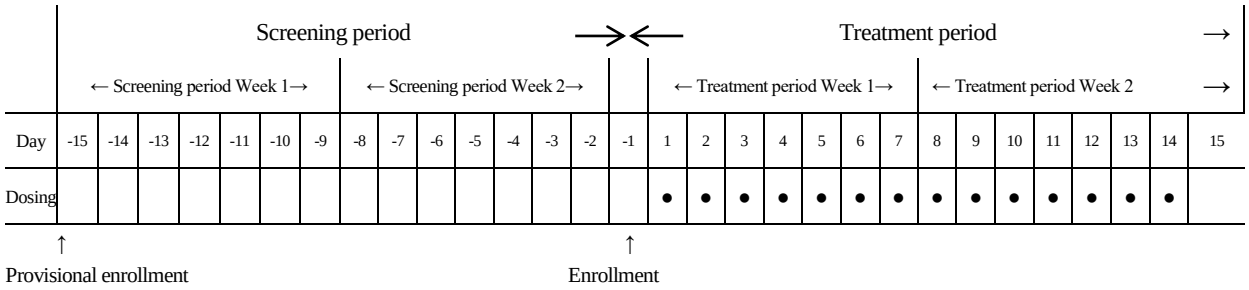
(1) Screening period

2 weeks

(2) Treatment period and method

After completing the 2-week screening period, the study drug will be started on the day after the day of enrollment.

AJG533 at 10 mg or placebo will be orally administered once daily approximately 30 minutes before breakfast for 14 days.



●: Study drug administration

[Rationales for the selection]

The rationales for the selection of the screening period, dosage and mode of administration, treatment method, and duration of treatment are presented below.

(i) Screening period

In order to include appropriate patients with chronic constipation and to perform the evaluation by unifying conditions for use of concomitant medications, etc. since the primary endpoint in this study is the change in bowel movement frequency from baseline to after the treatment, a 2-week screening period was chosen as a period for investigating the bowel movement frequency immediately before the start of the treatment.

(ii) Dosage

When 5, 10, or 15 mg of AJG533 or placebo was orally administered in patients with chronic constipation once daily about 30 minutes before breakfast for 14 days in the Phase II clinical study (AJG533/ET1), the primary endpoint, or the change in SBM from screening period Week 2 to treatment period Week 1 increased dose dependently: it was 2.60±2.89, 3.50±2.96, 5.66±4.15, and 5.59±3.51 in the placebo, 5 mg, 10 mg, and 15 mg groups, respectively. The change was comparable between the 10 mg and 15 mg groups and significantly larger in the 10 mg and 15 mg groups than the placebo group. Many of the other efficacy endpoints used as a secondary endpoint showed significant improvement in the 10 mg and 15 mg groups as compared to the placebo group. The mean stool consistency in the 10 mg group as evaluated using the Bristol Stool Form Scale was 2.25 ± 1.11, 4.00 ± 0.94, and 4.25 ± 1.03 at screening period Week 2, treatment period Weeks 1 and 2, respectively. These results show that stool consistency at Weeks 1 and 2 was close to 4, which indicates ideal stool consistency.

Safety evaluation showed that none of the groups had any serious or severe TEAE. Although 4 of the 43 subjects (9.3%) in the 5 mg group discontinued the study due to TEAEs, the rate of subjects who discontinued the study due to TEAEs was 2.6% (1/39) in the 10 mg group and 4.9% (2/41) in the 15 mg group, indicating that the rate was not dose dependent. Common TEAEs included abdominal pain and diarrhoea, which were assumed to be an extension of the pharmacologic effect. Their incidence was not correlated with the dose.

TRAEs that developed in more than 1 subject in any group were limited to abdominal pain, diarrhoea, and abdominal distension. There were no other characteristic adverse events of the study drug.

Based on these results, the recommended clinical dose of AJG533 was determined to be 10 mg.

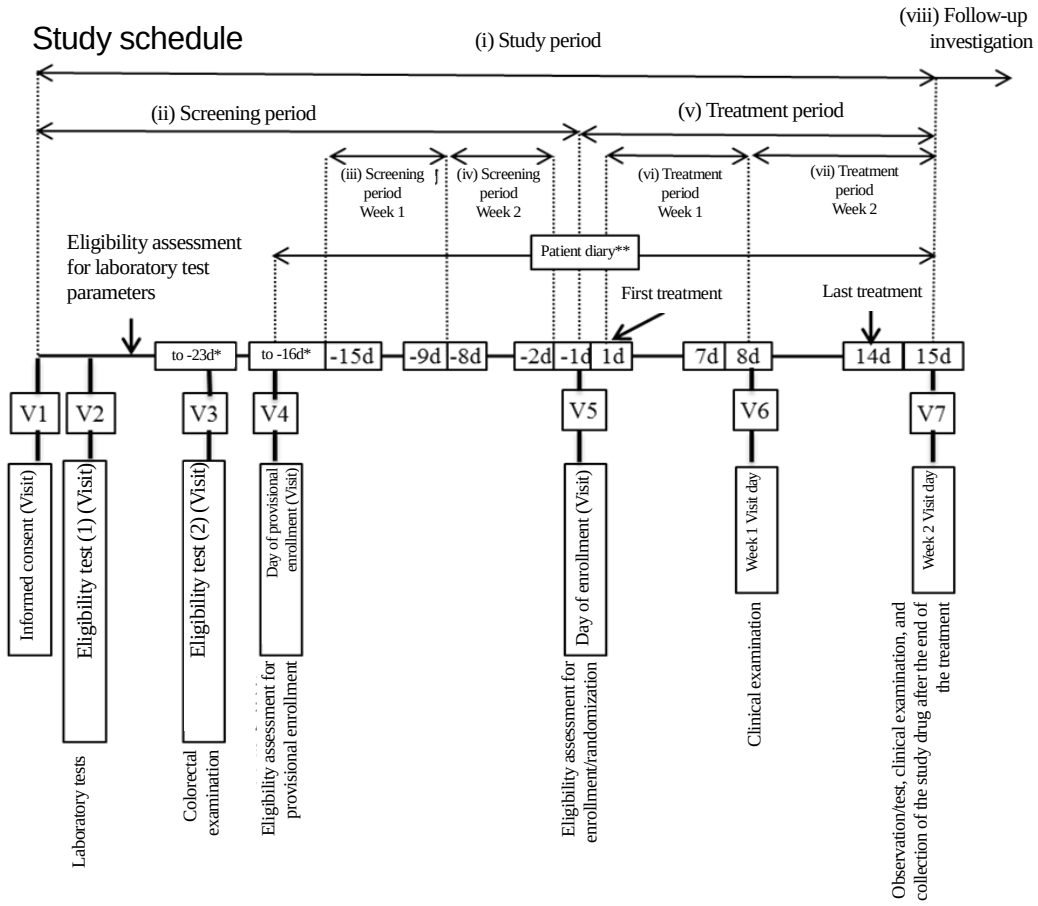
(iii) Treatment method and duration of treatment

It was confirmed in the Phase I clinical study in Japanese patients with chronic constipation (AJG533/CP1) that C_{max} was higher when AJG533 was administered without breakfast than when it was administered before breakfast. Because AJG533 directly acts in the intestinal tract and its blood concentration does not influence drug activity, the administration before breakfast associated with less transfer to blood was selected. It was confirmed in the subsequent Phase II clinical study in patients with chronic constipation (AJG533/ET1) that AJG533 as given once daily before breakfast improved constipation symptoms. Additionally, considering that the action mechanism of the drug is to inhibit bile acid reabsorption in the terminal ileum, administering AJG533 at breakfast, during which bile acid secretion in the digestive tract is estimated to be maximized in a day by the meal, is reasonable to ensure the maximum effect of the drug.

In the Phase II clinical study in patients with chronic constipation (AJG533/ET1), the change in SBM frequency in the 10 mg group from screening period Week 2 was almost constant in treatment period Weeks 1 and 2: it was 5.66 ± 4.15 and 5.16 ± 3.43, respectively. Similarly, there were no notable changes between treatment period Weeks 1 and 2for the secondary endpoints, change in SBM frequency at treatment period Week 2 from screening period Week 2. TEAEs were limited to those that developed as an extension of the pharmacological effect, such as abdominal pain and diarrhoea, and there were no other notable TEAEs. Most of TEAEs developed for the first time early after the start of treatment.

The primary endpoint of the present study is the change in SBM frequency from screening period Week 2 to treatment period Week 1. As described above, there was no major change in efficacy or safety between treatment period Weeks 1 and 2. It is therefore estimated that the sudden change in efficacy or safety at Week 3 or later is unlikely. Therefore, the duration of the treatment was set at 2 weeks, as in the Phase II clinical study (AJG533ET1).

7.1.4 Study schedule



*: Colonoscopy or radiographic contrast enema for reviewing eligibility will be performed by 8 days (Day -23) before the start of the screening period after obtaining informed consent and reviewing eligibility for the other items. After confirming eligibility, the patient will be provisionally enrolled by Day -16.

**: The use of laxatives and rescue medication and bowel movement status will be recorded from the day of the provisional enrollment. Eligibility will be assessed based on the records from Day -15 (0:00) to Day -2 (24:00) and the patient enrolled based on the results.

[Study period]

(i) Study period: Period from the receipt of the informed consent to the last observation/test (or the day when the contact specified in "12.2 Discontinuation procedure (4)" is made, if the observation/test at discontinuation cannot be performed)

(ii) Screening period: Period from the receipt of informed consent to enrollment

(iii), (iv) Screening period: Period from 0:00 on Day -15 to 24:00 on Day -2

(v) Treatment period: Although the study drug is administered for 14 days, the last observation of bowel movement status will be performed on Day 15 after the start of the study drug because bowel movement status at 14 days (336 hours) is investigated using the start of the study drug as the starting point.

[Follow-up investigation]

(viii) Follow-up investigation: When any TEAE does not recover to the normal or pre-treatment level at the test/observation at the end of the treatment period or at discontinuation, it will be followed with observations or tests. Such observations or tests (follow-up investigation) will be performed for 2 weeks after the end of the treatment period. However, even when any TEAE does not recover to the normal or

pre-treatment level, the follow-up may be terminated by documenting the reason for termination if the investigator or subinvestigator determines that no further follow-up investigation is necessary.

7.2 Study period (whole study)

October 30, 2015 to June 30, 2016 (scheduled date of the conclusion of the first study agreement to the date of the last visit of the last subject)

7.3 Endpoints

7.3.1 Primary Efficacy Variable

Change in SBM* frequency from screening period Week 2 to treatment period Week 1

*: Defecation without laxatives/enema or digital evacuation. Defecation within 24 hours after the use of a laxative or rescue medication will not be deemed as SBM in this study.

[Rationales for the selection]

Weekly bowel movement frequency was set as the efficacy endpoint because weekly bowel movement frequency is specified in the criteria for functional constipation in ROME III. Since it was considered inappropriate to include defecation induced by the use of a rescue medication in efficacy evaluation, SBM frequency was set. Because AJG533 was expected to quickly exert its effect, it was considered appropriate to specify the change in SBM frequency from screening period Week 2 to Week 1 after the start of the study treatment as the primary efficacy endpoint.

The change in SBM frequency from the screening period to Week 1 of the treatment period was also used as the primary endpoint in the clinical study of the approved and marketed drug, Lubiprostone 24 μg.

7.3.2 Secondary efficacy endpoints

- Change in SBM frequency from of the screening period to Week 2 of the treatment period
- Change in SBM frequency from the screening period to the treatment period
- Changes in CSBM* frequency from screening period Week 2 to treatment period Weeks 1 and 2

  *: SBM without sensation of incomplete evacuation
- Change in CSBM frequency from the screening period to the study drug treatment period
- Proportion of patients who experienced SBM within 24 hours/48 hours after the start of the study treatment
- SBM and CSBM responser ** rates at treatment period Weeks 1 and 2

**: A responder will be defined as a patient with weekly SBM or CSBM frequency of ≥ 3 and weekly SBM or CSBM frequency improved by ≥ 1 from Screening Period Week 2.

- Time to the first SBM
- Use of rescue medication

- Stool consistency as measured by Bristol Stool Form Scale
- Evaluation of weekly-based severity of constipation for treatment period Week 1 and Week 2

[Rationales for the selection]

This is a generally accepted endpoint used for recently developed constipation drugs and evaluated in the Phase II clinical study in patients with chronic constipation (AJG533/ET1).

7.3.3 Safety endpoints

- TEAEs
- Laboratory test parameters
- Vital signs

7.4 Method for allocation (randomization) of patients to treatment groups

Each subject confirmed eligible for the study will be assigned to an appropriate treatment group according to the allocation table prepared by the permuted block method.

7.5 Blinding Method

Blinding will be made using the study drug and comparator indistinguishable from it.

7.6 Maintenance of blinding

7.6.1 Method for maintaining blinding

(1) The study drug allocation manager will confirm that the study drug and comparator (placebo tablet) are indistinguishable from each other in appearance, weight, and package, and then blind the study drug according to the Inventory File and study drug allocation procedure.

(2) The study drug allocation manager, subject enrollment manager, and sponsor will take appropriate measures to ensure that the study treatment will be blinded for all those involved until key code breaking.

(3) At the key code breaking, the subject enrollment manager will confirm that Allocation Table and Inventory File provided by the study drug allocation manager are sealed, and that the study treatment is blinded throughout the study period. The subject enrollment manager will also check the cases of emergency unblinding with the EDC system, thereby confirming that blinding is maintained throughout the study period.

7.6.2 Preparation and storage of Inventory File and Allocation Table

The study drug allocation manager will provide the subject enrollment manager with the Inventory File immediately after the study treatment is blinded. The subject enrollment manager will prepare the Allocation Table. The subject enrollment manager will import the Inventory File and Allocation Table to the EDC system, seal them, and strictly store them until key code breaking. Emergency unblinding will be performed using appropriate EDC system functions. It is prohibited to unblind the study treatment without following the predetermined procedure (see "11.3 Emergency unblinding procedure").

7.6.3 Reporting of LDL-cholesterol and HDL-cholesterol measurements

The measurements of LDL-cholesterol and HDL-cholesterol after the start of the study drug treatment (at Visit 7 or discontinuation) will be disclosed to those involved in the study including the investigator, subinvestigator, and sponsor after key code breaking.

In addition, the measurements of LDL-cholesterol or HDL-cholesterol will be prohibited in tests except for this study during the period from enrollment to Visit 7 or until discontinuation except for medically compelling cases.

[Rationales for the selection]

AJG533 treatment may decrease LDL-cholesterol. Because comparing the change in LDL-cholesterol before and after the study drug treatment between subjects might unblind the study treatment, the disclosure of the measurements of LDL-cholesterol and HDL-cholesterol after the start of the study drug (at Visit 7 or discontinuation) was restricted until key code breaking and the measurement of LDL-cholesterol and HDL-cholesterol not specified in the protocol was prohibited after enrollment.

7.6.4 Key code breaking

The subject enrollment manager will break the key code after all electronic case report forms are prepared and data are fixed.

8. Selection and Enrollment of Subjects

The selection of prospective patients to whom the explanation for informed consent is given, acquisition of consent, confirmation of eligibility, and enrollment of subjects (those who provided consent) will be performed in accordance with the following procedures.

8.1 Screening

The investigator or subinvestigator will review whether or not prospective subjects meet the inclusion criteria and do not meet the exclusion criteria based on available information and select prospective subjects to whom the explanation for informed consent will be given. Screening is not intended for the final confirmation of eligibility,

but should be performed as accurate as possible to minimize patients found ineligible after informed consent is obtained. The investigator or subinvestigator will prepare a list of patients who receive the explanation for obtaining consent (subject screening list/register).

8.2 Informed consent

The investigator or subinvestigator will provide the explanation for informed consent in accordance with Sections "5.3 Timing and method for receiving informed consent" and "5.4 Points to consider for obtaining informed consent" of the protocol, and obtain written consent from prospective subjects.

8.3 Eligibility Assessment

The investigator or subinvestigator will thoroughly collect information necessary for confirming eligibility and make the final assessment of subject eligibility.

8.3.1 Tests for confirming eligibility

After receiving consent, the tests/observations specified in Section "10.2 Investigation items for eligibility assessment" will be performed.

For the exclusion criteria concerning diseases other than hepatic and renal disorders, subjects without a diagnosis of the diseases or subjects who have not received any therapeutic interventions for the diseases are determined eligible. In addition to the presence or absence of a diagnosis of liver disorder or renal disorder, laboratory values shall be investigated to confirm that the patient does not meet the exclusion criteria.

For patients who are not confirmed to have no organic lesion in the large intestine by colonoscopy or radiographic contrast enema performed within 5 years before provisional enrollment, colonoscopy or radiographic contrast enema shall be carried out for reviewing eligibility to confirm that they do not meet the exclusion criterion. Women of childbearing potential will receive pregnancy test to confirm that they are not pregnant.

The bowel movements status during the 2-week screening period in the patient diary will be checked before enrollment to confirm that applicable exclusion criteria are not met.

8.3.2 Eligibility Assessment

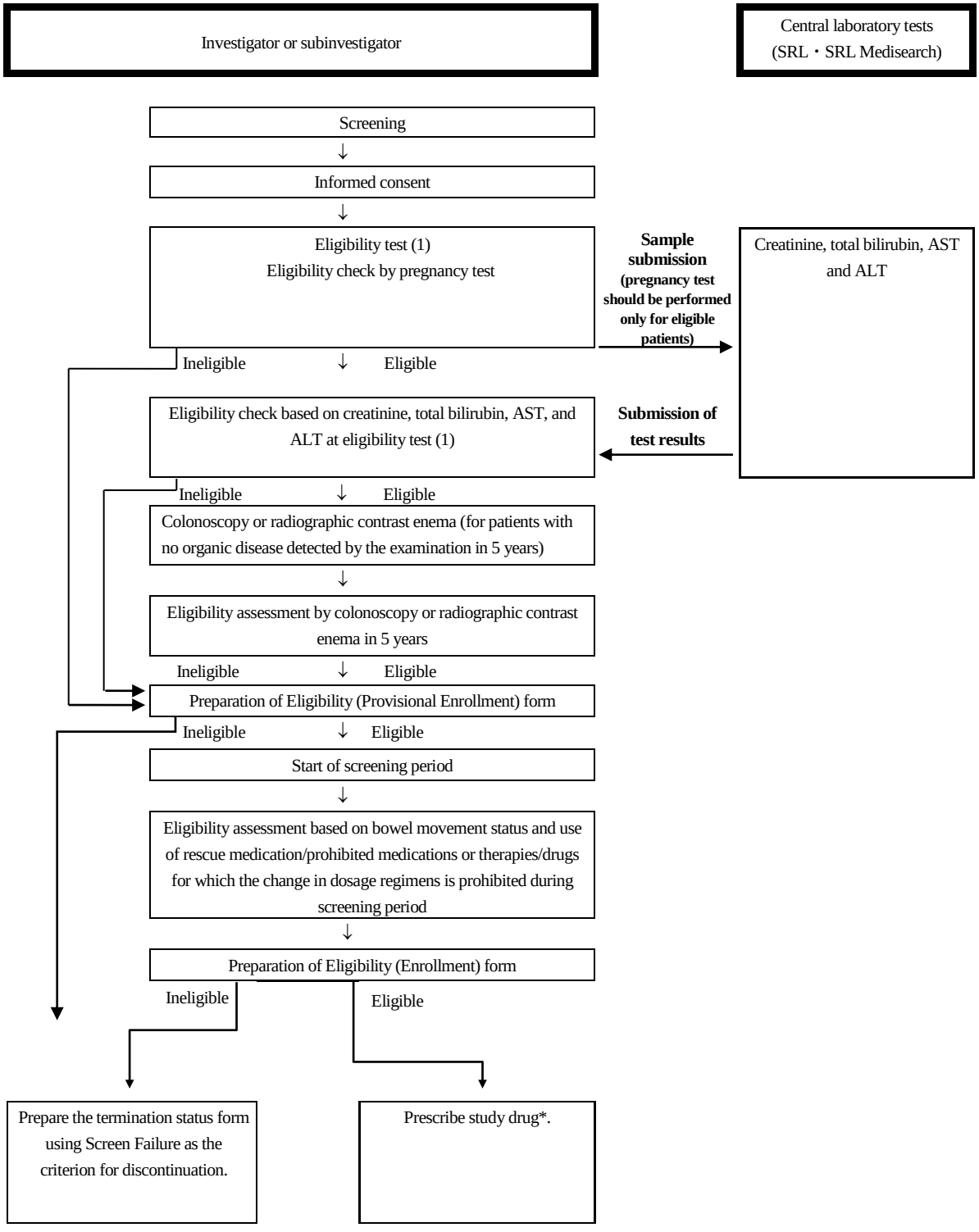
The investigator or subinvestigator will closely examine the test/observation results and make the final assessment of eligibility based on whether or not the prospective subject meets the inclusion criteria and does not meet the exclusion criteria.

8.4 Subject enrollment

The investigator or subinvestigator will enter and save enrollment-related information to the Eligibility form of eCRF at the review of laboratory test results of the eligibility test (1) and at the provisional enrollment and

enrollment for each subject who provides his/her consent. An alert will be displayed when the form is not completed or any parameter fails to meet the inclusion criteria or meets the exclusion criteria. The EDC system will select and display the drug number allocated to the subject from the drug numbers allocated to the study site. The investigator or subinvestigator will prescribe the study drug with the allocated drug number and enter the drug number to the subject screening list/register. If allocation is performed for an ineligible patient (with 1 or more alert), the system screen will display that the allocation criteria are not met and the system will not allocate any drug number. In that case, screening failure will be entered in the discontinuation criteria section.

[Subject enrollment flow chart]



* Patients evaluated as eligible will be enrolled.

8.5 Information to be reported to other departments/hospitals

If any subject is being treated by another physician, the investigator or subinvestigator will inform the physician, under the consent of the subject, of the participation of the subject in the clinical study and of concomitant drugs (therapies) to be restricted during the study.

8.6 Matters to be complied by patients

The investigator or subinvestigator will explain how to take the study drug, examination procedures, and how to handle the patient diary with special attention to the following instructions before prescribing the study drug.

(1) Subjects will visit the study site on predetermined days to undergo specified tests and examinations.

(2) Subjects will receive the study drug once daily at approximately 30 minutes before breakfast for 14 days from the next day of the day of enrollment, as instructed by the investigator or subinvestigator.

(3) Subjects may use the rescue medication when they have no bowel movement for at least 72 hours during the period from the start of the screening period to the last observation/test. Subjects will report the use of the rescue medication to the investigator, subinvestigator, or study collaborator in advance. Subjects may not use the rescue medication in 24 hours before and in 48 hours after the start of the study drug.

(4) Subjects will enter the following events to the patient diary as they arise.
Subjects will record the use of laxatives and rescue medication and bowel movement status from the day of the provisional enrollment.

(i) Events to be recorded from the provisional enrollment to the day before the start of the screening period

- Date and time of each bowel movement, stool consistency (Bristol Stool Form Scale), and sensation of incomplete evacuation
- Date and time of the use of laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and/or intestinal tract cleaning agents (including commerically available drugs)

(ii) Events to be recorded after the start of the screening period

- Date and time of each bowel movement, stool consistency (Bristol Stool Form Scale), and sensation of incomplete evacuation
- Date and time of taking the first dose of the study drug
- Date and time of using the rescue medication
- Severity of constipation (weekly) and date of evaluation

(5) That the patient diary should be brought to the hospital at each visit

(6) Subjects will bring any unused study drugs at Visit 7 or at discontinuation.

(7) Subjects will bring any unused rescue medication at Visit 5 or 7 or at discontinuation.

(8) Subjects should not change lifestyle habits such as a dietary life and exercise patterns during the study period.

(9) Subjects will report the use of drugs including those prescribed at other hospitals or purchased at pharmacies, supplements, and therapies to the investigator, subinvestigator or study collaborator.

(10) Subjects will comply with the instructions on the prohibited medications/therapies or drugs for which changing the dosage regimens is prohibited, as given by the investigator or subinvestigator.

(11) Women of childbearing potential will practice contraception using appropriate methods under the instructions of the investigator or subinvestigator during the period from informed consent to the last observation/test.

9. Concomitant Therapies

All used drugs and concomitant therapies will be recorded in the eCRF.

9.1 Prohibited Medications in Concomitant Therapies

9.1.1 Prohibited medications and therapies

The use of the following medications and therapies that may affect this study will be prohibited during the period from the start of the screening period to the last observation/test (for withdrawals, until the completion of observation, tests and investigations performed at discontinuation). However, bisacodyl suppositories 10 mg may be used based on the rules set forth in Section 9.1.2 Restricted concomitant medications (therapies), (1) Rescue medication.

The use of laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and intestinal tract cleaning agents will be investigated and recorded from 3 days before to the day before the start of the screening period. The use of the following drugs and therapies will be investigated and recorded from the start of the screening period to the last observation/test.

- Different types of laxatives (e.g. magnesium oxide preparations, sodium picosulfate, and sennoside)
- Chinese herbal medicines indicated for constipation (e.g., daio-kanzo-to, choi-joki-to, and dai-saiko-to)
- Drugs for the treatment of irritable bowel syndrome (IBS) (e.g., ramosetron hydrochloride, polycarbophil calcium, and trimebutine maleate)
- 5-HT_3 antiemetics
- Enteric movement accelerating agents (e.g., mosapride citrate, metoclopramide, and domperidone)
- Macrolide antibiotics (e.g., erythromycin, roxithromycin, and azithromycin)
- Antidepressants, antipsychotics, antianxiety agents, and tranquilizers (excluding those used for the treatment of insomnia)
- Anticholinergic drugs (excluding topical agents)
- Nonsteroidal anti-inflammatory drugs (excluding topical agents)
- Supplements, etc. for improving constipation

- Enema and intestinal lavage
- Intestinal tract cleaning agents
- Drugs affecting the amount of bile acid (e.g., colestimide and ursodeoxycholic acid)
- Other study drugs
- Constipation therapies such as biofeedback
- Digital evacuation

[Rationales for the selection]

These are prohibited because of the potential effect on the efficacy evaluation of AJG533.

9.1.2 Restricted concomitant medications (therapies)

Restricted medications are described below.

(1) Rescue medication

Bisacodyl suppositories 10 mg prescribed for this study may be used under the following conditions.

Bisacodyl suppositories 10 mg may be utilized as a rescue medication only when there is no bowel movement for at least 72 consecutive hours from the start of the screening period to the last observation/test. When there is no bowel movement even after using one dose of bisacodyl suppositories 10 mg, the investigator will determine whether or not the subject can continue the study. When there is no bowel movement for at least 72 consecutive hours after having a bowel movement following the use of the rescue medication, one dose of the rescue medication may be used again.

However, the use of the rescue medication within 24 hours before and 48 hours after the start of the study treatment is prohibited.

The date and time of the use of the rescue medication from the screening period to the last observation/test, if any, will be recorded in the eCRF.

[Rationales for the selection]

Since this is a clinical study of a drug for the treatment of chronic constipation, various types of laxatives are included in the prohibited concomitant medications. Nonetheless, because the screening period is as long as 14 days and this is a placebo-controlled study, the use of the rescue medication is permitted in consideration of the possibility of marked worsening of constipation symptoms during the study. However, the use of the rescue medication within 24 hours before and 48 hours after the start of the treatment is prohibited because it may greatly affect the efficacy evaluation of AJG533.

(2) Drugs of which changes in dosage regimens are prohibited

When any of the following drugs is used at the time of receiving the informed consent and needs to be continued during the study period, the change in its dosage regimen is prohibited during the study period from the time of the informed consent to the last observation/test.

For the drugs of which changing the dosage regimen is prohibited, the name of the drug used from the day of the informed consent to the last observation/test, period of use, and dosage regimen will be investigated and recorded.

- Hypnotics for the treatment of insomnia
- Calcium antagonists
- Iron preparations

[Rationales for the selection]

The change in the dosage regimen of hypnotics, calcium antagonists, or iron preparations was set because it might affect the efficacy evaluation of AJG533.

9.1.3 Guidance for subjects

Subjects will be instructed to avoid greatly changing their dietary habits and exercise patterns from the start of the screening period to the end of the study (or to the day of the contact specified in "12.2 Discontinuation procedures (4)" if the observations/tests at the discontinuation cannot be performed).

10. Observation, Test and Investigation Items and Schedule

10.1 Outline of observation, test and investigation items and schedule

The observation, test and investigation items in this study are presented in Tables 10-1a and 10-1b. The investigation items and schedule are presented in Table 10-2. The acceptable ranges at each Visit are presented in Table 10-3.

Table 10-1a Observation, test and investigation items related to eligibility confirmation

	Contents
Subject background	Date of birth, inpatient/outpatient status, medical history (including surgical history)*1 and complications *2
Observation/investigation items	Subjective symptoms, objective findings, and use of concomitant medications
Vital signs	Blood pressure (systolic and diastolic blood pressures) and pulse rate
Hematology	White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count
Blood biochemistry	Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin, urea nitrogen, creatinine, uric acid, LDL-cholesterol, HDL-cholesterol, total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl, and Ca
Urinalysis (qualitative)	Glucose, protein, urobilinogen, occult blood, pregnancy test (women only) *3
Items related to bowel movements	Bowel movement status (date and time, stool consistency as measured by Bristol Stool Form Scale) and use of laxatives*4 and rescue medication
Colorectal examination (colonoscopy or radiographic contrast enema)	Colorectal examination will be performed by 8 days before the start of the screening period for subjects for whom the absence of organic disease has not been confirmed with the examination in 5 years before the provisional enrolment.

*1: Medical history: Medically important symptoms or diseases (e.g., those related to the exclusion criteria) that developed within 5 years and cured before the start of the study treatment All laparotomy histories will be recorded.

*2: Complications: Diseases present or symptoms or signs observed at the start of the study treatment and considered as clinically important

*3: Pregnancy test will be performed at Visit 2 only in women of childbearing potential.

*4: Laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and intestinal tract cleaning agents (including commercially available drugs) used on the day before the start of the screening period will also be investigated.

Table 10-1b　Observation, test, and investigation items

	Contents
Subject background	Gender, birth date, race, medical history (including surgical history)[*1], complications[*2], and presence or absence of constipation-predominant IBS
Observation/investigation items	Study drug compliance, use of concomitant medications, subjective symptoms, objective findings, and study completion
Vital signs	Height, body weight, blood pressure (systolic and diastolic blood pressures), and pulse rate
Hematology	White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count
Blood biochemistry	Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin, urea nitrogen, creatinine, uric acid, LDL-cholesterol[*3], HDL-cholesterol[*3], total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl, and Ca
Urinalysis (qualitative)	Glucose, protein, urobilinogen and occult blood
Items related to bowel movements	Bowel movement status (date and time, stool consistency as measured by Bristol Stool Form Scale, and sensation of complete evacuation), constipation severity assessment (date of assessment and severity assessment), and use of the rescue medication

*1: Medical history: Medically important symptoms or diseases (e.g., those related to the exclusion criteria) that developed within 5 years and cured before the start of the study treatment All laparotomy histories will be recorded.

*2: Complications: Diseases present or symptoms or signs observed at the start of the study treatment and considered as clinically important

*3: LDL-cholesterol and HDL-cholesterol at Visit 7 and at discontinuation will be investigated in a blinded manner.

Table 10-2 Investigation items and schedule

Study period

Screening period

	Informed consent	Eligibility test (1)	Confirmation of laboratory test eligibility assessmen	Eligibility test (2) *2	Day of provisional enrollment		Day of enrollment	Starting day of study treatment		Week 1 Visit day		Week 2 Visit day /at disconti-nuation
visit	1	2		3	4		5			6		7
Test/observation time points (day)				to -23	to -16		-1	1		8	14	15
Informed consent	○											
Enrollment and confirmation of eligibility assessment results *1			○		○		○					
Medical history *3, complications *4, presence or absence of constipation-predominant IBS							○					
Clinical examination		○		○	○		○			○		○
TEAEs								←				→
Subject background (gender, birth date, inpatient/outpatient status, and race)		○										
Vital signs (height)							○					
Vital signs (body weight)							○					○
Vital signs (blood pressure, pulse rate)		○					○					○
Colonoscopy or radiographic contrast enema *2				○								
Study drug compliance								←				→
Use of rescue medication (patient diary)						←						→
Use of laxatives *5 (patient diary)					↔							
Use of concomitant medications (patient diary)	←											→
Bowel movement status *6 (patient diary)					←							→
Constipation severity evaluation (patient diary)							○			○		○
Hematology		○					○					○
Blood biochemistry (i) (excluding LDL-C and HDL-C)		○					○					○
Blood biochemistry (ii) (LDL-C and HDL-C)		○					○					○
Pregnancy test *7		○										
Urinalysis (qualitative)		○					○					○
Blood collection volume (m_)		7mL					7mL					7mL
Urine collection volume (m_)		10mL					10mL					10mL

○: To be performed

*1: Confirmation of laboratory test eligibility assessment results: Eligibility assessment results will be confirmed for renal and hepatic functions in the eligibility test (1). If eligible, the eligibility test (2) or provisional enrollment will be implemented. If ineligible, the patient will be registered as ineligible.

*2: Colonoscopy or radiographic contrast enema will be performed by 8 days before the start of the screening period (Day -23) for patients for whom the absence of organic disease has not been confirmed with the examination in 5 years before the provisional enrolment.

*3: Medical history: Medically important symptoms or diseases that developed within 5 years and cured before the start of the study treatment. However, all laparotomy histories will be recorded.

*4: Complications: Diseases present or symptoms or signs observed at the start of the study treatment and considered as clinically important

*5: Laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and intestinal tract cleaning agents（including commercially available drugs) used from the day of provisional enrollment to the day before the start of the screening period will be recorded.

*6: The bowel movement status from the day of provisional enrollment will be recorded.

*7: To be performed only in women of childbearing potential.

Table 10-3 Acceptable range at each visit

Visit	5	6	7 *1
No. of days from the start of study treatment	- 1 day	8 days	15 days
Acceptable range (day)	-1	8 to 10	15 to 17

*1: In principle, the tests at discontinuation will be performed within 2 weeks after the date of the last dose.

Note: Any deviation from these acceptable ranges is considered as a protocol deviation. The acceptable ranges for data adoption are presented in Section "13.3 Data Handling."

10.2 Investigation items related to eligibility assessment

The eligibility of patients who provide informed consent will be assessed and eligible patients will be provisionally enrolled by Day -16 before the start of the study treatment. The screening period will be started to assess eligibility based on bowel movements from Days -15 (0:00) to -2 (24:00) in the patient diary. Patients assessed as will be enrolled.

<<Before provisional enrollment>>

(1) Laboratory test (eligibility test (1)): Visit 2

Laboratory tests related to the inclusion and exclusion criteria will be performed.

The following parameters other than those of pregnancy test will be performed at SRL, Inc.

Hematology:	White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count
Blood biochemistry:	Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin, urea nitrogen, creatinine, uric acid, LDL-cholesterol, HDL-cholesterol, total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl, and Ca
Urinalysis (qualitative):	Glucose, protein, urobilinogen and occult blood
Urinalysis (qualitative):	Pregnancy test (women of childbearing potential only)

Samples will be collected by SRL Medisearch Inc. or SRL, Inc. and sent to SRL, Inc. Samples will be refrigerated before collection. Measured samples will be stored at SRL Medisearch Inc. until case fixation.

Measurement results will be provided as electronic data to the study site and sponsor and sent as test slips from SRL Medisearch to the study site by mail.

Women of childbearing potential will collect an urine sample at Visit 2, which will be used for the dipstick pregnancy test at the study site.

(2) Colonoscopy or radiographic contrast enema (eligibility test (2)): Visit 3

Colonoscopy or radiographic contrast enema for confirming eligibility will be performed for patients for whom the absence of organic disease has not been confirmed with the examination in 5 years before the provisional enrolment. In that case, colonoscopy or radiographic contrast enema will be performed by 8 days

before the start of the screening period (Day -23) after the eligibility for the other items is confirmed after informed consent.

(3) Other investigation items

Subject background (gender and inpatient/outpatient status) and items related to the inclusion and exclusion criteria other than laboratory test parameters (including medical history and complications) will be investigated and recorded in eCRF.

<< Before enrollment >>

(1) Items related to bowel movements from 3 days to the day before the start of the screening period

The items related to bowel movements (date and time and stool consistency as measured by the Bristol Stool Form Scale) will be investigated for 3 days from 3 days to the day before the start of the screening period (0:00 on Day -18 to 24:00 on Day -16) and recorded in the eCRF.

The items related to bowel movements will be recorded in the patient diary by subjects from the day of provisional enrollment.

(2) Use of laxatives on the day before the start of the screening period

The use (date and time) of laxatives, Chinese herbal medicines indicated for constipation, supplements, etc. for improving constipation, enema, intestinal lavage, and intestinal tract cleaning agents (including commercially available drugs) on the day before the start of the screening period (0:00 on Day -16 to 24:00 on Day -16) will be investigated and recorded in the eCRF.

The use of laxatives will be recorded in the patient diary by subjects from the day of provisional enrollment.

(3) Items related to bowel movements and use of rescue medication during screening period

The items related to bowel movements (date and time and stool consistency as measured by the Bristol Stool Form Scale) and use of the rescue medication for the 2-week screening period (0:00 on Day -15 to 24:00 on Day -2) will be investigated and recorded in eCRF. The items related to bowel movements and use of the rescue medication will be recorded in the patient diary by subjects after the end of the provisional enrollment.

(4) Use and implementation of prohibited concomitant drugs and therapies during screening period

The use and implementation of prohibited concomitant drugs and therapies during the 2-week screening period (0:00 on Day -15 to 24:00 on Day -2) will be investigated and recorded in the eCRF.

10.3 Subject background (investigation items for subject characteristics)

[Investigation time point: Visits 2 and 5]

The following items will be investigated and recorded in the eCRF.

Investigation items at Visit 2

(i) Gender

(ii) Birth date

(iii) Race

Investigation items at Visit 5

(i) Medical history (including a history of laparotomy) [BEFORE]

(ii) Complications [DURING/AFTER]

(iii) Vital signs (height)

(iv) Presence or absence of constipation-predominant IBS (whether or not the diagnostic criteria for irritable bowel syndrome [IBS]* are met)

*: Abdominal pain or abdominal discomfort occurs on at least 3 days per month during the last 3 months and meets at least 2 of the following criteria:

1. Relieved with defecation
2. Starts as a change in bowel movement frequency.
3. Starts as a change in stool form (appearance).

The symptom has to develop at least 6 months before diagnosis and meets the criteria for the last 3 months.
Abdominal discomfort means an unpleasant sensation other than abdominal pain.

10.4 Use of study drug

[Investigation time points: Visits 5 and 7 (or at discontinuation)]

The quantities of the study drug prescribed and collected and date and time of the first dose of the study drug will be investigated and recorded in the eCRF. The patient will record the date and time of the first dose of the study drug in the patient diary.

10.5 Use and implementation of concomitant drugs and therapies

[Investigation period: from the day of the informed consent to the day of the last observation/test (or discontinuation)]

The use of concomitant drugs (name of drugs, reasons for the concomitant use, and period of treatment) after the day of informed consent will be investigated and the information obtained after the start of the study treatment will be recorded in the eCRF for drugs other than prohibited concomitant drugs or drugs for which the

concomitant use is restricted. The use until the date of the contact specified in "12.2 Discontinuation procedure (4)" will be recorded in the eCRF if the observations/tests at discontinuation cannot be performed.

10.6 Use of rescue medication

[Investigation period: From the starting day of the screening period to the day of the last observations/tests (or discontinuation)]

The date and time of using the rescue medication from the starting day of the screening period to the day of the last observations/tests (or discontinuation) will be investigated and recorded in the eCRF. The use of the rescue medication will be recorded in the patient diary by subjects.

10.7 Efficacy investigation items

[Investigation period: From the day of provisional enrollment to the day of the last observations/tests (or discontinuation)]

Endpoints related to bowel movements during the treatment period (date and time, stool consistency as measured by Bristol Stool Form Scale, sensation of complete evacuation, and the severity of constipation) shall be investigated and recorded in the eCRF.

(1) Bowel movement status

The patient shall assess each bowel movement and record it in the patient diary.

(i) Date and time of defecation

(ii) Stool consistency (assessment on a 7-grade scale) based on the Bristol Stool Form Scale

1: Separate hard lumps, like nuts (hard to pass)

2: Sausage shaped, but lumpy

3: Like a sausage, but with cracks on its surface

4: Like a sausage or snake, smooth and soft

5: Soft blobs with clear cut edges (passed easily)

6: Fluffy pieces with ragged edges, a mushy stool

7: Watery, no solid pieces, entirely liquid

(iii) Sensation of complete evacuation (Level of sensation of incomplete evacuation)

0: Sensation of incomplete evacuation absent

1: Sensation of incomplete evacuation present

(2) Severity of constipation (assessment on 5-grade scale)

Subjects will evaluate the severity of constipation every week and record it in the patient diary.

0: None (no symptom of constipation at all)

1: Mild (minor symptoms of constipation)

2: Moderate (non-major symptoms of constipation)

3: Severe (severe constipation and have difficulty defecating or little sensation of evacuation)

4: Very severe (intractable constipation, almost no defecation or almost no sensation of evacuation)

10.8 Safety investigation items

(1) Laboratory tests

The following measurements will be performed at SRL, Inc.

Hematology:	White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count
Blood biochemistry:	Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin, urea nitrogen, creatinine, uric acid, LDL-cholesterol, HDL-cholesterol, total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl, and Ca
Urinalysis (qualitative):	Glucose, protein, urobilinogen and occult blood

Samples will be collected by SRL Medisearch Inc. or SRL, Inc. and sent to SRL, Inc. Samples will be refrigerated before collection. Measured samples will be stored at SRL Medisearch Inc. until case fixation. Measurement results will be provided as electronic data to the study site and sponsor and sent as test slips from SRL Medisearch to the study site by mail. However, the measurement results of LDL-cholesterol and HDL-cholesterol after the start of the study treatment (at Visit 7 or at discontinuation) will be disclosed to the persons involved in the study including the investigator, subinvestigator, and sponsor after key code breaking.

(i) Hematology

White blood cell count, red blood cell count, hemoglobin content, hematocrit, and platelet count

[Investigation time points: Visits 2, 5, and 7 (or at discontinuation)]

Two mL of whole blood will be drawn in a separately distributed EDTA-2K blood collection tube. Subject ID code and date of blood collection will be documented on its label. The sample will be refrigerated until collection.

(ii) Blood biochemistry (i)

Total protein, albumin, AST (GOT), ALT (GPT), γ-GTP, ALP, LDH, LAP, total bilirubin , urea nitrogen, creatinine, uric acid, total cholesterol, triglyceride, CRP, CK (CPK), Na, K, Cl and Ca

[Investigation time points: Visits 2, 5, and 7 (or at discontinuation)]

Blood collection for blood biochemistry (i) and (ii) will be performed using one serum separator blood collection tube and processed as follows:

Five mL of whole blood will be drawn in a separately distributed serum separator blood collection tube. Separated serum will be dispensed in a designated container, and subject ID code and date of blood collection

will be documented on its label. The sample will be refrigerated until collection. Only after the administration of the study drug (at Visit 7 or discontinuation), serum will be dispensed in two tubes, and subject ID code and the date of blood collection will be documented on each label. The samples will be refrigerated until collection.

(iii) Blood biochemistry (ii)

LDL-cholesterol and HDL-cholesterol

[Investigation time points: Visits 2, 5, and 7 (or at discontinuation)]

Blood collection for blood biochemistry (i) and (ii) will be performed using one serum separator blood collection tube and processed as follows:

Five mL of whole blood will be drawn in a separately distributed serum separator blood collection tube. Separated serum will be dispensed in a designated container, and subject ID code and date of blood collection will be documented on its label. The sample will be refrigerated until collection. Only after the administration of the study drug (at Visit 7 or discontinuation), serum will be dispensed in two tubes, and subject ID code and the date of blood collection will be documented on each label. The samples will be refrigerated until collection.

The measurements of LDL-cholesterol and HDL-cholesterol after the start of the study drug treatment (at Visit 7 or discontinuation) will be disclosed to those involved in the study including the investigator, subinvestigator, and sponsor after key code breaking.

(iv) Urinalysis (qualitative)

Glucose, protein, urobilinogen and occult blood

[Investigation time points: Visits 2, 5, and 7 (or at discontinuation)]

Ten mL of urine will be collected in a separately distributed poly Spitz tube, and subject ID code and date of collection will be documented on its label. The sample will be refrigerated until collection.

(v) Pregnancy test (women of childbearing potential only)

[Investigation time point: Visit 2]

Urine will be collected and tested using a separately distributed pregnancy test dipstick.

[Rationales for the selection]

These observation/test items include general items to allow the selection of subjects who can participate in the study and those to allow the selection of subjects with chronic constipation with reference to the diagnostic criteria of functional constipation in Rome III. Generally measured laboratory test parameters are included as a safety index, considering the results of the Phase I and II studies. The measurement of LDL-cholesterol is included because the study drug may influence blood LDL-cholesterol through its effect to inhibit the resorption of bile acid in the ileum. Because the comparison of the change before and after the study treatment may break blinding, the disclosure of the LDL-cholesterol and HDL-cholesterol measurement results after the start of the study treatment (at Visit 7 or discontinuation) is restricted before code breaking and the measurement of

LDL-cholesterol and HDL-cholesterol on the days not specified by the protocol is prohibited during the treatment period.

(2) Vital signs

Blood pressure (systolic and diastolic blood pressure) and pulse rate will be measured at Visits 2, 5 and 7. Body weight will be measured at Visits 5 and 7. Vital signs measured at Visits 5 and 7 will be recorded in the eCRF.

[Rationales for the selection]

Generally measured vital sign parameters are included.

(3) TEAEs

TEAEs will be assessed in accordance with the criteria in Items (i) to (iv). The following information on TEAEs will be investigated and recorded in the eCRF: TEAE term, date of onset, seriousness, severity, causal relationship with the study drug, actions taken for the study drug, and outcome (date of the outcome for "Recovered" or "death").

The TEAE term will be recorded based on diagnosis. If no diagnosis is available for a TEAE, its sign or symptom will be used to represent it. The outcome at the last observation/test will be recorded. For TEAEs for which the outcome at the last observation/test is not "Recovered," the outcome information in the eCRF will be revised if it is changed in 2 weeks. The situation at the contact specified in "12.2 Discontinuation procedure (4)" will be recorded in the eCRF if the last observation/test cannot be performed.

(i) Definitions of terms

Definition of TEAE

A treatment-emergent adverse event (TEAE) is defined as any unfavorable and unintended disease or its sign (including an abnormal laboratory value) in a subject who received the study drug, irrespective of the causal relationship with the study drug.

Guidance on the "Ministerial Ordinance on Good Clinical Practice," Notification No. 1228-7 of the Evaluation and Licensing Division (ELD), Pharmaceutical and Food Safety Bureau (PFSB) dated December 28, 2012

Symptoms, signs, or clinically relevant, unfavorable changes in laboratory test parameters that newly developed or worsened from the start of the study drug to the last observation/test point or, if the observations/tests at discontinuation cannot be performed, to the day of the contact specified in "12.2 Discontinuation procedure (4)" will be handled as a TEAE in this study.

TEAEs include:

- Newly developed symptoms or diseases (unfavorable events caused by drug-drug interactions or

overdosing)

- Changes in test values (e.g., laboratory tests and electrocardiography [ECG]) that are determined to be "clinically relevant, unfavorable changes." In other words, these include cases for which the investigator or subinvestigator requires therapeutic measures or medical practice due to the worsening of test values, etc. or the investigator or subinvestigator judges that the change is beyond the range of physiological fluctuations of the subject.
- Exacerbation of the target disease
- Worsening of complications
- Accidents

The followings will not be deemed as TEAEs:

- Lack of efficacy
- Surgery or intervention pre-scheduled before the start of the study treatment
- Abnormal test values (e.g., laboratory tests and ECG) associated with TEAEs

Definition of "TESAE"

Treatment-emergent serious adverse events (TESAEs) refer to any of the following cases that
(i) Death
(ii) Life-threatening event (when the patient is at risk of death when the event occurs*)
(iii) Hospitalization or prolongation of existing hospitalization (including the case where the subject is admitted to the hospital for treatment but receives no particular therapeutic intervention*)
(iv) Disability (associated with dysfunction interfering with activities of daily living* or resulting in persistent or significant disability/dysfunction*)
(v) Serious essentially comparable with those specified in the above (i) to (iv) (clinically important events requiring therapeutic interventions to prevent the outcomes specified in (i) to (iv) and (vi)
(vi) Congenital anomaly (the case where birth defect due to the exposure to a drug before or during pregnancy is suspected*)

Source: Article 273 of the Enforcement Regulations of "Act on Securing Quality, Efficacy and Safety of Pharmaceuticals, Medical Devices, Regenerative and Cellular Therapy Products, Gene Therapy Products, and Cosmetics"

"Clinical Safety Data Management: Definitions and Standards for Expedited Reporting"

Notification No. 227 from the Director of the ELD, Pharmaceutical Affairs Bureau (PAB) dated March 20, 1995

*: "Revision of the Q&A on Reporting of Adverse Reactions"

Administrative Note of the Safety Division (SD) and ELD, PFSB dated February 26, 2014

Definition of "adverse drug reaction"

Adverse drug reaction

Adverse drug reaction is defined as follows for the study drug (excluding commercially available drugs used as a comparator)

Adverse drug reaction is defined as any harmful, unintended reaction to the study drug (including laboratory test abnormality), irrespective of the dose. This means that adverse drug reaction is a reaction for which the causal relationship with the study drug is at least reasonably possible and cannot be ruled out. The causal relationship with the study drug can be assessed, considering the following factors: resolution of the reaction after the drug is withdrawn, recurrence of the reaction after the drug is resumed, causal relationship that has been established for the study drug or related drugs, absence of confounding risk factors, consistency with exposure level and period, accurate medical history that definitely supports the involvement of the study drug, and absence of reasonable possibility that the reaction results from concomitant drugs.

Adverse drug reaction is defined as follows for commercially available drugs.

Adverse drug reaction is defined as any harmful, unintended reaction (including laboratory test abnormality) to the drug in an usual dose range used to prevent, diagnose, or treat disease or adjust physiological functions. This means that adverse drug reaction is a reaction for which the causal relationship with the drug is at least reasonably possible and cannot be ruled out.

Guidance on the "Ministerial Ordinance on Good Clinical Practice," Notification No. 1228-7 of the Evaluation and Licensing Division (ELD), Pharmaceutical and Food Safety Bureau (PFSB) dated December 28, 2012

(ii) Severity of TEAEs

Table 10-4 Assessment criteria for severity of TEAEs

Category	Assessment criteria
[MILD]	An event that does not interfere with activities of daily living (e.g., Even if a TEAE is present, being able to go to work or do household chores)
[MODERATE]	An event that interferes with activities of daily living (e.g. a TEAE practically prevents performing work or household chores)
[SEVERE]	An event that prevents activities of daily living

(iii) Causal relationship with study drug

Table 10-5 Assessment criteria for causal relationship with study drug

Category	Assessment criteria
[RELATED]	The case not corresponding to "not related"
[NOT RELATED]	An event considered not temporally related (including the clinical course after the withdrawal of the study drug). Or event considered to result from other factors including the primary disease, complications, concomitant drugs, and concomitant treatments

(iv) Outcome

Table 10-6 Assessment criteria for outcome

Category	Assessment criteria
[RECOVERED]/ [RESOLVED]	• Disappearance or resolution of symptoms or findings • Normalization of test values or recovery to the baseline level before the study treatment
[RECOVERING] / [RESOLVING]	• Severity reduced by at least one level • Substantial disappearance of symptoms or findings • Improvement of test values that are not "normalized or recovered to baseline levels"
[NOT RECOVERED] / [NOT RESOLVED]	• No change in symptoms, findings, or test values • Worsening of symptoms, findings, or test values on the last observation day, as compared to the level at onset • Irreversible congenital anomaly
[RECOVERED / RESOLVED WITHSEQUELAE]	• Dysfunction that interferes with activities of daily living
[FATAL]	• Death that is found to be directly related to the TEAE The term, "directly related," means that the TEAE caused death or clearly contributed to death.
[UNKNOWN]	• The clinical course after the day of onset could not be followed as specified in the protocol due to transfer to another hospital or moving.

(Note) The outcome of a TEAE assessed (judged or estimated) as not being the direct cause of death in the same patient will be recorded as "Unknown," not "Death."

(v) Actions taken to administration of study drug

Table 10-7 Assessment criteria for actions to administration of study drug

Category	Assessment criteria
[DOSE INCREASED]	• The dose of the study drug was increased as an action to the TEAE.
[DOSE NOT CHANGED]	• The dose was not changed after the TEAE developed. • The study drug was discontinued for other reasons when the TEAE developed. • The TEAE was found during the tests/observations performed after discontinuance.
[DOSE REDUCED]	• The dose of the study drug was reduced as an action to the TEAE.
[DRUG INTERRUPTED]	• The study drug was temporarily discontinued as an action to the TEAE and then resumed.
[DRUG WITHDRAWN]	• The study drug was permanently discontinued as an action to the TEAE.
[NOT APPLICABLE]	• The study drug was completed or permanently discontinued when the TEAE developed.
[UNKNOWN]	• The subject was lost to follow-up and the clinical course after the development of the TEAE was unknown.

(Note) The decision by the subject is included.

10.9 Study completion

Study completion (date of the last dose of the study drug and premature discontinuation) will be investigated and recorded in the eCRF for all the subjects confirmed eligible for the study by the eligibility assessment results at the provisional enrollment (Visit 4). For subjects who prematurely discontinued the participation in the study, the discontinuation will be recorded in the eCRF according to "12.2 (5) Discontinuation procedure."

11. Ensuring Subject Safety

11.1 Basic matters

The investigator and subinvestigators will perform the study, paying attention to ensuring subject safety. The investigator and subinvestigators will ensure subject safety by reducing the dose of the study drug or temporarily or permanently discontinuing the study treatment and providing appropriate medical care for subjects experiencing TEAEs.

11.2 Actions to TEAEs

The investigator and subinvestigator will handle any symptom or sign that develops or worsens after the start of the study drug or any clinically relevant, unfavorable change in a laboratory test parameter that occurs after the start of the study drug as a TEAE (see "10.8 (3) Adverse Events") and, in principle, follow it until it is resolved to the normal or pre-treatment level.

11.2.1 Actions taken to TEAEs

(1) The director of the study institution and sponsor will give appropriate medical care to TEAEs that occur in subjects during or after the study.

(2) The investigator or subinvestigator will inform subjects when any TEAE that requires medical treatment occurs.

(3) The investigator or subinvestigator will give appropriate medical care to each TEAE. The investigator or subinvestigator will refer subjects with TEAEs to specialist physicians, as required.

(4) The investigator or subinvestigator will unblind the study treatment according to "11.3 Emergency Unblinding Procedure" when it is necessary to identify the treatment (study drug) given to a subject.

11.2.2 Follow-up of TEAEs

TEAEs with an outcome of Recovering or Not Recovered at the end (discontinuation) of the study will be followed in principle until they recover to the pretreatment level and the process documented. Follow-up will be continued in principle for 2 weeks after the end of the treatment period. TEAEs with an outcome of Unknown at the end (discontinuation) of the study will not be followed, but the reason for the outcome recorded.

The investigator or subinvestigator may stop following TEAEs that have not recovered to the normal or pre-treatment level during the follow-up period when it is considered no longer necessary to follow them to ensure subject safety, and document the reason for discontinuing the follow-up.

11.2.3 Reporting of serious adverse events by investigator

(1) Emergency reporting

The investigator will report any serious adverse event to the sponsor orally or using phone or fax within 24 hours of knowing it. The investigator will then report the serious adverse event to the director of the study site and sponsor using the "Serious Adverse Event Report (first report)" (Unified form 12-1) within 3 days of knowing it. The investigator may use another form specified at the study site to report serious adverse events. In that case, the investigator will confirm that the form specified at the study site contains all the information specified in the unified form. If not, the investigator will add necessary information.

(2) Detailed reporting

The investigator will report any serious adverse event to the director of the study site and sponsor using the "Serious Adverse Event Report (xxth report)" (unified forms 12-1 and 12-2) within 10 days of knowing it. The investigator may use another form specified at the study site to report serious adverse events. In that case, the investigator will confirm that the form specified at the study site contains all the information specified in the unified form. If not, the investigator will add necessary information.

11.3 Emergency unblinding procedure

The investigator or subinvestigator will inform the sponsor of the following information when it is deemed necessary to urgently unblind the study treatment to ensure subject safety.

1. Name of study site/department
2. Name of investigator (subinvestigator)
3. Subject ID code
4. Drug number for which unblinding is requested
5. Reason for unblinding

For unblinding due to TEAE: Name of TEAE, date of discontinuation judgment, reason for discontinuation, symptomatic therapy given, and actions taken/clinical course

The investigator will fill in the predetermined sections of the eCRF and unblind the study treatment.

The detailed procedure for unblinding is separately specified in the Emergency Unblinding Procedure.

11.4 Anticipated Adverse Drug Reactions (Including Abnormal Changes in Laboratory Test Parameters)

The adverse drug reactions reported for AJG533 are described below.

Japanese clinical study results

(1) Phase I clinical study in patients with chronic constipation (2.5, 5, 10, 15, or 20 mg of the study drug or placebo was administered in a single dose or once daily for 14 days)

AJG533 (2.5, 5, 10, 15, or 20 mg) or placebo was administered in a single dose in 60 subjects. After single-dose safety was confirmed in each treatment group, AJG533 at 2.5, 5, 10, 15 or 20 mg or placebo was orally administered once daily for 14 days in 60 subjects.

(i) Single-dose administration (without breakfast)

Adverse drug reactions were observed in 26 of the 60 subjects (10 subject/group), including 4 events in 2 subjects in the placebo group, 4 events in 3 subjects in the 2.5 mg group, 7 events in 5 subjects in the 5 mg group, 4 events in 3 subjects in the 10 mg group, 18 events in 10 subjects in the 15 mg group, and 3 events in 3 subjects in the 20 mg group. There were no severe or serious adverse drug reactions. All the observed adverse drug reactions were mild and evaluated as Recovering or Recovered.

The adverse drug reactions included abdominal distention (1, 2, 2, and 1 subject in the placebo, 5 mg, 15 mg, and 20 mg groups, respectively), lower abdominal pain (1, 1, 1, 1, and 3 subjects in the placebo, 2.5 mg, 5 mg, 10 mg, and 15 mg groups, respectively), constipation (1 subject in the 15 mg group), diarrhoea (3 subjects in the 5 mg group and 7 subjects in the 15 mg group), flatulence (1 subject in the 5 mg group), proctalgia (1 subject in the 15 mg group), abnormal gastrointestinal sounds (1 subject in the 2.5 mg group), headache (1 subject in the 20 mg group), hot flush (1 subject in the 20 mg group), rash (1 subject in the 10 mg group), dysmenorrhoea (1 subject in the 15 mg group), feeling hot (1 subject in the 15 mg group), thirst (1 subject in the 15 mg group), blood urine present (1 subject in the 2.5 mg group), red blood cells urine positive (1 subject in the 2.5 mg group), alanine aminotransferase increased (1 subject in the 10 mg group), and aspartate aminotransferase increased (1 subject in the 10 mg group).

(ii) Single-dose administration (30 minutes before breakfast)

Adverse drug reactions were observed in 28 of the 60 subjects (10 subject/group), including 6 events in 5 subjects in the placebo group, 5 events in 4 subjects in the 2.5 mg group, 10 events in 8 subjects in the 5 mg group, 2 events in 2 subjects in the 10 mg group, 8 events in 6 subjects in the 15 mg group, and 7 events in 3 subjects in the 20 mg group. There were no severe or serious adverse drug reactions. All the observed adverse drug reactions were mild and evaluated as Recovered.

The adverse drug reactions included abdominal distention (2, 2, 1, and 2 subjects in the placebo, 2.5 mg, 15 mg, and 20 mg groups, respectively), lower abdominal pain (3, 2, 4, 1, 1, and 1 subject in the placebo, 2.5 mg, 5 mg, 10 mg, 15 mg, and 20 mg groups, respectively), diarrhoea (1, 4, and 5 subjects in the placebo, 5 mg, and 15 mg groups, respectively), vomiting (1 subject in the 20 mg group), headache (1 subject in the 20 mg group), presyncope (1 subject in the 2.5 mg group), eye pain (1 subject in the 15 mg group), dermatitis (1 subject in the 20 mg group), rash (1 subject in the 10 mg group), skeletal muscle stiffness (1 subject in the 20 mg group), and alanine aminotransferase increased (1 subject in the 5 mg group).

(iii) Repeated-dose administration (30 minutes before breakfast)

Adverse drug reactions were observed in 46 of the 59 subjects (10 subject/group), including 14 events in 5 subjects in the placebo group, 16 events in 8 subjects in the 2.5 mg group, 28 events in 8 subjects in the 5 mg group, 23 events in 8 subjects in the 10 mg group, 21 events in 8 subjects in the 15 mg group, and 33 events in 9 subjects in the 20 mg group. There were no severe or serious adverse drug reactions. All the observed adverse drug reactions were mild except 2 moderate events and evaluated as recovering or recovered.

The adverse drug reactions included abdominal distention (3 in the placebo group, 3 in the 2.5 mg group, 3 in the 5 mg group, 2 in the 10 mg group, 4 in the 15 mg group, and 6 in the 20 mg group), lower abdominal pain (2 in the placebo group, 4 in the 2.5 mg group, 3 in the 5 mg group, 3 in the 10 mg group, 4 in the 15 mg group, and 5 in the 20 mg group), upper abdominal pain (2 in the 5 mg group, 2 in the 10 mg group, and 2 in the 20 mg group), abdominal discomfort (1 in the placebo group, 1 in the 5 mg group, 1 in the 15 mg group, and 1 in the 20 mg group), diarrhoea (1 in the 2.5 mg group, 4 in the 5 mg group, 4 in the 10 mg group, 4 in the 15 mg group, and 7 in the 20 mg group), constipation (1 in the placebo group and 1 in the 20 mg group), abnormal gastrointestinal sounds (1 in the 20 mg group), defecation urgency (1 in the 2.5 mg group), flatulence (1 in the 2.5 mg group and 1 in the 5 mg group), haemorrhoids (1 in the 15 mg group), nausea (1 in the 5 mg group), hordeolum (1 in the placebo group), dysphoria (1 in the 2.5 mg group), headache (1 in the 5 mg group), periodic limb movement disorder (1 in the 10 mg group), epistaxis (1 in the 10 mg group), hiccups (1 in the 5 mg group), heat rash (1 in the 15 mg group), skin exfoliation (1 in the 10 mg group), urticaria (1 in the 10 mg group), back pain (1 in the placebo group), dysmenorrhoea (1 in the 2.5 mg group), asthenia (1 in the 20 mg group), thirst (1 in the 20 mg group), chest pain (1 in the 10 mg group), alanine aminotransferase increased (1 in the 2.5 mg group, 1 in the 5 mg group, 3 in the 15 mg group, and 1 in the 20 mg group), aspartate aminotransferase increased (1 in the 5 mg group, 2 in the 15 mg group, and 1 in the 20 mg group), eosinophilia (1 in the 15 mg group and 1 in the 20 mg group), blood triglycerides increased (1 in the 10 mg group), and blood uric acid increased (1 in the 5 mg group).

(2) Phase II clinical study in patients with chronic constipation (5, 10, or 15 mg of the study drug or placebo was administered once daily for 14 days)

Adverse drug reactions were observed in 34 of 163 subjects, including 4 events in 2 of 40 subjects in the placebo group, 26 events in 14 of 43 subjects in the 5 mg group, 18 events in 11 of 39 subjects in the 10 mg group, and 11 events in 7 of 41 subjects in the 15 mg group. There were no severe or serious adverse drug reactions. All the observed adverse drug reactions were mild except 4 moderate events and evaluated as Recovering or Recovered.

The adverse drug reactions included abdominal pain (10, 10, and 5 subjects in the 5 mg, 10 mg, and 15 mg groups, respectively), diarrhoea (4, 2, and 3 subjects in the 5 mg, 10 mg, and 15 mg groups, respectively), abdominal distension (3 and 1 subject in the 5 mg and 15 mg groups, respectively), lower abdominal pain (1, 1, and 1 subject in the placebo, 10 mg, and 15 mg group, respectively), upper abdominal pain (1 and 1 subject in the placebo and 10 mg group, respectively), defecation urgency (1 subject in the 5 mg group), nausea (1 and 1 subject in the 5 mg and 10 mg groups), vomiting (1 subject in the 10 mg group), dizziness (1 subject in the 5 mg group), headache (2 subjects in the 10 mg group), loss of consciousness (1 subject in the 5 mg group), yawning (1 subject in the 5 mg group), abnormal hepatic function (1 subject in the 5 mg group), pruritus (1 subject in the 5 mg group), feeling abnormal (1 subject in the 5 mg group), malaise (1 subject in the 10 mg group), thirst (1 subject in the placebo group), blood alkali phosphatase increased (1 subject in the 15 mg group), eosinophil count increased (1 subject in the 5 mg group), and white blood cell count increased (1 subject in the placebo group).

Foreign clinical study results

(1) Phase I clinical study in healthy adult males (0.1, 0.5, 2.5, or 5 mg of the study drug or placebo was administered in a single dose or 0.25 mg of the study drug or placebo was administered once daily for 7 days)

The study drug was administered in a single dose in 30 subjects (10, 4, 4, 4, and 8 in the placebo, 0.1 mg, 0.5 mg, 2.5 mg, and 5 mg groups, respectively) and in repeated doses in 8 subjects (2 in the placebo group and 6 in the 0.25 mg group).

TEAEs that developed when the study drug was administered in a single dose included abdominal distension, abdominal pain, upper abdominal pain, anorectal disorder, defecation urgency, diarrhoea, flatulence, frequent bowel movements, nausea, proctitis, rectal discharge, gastrointestinal motility disorder, application site erythema, fatigue, bloody discharge, nasopharyngitis, rhinitis, anorexia, neck pain, dizziness, headache, and irritated sensation of pharynx. Those that developed in repeated doses included diarrhoea, flatulence, haematochezia, nausea, periproctitis, vomiting, gastrointestinal motility disorder, nasopharyngitis, headache, paraesthesia, nightmare, and dysphonia.

Although one serious TEAE developed as proctitis in 1 subject in the single-dose 5 mg group, most of TEAEs were classified into gastrointestinal disorders, mild in severity, and not clinically relevant.

(2) Phase I clinical study in patients with chronic idiopathic constipation (0.1, 0.3, 1.0, 3.0, or 10 mg of the study drug or placebo was administered once daily for 14 days)

Adverse drug reactions were observed in 8 of 30 subjects (5 subject/group), including 3 subjects in the placebo group, 1 subject in the 0.1 mg group, 2 subjects in the 0.3 mg group, 1 subject in the 1 mg group, and 1 subject in the 10 mg group. There were no serious adverse drug reactions or those resulting in the discontinuation of the study. The adverse drug reactions included abdominal distention in 3 subjects (1 subject each in the placebo, 0.3 mg, and 10 mg groups), abdominal pain in 2 subjects (1 subject each in the placebo and 0.3 mg groups), nausea in 2 subjects (1 subject each in the placebo and 0.1 mg groups), dyspepsia in 1 subject (0.3 mg group), and rectal tenesmus in 1 subject (1 mg group).

(3) Late Phase II clinical study in patients with chronic idiopathic constipation (5, 10, or 15 mg of the study drug or placebo was administered once daily for 56 days)

Adverse drug reactions were observed in 15.2% (7/46 subjects) of the placebo group, 29.2% (14/48 subjects) of the 5 mg group, 29.8% (14/47 subjects) of the 10 mg group, and 39.6% (19/48 subjects) of the 15 mg group. None of the adverse drug reactions were considered serious. TEAEs resulted in the discontinuation of the study drug in 1 subject in the placebo group, 3 subjects in the 5 mg group, 4 subjects in the 10 mg group, and 7 subjects in the 15 mg group.

Common adverse drug reactions (developed in 5% or higher of any group) included abdominal pain in 23 subjects (5, 5, and 13 in the 5 mg, 10 mg, and 15 mg groups, respectively), diarrhoea in 14 subjects (1, 4, 3, and 6 subjects in the placebo, 5 mg, 10 mg, and 15 mg groups, respectively), flatulence in 12 subjects (3, 4, 3, and 2 subjects in the placebo, 5 mg, 10 mg, and 15 mg groups, respectively), abdominal distension in 10 subjects (1, 4, 2, and 3 subjects in the placebo, 5 mg, 10 mg, and 15 mg groups, respectively), and nausea in 9 subjects (2, 2, 2, and 3 subjects in the placebo, 5 mg, 10 mg, and 15 mg groups, respectively).

(4) Phase II clinical study in patients with functional constipation to evaluate effect on gastrointestinal and colonic motor function (15 mg or 20 mg of the study drug or placebo was administered once daily for 14 days)

Adverse drug reactions were observed in 61.5% (8/13 subjects) of the placebo group, 58.3% (7/12 subjects) of the 15 mg group, and 72.7% (8/11 subjects) of the 20 mg group. None of the adverse drug reactions were considered serious. Adverse drug reactions resulted in the discontinuation of the study included diarrhoea in 3 subjects (1 in the placebo group and 2 in the 20 mg group). Diarrhoea was resolved soon after it developed or the study was discontinued in 2 of the 3 subjects. The remaining subject in the 20 mg group also developed a severe, non-serious adverse drug reaction of abdominal pain, which was resolved within the day it developed.

Common adverse drug reactions (developed in 2 or more subjects in any group) included abdominal pain in 10 subjects (5 and 5 in the 15 mg and 20 mg groups, respectively), headache in 10 subjects (5, 2, and 3 in the placebo, 15 mg, and 20 mg groups, respectively), diarrhoea in 6 subjects (1 and 5 subjects in the placebo and 20 mg groups, respectively), nausea in 4 subjects (2 and 2 subjects in the placebo and 20 mg groups, respectively), flatulence in 3 subjects (2 and 1 subject in the placebo and 20 mg groups, respectively), and abnormal gastrointestinal sounds in 3 subjects (2 and 1 subject in the 15 mg and 20 mg groups, respectively).

(5) Phase I clinical study in healthy male adults to examine absorption, distribution, metabolism, and excretion (5 mg in a single dose)

Adverse drug reactions were observed in 5 of the 6 subjects (83.3%), including abdominal discomfort (3 subjects) and diarrhoea (3 subjects). There were no serious adverse drug reactions or those resulting in the discontinuation of the study.

(6) Phase I clinical study in healthy adult males and females to examine drug-drug interaction (10 mg in a single dose)

A total of 45 adverse drug reactions were observed in 18 of the 25 subjects (72%). Common adverse drug reactions (2 or more events in any group) included diarrhoea (12 events in 8 subjects), abdominal pain (9 events in 9 subjects), somnolence (9 events in 7 subjects), headache (5 events in 3 subjects), abdominal discomfort (4 events in 4 subjects), and dizziness (2 events in 2 subjects).

(7) DSUR Safety Information

Ferring Pharmaceuticals prepared the second DSUR (Development Safety Update Report from January 22, 2014 to January 21, 2015) in March 2015.

Elobixibat was administered in 247 subjects including 191 patients with chronic idiopathic constipation in the following 6 studies: foreign Phase I study (D1240C00001 study), foreign late Phase I study (A3309-001 study), foreign late Phase II study (A3309-002 study), foreign Phase II study (A3309-003 study), foreign Phase I study (A3309-004 study), and early Phase II study in patients with dyslipidemia (A3309-005 study).

Three TESAEs occurred in the 247 subjects who received elobixibat. Two of the 3 events (haemorrhagic intestinal diverticulitis in the 5 mg group and breast cancer in the 10 mg group) occurred in the foreign late Phase II study in patients with chronic idiopathic constipation (A3309-002 study). Both of the events were considered not related to the study drug. The remaining 1 event occurred as proctitis in a subject treated with a single dose of 5 mg in the foreign Phase I study in healthy male subjects (D1240C00001 study) (the relationship with the study drug was not evaluated in the study and the adverse event was classified as an adverse drug reaction in the DSUR). There were no serious TEAEs resulting in death.

Ongoing studies during the reporting period included the foreign Phase I study (000132 study) and 3 studies in the Echo Program [*]. Elobixibat was administered in 25 healthy subjects in the foreign Phase I study (000132 study) and 607 patients with chronic idiopathic constipation in the Echo Program.

There were no serious TEAEs in the foreign Phase I study (000132 study).

A total of 24 subjects in the Echo Program experienced serious TEAEs, including 7 subjects in the Echo 1 study (3 [2.4%] in the 10 mg group, 3 [2.4%] in the 5 mg group, and 1 [0.8%] in the placebo group), 5 subjects in the Echo 2 study (5 [1.6%] in the placebo group), and 12 (2.9%) in the Echo 3 study. Of the serious TEAEs, only constipation in 1 subject in the Echo 3 study (verbatim term: worsening of constipation) was considered possibly related to the study drug. One subject in the Echo3 study had a serious TEAE resulting in death (aortic dissection), which was considered not related to the study drug.

[*] Echo Program: Three studies in patients with chronic idiopathic constipation including Phase III studies (Echo 1 and 2 studies) and long-term administration study performed as the extension trial of the preceding studies The Echo 1 and 2 studies were prematurely terminated due to a distribution issue with the trial medication. Subjects enrolled in the studies were transferred to the partially modified Echo 3 study.

#Echo 1 study (000079 study): Double blind comparative study in which elobixibat (5 or 10 mg) or placebo is administered for 26 weeks (target sample size: 840 subjects)

#Echo 2 study (000080 study): Double blind comparative study in which elobixibat (5 or 10 mg) or placebo is administered for 12 weeks and then subjects in the 5 mg group receives placebo or 5 mg, those in the 10 mg group receive placebo or 10 mg, and those in the placebo group receive 10 mg for 4 weeks (target sample size: 840 subjects)

#Echo 3 study (000081 study): Open study in which subjects enrolled in the Echo 1 or 2 study receive 10 mg of elobixibat for 52 weeks (dose may be reduced to 5 mg; target sample size: about 900 subjects)

12. Discontinuation Criteria and Procedure by Subjects

12.1 Discontinuation criteria

The investigator or subinvestigator will discontinue the participation in the study for subjects who meet any of the following criteria after enrollment. The investigator or subinvestigator will promptly notify the sponsor of the date of discontinuation judgment * and reason for discontinuation when any enrolled subject discontinues the study.

(1) Screen failure: Subjects are not enrolled because of ineligible test results.
(2) Withdrawal by subject: The subject requests the withdrawal from the study for his/her personal reasons including moving and hospital transfer (the discontinuation due to a TEAE or lack of efficacy should not be classified as withdrawal by subject.
(3) Adverse event: The investigator or subinvestigator decides that the subject can no longer receive the study drug due to an adverse event or the subjects requests the withdrawal from the study due to an adverse event.
(4) Protocol deviation: The subject is found to be ineligible for the study after enrollment.
(5) Lack of efficacy: The target disease is not worsened, but the continued participation in the study will pose an unacceptable risk to the subject due to inadequate clinical efficacy.
(6) Study terminated by sponsor: The whole study is terminated.
(7) Lost to follow-up: The contact with the subject is lost and the subject cannot continue the study.
(8) Others: The subject cannot comply with the protocol or the investigator or subinvestigator decides that the subject should discontinue the study for reasons other than the above.

*: The day of discontinuation judgment is defined as the day when the investigator or subinvestigator judges that the subject meets any of the discontinuation criteria. If the investigator or subinvestigator recognizes that the subject meets any of the discontinuation criteria ex post facto due to emergency or for other reasons, the day the situation arises is considered as the day of discontinuation judgment.

[Rationales for the selection]

(2): The criterion was set to respect the free will of subjects. (3) and (5): The criteria were set as general considerations to ensure subject safety. (4): The criterion was set to comply with the protocol. (6): The criterion was set assuming the discontinuation of the whole study. (8): The criterion was set assuming the case not meeting the criteria (1) to (7).

12.2 Discontinuation procedure

(1) When it is revealed that the patient met the criteria for discontinuation, the investigator or subinvestigator should explain such a fact to the concerned patient and withdraw him/her from the study. After withdrawal, appropriate therapeutic measures such as an alternative treatment should be implemented.

(2) The investigator or subinvestigator will ask subjects who withdraw from the study after receiving the study treatment to receive all the observations and tests specified at the end of the study as those at discontinuation, wherever possible. The investigator or subinvestigator will make efforts to have safety observations and tests wherever possible even if the observations and tests at the end of the study cannot be performed.

(3) When the patient is withdrawn from the study due to safety problems such as onset of a TEAE, the investigator or subinvestigator should promptly take appropriate therapeutic measures.

(4) When it is found that the patient cannot make visits due to his/her convenience after study initiation, the investigator or subinvestigator should verify the patient's health, check the reason for not making visits, and investigate safety using letters, FAX, telephone or other relevant means. If the treatment has been started, the details of the investigation should be recorded in writing.

(5) The investigator or subinvestigator will enter the following information in the eCRF for subjects who discontinue the study after the start of the study treatment: date when the last observations/tests are performed at discontinuation or, if the observations/tests cannot be performed, date when the investigator or subinvestigator makes contact according to Step (4) of "12.2 Discontinuation Procedure" and applicable discontinuation criteria (detailed reason for discontinuation for subjects who meet the discontinuation criterion (8)).

13. Statistical Analysis

13.1 Sample size

Total of 120 randomized subjects

AJG533 10 mg group	60 subjects
AJG533 placebo group	60 subjects

Total of 108 subjects evaluable for primary endpoint

AJG533 10 mg group	54 subjects
AJG533 placebo group	54 subjects

[Rationales for the selection]

The primary endpoint of the Japanese Phase II clinical study (AJG533/ET1), or the change in SBM frequency from screening period Week 2 to treatment period Week 1 (n, LSMeans, [SE]) was (n = 38, 5.63 [0.68]) in the AJG533 10 mg group and (n = 40, 2.63, [0.46]) in the placebo group, with an inter-group difference (LSMeans, [SE]) of (3.00, [0.82]). It has been reported that the target of demonstrating the superiority to the control or standard therapy may not be achieved by performing a new study using only point estimates of efficacy from earlier, relatively small studies because of the scale of the study. Further, several methods for setting sample size have been proposed. Therefore, the one-sided 80% confidence interval of the difference between the placebo and AJG533 10 mg groups was calculated and the lower limit of the interval was considered as the difference in the population mean of the study. The standard deviation of each group was specified based on the results of the AJG533/ET1 study.

The lower limit of the one-sided 80% confidence interval was 2.30 for the difference in the change before and after treatment between the placebo and AJG533 10 mg groups. The sample size required for the study was calculated at 54 subjects per group when it was calculated by t-test in unequal variance, assuming that the population standard deviations of the placebo and AJG533 10 mg groups were 2.90 and 4.24, respectively and assuming a two-sided significance level of 5%, statistical power of 90%, and allocation ratio of 1 to 1 (AJG533 10 mg group to placebo group). Therefore, it was decided that 54 subjects per group were necessary for evaluating the primary endpoint. Assuming that 10% of enrolled subjects would prematurely drop out of the study, it was decided that 60 subjects would be randomized to each group in the study, with the total number of subjects being 120.

13.2 Analysis sets

13.2.1 Definitions of analysis sets

The following 3 analysis sets will be defined:

(i) Full Analysis Set (FAS)

The population of all patients who received at least one dose of the study drug and have any observed efficacy data is defined as the "full analysis set (FAS)."

(ii) Per Protocol Set (PPS)

Per protocol set (PPS) is defined as the set of subjects in FAS excluding those who meet any of the following criteria.

1) Subjects who do not meet any of the inclusion criteria or meet any of the exclusion criteria
2) Subjects who received the study drug with a different drug number from the allocated number
3) Subjects with problems including the use of prohibited medications, poor treatment compliance, lost to follow-up, and missing data *)

*): The detailed criteria for adoption are specified separately in the "Case and Data Handling Standards."

(iii) Safety analysis set

The population of all patients who received at least one dose of the study drug is defined as the "safety analysis set."

13.2.2 Analysis sets to be used for each evaluation

(i) Analysis set for efficacy evaluation

The FAS will be the primary analysis set.

(ii) Analysis set for safety evaluation

Analyses using the "safety analysis set" will be carried out.

13.3 Data Handling

Standards for data handling are stipulated in the separately prepared Statistical Analysis Plan and Case and Data Handling Standards. In principle, efficacy data collected shall be used for summary and analysis.

Missing values of tests or data not adopted after the handling according to the Case and Data Handling Standards and Statistical Analysis Plan shall be excluded from the statistical tests or estimations for evaluation. Missing values of the primary endpoint will be subjected to the sensitivity analysis for the handling of missing data to examine the stability of the study results.

Because the efficacy evaluation will be performed based on the patient diary, efficacy data at each time point will be handled as follows:

1.	screening period Week 1:	Defined as a period of 168 hours from 0:00 on Day -15 to 24:00 on Day -9, assuming the date and time of the first dose of the study drug as the starting point (Day +1).
2.	screening period Week 2:	Defined as a period of 168 hours from 0:00 on Day -8 to 24:00 on Day -2, assuming the date and time of the first dose of the study drug as the starting point (Day +1).
3.	treatment period Week 1:	Defined as a period for 7 days (168 hours), assuming the date and time of the first dose of the study drug as the starting point (Day +1).
4.	treatment period: Week 2	Defined as a period for 7 days (168 hours) from Day 8, assuming the date and time of the first dose of the study drug as the starting point (Day +1).

13.4 Objectives of Analyses

The primary objective of statistical analysis is to indicate the presence/absence of statistical difference in the primary endpoint, change in SBM frequency, between the placebo and AJG533 10 mg groups by calculating the test statistics and significance probability (p-value) of the primary endpoint.

The secondary objective is to estimate the intergroup differences in various endpoints supporting the primary endpoint.

13.5 Analyses about study progress and subject background

13.5.1 Study progress

The disposition of subjects (including completion status, reasons for discontinuance, and treatment period) will be summarized for all the subjects who provided consent. Protocol deviations will be summarized for subjects who received the study drug. In addition, the treatment status will be analyzed and tabulated using the FAS and safety analysis set.

13.5.2 Subject background

The disposition of all the subjects who provided consent and all the analysis sets will be presented. Using the FAS, PPS and safety analysis set, the status of demographic variables and other baseline data in each group will be presented using frequency distribution tables and summary statistics.

13.6 Analyses for efficacy evaluation

Unless otherwise specified, the significance level and confidence interval of two-sided statistical tests will be 5% and 95%, respectively.

13.6.1 Primary endpoint

13.6.1.1 Primary analysis

Primary analysis will be performed in FAS. The primary endpoint will be defined with the following formula based on SBM frequency in treatment period Week 1 of AJG533 and screening period Week 2.

(Change in SBM frequency) = (SBM frequency in Treatment Period Week 1) - (SBM frequency in Screening Period Week 2)

For the analysis, a normal distribution is assumed for "the change in SBM frequency," and the analysis of covariance (ANCOVA) with the SBM frequency for Screening Period Week 2 as the covariate will be applied. The estimated "difference in the change in SBM frequency" adjusted by ANCOVA, standard error, 95% confidence interval, and p-value will be calculated.

Null hypothesis $H_{ij0}: \mu_i=\mu_j$ (i = placebo group, j = AJG533 10 mg group)
Alternate hypothesis $H_{ij1}: \mu_i\neq\mu_j$, where μ is defined as adjusted mean in this section.

13.6.1.2 Secondary analyses

(1) Sensitivity analysis of primary analysis

The same analyses as the primary analysis will be done in PPS.

(2) Adjusted analysis

Adjusted analysis will be performed in FAS by adding any of the following factors to the primary analysis model as a covariate. The items and categories will be reexamined based on the results of the blinded review and presented in the Statistical Analysis Plan.

1) Gender
2) Age
3) BMI [less than 25kg/m^2, 25kg/m^2 or higher]
4) Binary data on subjects who meet IBS diagnostic criteria and those who do not
5) Use of rescue medication in screening period Week 2
6) Constipation severity evaluation in screening period Week 2
7) Stool consistency as measured by Bristol Stool Form Scale in screening period Week 2

(3) Subgroup analysis

Subgroup analysis using the following items as a stratification factor will be performed. The items and categories will be reexamined based on the results of the blinded review and presented in the Statistical Analysis Plan.

1) Gender
2) Age
3) BMI [less than 25 kg/m^2, 25 kg/m^2 or higher]
4) Complications
5) Medical history
6) Binary data on subjects who meet IBS diagnostic criteria and those who do not
7) Use of rescue medication in screening period Week 2
8) Constipation severity evaluation in screening period Week 2
9) Stool consistency as measured by Bristol Stool Form Scale in screening period Week 2

13.6.2 Secondary endpoints

The belowmentioned endpoints will be analyzed using the FAS and PPS.

13.6.2.1 Change in SBM frequency from screening period Week 2 to treatment period Week 2

The change in SBM frequency will be determined for each subject to calculate summary statistics by groups.

13.6.2.2 Change in SBM frequency from the screening period to the treatment period

The change in SBM frequency for 14 days will be determined for each subject to calculate summary statistics by groups.

13.6.2.3 Changes in CSBM frequency from screening period Week 2 to treatment period Week 1 and Week 2

The change in CSBM frequency will be determined for each patient to calculate summary statistics by groups.

13.6.2.4 Change in CSBM frequency from the screening period to the treatment period

The change in CSBM frequency for 14 days will be determined for each subject to calculate summary statistics by groups.

13.6.2.5 Proportion of patients who experience spontaneous bowel movements within 24 or 48 hours after the start of the study drug

The rate of subjects who experience spontaneous bowel movements within 24 or 48 hours after the first dose will be calculated by groups to test the difference between the groups by $\chi 2$ or Fisher's exact test.

13.6.2.6 SBM and CSBM responder rates for treatment period Weeks 1 and 2

Each responder rate and its 95% confidence interval will be calculated for each treatment group.

13.6.2.7 Time to the first SBM

The time to the first SBM will be estimated with the Kaplan-Meier method and tested with the Log-rank test.

13.6.2.8 Use of rescue medication

The rate of subjects who use the rescue medication will be calculated by groups.

13.6.2.9 Stool consistency as measured by Bristol Stool Form Scale

The mean stool consistency will be handled as a continuous variable to calculate summary statistics by groups. The median stool consistency during the evaluation period will be calculated to prepare a frequency table of each score. A frequency table in which stool consistency is categorized into [1, 2], [3, 4, 5], or [6,7] will be then prepared.

13.6.2.10 Evaluation of weekly-based severity of constipation for treatment period Week 1 and Week 2

The evaluation of the severity of constipation will be tabulated and analyzed in each treatment group.

13.6.3 Multiple comparison/multiplicity

This study will not have any multiplicity issue because a pairwise comparison of the primary endpoint, change in SBM frequency, between the placebo and AJG533 10 mg groups is planned.

13.7 Analysis for safety evaluation

13.7.1 Coding of TEAEs

TEAEs will be coded using MedDRA. Primary system organ classes (SOCs) will be adopted.

13.7.2 Incidence of TEAEs

For TEAEs, the number of patients with TEAEs and incidence by causal relationship with the stydy drug will be calculated for each SOC and Preferred Term (PT), and CIs of the differences in the incidence between the placebo and AJG533 groups will be tabulated for each treatment group. As necessary, subgroup analyses according to demographic characteristics, other patient background characteristics, etc. will be implemented. In addition, in accordance with criteria defined in the statistical analysis plan, for common TEAEs, TEAEs by severity, death, TESAEs other than death, TEAEs leading to discontinuation, and clinically important TEAEs, the same tabulation will be performed.

13.7.3 Analysis on TEAE onset timing

The number of subjects with TEAEs will be summarized for each PT and SOC by groups to determine the median “number of days before the first onset” in the group of subjects who developed each TEAE. Also, the number of patients with TEAEs resolved by the last visit day will be tabulated, and median values of the

"number of days from onset to recovery" will be calculated in a population of patients who developed applicable TEAEs.

13.7.4 Laboratory test parameters and vital signs

Summary statistics will be calculated for each laboratory test parameter and vital sign (except urinalysis) by groups and evaluation time points. A boxplot will be prepared for each parameter by groups. For urinalysis, a shift table indicating the test results on the day of enrollment and at Week 2 visit/discontinuation will be prepared by groups.

13.7.5 Abnormal changes in laboratory test parameters

For each laboratory test item at each evaluation time point after the start of the treatment, the number of patients with abnormal changes in accordance with criteria defined in the "statistical analysis plan" will be tabulated by treatment group.

13.8 Interim Analysis

No interim analysis will be performed in this study.

14. Protocol Compliance, Deviations or Amendments, and Revision

14.1 Protocol compliance

(1) The sponsor will provide the study protocol and investigator's brochure, etc. to the prospective investigator. Based on the materials and information provided, the prospective investigator will fully examine and discuss the ethical and scientific validity of conducting the study with the sponsor and agree with the sponsor on the protocol and protocol compliance. To witness the agreement, the investigator and sponsor will write their names and put their seal or write their signatures and enter the date on the Statement of Agreement on Study Protocol.

(2) The investigator or subinvestigator shall not deviate the protocol (except emergency deviations) or make changes in it without the prior written agreement between the investigator and sponsor and prior written approval of the institutional review board based on the prior review.

14.2 Protocol deviations or amendments

(1) The investigator or subinvestigator will document any deviations from the protocol, irrespective of its reason. The investigator will prepare a document describing the reason and immediately submit it to the director of

the study site and sponsor for deviations made for avoiding urgent risks or for medically unavoidable reasons.

(2) The investigator or subinvestigator may deviate from or change the protocol without the prior written agreement with the sponsor and prior approval of the IRB if there are medically unavoidable reasons including the avoidance of urgent risks to subjects (deviations at emergency). In that case, the investigator will submit the deviations or changes made, their reasons, and, if deemed appropriately, draft revision of the protocol, to the sponsor and director of the study site as soon as possible. The investigator will obtain the approval of IRB via the director of the study site and obtain the written approval of the director of the study site and written agreement with the sponsor via the director of the study site.

(3) The investigator shall promptly submit a report to the sponsor, director of the study site, and institutional review board via the director of the director of the study site for any change in the study activities that may have a serious effect on the conduct of the study or increase the risk to subjects (including serious deviations).

14.3 Protocol revision

(1) The sponsor will provide the investigator with the revised protocol if the protocol is revised.

(2) Based on the materials and information provided, the investigator will fully examine and discuss the ethical and scientific validity of conducting the study with the sponsor and agree with the sponsor on the revised protocol and protocol compliance. To witness the agreement, the investigator and sponsor will write their names and put their seals or write their signatures and enter the date on the Statement of Agreement on Study Protocol. This also applies to the case when the protocol is revised according to the directions of the director of the study site based on the IRB's opinions within the acceptable range for the sponsor.

(3) The sponsor will immediately submit the revised protocol to the director of the study site and obtain the approval of the IRB via the director of the study site. Upon approval, the investigator and subinvestigators will perform the study according to the revised protocol.

15. Study Completion or Termination and Suspension

15.1 Study completion

Upon completion of the study, the investigator will immediately report it and provide the written summary of the study results to the director of the study site. The director of the study site will immediately report it and provide the written summary of the study results to the IRB and sponsor.

15.2 Study termination or suspension procedure by sponsor

The sponsor will examine whether to continue the study based on the study protocol for the whole study or study at specific study sites when any of the followings arises.

(1) The sponsor knows important information to properly perform the study including that on the quality, efficacy, and safety of the study drug.
(2) It becomes necessary to revise the protocol and a study site(s) cannot accept the revision.
(3) The director of a study site directs the revision of the protocol and the sponsor cannot approve it.
(4) A study site makes a serious or continuous violation of GCP, study protocol, or study agreement.
(5) The IRB of a study site decides that the study should not be continued and the director of the study site directs the discontinuation of the study.
(6) The investigator decides that the study cannot be continued.

The sponsor will immediately notify the director of each study site of the termination or suspension of the study and its reason in writing when the sponsor decides to discontinue or suspend the study.

Upon receipt of the notification to discontinue or suspend the study from the sponsor, the director of the study site will immediately notify the investigator and IRB of it and its reason in writing.

Upon receipt of the notification to discontinue or suspend the study from the sponsor via the director of the study site, the investigator will immediately notify subjects of it, provide appropriate medical care, and take other necessary measures. The “12.2 Discontinuation procedure” will be followed for the actions to be taken to subjects if the study is discontinued.

15.3 Study termination or suspension procedure at study sites

(1) The investigator will immediately report the
the termination or suspension of the study in writing and provide a written explanation on it to the director of the study site when the investigator decides to discontinue or suspend the study.
(2) Upon receipt of the notification to discontinue or suspend the study from the investigator via the director of the study site, the sponsor will request the written detailed explanation on the termination or suspension from the director of the study site.

16. Electronic Case Report Form (eCRF)

16.1 Precautions for preparing and correcting eCRF

(1) The investigator or subinvestigator will prepare eCRFs using the EDC system according to the EDC Operation Manual and Guide for Preparing and Correcting eCRFs. eCRFs will be prepared for subjects who provide the informed consent.

(2) The investigator may instruct the pre-designated study collaborator to assist the preparation of eCRFs. The assistance for the preparation of eCRFs by the study collaborator is limited to the data transcription from source materials.

(3) The investigator, subinvestigator, or study collaborator will fill in the EDC form.

(4) The investigator, subinvestigator, or study collaborator will add data or correct the data in the form according to the alert [1], if any, displayed in saving the data.

(5) The investigator, subinvestigator, or study collaborator will change the entry status to "Completed" when the data entry including the handling of the alert is completed.

(6) The investigator, subinvestigator, or study collaborator will take appropriate measures, such as data correction and response to queries when the monitor or person in charge of study quality control data control issues a query [2].

(7) The investigator will confirm that eCRFs locked to prevent changes are accurate and complete for each subject, write an electronic signature using an appropriate EDC system function, and submit it to the sponsor.

(8) When it becomes necessary to make changes to an eCRF submitted to the sponsor, the investigator will request the sponsor to unlock the eCRF and then make changes. The corrected eCRF will be re-locked. The investigator will confirm that the changes made to the eCRF are accurate and complete, write an electronic signature again, and submit it to the sponsor.

(9) The investigator will keep a copy of each eCRF containing input data, correction history, electronic signature information, query communication records in a non-rewritable electronic medium (e.g. CD-R).

(10) When the investigator finds any inconsistency between an eCRF and corresponding source materials, the investigator will prepare a material explaining its reason, submit it to the sponsor, and keep its copy.

1. Warning text displayed on the screen by the automatic logical-check of the entries to the eCRF when they are saved
2. Queries issued, read, and answered on the EDC system

16.2 Identification of Items for Which Data in eCRF Should be Handled as Raw Data

Source data will be identified with the Source Material Identification List of each study site. Raw data will be recorded in materials other than eCRF so that the entries to the EDC system will not be handled as raw data.

16.3 Identification of Original eCRF

The contents recorded on the ASP server will be considered as the original eCRF while the EDC system is operated.

After the operation of the EDC system is completed, the contents recorded on the non-rewritable electronic medium (e.g. CD-R and DVD-R) delivered from the EDC vendor (or data management contractor) to the sponsor will be considered as the original eCRF.

17. Source Document Verification

The director of the study site and investigator will accept the monitoring and audits by the sponsor and inspections by the IRB and regulatory authorities and provide all the study related records including source materials for source document verification. The director of the study site and investigator have to pay careful attention to protect the privacy of subjects throughout source document verification.

18. Quality Control and Assurance of Clinical Study

18.1 Quality control

(1) The monitor will confirm that the study is properly conducted in compliance with the protocol and GCP, and documents to be retained are completely archived at the study sites through monitoring.

(2) The monitor will crosscheck study-related records such as source materials against eCRFs to confirm consistency. The persons in charge of data management and statistical analysis will check and confirm data at each stage of data handling.

(3) The person in charge of study quality control will inspect and confirm at each phase of the study that the study is performed in compliance with GCP, protocol, and study implementation procedures of the sponsor.

18.2 Quality assurance

To assure that the study is conducted and its data are prepared, recorded, and reported in compliance with the protocol, GCP, and study implementation procedures of the sponsor, the auditor independent of study-related departments including the monitoring department will inspect the internal procedures of the sponsor and procedures of study sites, as required, to confirm that the quality of the study is appropriately controlled.

19. Ethical Conduct of the Study

19.1 Institutional Review Board

(1) This study will be reviewed by the institutional review board (IRB) at each study site for the validity of conducting the study from ethical, scientific, and medical/pharmaceutical viewpoints.

(2) The investigator at each study site will start the study according to the directions and decisions of the director of the study site made based on the approval of the study by IRB.

(3) The director of the study site will be reviewed by IRB for the validity of continuing the study when any of the following events arises.

(i) The investigator reports a serious adverse event.

(ii) The sponsor provides a "new safety report."

(iii) The investigator revises the informed consent form and patient information leaflet based on the information that may affect the willingness of subjects to continue the study.

(iv) The study continues for more than 1 year.

(v) The director recognizes the need of the review for other reasons.

19.2 Protection of human rights of subjects

The investigator and subinvestigators will carefully examine the validity of inviting a potential subject to participate in the study, considering the health status, symptoms, age, gender, ability to give consent, dependence on the investigator, etc., and participation in other clinical studies, based on the protection of human rights and compliance with the inclusion and exclusion criteria.

The subject ID code will be used to enroll subjects and identify subjects on eCRFs. Full consideration will be given to the protection of the privacy of subjects including their name and disease during the source document verification of study-related source materials and informed consent documents and at the publication of the study results.

20. Record Storage

(1) The director of the study site will store study-related documents or records specified by GCP including source materials, agreements, informed consent form and patient information leaflet, protocol, and records related to study drug management and other study activities until the following (i) or (ii), whichever is later. However, the director of the study site will consult with the sponsor about storage period and method if the sponsor requests storage for a longer period. The director of the study site will designate persons in charge of the storage of different records.

(i) Date of the manufacturing/marketing approval (or approval of partial changes in the approval) of the study drug (or date at 3 years after the notification of termination if the development of the study drug is terminated)

(ii) Date at 3 years after the termination or end of the study

(2) The person who established IRB will store applicable standard operating procedures, records on the designation of board members, member list (including the information on qualification, occupation, and affiliation), submitted documents, meeting minutes, and other study related records including correspondences until the following (i) or (ii), whichever is later. However, the director of the study site will consult with the sponsor about storage period and method if the sponsor requests storage for a longer period.

(i) Date of the manufacturing/marketing approval (or approval of partial changes in the approval) of the study drug (or date of the notification of termination if the development of the study drug is terminated)

(ii) Date at 3 years after the termination or end of the study

(3) The sponsor will store study related documents and records until the following (i) or (ii), whichever is later.

(i) Date at 5 years after the date of the manufacturing/marketing approval (or approval of partial changes in the approval) of the study drug (or date at 3 years after the notification of termination to the director of the study site if the development of the study drug is terminated) or date of the end of re-examination

(ii) Date at 3 years after the termination or end of the study

(4) If the sponsor decides that the director of the study site or person who established IRB does not need to store study related documents or records any more, the sponsor will notify the decision to the director of the study site or person who established IRB via the director of the study site.

21. Monetary Payment and Insurance

21.1 Monetary payment

If money is paid to subjects to reduce the financial burden to participate in the study, the amount and method of the payment will be reviewed and approved by IRB according to applicable predetermined standard operating procedures of the study site. The expenses to be reimbursed to subjects will be paid by the sponsor via the study site according to the procedure specified in the agreement, etc.

The sponsor will bear the following costs as the costs not covered by the health insurance and concomitant medical expense payment *.

1. Period after the informed consent to the day before the starting day of the study drug treatment

- Costs for study related examinations and imaging diagnoses
- Costs for rescue medication (bisacodyl suppositories)

2. Period from the starting day to last day of the study drug treatment

- Costs for all examinations and imaging diagnoses
- Costs for rescue medication (bisacodyl suppositories)
- Costs for drugs of the same type or with the same indications as the study drug

3. Period from the next day of the last day of the study drug treatment or later

- Costs for examinations and imaging diagnoses for following adverse events
- Costs for study related examinations and imaging diagnoses

* Notification No. 0929002 from Medical Economics Division, Health Insurance Bureau dated September 29, 2006: Partial Amendments of "Considerations Associated with Establishment of Medical Fee Calculation System"

JPMA Notification No. 101 dated February 27, 2007: Interpretation of Expenses to be Borne by Sponsor in Uninsured Concomitant Treatment Cost System

21.2 Compensation for Health Hazards and Insurance

(1) If any study related health hazard arises in subjects, the study site shall immediately give appropriate medical care and take other necessary measures. The medical costs for such health hazards payed by subjects will be reimbursed by the sponsor.

(2) If any study related health hazard arises in subjects and the sponsor is liable for compensation, the sponsor will pay compensation within the scope of responsibilities.

(3) The compensation in the previous section shall be paid according to the compensation system of the sponsor, with reference to the payment by the Relief System for Sufferers from Adverse Drug Reactions (for residual disabilities, the first to third levels as specified in the disability certification standards of the National Pension/Employees' Pension Insurance System will apply).

(4) The sponsor will not pay compensation to health hazards for which the causal relationship with the study drug is denied, health hazards for which the sponsor, study site, or third parties (those other than subjects, sponsor, and medical institutions) are liable for indemnification, or intentional health hazards by subjects. However, the sponsor will pay compensation when the indemnification remains unclear within a reasonable period of time from the request of the compensation. In that case, when the sponsor has paid compensation to a subject for a health hazard for which the study site or third party is liable, the sponsor can request the reimbursement within the amount of the payment to the study site or third party. The compensation for disability will be reduced or not paid if the disability results from a serious fault of the subject.

(5) The indemnification liability to health hazards related to the study shall be borne by the sponsor unless it should be attributable to the study site. For the indemnification liability for which where responsibility lies is unknown, the sponsor and study site shall sincerely confer with each other and solve the problem.

(6) When a health hazard related to the study occurs in a subject and may cause a conflict with the subject or third party, the study site shall immediately report it to the sponsor and the study site and sponsor shall cooperate to resolve the conflict.

(7) The sponsor shall take appropriate measures including insurances to perform the indemnification and compensation responsibilities for subject health hazards related to the study.

22. Arrangements for Publication

(1) The investigator, subinvestigator, or all those involved in the study at the study site shall obtain the written prior approval of the sponsor if he/she intends to publish the information obtained from the study at scientific conferences or journals, etc.

(2) The sponsor will reserve the right to use the information obtained from the study for the application for the manufacturing/marketing approval of the study drug (or approval of partial changes) without restrictions. The sponsor will also reserve the right to use the information in the product information summary, etc.

23. Study Implementation Structure

This study will be conducted by the organization composed of the following members.

23.1 Sponsor

23.1.1 Sponsor

Name of Sponsor: Ajinomoto Pharmaceuticals Co., Ltd.

Location: 2-1-1 Irifune, Chuo-ku, Tokyo 104-0042

23.1.2 Person who has the right to sign the protocol as the representative of the sponsor

See Attached Document 1.

(Main duties)

- To sign the protocol as the representative of the sponsor.
- To take overall responsibility for the study-related activities carried out by the sponsor.
- To issue and revise the protocol.
- To issue and revise the investigator's brochure.
- To prepare the clinical study report.

23.1.3 Medical expert

See Attached Document 1.

(Main duties)

- To promptly provide advice on the following matters implemented by the sponsor from a medical point of view:
  - Drawing up and revising the protocol and investigator's brochure
  - Selection of study sites and investigators
  - Preparation of draft informed consent form and patient information leaflet
  - Continuous assessment of safety information related to the study drug (e.g., actions taken against treatment emergent serious adverse events [TESAEs])
  - Medical evaluation of data obtained from the study (appropriateness of evaluations)
  - Preparation of the clinical study report
- To make a medical judgment on the causal relationship of health hazards with the clinical study
- To sign the clinical study report.

- To give advice to the study sites, as necessary.
- To attend study meetings, as necessary.
- To meet with the regulatory authorities, as necessary.

23.1.4 Monitors

See Attached Document 1.

(Main duties)

- To verify the implementation status of the clinical study through means such as vising the study sites.
- To deliver and collect the study drug
- To check that the contents of the eCRF are correct and consistent with source documents.
- To collect and provide information
- To perform source document verification
- To verify the archiving status (e.g., archiving place and record archiving manager) of study-related documents or records.

23.1.5 Quality Control of Study

See Attached Document 1.

(Main duties)

- To check that the sponsor is conducting the study in accordance with the study implementation procedures

23.1.6 Data Management

See Attached Document 1.

(Main duties)

- To design, construct, inspect, operate, and maintain the computer system for data management
- To properly perform data management activities (CRF check, data input, and data fixation)
- To prepare data sets for analysis by reflecting case and data handling
- To manage outsourced activities upon discussion with the person in charge of data management at the data management contractor

23.1.7 Statistical analysis

See Attached Document 1.

(Main duties)

- To prepare the statistical analysis plan and statistical analysis report
- To prepare the statistical aspects of the protocol

- To be responsible for the statistical analysis in the clinical study report
- To manage outsourced activities upon discussion with the statistical analyst at the statistical analysis contractor

23.1.8 Medical Statistics Advisor

See Attached Document 1.

(Main duties)

To verify statistical aspects of the clinical study.

23.1.9 Audit

See Attached Document 1.

(Main duties)

- To conduct audits in accordance with the procedures for the implementation of audits issued by the sponsor.

23.1.10 Study drug control

See Attached Document 1.

(Main duties)

- To manage the study drug in accordance with the procedures for the implementation of clinical studies issued by the sponsor.

23.2 Contract Research Organizations

23.2.1 Laboratory testing institution

See Attached Document 2.

(Main duties)

- To collect samples submitted by each study site
- To measure samples of central laboratory test parameters, prepare test reports, and notify the test results to each study site and sponsor
- To give consideration to keep the name of subjects undisclosed in the test results submitted to the sponsor
- To store samples after testing

23.2.2 Study drug allocation manager

See Attached Document 2.

(Main duties)

- To allocate to the study drug
- To prepare study drug inventory files
- To randomly collect storage samples
- To confirm indistinguishability

23.2.3 Subject enrollment manager

See Attached Document 2.

(Main duties)

- To prepare the allocation table
- To import the allocation table and inventory files to the EDC system
- To take measures against the misuse of the study drug and product failure, such as the re-allocation of the drug number
- To confirm that the study drug is kept blinded until key code breaking
- To break the key code

23.2.4 Contract Research Organization for data management

See Attached Document 2.

(Main outsourced duties)

- To properly develop and maintain systems (including allocation and emergency key opening systems), manage study data (CRF check, data input, and data fixation), and prepare analysis datasets

23.2.5 Contract research organization for statistical analysis

See Attached Document 2.

(Main outsourced duties)

- To properly perform statistical analysis activities (preparation of the Statistical Analysis Plan, analysis programs, and Statistical Analysis Report) in accordance with applicable SOPs

23.2.6 Contract research organization for study drug storage and management

See Attached Document 2.

(Main outsourced duties)

- To properly transport and storage the study drug (transportation, collection, entering and dispatching warehouse, and storage) according to applicable SOPs

23.3 Study sites and Investigators

See Attached Document 3.

(main duties of investigator)

- To conduct the study in compliance with the protocol and GCP
- To review the protocol (including revisions) and make an agreement with the sponsor, and prior to the initiation of the study, obtain the approval of the director of the study site based on the approval of the IRB
- To prepare and revise the informed consent form and patient information leaflet in cooperation with the sponsor and obtain approval of IRB
- When important study-related activities are assigned to the subinvestigators or study collaborator, to prepare a list of the contents of assigned activities and assigned persons, and obtain prior approval of the head of the study site.
- To select subjects who meet the criteria of the protocol and prepare the subject screening list/register
- To give an explanation on the details of the clinical study to the subjects using a document and obtain their written consent for participation in the study.
- To check whether or not subjects are being treated by other physicians, and if yes, notify, upon their consent, the physicians that they will take part in the study, and obtain information on drugs and treatment being given
- To review the clinical study agreement
- To give adequate information on the study to the subinvestigators and study collaborators and instruct and supervise them
- To take responsibility for all medical decisions related to the study
- To record protocol deviations, if any
- In case there was a protocol deviation to eliminate an immediate hazard to the subject or due to another medically compelling reason, to immediately report it in writing to the head of the study site and the sponsor, and retain its copy.
- When emergency reporting on TESAEs or other relevant matters is necessary, to immediately report them in writing to the head of the study site and the sponsor.
- To check and confirm eCRFs prepared by subinvestigators
- To prepare accurate and complete eCRFs, electronically sign them, and submit them to the sponsor
- When the study is completed and the investigator him-/herself prematurely terminated or interrupted the study, to report it in writing to the head of the study site.
- To report in writing a summary of the actual status of the study to the head of the study site once a year or more frequently upon request of the IRB.
- To collaborate in source document verification during monitoring and audits
- To retain study-related documents or records to be kept by the investigator

- To prepare necessary documents and submit them to the sponsor if any health hazard occurs
- To retain study related important correspondence records with the sponsor including letters, meeting minutes, and telephone communications
- To unblind the study treatment in an emergency

24. References

1) ROME III Japanese Translation. Translated by Fukushi Shin, Hongo Michio, and Matueda Kei. Tokyo, Kyowa Kikaku, 2008

2) Uesaka Hiroyuki: Perspective and Method for Deciding Sample Size in Clinical Studies. Biometrics, 2003, 17-41, Analysis Sets