Official Title: A Multicenter, Randomized, Double-Blind, Placebo Controlled Phase Ib Study to Investigate the Safety, Tolerability, Pharmacokinetics, Preliminary Efficacy, and Pharmacodynamics of Subcutaneously Administered RO7049665 in Participants With Active Ulcerative Colitis

NCT Number: NCT03943550

Document Date: Protocol Version Version 5: 20 Nov 2020

PROTOCOL

TITLE:	**A MULTICENTER, RANDOMIZED, DOUBLE-BLIND, PLACEBO CONTROLLED PHASE IB STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, PHARMACOKINETICS, PRELIMINARY EFFICACY, AND PHARMACODYNAMICS OF SUBCUTANEOUSLY ADMINISTERED RO7049665 IN PARTICIPANTS WITH ACTIVE ULCERATIVE COLITIS**
PROTOCOL NUMBER:	WP40161
VERSION:	5
EUDRACT NUMBER:	2017-004599-74
IND NUMBER:	137,492
TEST PRODUCT:	RO7049665
SPONSOR:	F. Hoffmann-La Roche Ltd
DATE FINAL:	Version 1: 15 December 2017
	Version 2: 08 May 2018
	Version 3: 15 October 2018
	Version 4: 29 May 2019
DATE AMENDED:	Version 5: See electronic date stamp below

FINAL PROTOCOL APPROVAL

Date and Time (UTC)	Title	Approver's Name
20-Nov-2020 17:19:45	Company Signatory	[redacted]

PROTOCOL ACCEPTANCE FORM

TITLE:	**A MULTICENTER, RANDOMIZED, DOUBLE-BLIND, PLACEBO CONTROLLED PHASE IB STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, PHARMACOKINETICS, PRELIMINARY EFFICACY, AND PHARMACODYNAMICS OF SUBCUTANEOUSLY ADMINISTERED RO7049665 IN PARTICIPANTS WITH ACTIVE ULCERATIVE COLITIS**
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I agree to conduct the study in accordance with the current protocol.

_____

Principal Investigator's Name (print)

_____ _____

Principal Investigator's Signature Date

Please keep the signed original form in your study files, and return a copy to your local Study Monitor.

PROTOCOL AMENDMENT, VERSION 5:
RATIONALE

Protocol WP40161 Versions 4 has been amended and the rationale for each of the proposed changes is listed below.

- New dose level: dose level 3, 15 mg, Q2W
  In order to fully characterize the pharmacokinetic (PK), pharmacodynamics and safety profile of RO7049665, the Sponsor proposes to escalate to 15 mg RO7049665, maintaining the current biweekly (every 2 weeks [Q2W]) dose-administration regimen. The safety and tolerability of participants who have received doses up to 7.5 mg Q2W RO7049665 to date is considered to be acceptable with the majority of the adverse events (AEs) being mild to moderate. One participant has had AEs of injection-related reaction (IRR) and hypersensitivity (Grade 2). Eosinophil blood cell counts increase has occurred in the majority of treated participants, with one participant so far at 7.5 mg Q2W experiencing a severe AE of eosinophilia (Grade 3). The Grade 3 eosinophilia resolved with treatment and the participant was found to have preexisting untreated thyroiditis. Although there are dose-dependent effects on the eosinophil blood cell counts, to date no patient in study WP40161 has met the stopping criterion. The RO7049665 exposure from 15 mg Q2W dosing is expected to remain at least 2-fold below the corresponding exposure at the no-observed-adverse-effect level (NOAEL) of the 13-week Good Laboratory Practice toxicity study in non-human primates. Expected increases of regulatory T cells (T_{regs}) and effector T cells (T_{effs}) counts in blood are predicted to remain within acceptable limits. There is no clear relationship between anti-drug antibodies (ADA) and dose based on the current data (Sections 2.2.2.1., 2.3, 4.1, 4.3, Table 5 of Section 6.1).

- Removal of stopping criterion (absolute T_{reg} cell count)
  The stopping criterion relating to an absolute T_{reg} increase of $>$ 450 cells/μL within a dose level was previously implemented in response to a request from the Paul-Ehrlich-Institute (the German Health Authority) prior to availability of any multiple dose data with RO7049665 and in the absence of data on potential accumulation (Section 4.1.3.). Study WP40161 recently met this stopping criterion at the 7.5 mg dose level as described in Version 4 of the study protocol. An increase in T_{reg} cells reflects the primary mode of action of RO7049665 and is thus an expected and a desired effect. The current cutoff of $>$ 450 cells/μL is **not** considered by the Sponsor to be a safety limit. To the Sponsor's knowledge there is no published threshold beyond which T_{reg} numbers in humans are considered a risk to human health or lead to a clinically meaningful impairment of the normal immune response. Recent data from the ongoing study WP40161 supports the absence of AEs related to increased T_{reg} counts and there has been no T_{reg} accumulation observed with Q2W dosing. Therefore, the stopping criterion has been removed from the protocol.

- Extended safety observation period for the new dose level
  The safety observation period of the first 3 participants at a new dose level remains the same while the observation period between the third and the fourth participants of the dose level has been extended to (a minimum of) 14 days. This

longer interval between first doses will allow the review of the safety laboratory data prior to enrolling and dosing subsequent participants (Section 2.3 and 4.1).

- Safety-management changes
  The protocol has been updated following IRR and hypersensitivity AEs in one participant receiving 7.5 mg Q2W RO7049665/ placebo to include the management of participants who experience these type of reactions (Sections 8.3.6. and 8.3.8). This is in line with a previously issued Dear Investigator Letter and includes a new individual participant stopping criterion (Section 4.1.4. and 4.1.5) and suggested pre-medication (Sections 6.5.1. and 8.3.8).

- Change in sample size
  Due to the new dose level the current sample size needs to be increased as the first cohorts have enrolled 10 or more participants per cohort. The sample size has therefore been adjusted to up to 65 participants (Section 9.2).

- Alignment of assessments and additional safety monitoring (study day 50)
  For additional safety monitoring measures at 15 mg Q2W, and to ensure that the eosinophil blood cell count can be followed in parallel to the blood flow cytometry, safety laboratory assessments (includes hematology, blood biochemistry and urinalysis) have been included at study Day 50 for participants receiving 15mg RO7049665 Q2W (Schedule of Activities [SoA], Section 1.3).

- Updates in case of temporary interruptions to study treatment administration
  The temporary interruption and study assessments and procedure sections (Sections 7.1.1. and 8) have been updated for greater clarity and to allow the participants in case of a dose interruption to continue the visit schedule, although assessments and sampling specifically related to dosing can be omitted.

- Other minor modifications
  Additional minor changes have been made to improve clarity and consistency, including an update of background sections of the protocol in line with recent data of the study and changes to the Investigator Brochure, and an update of SoAs, study figure (Figure 1) and SoA footnotes. Time windows when collecting PK samples, vital signs and electrocardiograms post-dose have been added as per site feedback. Additionally it has been clarified that ova and parasite stool examination at screening may be done locally at sites having the possibility. Following the outbreak of coronavirus disease 2019, a statement on impact to study WP40161 and the measures taken to date have been added to the benefit and risk section (Section 2.3).

Substantial new information appears in *Book Antiqua* italics. This amendment represents cumulative changes to the original protocol.

The Master ICF has been updated to accommodate the changes of the study protocol.

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LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation	Definition
5-ASA	5-aminosalicylate
6-MP	6-mercaptopurine
ADA	Anti-drug antibody
AE	Adverse event
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
ANCOVA	Analysis of Covariance
ANOVA	Analysis of Variance
ASA	Aminosalicylate
AST	Aspartate aminotransferase
AUC	Area under the curve
AUC_{inf}	Area under the serum concentration-time curve extrapolated to infinity
AUC_{tau}	Area under the serum concentration-time curve over the dosing interval
BMI	Body mass index
BP	Blood pressure
C_{max}	Maximum serum concentration observed
CL/F	Apparent clearance
CLS	Capillary leak syndrome
COA	Clinical outcome assessments
CRF	Case report form
CRO	Contract research organization
CSR	Clinical study report
CTCAE	Common terminology criteria for adverse events
DNA	Deoxyribonucleic acid
EC	Ethics Committee
ECG	Electrocardiogram
eCRF	Electronic case report form
EDC	Electronic data capture
EEA	European Economic Area
EIH	Entry-in-Human
ELISA	Enzyme-linked immunosorbent assay
ESF	Eligibility screening form
EU	European Commission
FDA	Food and Drug Administration

Abbreviation	Definition
FMT	Fecal microbiota transplant
GGT	Gamma-glutamyl transferase
GLP	Good laboratory practice
GS	Geboes Score
HBsAg	Hepatitis B surface antigen
HBcAb	Total hepatitis B core antibody
HCV	Hepatitis C virus
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human immunodeficiency virus
ICF	Informed Consent Form
ICH	International Conference on Harmonisation
IEC	Independent Ethics Committee
IFN	Interferon
IHC	Immunohistochemistry
IL	Interleukin
IL-2R	Interleukin-2 receptor
IMP	Investigational medicinal product
IND	Investigational New Drug (application)
INR	International normalized ratio
IRB	Institutional Review Board
IRR	Injection-related reaction
ISR	Injection-site reaction
IVRS/IWRS	Interactive (voice/web) response system
LPLO	Last participant, last observation
LRG	Leucine-rich alpha-2 glycoprotein
MAD	Multiple ascending doses
MCS-ES	Endoscopy Subscore of the Mayo Clinic Score
MP	6-Mercaptopurine
NCI	National Cancer Institute
NCI-CTCAE	National Cancer Institute Common Terminology Criteria for Adverse Events
NGS	Next generation sequencing
NHI	Nancy Histology Index
NK	Natural killer cell
NOAEL	No-observed-adverse-effect level
NSAESI	Non-serious adverse event of special interest
OTC	Over-the-counter

Abbreviation	Definition
pANCA	Perinuclear anti-neutrophil cytoplasmic antibody
PD	Pharmacodynamic
PK	Pharmacokinetic
PR	PR Interval
pSTAT5	Phosphorylated signal transducer and activator of transcription 5
RHI	Robarts Histology Index
QRS	QRS complex
QT	QT interval
QTc	QT corrected for heart rate
QTcF	QT corrected for heart rate using the Fridericia's correction factor
RBC	Red blood cell
RBR	Research biosample repository
RNA	Ribonucleic acid
RR	RR interval
SAD	Single ascending dose
SAE	Serious adverse event
SC	Subcutaneous
SoA	Schedule of activities
SOC	Standard of care
SOP	Standard operating procedure
SUSAR	Suspected unexpected serious adverse reactions
TOC	Test of cure
$T_{1/2}$	Half-life
T_{eff}	Effector T cell
T_{max}	Time to maximum concentration
T_{reg}	Regulatory T cell
TNF	Tumor necrosis factor
UC	Ulcerative colitis
UCEIS	Ulcerative colitis endoscopic index of severity
ULN	Upper limit of normal
V/F	Apparent volume of distribution
WBC	White blood cell
WES	Whole exome sequencing
WGS	Whole genome sequencing

1. PROTOCOL SUMMARY

1.1 SYNOPSIS

PROTOCOL TITLE:	**A MULTICENTER, RANDOMIZED, DOUBLE-BLIND, PLACEBO CONTROLLED PHASE IB STUDY TO INVESTIGATE THE SAFETY, TOLERABILITY, PHARMACOKINETICS, PRELIMINARY EFFICACY, AND PHARMACODYNAMICS OF SUBCUTANEOUSLY ADMINISTERED RO7049665 IN PARTICIPANTS WITH ACTIVE ULCERATIVE COLITIS**
SHORT TITLE	**Double-Blinded, Placebo-Controlled Phase 1b Study for Safety, PK, efficacy, PD of RO7049665 in Patients with UC**
PROTOCOL NUMBER:	WP40161
VERSION:	5
TEST PRODUCT:	RO7049665
PHASE:	Ib

RATIONALE

RO7049665 is a novel dimeric interleukin 2 (IL-2) mutein that is being developed as a therapy for autoimmune conditions, including ulcerative colitis (UC). It is expected to treat the impaired regulatory T cell (T_{reg}) numbers and/or function associated with autoimmune diseases without the broad based immuno-stimulatory effects of IL-2. Specifically, RO7049665 was engineered to address and overcome the pharmacologic, safety and functional liabilities of Proleukin© (aldesleukin), a recombinant human IL-2 therapy currently approved for the treatment of renal cell carcinoma and malignant melanoma.

This study (WP40161) will build upon experience gained in the healthy volunteer entry-in-human (EIH) single ascending dose (SAD) study (WP39826). In study WP40161, RO7049665 will be administered in multiple doses and to participants with an autoimmune condition for the first time. The principal aim of this study is to evaluate the safety and tolerability of RO7049665 in participants with active ulcerative colitis. In addition, the pharmacokinetics (PK), preliminary efficacy, pharmacodynamics (PD) and immunogenicity will also be explored. The study results will be used to support further clinical development of RO7049665.

OBJECTIVES AND ENDPOINTS

Objectives	Endpoints
Primary	
• To evaluate the safety and tolerability of repeat dosing of subcutaneous (SC) injections of RO7049665 in participants with active ulcerative colitis (UC).	• Incidence of dose-limiting or intolerable treatment-related adverse events (AEs). • Incidence, severity, and relationship of treatment-emergent AEs. • Incidence of abnormal laboratory findings. • Incidence of abnormal vital signs and electrocardiogram (ECG) parameters.
Secondary	
• To investigate the multiple-dose pharmacokinetics of RO7049665.	• Pharmacokinetic parameters of RO7049665.
• To investigate the effects of multiple doses of RO7049665 on changes in endoscopic appearance of the mucosa of the sigmoid colon.	• Change from baseline in the Endoscopy Subscore of the Mayo Clinic Score (MCS-ES) at Day 29 and Day 57. • Change from baseline in the Ulcerative Colitis Endoscopic Index of Severity (UCEIS) at Day 29 and Day 57.
• To investigate the effects of multiple doses of RO7049665 on changes in histologic appearance of mucosa in biopsies from the sigmoid colon.	• Change from baseline to Day 29 and Day 57 in histology score of sigmoid colon biopsies (Geboes Score [GS], Robarts Histology Index [RIH], or Nancy Histology Index [NHI]).
• To investigate the effects of multiple doses of RO7049665 on changes in disease activity.	• Change from baseline in the Mayo Clinic Score (MCS) at Day 29 and Day 57.
• To evaluate the immunogenicity of RO7049665.	• Incidence of anti-drug antibodies (ADAs).
• To investigate the effects of multiple doses of RO7049665 on immune cells in the periphery and in mucosal biopsies of the sigmoid colon.	• Quantitative changes in WBCs (T_{regs}, T_{effs}, natural killer [NK] cells, B cells and eosinophils).

OVERALL DESIGN

Study Design

- Study WP40161 is a multicenter, randomized, adaptive, and placebo-controlled study to investigate the safety, tolerability, pharmacokinetics (PK), preliminary efficacy, and pharmacodynamics (PD) of multiple ascending doses (MAD) of RO7049665 on top of standard of care (SOC) in participants with endoscopic evidence of moderate to severely active UC. Participants may be enrolled if they have a history of anti-TNFα or anti-integrin therapy but may not be actively treated with these agents during the study.
- Investigator and participant blind.
- Participants within each cohort will be randomized in a 4:1 ratio to receive multiple SC injections of RO7049665 or placebo, respectively. Randomization occurs on Study Day -1, after a maximum 35-day screening period.

Treatment Groups and Duration

The investigational medicinal products are: RO7049665 and Placebo.

RO7049665 (5 mg/mL) and Placebo will be provided by the Sponsor in 2 mL colorless glass vials. Both must be prepared for SC injection in the abdomen under appropriate aseptic conditions and the solution for injection must be filtered prior to use. Both should be used immediately.

The dosing interval will be every 2 weeks (Q2W) for 4 doses. Participants enrolled in dose level 1 cohorts will receive 3.5 mg of RO7049665 (or placebo), participants enrolled in dose level 2 cohorts will receive 7.5 mg of RO7049665 (or placebo)*, and participants enrolled in the cohorts of dose level 3 will receive 15 mg of RO7049665 (or placebo)*, based on data from study WP39826 (EIH study in healthy volunteers) *and review of data from completed cohorts in this study*.

Safety and tolerability, ADA, PK, and selected PD data from at least 8 weeks of treatment in at least 5 participants (4 active, 1 placebo) at *a* dose level will be reviewed prior to enrolling participants into the *next higher* dose level *for the first time* (i.e., 7.5 mg *and 15 mg*).

Dose escalation and dose stopping rules are described in the protocol body.

Length of Study

The total duration of the study for each participant will be approximately 19 weeks, divided as follows:

- Screening: Up to 5 weeks (35 days).
- Dosing Period: Day 1 to Day 43.
- In-clinic period: From Day −1 to Day 2 for the first dose, from Day 15 to Day 16 for the second dose, from Day 29 to Day 30 for the third dose, and from Day 43 to Day 44 for the last dose.
- Final follow-up: Day 99 ± 3 days (8 weeks post last dose).

End of Study

A participant is considered to have completed the study if he/she has completed all phases of the study including the last scheduled procedure shown in the Schedule of Activities.

The end of the study is defined as the date when the last participant last observation (LPLO) occurs. LPLO is expected to occur approximately 56 (± 3) days after last study drug administration.

Internal Monitoring Committee:

There will be a dedicated group of individuals from the Sponsor functioning as an internal monitoring committee (IMC), which will be independent to the project. The roles, responsibilities, membership, scope of activities, time of meetings and communication plan for the IMC will be documented in an appropriate charter.

PARTICIPANT POPULATION

This study will enroll *up* to *65* participants with UC between 18 and 70 years of age, inclusive, with evidence of moderate to severely active disease (MCS ≥ 5: MCS-ES ≥2, stool frequency subscore ≥ 1, and rectal bleeding subscore ≥ 1), who have failed at least one prior standard of care therapy.

Inclusion Criteria

Participants are only eligible to be included in the study if all of the following criteria apply:

Informed Consent

1. Able and willing to provide written informed consent and to comply with the study protocol according to International Conference on Harmonisation (ICH) and local regulations. Informed consent may be obtained up to 3 months prior to randomization. A signed Informed Consent Form must be available from the participant before starting any study-specific assessments.

Age

2. Between 18 to 70 years of age inclusive, at the time of signing the informed consent.

Type of Participants and Disease Characteristics

3. Diagnosed with UC for at least 12 weeks prior to screening
4. Screening colonoscopy for colorectal cancer conducted within the prior two years if:
   - History of pancolitis and disease duration $\geq$ 8 years, or
   - History of left-sided colitis and disease duration $\geq$ 12 years.
5. Evidence of disease activity at time of screening as measured by MCS $\geq$ 5, with MCS-ES $\geq$ 2, extending at least 10 cm from the anal verge, stool frequency subscore $\geq$ 1, and rectal bleeding subscore $\geq$ 1.
6. Insufficient clinical response to standard of care (SOC) therapy or intolerance to SOC, where SOC may include (but not limited to):
   - Glucocorticoids (Prednisolone at least 0.75 mg/kg/day or equivalent for at least 4 weeks).
   - Treatment with JAK inhibitors of at least 8 weeks duration.
   - Immunomodulators (e.g., azathioprine, methotrexate, oral 5-aminosalicylate [5-ASA] or 6-mercaptopurine) at a standard therapeutic dose for at least 12 weeks.
   - Therapy with an anti-TNFα monoclonal antibody of at least 3 months duration, or intolerance due to side effects of medication (not including anaphylaxis).
   - Treatment with anti-integrin therapy of at least 14 weeks duration.

Weight

7. Body mass index (BMI) within the range of 18-35 kg/m2 (inclusive).

Sex

8. Male and female participants

   The contraception and abstinence requirements are intended to prevent exposure of an embryo to the study treatment. The reliability of sexual abstinence for male and/or female enrollment eligibility needs to be evaluated in relation to the duration of the clinical study and the preferred and usual lifestyle of the participant. Periodic abstinence (e.g., calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are not acceptable methods of contraception.

   Female participants:

   Compliance with contraception requirements:

   - Women of non-child bearing potential (WONCBP) may be enrolled. A woman is considered to be of childbearing potential if she is postmenarchal, has not reached a post-menopausal state ($\geq$ 12 continuous months of amenorrhea with no identified cause other than menopause), and has not undergone surgical sterilization (removal of ovaries and/or uterus).

For women of childbearing potential (WOCBP):

- May enroll if not pregnant and not breastfeeding.
- Agree to remain abstinent (refrain from heterosexual intercourse) or use contraceptive methods that result in a failure rate of < 1% per year during the treatment period and for at least 56 days after the last dose of study drug.

  Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal occlusion/ligation, male sterilization, proper use of hormonal contraceptives that inhibit ovulation, hormone-releasing intrauterine devices, and copper intrauterine devices.
- Have a negative pregnancy test (blood) within the 35 days prior to the first study RO7049665/placebo administration.

For men:

During the treatment period and for at least 56 days after the last dose of RO7049665/placebo, agreement to:

- Remain abstinent (refrain from heterosexual intercourse) or use contraceptive measures such as a condom plus an additional contraceptive method that together result in a failure rate of < 1% per year, with partners who are women of childbearing potential (WOCBP).

  With pregnant female partners, remain abstinent (refrain from heterosexual intercourse) or use contraceptive measures such as a condom to avoid exposing the embryo.
- Refrain from donating sperm for 56 days after the last dose of RO7049665 or placebo.

Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1. Diagnosis of Crohn's disease or indeterminate colitis
2. History of infection with, hepatitis B, human immunodeficiency virus (HIV), active hepatitis C virus (HCV) infection, or other chronic infection.
3. Active infections requiring systemic therapy with antibiotic, antiviral or antifungal or febrile illness within 7 days before Day -1.
4. History of primary or acquired immunodeficiency
5. History of chronic pulmonary disease with resultant abnormal pulmonary function
6. History of clinically significant cardiac or cardiovascular disease or uncontrolled hypertension
7. Evidence of infection with *Clostridium difficile* or other intestinal pathogen within 35 days from the start of screening.
8. Evidence of colonic dysplasia that cannot be completely removed
9. Females: Pregnant or lactating.
10. History of and/or current suicidal ideations
11. Any condition or disease detected during the medical interview/physical examination that would render the patient unsuitable for the study, place the patient at undue risk or interfere with the ability of the patient to complete the study in the opinion of the investigator, including malignant disease
12. Symptomatic herpes zoster within 3 months prior to screening.
13. History of tuberculosis or a positive Quantiferon® Gold test.

14. History of clinically significant severe drug allergies, multiple drug allergies, allergy to any constituent of the product, or intolerance to topical corticosteroids.
15. Lymphoma, leukemia, or any malignancy within the past 10 years, except for basal cell or squamous epithelial carcinomas of the skin that have been resected with no evidence of metastatic disease for 3 years and in situ carcinoma of the cervix that was completely removed surgically.
16. History or presence of clinically significant ECG abnormalities before study drug administration (e.g., PQ/PR interval $\geq$ 220 ms, QT corrected for heart rate using Fridericia's correction factor (QTcF) < 350 ms or $\geq$ 450 ms) or clinically significant cardiovascular disease (e.g., cardiac insufficiency, coronary artery disease, cardiomyopathy, congestive heart failure, family history of congenital long QT interval syndrome, family history of sudden death).
17. Fecal Microbiota Transplant (FMT), defined as receipt of any product derived from the feces of another human and administered per oral, per nasogastric or nasoduodenal, or per rectum within the last 6 months.

Prior/Concomitant Therapy

18. Use of calcineurin inhibitors (e.g., tacrolimus, cyclosporine), vedolizumab, anti-TNF-α therapeutic or any other immune system–targeted biological therapy within 12 weeks or 5 half-lives, whichever is longer, prior to screening
19. Rectal therapy with 5-ASA or corticosteroids within 2 weeks of screening
20. Use of any investigational drug within 12 weeks of screening
21. Leukocyte apheresis within 12 weeks of screening
22. Live vaccine(s) within one month prior to screening, or plans to receive live vaccines during the study or within 28 days of the last dose.

Prior/Concurrent Clinical Study Experience

23. Donation of blood or blood products in excess of 500 mL within 3 months of screening.
24. Exposure to more than 4 investigational treatments within 12 months prior to Day 1.
25. Current enrollment or past participation within the 90 days immediately preceding signing of informed consent in this or any other clinical study involving an investigational study treatment or any other type of interventional medical research.

Diagnostic Assessments

26. Abnormal hematologic values:
    - Anemia (Hgb $<$ 9 g/dL)
    - Leukocytosis (WBC $\geq 2\times$ ULN)
    - Thrombocytopenia (platelet count $<$ 100,000/μL)
    - Thrombocytosis (platelet count $\geq 2 \times$ ULN)
    - Eosinophilia (eosinophil count $\geq 2 \times$ ULN)
27. Abnormal hepatic enzyme or hepatic function values:

- Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), alkaline phosphatase, or Gamma-glutamyl transferase (GGT) $\geq 2 \times$ ULN
- Total bilirubin $\geq 2 \times$ ULN
- International normalized ratio (INR) ≥ 1.2
- Albumin < 3g/dL

28. Positive HIV antibody test.
29. Presence of hepatitis B surface antigen (HBsAg) or positive for total hepatitis B core antibody (HBcAb), or positive hepatitis C by PCR test result at screening or within the 3 months prior to starting study treatment.

Other Exclusions

30. History of regular alcohol consumption within 2 months of screening defined as:

    An average weekly intake of $>$ 14 drinks for males or $>$ 7 drinks for females. One drink is equivalent to 12g of alcohol: 12 ounces (360 mL) of beer, 5 ounces (150 mL) of wine or 1.5 ounces (45 mL) of 80 proof distilled spirits.
31. Any suspicion or history of alcohol abuse and/or suspicion of regular consumption of drug of abuse
32. Patients under judicial supervision, guardianship or curatorship

NUMBER OF PARTICIPANTS

The study will enroll *up* to *65* participants to assess the safety and tolerability of repeated dosing of RO7049665 (approximately 10 participants, randomized in the ratio of 4:1 RO7049665 to placebo, per cohort).

CONCOMITANT MEDICATIONS

Permitted Therapy

All concomitant medications throughout the duration of the study should be recorded in the case report form. Use of the following therapies will be permitted, as specified below:

- Paracetamol is allowed up to a maximum dose of 2 g/day up to 48 hours prior to each RO7049665 or placebo dosing, not to exceed 4 g total during the week prior to each dosing.
- Participants who use the following therapies prior to screening and are on a stable regimen should continue their use:
  - Oral contraceptives, hormone-replacement therapy, or other maintenance therapy.
- Participants who are taking non-biologic SOC therapy for UC during the study will remain on their SOC therapy. Specific instructions for each SOC is provided below:
  - Stable therapy for at least 2 weeks prior to screening with:
    - oral 5-ASA, or
    - oral glucocorticoid (not to exceed 20 mg/day prednisone equivalent at start of trial, may be weaned after dosing with study drug has been initiated).
  - Stable therapy for at least 8 weeks with:
    - azathioprine, or
    - 6-mercaptopurine (MP), or
    - methotrexate (and folic acid)

- *In the event of a systemic injection-related reaction (IRR) or systemic hypersensitivity AE: participants may receive pre-medication with paracetamol and antihistamines (as per local guidelines) prior to the next administration of study drug.*

Prohibited Therapy

As a general rule, no concomitant medication will be permitted, with the exception of medications to treat AEs and those specified as permitted therapies, unless the rationale for exception is discussed and clearly documented between the Investigator and the Sponsor.

Use of the following therapies will be prohibited during the study and for at least 14 days or at least 5 half-lives prior to initiation of study treatment, whichever is longer unless otherwise specified below:

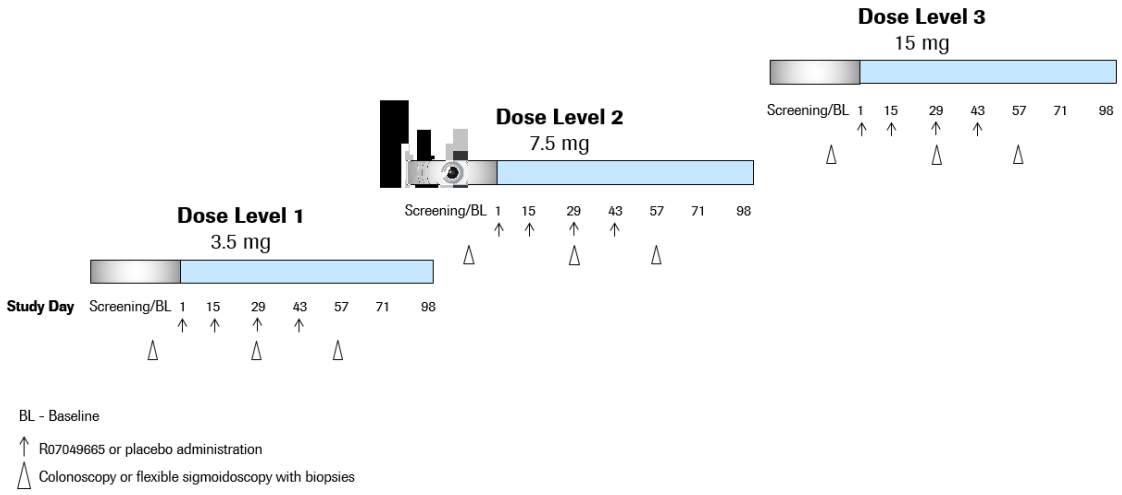
- Any rectal therapy
- Doses of glucocorticoids exceeding 20 mg/day of prednisone or equivalent
- Non-steroidal anti-inflammatory medications (e.g.: ibuprofen, naproxen) due to a propensity for these treatments to worsen inflammation in ulcerative colitis.

Calcineurin inhibitors (e.g., tacrolimus, cyclosporine) and any immune system targeted biological therapy within 12 weeks or 5 half-lives, whichever is longer, before the start of screening.

1.2 SCHEMATIC OF STUDY DESIGN

An overview of the study design is provided in Figure 1.

Figure 1 Overview of Study Design



The doses for dose level 1 (3.5 mg) and dose level 2 (7.5 mg) are based on results from the entry-in-human (EIH) Study WP39826, which supported safety, pharmacokinetics, and pharmacodynamics in healthy volunteers in single dosing. *Dose level 3 (15 mg) is based on initial analyses of results from repeat dosing with 3.5 and 7.5 mg in participants with ulcerative colitis in this study.*

1.3 SCHEDULE OF ACTIVITIES

The schedule of activities is provided in Table 1 and Table 2.

Table 1 Schedule of Activities – Main Table

Activity	Screening / Baseline[a]	Dose 1			Dose 1 Outpatient Visits				Dose 2		Outpatient Phone Call	Outpatient visit	Dose 3		Outpatient Phone Call	Outpatient visit
Overall Study Day	D -36 to D -2	D -1	Day 1	Day 2	Day 4	Day 6	Day 8	Day 10	Day 15	Day 16	Day 19	Day 22	Day 29	Day 30	Day 33	Day 36
Dose/Day		Dose 1/Day -1	Dose 1/Day 1	Dose 1/Day 2	Dose 1/Day 4	Dose 1/Day 6	Dose 1/Day 8	Dose 1/Day 10	Dose 2/Day 1	Dose 2/Day 2	Dose 2/Day 5	Dose 2/Day 8	Dose 3/Day 1	Dose 3/Day 2	Dose 3/Day 5	Dose 3/Day 8
Time Relative (h)	***	***	0	24	72	120	168	216	336	360	432	504	672	696	768	840
Visit Window							(+/-1)	(+/-1)			(+/-1)	(+/-1)	*(+/-1)*		(+/-1)	(+/-1)
Assessments																
Informed Consent[b]	X															
Demography	X															
Medical History	X															
Randomization[c]		X														
Physical Examination[d]	X	X	X	X					X				X			
Vital Signs[e]	X	X	2	X					X	X			X	X		
Eligibility	X	X														
Tuberculosis Screening Test[f]	X															
Clinical Genotyping[g]			X													
ECG-12 lead[h]	X		2	X					X				X			
Hematology[i]	X	X		X			X		X			X	X			X
Blood Chemistry[i]	X	X		X			X		X			X	X			X
Urinalysis[i]	X	X		X			X		X			X	X			X
Serology and viral PCR[j]	X															
Pregnancy Testing[k]	X	X							X				X			
Coagulation	X															
Alcohol breath or blood test	X	X														
Drugs of abuse[l]	X	X														
Colonoscopy or flexible sigmoidoscopy[m]	X												X			
Colon Biopsy	X												X			
Stool *Clostridium difficile*[n]	X															
Stool Ova and Parasites	X															
In-house period		X	X	X					X	X			X	X		
Drug/Placebo Administration			X						X				X			
Standard meal[o]		X	X	X					X	X			X	X		
PK sampling[y]			3	X	X	X	X	X	2			X	X			X
Blood flow cytometry		X	X	X		X	X	X	X			X	X			X
Blood epigenetic testing			X			X	X	X	X							
Soluble PD biomarkers[p]			X	X	X	X	X		X				X			
Soluble disease biomarkers[q]			X	X					X				X			
sCD25[w]			X	X		X	X		X				X			
Fecal Calprotectin[v]			X						X							
Abdominal skinfold thickness		X														
Anti-drug antibodies		X					X		X			X	X			
Local pain/Skin reactivity assessments[r]			X						X				X			
Clinical Outcome Assessment[s]	X		X				X		X			X	X			X
Safety Phone Call[t]											X				X	
Safety biomarkers[u]																
Adverse Events	X															
Previous and Concomitant Treatments	X															

Table 1 Schedule of Activities – Main Table (cont.)

Activity	Dose 4		Dose 4 Outpatient Visits						Follow Up visit	Outpatient visit	Hypersensitivity/ IRR (≥ Grade 2)	Early Termination Visit
Overall Study Day	Day 43	Day 44	Day 46	Day 48	Day 50	Day 52	Day 57	Day 71	Day 99[w]	Unscheduled visit	Unscheduled visit	Unscheduled visit
Dose/Day	Dose 4/Day 1	Dose 4/Day 2	Dose 4/Day 4	Dose 4/Day 6	Dose 4/Day 8	Dose4/Day10	Dose 4/Day 15	Dose 4/Day 29				Unscheduled visit
Time Relative (h)	1008	1032	1080	1128	1176	1224	1344	1680	2376	Anytime	Anytime	Anytime
Visit Window					(+/- 1)	(+/-1)	(+/-1)	(+/-1)	(+/-3)			
Assessments												
Informed Consent[b]												
Demography												
Medical History												
Randomization[c]												
Physical Examination[d]	X								X	X	X	
Vital Signs[e]	2	X							X	X	X	X
Eligibility												
Tuberculosis Screening Test[f]												
Clinical Genotyping[g]												
ECG-12 lead[h]	2	X							X	X		X
Hematology[i]	X				X[x]		X	X	X	X	X	X
Blood Chemistry[i]	X				X[x]		X	X	X	X	X	X
Urinalysis[i]	X				X[x]		X	X	X	X	X	X
Serology and viral PCR[j]												
Pregnancy Testing[k]	X								X	X		X
Coagulation									X	X	X	X
Alcohol breath or blood test										X		
Drugs of abuse[l]										X		
Colonoscopy or flexible sigmoidoscopy[m]							X					
Colon Biopsy							X					
Stool *Clostridium difficile*[n]												
Stool Ova and Parasites												
In-house period	X	X										
Drug/Placebo Administration	X											
Standard meal[o]	X	X										
PK sampling[y]	3	X	X	X	X	X	X	X	X	X	X	X
Blood flow cytometry	X			X	X	X	X	X		X		
Blood epigenetic testing	X			X	X	X	X	X		X		
Soluble PD biomarkers[p]	X	X	X	X			X			X		
Soluble disease biomarkers[q]	X	X					X			X	X	
sCD25[w]	X	X		X	X			X	X	X		
Fecal Calprotectin[v]	X				X[v]							
Abdominal skinfold thickness												
Anti-drug antibodies	X						X	X	X	X	X	X
Local pain/Skin reactivity assessments[r]	X								X	X	X	X
Clinical Outcome Assessment[s]	X				X		X	X	X	X		X
Safety Phone Call[t]												
Safety biomarkers[u]											X	
Adverse Events	X											
Previous and Concomitant Treatments	X											

Table 1 Schedule of Activities – Main Table (cont.)

a) A 35-day maximum screening period will begin with the first assessment, not from consent.

b) Participants can be consented up to 3 months prior to randomization.

c) Randomization may be conducted on Day -1 or Day 1 prior to first dose.

d) A complete physical examination will be performed at screening (including height and weight), and abbreviated physical examinations at all other visits (no height measurement needed; weight only).

e) Single measurements of vital signs to be taken before blood collection for laboratory tests but after ECG collection when scheduled at the same time point. Includes single measurements of systolic and diastolic blood pressure, pulse rate, body temperature (tympanic) and respiratory rate. All measurements to be taken after the participant has rested in a supine position for at least 5 minutes. *Vital sign assessments post-dose can be done with a deviation of ± 15 minutes.*

f) QuantiFERON-TB Gold testing should be performed.

g) Clinical genotyping is mandatory in countries where permitted.

h) ECGs should be performed prior to any scheduled vital sign measurements and blood draws. 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2 - 5 minute interval) after the participant has been in a supine position for at least 10 minutes. Automated ECG intervals (PR [PQ], QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented. Measure baseline prior to endoscopy if endoscopy performed on the same day. *ECG assessments post-dose can be done with a deviation of ± 15 minutes.*

i) Safety tests can be repeated during screening period at discretion of Investigator.

j) Hepatitis B and HIV 1/2 antibody testing; HCV PCR.

k) Serum beta-human chorionic gonadotropin (β-HCG) at screening, urine on all other occasions (women of childbearing potential only).

l) Drugs of abuse screen to include at minimum: amphetamines, barbiturates, cocaine, opiates, and benzodiazepines. Additional testing optional to local standards.

m) Endoscopy to be performed in the morning prior to dose on Day 29.

n) *Clostridium difficile* ELISA.

Table 1 Schedule of Activities – Main Table (cont.)

o) Standard breakfast, lunch, dinner and snack to be provided at the times deemed convenient by the site.

p) For exploratory PD cytokines and other soluble markers.

q) LRG and ANCA screen. Based on outcome of ANCA screen, p-ANCA, c-ANCA, and atypical p-ANCA may be tested. LRG is optional.

r) And at additional regular intervals at the Investigator's discretion if an adverse event associated with local tolerability and pain is observed.

s) Partial Mayo Score.

t) Site personnel will contact the participant. If deemed required by the Investigator, participants can be asked to come to the site for an unscheduled visit.

u) IgE, tryptase *and* exploratory safety biomarkers may be tested in case of participants experiencing signs and symptoms associated with injection-related reactions *(IRRs)*. This includes hypersensitivity reactions.

v) The Day 50 stool sample for measuring fecal calprotectin can be taken +/- 2 days. The other stool samples should be collected +/- 1 day.

w) Participants positive for anti-drug antibodies (ADA) will be requested to return and have additional ADA and sCD25 samples to monitor off-treatment ADA persistence.

x) *Safety laboratory assessments on Day 50 are only for participants enrolled in dose level 3 (15 mg, Q2W).*

y) *PK samples collected within 24 hours post-dose, can be taken with a deviation of ± 15 minutes for samples taken up to 6 hours after dosing with RO7049665/ placebo, and for samples thereafter with a deviation of ± 30 minutes.*

Table 2 Schedule of Activities - Detailed Table

Visit	Day	Scheduled Time (h)	Vital Signs[b]	Physical Examination[c]	ECG-12 lead[d]	PK Sample[e]	ADA	Blood Flow Cytometry	Blood Epigenetic Testing	sCD25	Local Pain and Skin Reactivity
Dose 1 in-house period[a]	Day -1		x	x			x	x			
	Day 1	Predose	x		x	x		x	x	x	
		0									
		2	x								
		6		x		x					
		12			x	x					x
	Day 2	24	x	x	x	x		x		x	
Dose 1 outpatient visits	Day 4					x					
	Day 6					x		x	x	x	
	Day 8					x	x	x	x	x	
	Day 10					x		x	x		
Dose 2 in-house period	Day 15	Predose	x	x	x	x	x	x	x	x	
		0									
		12				x					x
	Day 16		x								
Dose 2 outpatient visit	Day 22					x	x	x			
Dose 3 in-house period	Day 29	Predose	x	x	x	x	x	x		x	
		0									
		12									x
	Day 30		x								
Dose 3 outpatient visit	Day 36					x		x			
Dose 4 in-house period	Day 43	Predose	x	x	x	x	x	x	x	x	
		0									
		2	x								
		6				x					
		12			x	x					x
	Day 44	24	x		x	x				x	
Dose 4 outpatient visits	Day 46					x					
	Day 48					x		x	x	x	
	Day 50					x		x	x	x	
	Day 52					x		x	x		
	Day 57					x	x	x	x		
	Day 71					x	x	x	x	x	
Follow Up visit	Day 99		x	x	x	x	x			x	x
Unscheduled visit	Anytime		x	x	x	x	x	x	x	x	x
Hypersensitivity/ IRR (≥ Grade 2)	Anytime		x	x		x	x				x
Early Termination Visit	Anytime		x		x	x	x				x

Table 2 - Schedule of Activities - Detailed Table – (cont.)

a) Randomization may be conducted on D-1 or D1 prior to first dose.

b) Single measurements of vital signs to be taken before blood collection for laboratory tests but after ECG collection when scheduled at the same time point. Includes single measurements of systolic and diastolic blood pressure, pulse rate, body temperature (tympanic) and respiratory rate. All measurements to be taken after the participant has rested in a supine position for at least 5 minutes. *Post-dose vital sign assessments can be done with a deviation of ± 15 minutes.*

c) A complete physical examination will be performed at screening (including height and weight), and abbreviated physical examinations at all other visits (no height measurement needed; weight only).

d) ECGs should be performed prior to any scheduled vital sign measurements and blood draws. 12-lead ECGs will be obtained in triplicate (three consecutive interpretable 12-lead ECGs within a 2-5 minute interval) after the participant has been in a supine position for at least 10 minutes. Automated ECG intervals (PR [PQ], QRS, QT, QTcF) and HR will be captured or calculated, and the changes in T-wave and U-wave morphology will be documented. Measure baseline prior to endoscopy if endoscopy performed on the same day. *Post-dose ECG assessments can be done with a deviation of ± 15 minutes.*

e) PK samples collected within 24 hours post-dose can be taken with a deviation of ± 15 minutes for samples taken up to 6 hours after dosing with RO7049665/ placebo, and for samples thereafter with a deviation of ± 30 minutes.

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2. INTRODUCTION

2.1 STUDY RATIONALE

RO7049665 is a novel dimeric interleukin 2 (IL-2) mutein that is being developed as a therapy for autoimmune conditions, including ulcerative colitis (UC). It is expected to treat the impaired regulatory T cell (T_{reg}) numbers or function associated with autoimmune diseases without the broad-based immunostimulatory effects of IL-2. Specifically, RO7049665 was engineered to address and overcome the pharmacologic, safety and functional liabilities of Proleukin® (aldesleukin), a recombinant human IL-2 therapy currently approved for the treatment of renal cell carcinoma and malignant melanoma.

Study (WP40161) has taken into consideration experiences gained in the healthy volunteer entry-in-human (EIH) single ascending dose (SAD) study WP39826. This is the first study where RO7049665 is administered in multiple doses and to participants with an autoimmune condition. The primary objective of this study is to evaluate the safety and tolerability of RO7049665 in participants with active UC. In addition, secondary objectives to be assessed are the pharmacokinetics (PK), preliminary efficacy, pharmacodynamics (PD) and immunogenicity. The study results will be used to support further clinical development of RO7049665.

The rationale for the study design is provided in Section 4.2.

2.2 BACKGROUND

2.2.1 Ulcerative Colitis

UC is a chronic, relapsing disease characterized by diffuse mucosal inflammation of the colon. Moderate to severe UC is a debilitating disease that can result in hospitalization for disease flares, potential bowel perforation or hemorrhage, and increased risk of colorectal cancer. Current goals of treatment include induction and maintenance of remission, improved quality of life, reduction in need for long-term corticosteroids, and minimization of cancer risk. Despite current medicinal and surgical treatments for UC (see Section 2.3), over 70% of patients with UC still lack a long-term efficacious therapy (Neurath 2017). Therefore, there is an urgent need for new treatments for UC in patients who do not respond to current treatments.

A loss of tolerance and a disruption in immune homeostasis are at the core of autoimmunity and have been strongly associated with reduced T_{reg} numbers and impaired function. In many autoimmune diseases such as UC, a numerically and/or functional deficiency of T_{reg}s is observed and considered a key factor for the failure to maintain immune-homeostasis and suppress auto-reactivity to self tissues. As a molecule critical for immune homeostasis, IL-2 plays a key role in maintaining a state of tolerance to “self” through its effects on the induction and activation of T_{regs}. Low-dose aldesleukin has shown some clinical efficacy in patients with graft-versus-host disease (GVHD), hepatitis C-induced vasculitis, and moderate-to-severe systemic lupus

erythematosus (SLE) and is currently being explored in patients with type 1 diabetes and UC.

2.2.2 RO7049665

RO7049665 is a novel IL-2 mutein immunocytokine. Compared to aldesleukin, RO7049665 has reduced binding affinity to IL-2 receptor (IL-2R)$\beta\gamma$ present on $CD4^+$ and $CD8^+$ effector T cells (T_{eff}) and natural killer (NK) cells, with no loss of binding affinity to IL-2Rα, a required component of IL-2R$\alpha\beta\gamma$ receptors present at the highest levels on T_{regs}. By virtue of its reduced affinity to IL-2R$\beta\gamma$ receptors and its T_{reg} selectivity, RO7049665 enhances the T_{reg}-supporting function of human IL-2 without the broad-based IL-2R$\beta\gamma$-mediated immunostimulatory effects of IL-2. RO7049665 therefore provides a novel selective immunotherapy to treat autoimmune diseases, including UC, with an expected improved safety profile compared to aldesleukin.

In a human whole blood assay, RO7049665 exhibited greatly reduced potency to induce pro-inflammatory cytokines compared to aldesleukin, with 600-fold lower potency for induction of Interferon (IFN)γ, Interleukin 6 (IL-6), tumor necrosis factor (TNF)α, and Interleukin 1 (IL-1)β and 5-fold lower potency for induction of Interleukin 8 (IL-8) compared to aldesleukin. A tissue cross-reactivity study of RO7049665 showed staining patterns consistent with known IL-2R expression profiles.

Observed toxicities in both 4-week and 13-week good laboratory practice (GLP) toxicology studies in cynomolgus monkeys treated with RO7049665 were consistent with increased pharmacological activity of IL-2, and included skin findings (at the injection site and generally), increased heart rate, changes in blood leukocyte numbers including increased eosinophils as well as microscopic findings of tissue infiltrates, and changes in lymphoid tissues. Findings were dose-dependent and had resolved or were trending to resolution following a 4-week recovery period. At all dose levels tested in both studies, there was no evidence for capillary leak syndrome (CLS), a known toxicity of high dose IL-2 therapy.

Following subcutaneous (SC) administration of RO7049665 in the 13-week GLP toxicology study, exposure to RO7049665 on Day 1 increased less than dose proportionally over the dose range of 0.2 mg/kg to 3 mg/kg. Rapid development of ADAs was observed at a low dose (0.2 mg/kg), leading to almost complete loss of exposure and loss of PD response by study Day 50 in the majority of animals. The no-observed-adverse-effect level (NOAEL) dose in the female animals was 0.2 mg/kg weekly that retained exposure to the drug until the dosing cycle at Day 22, with maximum serum concentration observed (C_{max}) and area under the curve $(AUC)_{0\text{-}168h}$ measured on Day 22 as 254 ng/mL and 8440 ng•h/mL respectively. At higher doses ($\geq$1 mg/kg), a subset of the animals (approximately 50%) did not develop high ADAs and remained exposed until the end of the study but developed generalized skin rash. Assessment of PD markers and immune-lineage cell counts in the 13 week GLP study confirmed induction of phosphorylated signal transducer and activator of

transcription 5 (pSTAT5) and other markers of activation of T_{reg}s, as well as expected changes of lymphocyte populations, including transient reduction followed by an increase of lymphocyte populations (total T cells, T_{regs}, $CD4^+$ T cells, $CD8^+$ T cells, NK cells, and B cells) and monocytes, with the increases of T_{reg}s being of greater magnitude than that of other T cell populations.

2.2.2.1 Previous Clinical Studies

Study WP39826

Study WP39826, the first clinical trial where RO7049665 was administered to humans, has completed dosing. The aim of this Entry-into-Human study was to investigate the safety, tolerability, PK and PD of single ascending doses of RO7049665 in a healthy male population. A total of 56 participants were administered single doses of RO7049665 or matching placebo injected subcutaneously in the abdomen.

A total of 9 doses were tested, ranging from 1.5 µg, which was the dose based on MABEL, up to 7500 µg. Thirty-eight participants (67.9%) received one SC injection of RO7049665, and 18 (32.1%) received placebo. Until Cohort 5, dose groups consisted of 3 participants on RO7049665 and 2 on placebo, split in a sentinel group of 2 participants (1 on active and 1 on placebo), and at least 48 hours later, the remaining participants of the group. The number of participants under active treatment was increased to 6 in Cohorts 6 to 9, to allow for a better characterization of the safety, PK and PD parameters. In Cohort 9, only 5 subjects were enrolled into the active treatment arm due to recruitment limitations.

Overall RO7049665 was well tolerated. A total of 121 treatment-emergent adverse events (TEAEs) were reported across the dose range tested, 94 (77.7%) in the active treatment groups, and 27 (22.3%) in the placebo group. Across all groups, the most frequently reported TEAEs were nasopharyngitis, injection site erythema, injection site pruritus, headache, diarrhea and myalgia. The majority of TEAEs were mild in intensity, with the exception of 5 events (1 event of gastroenteritis, 3 events of food poisoning, and 1 event of influenza-like illness) in 5 subjects on active treatment, rated as moderate in intensity but not related to the study drug by the Investigator of the study. There were no deaths or serious adverse events (SAE), and no withdrawals due to an adverse event (AE). There was no trend or pattern in the AEs observed with increasing doses or between active and placebo groups, with the exception of local injection site reactions (erythema, pruritus), observed in an increasing number of participants as the dose increased. All AEs related to the injection site were mild in intensity and resolved without treatment.

There were no trends observed in vital signs, electrocardiogram (ECG) intervals and lab safety results across the dose range tested.

Immunogenicity was assessed, and positive results for anti-drug antibody (ADA) were observed in a total of 12 participants starting from Cohort 5 (dose of 190 µg). ADAs were of early onset (detected in first samples on Day 8 post-dose), generally transient, and exhibited dose dependency with regard to titers and incidence. The presence of ADA had no apparent impact on the safety, PK and PD profiles. Further analysis was performed and concluded that the ADAs were not capable of neutralizing recombinant human IL-2.

Serum RO7049665 concentration reached peak level (C_{max}) approximately 12 hours post-dose and declined in an apparent biphasic manner, with a mean apparent terminal phase half-life of approximately 72 to 137 hours (see Table 3). Serum exposure levels of RO7049665 were below or close to the detection limit in the 1.5 and 5µg dose groups and thereafter increased with increasing doses in a highly variable manner. Overall, no obvious deviation from dose-linearity was noted. Sufficient concentration data for reliable estimates of non-compartmental parameters in terms of both AUC and C_{max} was available as of cohort 6 (650 µg) and above (see Table 3). Although the data showed moderate between-subject variability, no relevant departure from linearity was noted. The mean serum apparent systemic clearance of RO7049665 appeared dose-independent and ranged from 1.4 to 2.2 L/h.

RO7049665 disposition occurred with a mean $T_{1/2}$ of about 5 days, thus is expected to approach near-complete washout (C_{trough} < 10% of C_{max}) no later than 2 weeks post last dose. Accordingly, no substantial accumulation in reaching steady state is expected in a biweekly regimen.

At the highest dose of 7500 µg RO7049665, exposure was 33% and 65% of the exposure at the no-observed-adverse-effect level (NOAEL) (established in female monkeys on Day 22) in terms of C_{max} and $AUC_{0\text{-}168h}$, respectively.

Table 3 Serum Pharmacokinetic Parameters of RO7049665 after Single Subcutaneous Administration

Dose (μg)	n	C_{max} (ng/mL)	T_{max}[a] (h)	AUC_{inf} (ng • h/m)	CL/F (L/h)	V_Z/F (L)	$T_{1/2}$ (h)
Expressed as Geometric means (%CV)							
650	6	4.55 (49.4)	12.00 (6.00-24.00)	409 (43.1)	1.59 (43.1)	210 (28.2)	91.8 (52)
2200	6	11.0 (58.8)	12.03 (5.98-48.00)	1020 (37.7)	2.16 (37.7)	276 (17)	121 (23.2)
5000	5	46.5 (27.4)	12.00 (12.00-24.00)	2831 (26.9)	1.77 (26.9)	331 (25.6)	130 (18)
7500	6	68.7 (85.6)	12.00 (5.98-12.00	5266 (55.4)	1.42 (55.4)	281 (62.4)	137 (9.3)

AUC_{inf} = area under the serum concentration-time curve extrapolated to infinity; C_{max} = maximum serum concentration observed; CL/F = apparent clearance; ND = not determined; $T_{1/2}$ = terminal elimination half-life; T_{max} = time to maximum concentration; Vz/F = apparent volume of distribution.

a Median (Min–Max).

Pharmacodynamic data showed that RO7049665 expanded the T_{reg} cell pool, thereby delivering proof of pharmacology in a healthy population. The T_{reg} expansion was dose-dependent, and reached an absolute increase of 200 cells/μL at the maximum tested dose of 7500 μg, ranging from 123 to 297 cells/μL. This represented a mean-fold increase of 5.9 (2.9-7.6) from baseline. The peak was, on average, reached on Day 8, following a drop at 24 hours post-dose, and T_{reg} levels were back to baseline values at Follow-up (Day 57). Overall, the estimated therapeutic range of T_{reg} was achieved with the doses of 2200, 5000 and 7500 μg, eliciting a mean increase of 2, 4 and 6-fold, respectively, as compared to baseline.

There was no expansion of natural killer (NK) cells in any dose group, and no significant trends observed in eosinophils. There was no obvious effect on T_{eff} at doses up to 2200 μg. A possible weak trend for increase in T_{eff} count occurred with doses of 5000 μg and 7500 μg, with mean change from baseline on Day 12 reaching up to 1.5-fold increase from baseline.

Study WP40161

In this ongoing study WP40161, to date, multiple doses (i.e., up to 4) of 3.5 mg and 7.5 mg have been well tolerated, and they induced dose-dependent T_{reg} cell expansions up to 5-fold (mean at the highest dose of 7.5 mg) versus baseline.

The most frequently reported AEs were injection site reactions (ISRs) and eosinophilia. The majority of AEs reported were mild or moderate (Grade 1 or 2) in intensity except for a low number of events of severe (Grade 3) intensity, including anemia, eosinophilia and hyperkalemia. No events of CLS were reported. Hypersensitivity reactions (Grade 2)

with some, or all, symptoms of erythema, edema of the forearm, scapula and right arm, have been reported, as well as injection-related reactions (IRR) with some, or all, symptoms of myalgia, fatigue, chills and redness of face and neck. All events either have resolved without sequelae or are ongoing. There have been no deaths reported to date.

Immunogenicity was assessed, and positive results for ADA were observed in approximately 50% of participants. There have been no results to date indicating ADAs with neutralizing potential against endogenous IL-2. The ADAs were not associated with changes in PK parameters, and did not impact exposure or PD.

No significant abnormalities on vital signs or ECG intervals have been reported to date and, besides the changes on eosinophils, no significant laboratory safety results have been noted in participants with UC receiving up to 4 doses of 3.5 mg and 7.5 mg Q2W of RO7049665.

Detailed descriptions of the chemistry, pharmacology, pharmacokinetics, efficacy and safety of RO7049665 are provided in the RO7049665 Investigator's Brochure.

2.3 BENEFIT/RISK ASSESSMENT

UC is a chronic, relapsing disease characterized by diffuse mucosal inflammation of the colon. The precise etiology of UC is unknown; however, it is thought to be caused by an inappropriate inflammatory response to gut antigens in genetically predisposed individuals (Abraham and Cho 2009; Talley et al. 2011). The estimated incidence of UC in the United States is 9-12 cases per 100,000 persons per year, with an estimated prevalence of 205-240 cases per 100,000 persons (Danese and Fiocchi 2011). There is considerable variability in the incidence and prevalence of UC around the world.

Current management of UC includes initial use of aminosalicylates (e.g., sulfasalazine or mesalamine) and/or corticosteroids to induce remission or the use of biologic agents (e.g., tumor necrosis factor [TNF] inhibitors or integrin inhibitors) in patients with severe disease or in whom induction cannot be achieved with steroids. Once induction of remission has been achieved, maintenance therapy such as aminosalicylates, thiopurines (e.g., azathioprine or 6-mercaptopurine), or biologics (e.g., anti-TNF or anti-integrins) is given with the aim of maintaining disease control and avoiding long-term steroid use. Patients with severe acute disease or severe refractory disease may be considered for surgery (e.g., colectomy). Among multiple new molecular entities tested in patients with UC over the past two decades, only anti-TNF, anti-α4β7, and a pan-Jak inhibitor have demonstrated efficacy in patients with moderate to severe UC. Despite these recent additions to the treatment arsenal, over 70% of patients with moderate to severe UC still lack a long-term efficacious therapy (Neurath 2017).

Study WP40161 is the first study involving dosing of RO7049665 in participants with UC and the first study with repeat dosing. At the time of the commencement of Study WP40161, the safety data in healthy volunteers from the EIH Study WP39826 *were* available. Potential risks have been defined based on clinical experience in healthy

volunteers, *data from participants with UC in Cohorts 1 (3.5 mg) to 4 (7.5 mg; see Section 2.2.2.1*), non-clinical pharmacology and toxicology data in the relevant animal species, and theoretical risks based on expected PD effects and the safety profile of Proleukin®.

The eligibility criteria, design and procedures adopted are considered to be appropriate for the safe conduct of the planned study. Participants will be closely monitored for safety consistent with standard practices, and will be under close medical observation during the study.

The doses planned for dose level 1 (3.5 mg) and dose level 2 (7.5 mg) were derived from data in the WP39826 healthy volunteer study. Single doses of 7500 μg (7.5 mg) and below were safe and well tolerated in the subjects in WP39826, and induced dose-dependent T_{reg} expansions up to 6-fold (mean at the highest dose of 7500 μg) from baseline. *The dose planned for dose level 3 (15 mg) was based on results from repeat dosing with 3.5 mg and 7.5 mg in participants with UC in this study (WP40161); both the 3.5 mg and 7.5 mg doses have been well tolerated to date. The Sponsor believes it is important to fully characterize the safety profile and PK/PD relationship of RO7049665 in this study, which will also contribute to, and enable, an optimal design in future dose-setting studies.*

Identified potential risks and plans for their mitigation are as follows:

- Excessive Interleukin-2 Effects

  Excessive IL-2 effects have been reported in the Proleukin® label and include an increase in T_{eff}, increased eosinophilia, cytokine storm, CLS (symptoms include hypotension, mental status changes, dyspnea, pulmonary edema, reduced urine output and tachycardia), flu-like symptoms (such as fever, headache, muscle and joint pain, fatigue, nausea/vomiting/diarrhea, loss of appetite*),* dry and/or itchy skin or rash, liver and kidney failure, infections including sepsis and new onset or exacerbation of autoimmune disorders. These are considered less likely to occur with RO7049665 due to its selective effect on T_{regs}. Limited expansion of T_{eff} cells was observed at the two highest doses (5000 μg and 7500 μg) in the WP39826 study. There will be close monitoring of T_{eff} and eosinophils in blood, and of AEs suggestive of excessive IL-2 effects, with stopping criteria in place for CLS events, eosinophil counts *and in the event of systemic IRRs in a participant*. Exploratory safety cytokines such as IL-6, IL-8, TNF-α and IL-1β may be measured in an ad hoc fashion (e.g., in participants experiencing symptoms associated with *IRRs*).

- Hypersensitivity

  As with all biologic agents, there is a theoretical risk for hypersensitivity reactions, which can range from a mild rash to a life-threatening anaphylaxis. Hypersensitivity reactions may occur acutely following drug administration or have a delayed onset,

e.g., serum sickness reactions due to accumulation of immune complexes. No events suggestive of a systemic hypersensitivity reaction were observed following the receipt of single doses of RO7049665 in the WP39826 study. ADAs were observed which could increase the risk of developing hypersensitivity reactions with repeat dosing.

Patients will be closely monitored for clinical signs and symptoms suggestive of hypersensitivity reactions (both acute and non-acute). Patients developing hypersensitivity reactions will receive supportive care and treatment according to the institutional practice and local standard of care.

As mitigation, participants will receive all doses at an investigative site with resuscitation equipment available for immediate use and be housed at the site overnight. Dosing will also be staggered (see section on ADA below). *Additionally, stopping rules will be in place in the event of systemic hypersensitivity AEs in a participant and for* a *participant* experiencing a ≥ Grade 3 hypersensitivity reaction considered related to the study drug and believed to pose an unacceptable risk if the dose is repeated.

Additional assessments may be performed (e.g., IgE, tryptase, ADA, safety cytokines and further exploratory safety biomarkers) in participants experiencing signs and symptoms suggestive of a hypersensitivity reaction.

- ADA Formation

  The development of ADAs is a potential risk for all therapeutic protein products with the possibility of cross-reactivity being a particular concern for products that have endogenous counterparts. Twelve out of the 38 subjects (i.e., 32%) who received RO7049665 in the WP39826 study were positive for ADAs, with a trend for increasing incidence and titres at higher doses. ADAs raised to RO7049665 could be asymptomatic or could potentially cause hypersensitivity reactions, a decrease in drug exposure, neutralization of the therapeutic effect, or scavenging and neutralization of endogenous IL-2, thus causing an IL-2 deficiency state. Hypersensitivity reactions could range from rash to anaphylaxis or serum sickness. Absence of IL-2 has been associated with autoimmune conditions such as autoimmune thyroiditis or type 1 diabetes. The ADAs observed in WP39826 following single doses were not neutralizing to recombinant human IL-2 and had no apparent clinical consequences.

  This is the first repeat dose and first patient study for RO7049665 and the safety monitoring and risk mitigation measures listed below will be implemented:

  - Participants will be monitored closely following the receipt of each dose and housed at a clinical research site for at least 24 hours post-dosing.

- Staggered dosing will be employed. There will be a 5-day interval between receipt of the first dose *of* the first, second, and third *participants* treated at a specific dose level. *The fourth participant can receive the first dose 14 days after the first dose of the third participant, at the earliest. There will be* an interval of 48 hours between *administrations* of the first dose for subsequent *participants in each dose level.*

- Assessments of ADA development, their neutralising capability and the impact on continued dosing and dose escalation are planned at the following three timepoints:

  - Once the ADA results up to and including Day 22 (i.e., one week after the second dose) are available for the first five participants (4 active and 1 placebo) at the 3.5 mg dose level and subsequently at the 7.5 mg dose level (i.e., two separate assessments).

  - Once the ADA results up to and including Day 57 (i.e., two weeks after the fourth dose) are available for the first five participants (4 active and 1 placebo) at the 3.5 mg dose level. This is the dose escalation timepoint outlined in Section 4.1.2.

- Inclusion of individual *participants* and study level stopping criteria relating to hypersensitivity reactions and development of neutralising antibodies to recombinant human IL-2 (See Section 4.1.3).

- Signs and symptoms of autoimmune phenomena will be monitored and treated according to standard of care.

Concomitant therapy with immunomodulators such as azathioprine/6-mercaptopurine (6-MP)/methotrexate may decrease the risk of ADA and neutralizing IL-2 antibody formation. Patients who are already on one of these therapies will continue; patients who are not already on these therapies will not be proactively started on them. The mechanism of action of RO7049665 is to induce tolerance via increases in T_{reg} activity. This tolerance induction can potentially mitigate the risk of ADA and neutralizing IL-2 antibody formation.

- Increased Regulatory T Cells Causing Immunosuppression

  There is a theoretical risk of immunosuppression from increased T_{reg} cell count and an increased proportion of T_{regs} compared with total T cells. Infections, especially opportunistic infections, and malignancies are theoretical risks. Signs and symptoms of immunosuppression will be monitored and treated according to standard of care. A stopping criterion specific for T_{regs} increases has also been included.

- Injection Site Reactions

  Localized injection site reactions (including erythema, pruritus and swelling) of mild intensity and resolving without treatment were observed following dosing with RO7049665 in the WP39826 study. There was a higher incidence at higher doses and injection site erythema was the most frequently observed event. The erythema was first observed at 2 to 6 days after dosing and had a duration varying from 3 to 54 days until complete resolution. Assessments of local skin reactivity and pain following injection will be performed in this study to further characterize injection site reactions. Administration of serial doses in the same *participant* should be done in different quadrants of the abdomen.

- Non-clinical Finding of Heart Rate Increase

  Heart rate increases were observed in the 13-week good laboratory practice (GLP) study in cynomolgus monkeys. The heart rate increases showed complete reversibility in recovery. The effects seen on heart rate were likely linked to the acute phase response and/or elevated cytokine levels, both of which were more pronounced after Week 1 and thus are considered secondary to the pharmacologic action of the study drug. These effects were completely reversible. No trend towards increased heart rate was observed in the WP39826 clinical study after single dosing. Vital signs and ECGs will be monitored in this study. Further details are provided in the RO7049665 Investigator's Brochure.

- Procedure-Related Risks

  The procedures performed are flexible sigmoidoscopy (or possible colonoscopy at screening) with mucosal biopsies from the sigmoid colon, SC injections and blood draws. The risks associated with flexible sigmoidoscopy or colonoscopy with biopsies include bleeding, infection, and perforation of the intestine. SC injections may produce a local inflammatory reaction, which can range from a slight irritation to infection or possible necrosis. The risks associated with blood draws include hematoma, and rarely injury to neurovascular structures.

- Risks Related to Concomitant Medications

  Risks of Corticosteroid and RO7049665 Adjunctive Therapy

  Long-term use of corticosteroids is associated with pleiotropic effects, which are well described (Ford et al. 2011). An increased susceptibility to infection is a known side effect. Addition of RO7049665 could theoretically further increase the risk of infection. Current standard of care includes the addition of other immune-modulating therapeutics on a background of corticosteroid therapy, with the goal of therapy being to discontinue corticosteroids. Patients in this study may continue on a maximum dose of prednisone that will not exceed 20 mg/day for a maximum of 12 weeks of therapy. Corticosteroid dose could be decreased in response to steroid-associated side effects to minimize the toxicity of chronic

therapy with corticosteroids. Addition of RO7049665 to background corticosteroid therapy will follow standard of care practices since rapid discontinuation of corticosteroids is not acceptable due to the risk of adrenal insufficiency.

- Exacerbation of UC

  Patients with UC have a significant inflammatory burden from the ongoing disease. There is a potential risk of activating and expanding $CD25^+$ T_{eff} cells with RO7049665, or development of excessive IL-2 effects as described above.

  To mitigate this risk, participants will be housed for at least 24 hours after each dose and monitored closely (e.g., vital signs, ECG, physical exam). There will also be ongoing PD monitoring of immune cell subsets and optional ad hoc cytokine testing. Reassuringly, in vitro experiments with peripheral blood mononuclear cells from participants with UC demonstrated that after in vitro treatment with RO7049665, there was activation of only a very minor fraction of CD25+ T_{eff} (~3% of total $CD4^+$ cells), and only at an RO7049665 concentration that was ≥ 1,000 fold higher than the concentration needed to activate T_{regs} (for details, see the RO7049665 Investigator's Brochure). Limited expansion of T_{eff} cells was observed at the two highest doses (5000 μg and 7500 μg) in the WP39826 study. Thus, the risk of disease worsening due to T_{eff} activation in UC is considered minimal, while the potential benefit of increased T_{reg}s is the reversal of the inflammatory process. Participants in the study will be monitored closely for signs of disease worsening, and dosing in a participant will be stopped if persistent disease worsening is evident.

To ensure appropriate safety monitoring, the participants will be housed at the clinical research unit during each dosing period. Participants will receive a given dose of RO7049665 every 2 weeks for a total of 4 doses. Participants will be monitored for 24 hours in the clinical research unit after receipt of each dose.

An assessment was conducted to determine whether there is any impact of the coronavirus disease 2019 (COVID-19) pandemic on the benefit/risk assessment of this study protocol including, but not limited to, the patient population under study and study treatment being evaluated. On the basis of that assessment, no impact is anticipated and the existing safety monitoring and management guidelines, and risk mitigation measures provided in the study protocol are considered adequate.

Overall, the expected benefits of RO7049665 in the treatment of autoimmune conditions are greater than the expected risks. The level of risk for UC participants is considered acceptable as it is mitigated by careful monitoring, careful dose escalation, and study stopping criteria (See Section 4.1.2 and 4.1.3).

More detailed information about the known and expected benefits in the context of potential risks and reasonably expected AEs of RO7049665 is provided in the RO7049665 Investigator's Brochure.

3. OBJECTIVES AND ENDPOINTS

The objectives and corresponding endpoints are provided in Table 4.

Table 4 Objectives and Endpoints

Objectives	Endpoints
Primary	
• To evaluate the safety and tolerability of repeat dosing of subcutaneous (SC) injections of RO7049665 in participants with active ulcerative colitis (UC).	• Incidence of dose-limiting or intolerable treatment-related adverse events (AEs). • Incidence, severity, and causal relationship of treatment-emergent AEs. • Incidence of abnormal laboratory findings. • Incidence of abnormal vital signs and electrocardiogram (ECG) parameters.
Secondary	
• To investigate the multiple-dose pharmacokinetics of RO7049665.	• Pharmacokinetic parameters of RO7049665
• To investigate the effects of multiple doses of RO7049665 on changes in endoscopic appearance of the mucosa of the sigmoid colon.	• Change from baseline in the Endoscopy Subscore of the Mayo Clinic Score (MCS-ES) at Day 29 and Day 57. • Change from baseline in the Ulcerative Colitis Endoscopic Index of Severity (UCEIS) at Day 29 and Day 57.
• To investigate the effects of multiple doses of RO7049665 on changes in histologic appearance of mucosa in biopsies from the sigmoid colon.	• Change from baseline to Day 29 and Day 57 in histology score of sigmoid colon biopsies (Geboes Score [GS], Robarts Histology Index [RHI], or Nancy Histology Index [NHI]).
• To investigate the effects of multiple doses of RO7049665 on changes in disease activity.	• Change from baseline in the Mayo Clinic Score (MCS) at Day 29 and Day 57.
• To evaluate the immunogenicity of RO7049665.	• Incidence of anti-drug antibodies (ADAs).
• To investigate the effects of multiple doses of RO7049665 on immune cells in the periphery and in mucosal biopsies of the sigmoid colon.	• Quantitative changes in WBCs (T_{regs}, T_{effs}, natural killer [NK] cells, B cells and eosinophils).
Tertiary/Exploratory	
• To explore the association of genotype with PD, efficacy and safety of RO7049665.	• Correlation analysis of single nucleotide polymorphisms with PD, efficacy and AEs.

4. STUDY DESIGN

4.1 OVERALL DESIGN

Study WP40161 is a multicenter, randomized, adaptive, Investigator and participant blind, placebo-controlled study to investigate the safety, tolerability, PK, PD, and

preliminary efficacy of multiple ascending doses (MAD) of RO7049665 on top of standard of care (SOC) in participants with endoscopic evidence of moderate to severely active UC. The participants, investigators, and site personnel will be blinded to treatment assignments.

Since this is the first clinical study in which RO7049665 will be tested in participants diagnosed with UC and the first study in which multiple doses of RO7049665 will be administered, a dose escalation design will be used.

All participants in the study will have evidence of active disease on endoscopy at screening (MCS $\geq$5, Endoscopy subscore of the Mayo Clinic Score [MCS-ES] $\geq$ 2, stool frequency subscore $\geq$ 1, and rectal bleeding subscore $\geq$ 1). Participants will be eligible for the study if they are currently receiving non-biologic SOC therapy for UC and have not been treated with any biologic therapy within 3 months (or 5 half-lives, whichever is longer) before the start of screening.

Participants will be randomized in a 4:1 ratio to receive multiple SC injections of RO7049665 or placebo, respectively (approximately 8 on active and 2 on placebo per cohort). Participants will receive their first doses in a staggered manner, with a five-day interval between the *first dose of the* first, second, and third participants. *The fourth participant can receive the first dose 14 days after the first dose of the third participant, at the earliest. There will be* a 48-hour interval between the first dose of subsequent participants in each dose level. Based on emerging data, dose levels may be expanded.

The dosing interval will be every 2 weeks (Q2W), for 4 doses. The first dose level will be 3.5 mg, the second dose level will be 7.5 mg *and participants enrolled in the cohorts of dose level 3 will receive 15 mg of RO7049665 (or placebo)*. The doses are based on safety, tolerability, PK and PD data derived from study WP39826 (see Section 4.3) *and review of data from completed cohorts in this study*. Each dose level will have cohorts of approximately 10 participants, randomized in the ratio of 4:1 RO7049665 to placebo. Based on the emerging data there may be more than one cohort enrolled per dose level.

The second dose level may run in parallel with the first (see Section 1.2), after completion of recruitment into the lower dose level. However, prior to enrolling participants into the higher dose level, there will be an assessment of safety, tolerability, ADA, PK and selected PD data from at least 5 participants (4 on RO7049665, 1 on placebo) in dose level 1 who have received at least 8 weeks of treatment.

An overview of the study design is provided in Section 1.2.

4.1.1 Length of the Study

The total duration of the study for each participant will be approximately 19 weeks divided as follows:

- Screening: Up to 5 weeks (35 days).
- Dosing Period: Day 1 to Day 43.
- In-clinic period: From Day −1 to Day 2 for first dose, from Day 15 to Day 16 for the second dose, from Day 29 to Day 30 for the third dose, and from Day 43 to Day 44 for the last dose.
- Final follow-up: Day 99 ± 3 days (8 weeks post last dose).

4.1.2 Dose-Escalation Decision Criteria

The decision to escalate to the next dose level will be made following review of all relevant safety information collected, including available AEs, ECG, vital signs, clinical laboratory test results, ADA, selected PD data (quantitative changes in WBCs, T_{regs}, and eosinophils), and PK data collected up to at least 8 weeks in 5 participants (4 active and 1 placebo) at the immediately preceding dose level. The dose-escalation procedure for this study is based on data and decisions from study WP39826, where dose-escalation proceeded after a minimum of 3 participants on RO7049665 and 2 on placebo.

The decision to escalate will be made jointly by the Sponsor study team, the Sponsor's IMC, the Investigators, and any other person considered necessary to assist with the decision.

4.1.3 Stopping Rules Criteria

Dosing within a dose level will be stopped and no higher doses will be started if one of the circumstances or dose-limiting adverse events (DLAEs) listed below occurs in participants treated with RO7049665 within the same dose level, unless it is determined by the Investigator that the occurrence is not related to the study drug:

- Eosinophil count increase to > 5.0 x 10^9/L occurring in 3 or more participants in a single dose level.
- Grade 3 or higher (as defined by the Investigator) RO7049665-related AEs of the same type in 2 or more participants (See also Appendix 2).
- Grade 3 or higher RO7049665-related laboratory abnormalities of same type in 2 or more participants in a single dose level.
- Grade 2 or greater RO7049665-related changes in vital signs or ECGs (e.g., QTc > 500 ms, or > 60 ms longer than the pre-dose baseline) of same character in 2 or more participants in a single dose level.
- Other findings (e.g., AEs or lab abnormalities) that, at the joint discretion of the Sponsor's lead clinical scientist and safety science leader, the Sponsor's IMC and the Investigators, indicate the dosing or dose escalation should be stopped.
- Development of neutralizing antibodies to recombinant human IL-2 in one or more participants.

Dosing and dose escalation (alternatively, a lower dose level or less frequent dosing) may be resumed following further evaluation of the data and the benefit/risk profile, and

upon the joint agreement of the Sponsor's lead clinical scientist, safety science leader, the Sponsor's IMC, and the study Investigator, and only once the necessary Ethics Committee (EC) and health authority approvals have been received.

4.1.4 Individual Participant Stopping Criteria

Dosing will be stopped in a given individual participant if, compared with baseline, one of the following circumstances occurs:

- *A second systemic IRR or hypersensitivity AE of any grade occurs (see Sections 4.1.5 and 8.3.8).*
- Grade 3 or greater severe AE or SAE considered related to study drug, including but not limited to:
    - *IRR/ h*ypersensitivity reaction which is considered related to study drug and believed to pose an unacceptable risk if the dose is repeated.
    - CLS including severe, clinically significant respiratory distress syndrome, edema, effusions, or decrease in blood pressure
- Disease exacerbation requiring escalation of baseline treatment or other treatment strategies (pharmacological or surgical) signifying study treatment failure (See Section 7).
- Development of neutralizing antibodies to recombinant human IL-2 (also see study stopping criteria Section 4.1.3).
- Other findings that, at the discretion of the Investigator, indicate that dosing should be stopped.

Study closure information is provided in Appendix 1.

4.1.5 Communication Strategy

Information will be communicated during the study, as follows:

- For all participants, the Investigator(s) must confirm to Roche that the participant has been dosed and provide a brief summary of the status of the participant in terms of safety and tolerability to RO7049665/ placebo, communicated by email or telephone between 24 to 48 hours of first dose. Please refer to the Cohort Management and Dose Escalation Document.
- In the event of a DLAE, or an event or findings meeting the stopping criteria defined in Section 4.1.4 , the Investigators will contact the Sponsor immediately to discuss participant status and action taken/to be taken.
- *In the event of a first systemic IRR or systemic hypersensitivity the Investigator* ***must*** *contact the Sponsor prior to the next dose for discussion.*
- The Sponsor will rapidly inform all investigational sites of any urgent safety measures required to protect the clinical trial participants from any immediate hazard to their health and safety.
- Prior to dose-escalation, a dose-escalation meeting will be conducted by the Investigators, the Medical Monitor, and the Sponsor Clinical Team.

- Upon reaching the minimum criteria for dose escalation consideration (minimum of 5 participants having completed 8 weeks [DLAE period]) the Sponsor will organize a teleconference with the Investigators to discuss the safety and tolerability of RO7049665/ placebo. If the teleconference occurs prior to the end of the DLAE evaluation period, the Investigator will provide a final status prior to start of the next cohort.

  During each teleconference:

  - National Cancer Institute Common Terminology Criteria for Adverse Events (NCI-CTCAE) Version 5.0 Toxicities (see Appendix 2) will be discussed along with the results of PK data (if available), in addition to safety laboratory results and any other available data (as described in Section 4.1.2) that may assist the dose-escalation decision process.
  - Dose-escalation will only proceed to the next dose level if the Investigators and the Sponsor (including IMC decision) are satisfied with the safety profile of the previous dose level and agree to move to the next dose level.
  - If other RO7049665 dose levels are explored, it will be documented in writing and both the Sponsor and Investigators will approve the minutes of these meetings to confirm agreement.

In addition to these communications, the Sponsor and Investigators will be in regular contact throughout the study by email/telephone/fax, as per normal interactions during the conduct of a clinical study, and the Sponsor will arrange regular teleconferences and meetings to discuss study status.

The Sponsor will be available 24 hours a day to discuss any medical or study-related issues that may arise during the conduct of this study.

4.2 SCIENTIFIC RATIONALE FOR STUDY DESIGN

The study rationale is provided in Section 2.1.

4.2.1 Rationale for Study Population

This study will recruit male and female participants who have moderate to severe UC according to standard criteria and based on guidelines from the health authorities. While it is anticipated that the PK and PD of RO7049665 in participants with UC will be similar to that in healthy volunteers, the strength of the PD effect may differ due to the baseline immune dysregulation present in participants with UC. Similarly, it is important to assess the safety of the immunomodulatory approach of RO7049665 in participants with baseline immune dysregulation that differs meaningfully from healthy volunteers. This population will enable collection of PD data at the site of the disease (i.e., colonic biopsies) and for preliminary efficacy evaluations to be made. Therefore, enrollment of participants with UC is necessary to enable the evaluation of RO7049665 as a potential treatment of moderate to severe UC.

4.2.2 Rationale for Control Group

RO7049665 will be compared against placebo-concurrent control, with participants randomized in a 4:1 ratio to receive multiple SC injections of RO7049665 or placebo, respectively. We will employ a blinded placebo arm in order to derive a preliminary assessment of the safety risk of RO7049665 in participants with UC and for equipoise in discriminating drug-relatedness of AEs. A placebo group will similarly inform the analysis of key biomarkers, including histopathologic changes in colonic biopsy samples and biomarkers used to establish PD effect. As we are studying participants with active disease it will be critical to distinguish disease-related complications from intervention associated safety signals, as well as study-drug related changes in leukocyte and cytokine biomarkers. RO7049665 is an investigational agent for the treatment of autoimmune diseases and not an established therapy, and both placebo and active participants will receive current standard of care background therapy, which mitigates the ethical concerns of a placebo group.

4.2.3 Rationale for Biomarker Assessments

Key objectives of the mandatory biomarker strategy are to confirm and further investigate the expected effect of drug on target cells (expansion of T_{regs} in number), to identify a safe dose at which target PD effect can be observed, as well as to confirm the magnitude and kinetics of this effect after multiple dosing and in the particular study population.

In addition to characterizing expected target PD effects, monitoring of biomarkers for unwanted events such as expansion of effector cells (T cells, NK cells, and Eosinophils) is an important objective. This could add valuable information towards an estimate of the therapeutic window of this novel drug MoA as changes in these markers may provide a means to predict the occurrence of safety events, also potentially informing stratification of patients.

Additional objectives of the mandatory biomarker assessments are: (1) to investigate if systemic PD effects can be measured as surrogate markers for indicating tissue PD effects; (2) to identify which of the candidate systemic biomarkers shows highest correlation to tissue effects; (3) to determine if any tissue or systemic biomarkers can predict efficacy.

Colon biopsies will be obtained to characterize immune cell infiltration and histologic appearance. Additional measurements which may be performed on colon biopsy derived material are whole transcriptome or targeted gene expression analyses. The objective of these measurements is to confirm expected changes in T cell subsets and to identify further mechanistic markers such as changes in inflammatory signatures or markers of tissue healing. Colon biopsies may furthermore be subjected to bacterial DNA or RNA analysis to determine the composition of the mucosa-associated microbiome in patients.

These data are of potential value for patient stratification purposes or for treatment monitoring.

Blood samples will be collected to quantify relative and absolute WBC counts (including B-cells, T-cells, NK-cells, and eosinophils). Furthermore, markers for quantification and characterization of T_{regs} and T_{eff} subsets including, but not limited to, CD3, CD4, CD8, CD127, CD25, and FoxP3 will be determined.

Serum and plasma samples will be collected to measure cytokines or other soluble biomarkers mechanistically linked to RO7049665's mode of action. In addition, exploratory safety biomarkers may be measured in an ad-hoc fashion, (e.g., in participants experiencing symptoms associated with injection-related reactions).

Clinical blood genotyping data may be used to explore whether genotype may correlate with efficacy, influence the magnitude of PD effects or may impact the safety of RO7049665.

Exploratory disease biomarkers in serum and stool samples will be tested for their ability to stratify participants and to evaluate their potential for non-invasive response to treatment monitoring.

Additional PD biomarkers and methods to obtain further confidence on the mechanism of action of RO7049665 in participants may be tested. Such assessments may include, but are not limited to, determining specific methylation signatures in blood or colon biopsies to quantify T_{reg} or T_{eff} subsets. The purpose of this assessment is to explore alternative T cell subset quantification methods and to understand their potential usefulness for replacing established methods with limited throughput and logistical challenges in later stages of clinical development.

4.3 DOSE JUSTIFICATION

The starting dose and dose justification are based on data from the EIH study (WP39826) in healthy volunteers. The therapeutic range of RO7049665 is unknown but is predicted to fall between a 2-fold and 10-fold expansion of baseline T_{reg} cells (see RO7049665 Investigator's Brochure). Average UC patient T_{reg} counts are reported to lie within a range of 20 to 50 cells/μL. Assuming literature derived values can be confirmed, the targeted expansion of T_{regs} up to an approximate maximum of 450 cells/μL in this study is predicted to cover the efficacious T_{reg} expansion range.

Data from WP39826 demonstrated 4-fold expansion of T_{reg} cells in healthy volunteers after a single 5000 μg dose of RO7049665 (mean peak T_{reg} expansion to 155.2 cells/μL) and a mean 5.8-fold expansion of T_{reg} cells after a single 7500 μg dose (with a mean peak of approximately 200 cells/μL [range 123 – 297]). As the 2200 μg single dose in the SAD study elicited a mean 2-fold increase in T_{reg} cells, the starting dose of 3.5 mg (3500 μg) in study WP40161 is expected to produce a measurable PK effect (specifically,

the expansion or T_{reg} cells) however the overall expansion is projected to be at the low end of the hypothesized therapeutic target.

Single doses at these dose levels were safe and well tolerated in healthy volunteers with no SAEs and no DLAEs, with limited expansion of T_{eff} and eosinophils and no observed effect on NK cells. The maximum T_{reg} expansion was observed at Day 8 following the dose. In study WP40161, participants enrolled in dose level 1 cohorts will receive 3.5 mg of RO7049665 (or placebo) in multiple doses every 2 weeks, and the participants enrolled in *cohorts of* dose level*s* 2 *and 3* will receive 7.5 mg *and 15 mg respectively,* of RO7049665 (or placebo) in multiple doses every 2 weeks. Two doses above 3500 μg (3.5 mg) have been tested in single dosing in healthy volunteers in study WP39826 and were safe and well tolerated.

The averaged steady state exposure of RO7049665 following the top dose in this study is predicted not to exceed the NOAEL exposures in the monkey.

Assuming no effects of ADAs on PK or PD response, it is predicted that RO7049665 will result in an approximately 3-fold expansion in peak T_{reg} count at the proposed starting dose of 3.5 mg Q2W. This *starting* dos*e* regimen is predicted to have no effect on NK cells, and no to minimal effect on CD4+T_{eff} (<1.3 fold). Assuming *the dose was not stopped due to dose* stopping criteria, the dose could be escalated up to *1*5 mg Q2W for a more sustained T_{reg} expansion, to exceed >2 fold expansion over the dosing interval (and achieve peak T_{reg} at ~3.*7*-fold). There will be monitoring of T_{eff} cell counts and excessive IL-2 effects, as predictions indicate a trend for CD4+T_{eff} increase at the *15* mg dose (*~1.5* fold after the 4th dose).

Based on the safety results from the WP39826 study, *and initial results from this current study,* the proposed risk management and safety monitoring measures and the dose-escalation approach for this study, it is considered appropriate to study repeat dosing with RO7049665 up to a dose of *15* mg in participants with UC. Because safety and tolerability may be different between healthy volunteers and participants with UC, safety and tolerability will be reviewed on an individual basis and regular follow-ups are planned as detailed in the Schedule of Assessments.

Further details are provided in the RO7049665 Investigator's Brochure.

4.4 END OF STUDY DEFINITION

A participant is considered to have completed the study if he/she has completed all phases of the study including the last scheduled procedure shown in the Schedule of Activities (Section 1.3).

The end of the study is defined as the date when the last participant last observation (LPLO) occurs. LPLO is expected to occur approximately 56 (± 3) days after last study drug administration.

5. STUDY POPULATION

This study will enroll up to 65 participants with UC between 18 and 70 years of age, inclusive, with evidence of moderate to severely active disease (MCS $\geq$ 5, MCS-ES $\geq$ 2, stool frequency subscore $\geq$ 1, and rectal bleeding subscore $\geq$ 1), who have failed at least one prior standard of care therapy.

The study population rationale is provided in Section 4.2.1.

Prospective approval of protocol deviations from recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

5.1 INCLUSION CRITERIA

Participants are only eligible to be included in the study if all of the following criteria apply:

Informed Consent

1. Able and willing to provide written informed consent and to comply with the study protocol according to International Conference on Harmonisation (ICH) and local regulations. Informed consent may be obtained up to 3 months prior to randomization. A signed Informed Consent Form must be available from the participant before starting and study-specific assessments.

Age

2. Between 18 to 70 years of age inclusive, at the time of signing the informed consent.

Type of Participants and Disease Characteristics

3. Diagnosed with UC for at least 12 weeks prior to screening

4. Screening colonoscopy for colorectal cancer conducted within the prior two years if:
    - History of pancolitis and disease duration $\geq$ 8 years, or
    - History of left-sided colitis and disease duration $\geq$ 12 years.
5. Evidence of disease activity at time of screening as measured by MCS $\geq$ 5, with MCS-ES $\geq$ 2, extending at least 10 cm from the anal verge, stool frequency subscore $\geq$ 1, and rectal bleeding subscore $\geq$ 1.

6. Insufficient clinical response to standard of care (SOC) therapy or intolerance to SOC, where SOC may include (but not limited to):
    - Glucocorticoids (Prednisolone at least 0.75 mg/kg/day or equivalent for at least 4 weeks).

- Treatment with JAK inhibitors of at least 8 weeks duration.
- Immunomodulators (e.g., azathioprine, methotrexate, oral 5-aminosalicylate [5-ASA] or 6-mercaptopurine) at a standard therapeutic dose for at least 12 weeks.
- Therapy with an anti-TNFα monoclonal antibody of at least 3 months duration, or intolerance due to side effects of medication (not including anaphylaxis).
- Treatment with anti-integrin therapy of at least 14 weeks duration.

Weight

7. Body mass index (BMI) within the range of 18-35 kg/m^2 (inclusive).

Sex

8. Male and/or female participants

   The contraception and abstinence requirements are intended to prevent exposure of an embryo to the study treatment. The reliability of sexual abstinence for male and/or female enrollment eligibility needs to be evaluated in relation to the duration of the clinical study and the preferred and usual lifestyle of the participant. Periodic abstinence (e.g., calendar, ovulation, symptothermal, or post-ovulation methods) and withdrawal are not acceptable methods of contraception.

   Female participants:

   Compliance with contraception requirements:

   - Women of non-child bearing potential (WONCBP) may be enrolled. A woman is considered to be of childbearing potential if she is postmenarchal, has not reached a post-menopausal state (≥ 12 continuous months of amenorrhea with no identified cause other than menopause), and has not undergone surgical sterilization (removal of ovaries and/or uterus).

   For women of childbearing potential (WOCBP), as defined in Appendix 5:

   - May enroll if not pregnant and not breastfeeding.
   - Agree to remain abstinent (refrain from heterosexual intercourse) or use contraceptive methods that result in a failure rate of < 1% per year during the treatment period and for at least 56 days after the last dose of study drug.

     Examples of contraceptive methods with a failure rate of < 1% per year include bilateral tubal occlusion/ligation, male sterilization, proper use of hormonal contraceptives that inhibit ovulation, hormone-releasing intrauterine devices, and copper intrauterine devices (see Appendix 5).
   - Have a negative pregnancy test (blood) within the 35 days prior to the first study RO7049665/placebo administration.

For men:

During the treatment period and for at least 56 days after the last dose of RO7049665 or placebo, agreement to:

- Remain abstinent (refrain from heterosexual intercourse) or use contraceptive measures such as a condom plus an additional contraceptive method that together result in a failure rate of < 1% per year, with partners who are women of childbearing potential (WOCBP, as defined in Section 1 of Appendix 5).

  With pregnant female partners, remain abstinent (refrain from heterosexual intercourse) or use contraceptive measures such as a condom to avoid exposing the embryo.
- Refrain from donating sperm for 56 days after the last dose of RO7049665 or placebo.

5.2 EXCLUSION CRITERIA

Participants are excluded from the study if any of the following criteria apply:

Medical Conditions

1. Diagnosis of Crohn's disease or indeterminate colitis.
2. History of infection with hepatitis B, human immunodeficiency virus (HIV), active hepatitis C virus (HCV) infection, or other chronic infection.
3. Active infections requiring systemic therapy with antibiotic, antiviral or antifungal or febrile illness within 7 days before Day -1.
4. History of primary or acquired immunodeficiency.
5. History of chronic pulmonary disease with resultant abnormal pulmonary function.
6. History of clinically significant cardiac or cardiovascular disease or uncontrolled hypertension.
7. Evidence of infection with *Clostridium difficile* or other intestinal pathogen within 35 days from the start of screening.
8. Evidence of colonic dysplasia that cannot be completely removed.
9. Females: Pregnant or lactating.
10. History of and/or current suicidal ideations

11. Any condition or disease detected during the medical interview/physical examination that would render the patient unsuitable for the study, place the patient at undue risk or interfere with the ability of the patient to complete the study in the opinion of the investigator, including malignant disease.

12. Symptomatic herpes zoster within 3 months prior to screening.

13. History of tuberculosis or a positive Quantiferon® Gold test.

14. History of clinically significant severe drug allergies, multiple drug allergies, allergy to any constituent of the product, or intolerance to topical corticosteroids.

15. Lymphoma, leukemia, or any malignancy within the past 10 years, except for basal cell or squamous epithelial carcinomas of the skin that have been resected with no evidence of metastatic disease for 3 years and in situ carcinoma of the cervix that was completely removed surgically.

16. History or presence of clinically significant ECG abnormalities before study drug administration (e.g., PQ/PR interval $\geq$ 220 ms, QT corrected for heart rate using Fridericia's correction factor (QTcF), $<$350 or $\geq$ 450 ms) or clinically significant cardiovascular disease (e.g., cardiac insufficiency, coronary artery disease, cardiomyopathy, congestive heart failure, family history of congenital long QT interval syndrome, family history of sudden death).

17. Fecal Microbiota Transplant (FMT), defined as receipt of any product derived from the feces of another human and administered per oral, per nasogastric or nasoduodenal, or per rectum within the last 6 months.

Prior/Concomitant Therapy

18. Use of calcineurin inhibitors (e.g., tacrolimus, cyclosporine), vedolizumab, anti-TNFα therapeutic or any other immune system–targeted biological therapy within 12 weeks or 5 half-lives, whichever is longer, prior to screening.

19. Rectal therapy with 5-ASA or corticosteroids within 2 weeks of screening.

20. Use of any investigational drug within 12 weeks of screening.

21. Leukocyte apheresis within 12 weeks of screening.

22. Live vaccine(s) within one month prior to screening, or plans to receive live vaccines during the study or within 28 days of the last dose.

Prior/Concurrent Clinical Study Experience

23. Donation of blood or blood products in excess of 500 mL within 3 months.

24. Exposure to more than 4 investigational treatments within 12 months prior to Day 1.

25. Current enrollment or past participation within the last 90 days immediately preceding signing of informed consent in this or any other clinical study involving an investigational study treatment or any other type of interventional medical research.

Diagnostic Assessments

26. Abnormal hematologic values:

    - Anemia (Hgb < 9 g/dL)
    - Leukocytosis (WBC $\geq 2\times$ ULN)
    - Thrombocytopenia (platelet count $< 100{,}000/\mu L$)
    - Thrombocytosis (platelet count $\geq 2 \times$ ULN)
    - Eosinophilia (eosinophil count $\geq 2 \times$ ULN)

27. Abnormal hepatic enzyme or hepatic function values:

    - Alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase, or gamma-glutamyl transferase (GGT) $\geq 2 \times$ ULN
    - Total bilirubin $\geq 2 \times$ ULN
    - International normalized ratio (INR) ≥ 1.2
    - Albumin < 3 g/dL

28. Positive HIV antibody test.

29. Presence of hepatitis B surface antigen (HBsAg) or positive for total hepatitis B core antibody (HBcAb), or positive hepatitis C by PCR test result at screening or within the 3 months prior to starting study treatment.

Other Exclusions

30. History of regular alcohol consumption within 2 months of screening defined as:

    An average weekly intake of $>$ 14 drinks for males or $>$ 7 drinks for females. One drink is equivalent to 12g of alcohol: 12 ounces (360 mL) of beer, 5 ounces (150 mL) of wine or 1.5 ounces (45 mL) of 80 proof distilled spirits.

31. Any suspicion or history of alcohol abuse and/or suspicion of regular consumption of drug of abuse.

32. Patients under judicial supervision, guardianship or curatorship.

5.3 LIFESTYLE CONSIDERATIONS

5.3.1 Meals and Dietary Restrictions

During the period from screening to the follow-up visit when participants are not resident in the unit, participants will be advised about the following requirements:

- Coffee or tea consumption should be no more than 3 cups/day, and methylxanthine containing drinks must be $<$1 L/day.
- Alcohol consumption should be no more than 2 drinks/day.

  One drink is equivalent to 12g of alcohol: 12 ounces (360 mL) of beer, 5 ounces (150 mL) of wine or 1.5 ounces (45 mL) of 80 proof distilled spirits.

5.3.2 Caffeine, Alcohol, and Tobacco

Caffeine and tobacco should be avoided during the residential period.

Consumption of alcohol will not be allowed from 48 hours before dosing until the end of the residential period.

5.3.3 Activity

Light ambulatory activities will be permitted, with the level of activities kept as similar as possible on all days in the clinical research unit during the study. *Participants* are required to refrain from intense physical activity from 96 hours before screening visit until the final follow-up visit.

5.4 SCREEN FAILURES

Screen failures are defined as participants who consent to participate in the clinical study but are not subsequently randomized to study treatment/entered in the study.

The Investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure.

Individuals who do not meet the criteria for participation in this study (screen failure) may be re-screened. Re-screened participants should be assigned the same participant number as for the initial screening.

For re-screened participants, the following specific criteria must be met:

- Screening colonoscopy or flexible sigmoidoscopy for inclusion must be completed within 3 weeks prior to first dose.
- Serologies for hepatitis B, HIV, and PCR for HCV need not be repeated, and screening for latent tuberculosis infection need not be repeated if already done.
- Women of child bearing potential must repeat pregnancy testing.

- Screening laboratory evaluation beyond those itemized above must be completed within 35 days of the starting dose.
- For participants who were initially excluded due to active *Clostridium difficile*-related infection, participants must have completed therapy and have been stable off therapy for at least 7 days prior to repeat *Clostridium difficile* testing for Test of Cure (TOC). Those who test negative may be eligible for re-enrollment.

5.5 RECRUITMENT PROCEDURES

Participants will be identified for potential recruitment using pre-screening enrollment logs, clinical database and Independent Ethics Committees (IEC)/Institutional Review Board (IRB) approved newspaper/radio/social-media advertisements prior to consenting to take place on this study.

6. TREATMENTS

Study treatment is defined as any investigational treatment(s), marketed product(s), placebo, or medical device(s) intended to be administered to a study participant according to the study protocol.

The investigational medicinal product (IMP) required for completion of this study will be provided by the Sponsor. Study drug will be administered by the investigational staff (i.e., Investigator or study nurse) at the study center under supervision of the Investigator.

6.1 TREATMENTS ADMINISTERED

Table 5 summarizes the treatments administered.

Table 5 Summary of Treatments Administered

Study Treatment Name:	RO7049665	Placebo
Dosage Formulation:	Solution for injection	Solution for injection
Unit Dose Strength:	5.0 mg/mL	N/A
Dose:	Starting dose 3.5 mg (dose level 1), 7.5 mg (dose level 2), *and 15 mg (dose level 3)*	N/A
Route of Administration:	SC injection	SC injection
Dosing Instructions:	RO7049665 must be prepared for dosing under appropriate aseptic conditions and should be used immediately. The solution for injection must be filtered prior to use	Placebo must be prepared for dosing under appropriate aseptic conditions and should be used immediately. The solution for injection must be filtered prior to use
Packaging and Labeling:	RO7049665 and Placebo will be provided in 2 mL colorless glass vials. Each vial will be labeled as required per country requirement.	
Storage Conditions	For RO7049665 and Placebo, if not used immediately, the total storage time of the dose solution prior to administration should not exceed 24 hours to limit the risk of microbial growth in case of accidental contamination. The recommended storage condition for the dose solution is 2°C to 8°C, but dose solutions may be stored at room temperature for up to a maximum of 4 hours	
Manufacturer	F. Hoffmann-La Roche, Ltd.	F. Hoffmann-La Roche, Ltd.

Guidelines for dosage modification and treatment interruption or discontinuation are provided in Section 6.6 or Section 7, respectively.

Please see the RO7049665 Investigator's Brochure and Pharmacy Manuals for more details.

6.2 PREPARATION/HANDLING/STORAGE/ACCOUNTABILITY

Study drug packaging will be overseen by the Roche clinical trial supplies department and bear a label with the identification required by local law, the protocol number, drug identification and dosage.

The packaging and labeling of the study medication will be in accordance with Roche standard and local regulations.

The investigational site will acknowledge receipt of IMPs and confirm the shipment condition and content. Any damaged shipments will be replaced.

Upon arrival of the IMPs at the site, site personnel will complete the following:

- Check the IMPs for damage.
- Verify proper identity, quantity, integrity of seals and temperature conditions.
- Report any deviations or product complaints to the Monitor upon discovery.

The qualified individual responsible for dispensing the study treatment will prepare the correct dose according to the randomization/treatment assignment schedule and Pharmacy Manual.

The Investigator or delegate must confirm appropriate temperature conditions have been maintained during transit for all study treatment received and any discrepancies are reported and resolved before use of the study treatment.

Only participants enrolled in the study may receive study treatment and only authorized site staff may supply or administer study treatment. All study treatments must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the Investigator and authorized site staff.

The Investigator, Institution, or the Head of the Medical Institution (where applicable) is responsible for study treatment accountability, reconciliation, and record maintenance (i.e., receipt, reconciliation and final disposition records).

IMPs will either be disposed of at the study site according to the study site's institutional standard operating procedure (SOP) or returned to the Sponsor with the appropriate documentation. The site's method of IMP destruction must be agreed upon by the Sponsor. Local or institutional regulations may require immediate destruction of used IMP for safety reasons. The site must obtain written authorization from the Sponsor before any IMP is destroyed, and IMP destruction must be documented on the appropriate form.

Further guidance and information for the final disposition of unused study treatment are provided in the Pharmacy Manual or other specified location.

6.3 MEASURES TO MINIMIZE BIAS: RANDOMIZATION AND BLINDING

6.3.1 Method of Treatment Assignment

All participants will be centrally assigned to randomized study treatment using an Interactive Voice/Web Response System (IVRS/IWRS). Before the study is initiated, the

telephone number and call-in directions for the IVRS and/or the log in information and directions for the IWRS will be provided to each site.

Participants will be randomly assigned in a 4:1 ratio to receive multiple SC injections of RO7049665 or placebo, respectively. The randomization numbers will be generated by the Sponsor or its designee. The randomization list will be made available to the individual responsible for PK/PD sample bioanalysis, and to statisticians or programmers at Roche. PK/PD data can be received and cleaned on an ongoing basis. The data will be handled and cleaned in a secure area which is not accessible by any blinded SMT member.

Please refer to the Cohort Management and Dose Escalation Document for details regarding assignment of participants to treatment groups and cohorts.

6.3.2 Blinding

This study is double-blinded. For this study it means that the participant, the Investigator(s), and all individuals in direct contact with the participant at the investigative site will be blinded. Members of the Sponsor's project and study teams who do not have direct contact with the participant may be unblinded.

To allow informed recommendations or decisions regarding the dose-selection in this study, an integrated assessment of the safety, tolerability and available PK and/or PD will be made prior to each dose-decision.

If required, unblinded data (individual as well as at group level) may also be presented to the Drug Safety Committee or IMC or other experts of the Sponsor and at group level to the PI.

The IVRS/IWRS will be programmed with blind-breaking instructions. The blind may be broken if, in the opinion of the Investigator, it is in the participant's best interest to know the study treatment assignment. The Sponsor must be notified before the blind is broken unless identification of the study treatment is required for a medical emergency in which the knowledge of the specific blinded study treatment will affect the immediate management of the participant's condition (e.g., antidote available). In this case, the Sponsor must be notified within 24 hours after breaking the blind. The date and reason that the blind was broken must be recorded in the source documentation and case report form (CRF), as applicable.

6.4 TREATMENT COMPLIANCE

The qualified individual responsible for dispensing the study treatment will prepare the correct dose according to the randomization schedule. This individual will write the date dispensed and participant number on the study treatment vial label and on the Drug Accountability Record. This individual will also record the study treatment number received by each participant during the study.

6.5 CONCOMITANT THERAPY

6.5.1 Permitted Therapy

Any medication (including over-the-counter [OTC] or prescription medicines, approved dietary and herbal supplements, nutritional supplements) used by a participant from 30 days prior to screening until the follow-up visit must be recorded along with reason for use, dates of administration (including start and end dates) and dosage information (including dose and frequency).

The same information must be recorded for the following concomitant medications taken by a participant during the indicated period prior to screening until the follow-up visit:

- Immunomodulators used 12 weeks prior to screening

The Medical Monitor should be contacted if there are any questions regarding concomitant or prior therapy.

All concomitant medications should be reported to the Investigator and recorded on the Concomitant Medications electronic Case Report Form (eCRF).

All therapy and/or medication administered to manage AEs should be recorded on the Adverse Event eCRF.

Use of the following therapies will be permitted as specified below:

- Paracetamol is allowed up to a maximum dose of 2 g/day up to 48 hours prior to each RO7049665 or placebo dosing, not to exceed 4 g total during the week prior to each dosing. All concomitant medications throughout the duration of the study should be recorded in the case report form.
- *In the event of a systemic IRR or systemic hypersensitivity AE, pre-medicate with paracetamol and antihistamines (as per local guidelines) prior to the next administration of study drug (Section 8.3.8).*
- Participants who use the following therapies prior to screening and are on a stable regimen should continue their use:
  - Oral contraceptives, hormone-replacement therapy, or other maintenance therapy.
- Participants who are taking non-biological SOC therapy for UC during the study will remain on their SOC therapy. Specific instructions for each SOC is provided below:
  - Stable therapy for at least 2 weeks prior to screening with:
    - oral 5-ASA, or
    - oral glucocorticoid (not to exceed 20 mg/day of prednisone equivalent) at start of trial, may be weaned after dosing with study drug has been initiated).
  - Stable therapy for at least 8 weeks with

- azathioprine, or
- 6-MP, or
- methotrexate (and folic acid).

6.5.2 Prohibited Therapy

All medications (prescription and OTC) taken within 30 days of study screening will be recorded on the appropriate eCRF.

As a general rule, no concomitant medication will be permitted, with the exception of medications to treat AEs and those specified (Section 6.5.1), unless the rationale for exception is discussed and clearly documented between the Investigator and the Sponsor.

Use of the following therapies will be prohibited during the study and for at least 14 days or at least 5 half-lives prior to initiation of study treatment, whichever is longer unless otherwise specified below:

- Unless covered by the exceptions described in Section 6.5.1:
  - Any rectal therapy.
  - Doses of glucocorticoids exceeding 20 mg/day of prednisone or equivalent
  - Non-steroidal anti-inflammatory medications (e.g., ibuprofen, naproxen) due to a propensity for these treatments to worsen inflammation in ulcerative colitis.
- Calcineurin inhibitors (e.g., tacrolimus, cyclosporine) and any other immune system targeted biological therapy 12 weeks or 5 half-lives, whichever is longer, before the start of screening.

6.6 DOSAGE MODIFICATION

No dosage modification is permitted for any participant in the study.

Study drug may be temporarily halted and restarted, as described in Section 7.1 and Section 7.1.1.

6.7 TREATMENT AFTER THE END OF THE STUDY

The Sponsor does not intend to provide RO7049665 to participants after conclusion of the study or any earlier participant withdrawal.

7. DISCONTINUATION OF STUDY, *STUDY* TREATMENT, AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

An excessive rate of withdrawals (either participants discontinuing study treatment or withdrawing from the study) can render the study non-interpretable. Therefore, unnecessary withdrawal of participants should be avoided and efforts should be taken to motivate participants to comply with all the study specific procedures as outlined in this protocol.

Details on study and site closures are provided in *Section 4,* Appendix 1.

7.1 DISCONTINUATION OF STUDY TREATMENT

Dosing will be stopped in a given individual as described in Section 4.1.4.

Participants who discontinue study treatment prematurely will be asked to return to the clinic for a study completion/early termination visit (see Section 8.10.3) and may undergo follow-up assessments (see Section 8.10.4). See the SoA (Section 1.3) for data to be collected at the time of treatment discontinuation and follow-up and for any further evaluations that need to be completed.

The primary reason for premature study treatment discontinuation should be documented on the appropriate eCRF.

Reasons for discontinuation of study treatment (or withdrawal from the study) include, but are not limited to, the following:

- Participant withdrawal of consent at any time.
- Any medical condition that the Investigator or Sponsor determines may jeopardize the participant's safety if he or she continues in the study.
- Investigator or Sponsor determination that treatment discontinuation is in the best interest of the participant.
- Pregnancy.

7.1.1 Temporary Interruption

Before permanently discontinuing study treatment (regardless of whether initiated by the participant, the Investigator or Sponsor), an interruption should be considered. Participants who have temporarily interrupted study treatment should be considered to restart as soon as medically justified in the opinion of the Investigator after discussion with Sponsor and would *revert to the protocol-specified SoA (see Section 1.3) as soon as dosing of study treatment is reinitiated.*

7.2 PARTICIPANT DISCONTINUATION/WITHDRAWAL FROM THE STUDY

Participants have the right to voluntarily withdraw from the study at any time for any reason.

In addition, the Investigator has the right to withdraw a participant from the study for medical conditions that the Investigator or Sponsor determines may jeopardize the participant's safety if he/she continues in the study.

If possible, information on reason for withdrawal from the study should be obtained. The primary reason for withdrawal from the study should be documented on the appropriate eCRF. Participants will not be followed for any reason after consent has been withdrawn.

When a participant voluntarily withdraws from the study, or is withdrawn by the Investigator, samples collected until the date of withdrawal will be analyzed, unless the participant specifically requests for these to be discarded or local laws require their immediate destruction. However, if samples have been tested prior to withdrawal, results from those tests will remain as part of the overall research data. A participant's withdrawal from this study does not, by itself, constitute withdrawal of specimens donated to the Research Biosample Repository (see Section 8.8.2).

Participants who withdraw from the study for safety reasons will not be replaced. Participants who withdraw from the study for other reasons may be replaced.

See SoA (Section 1.3) for data to be collected at the time of study discontinuation and at safety and follow-up visits, and for any further evaluations that need to be completed.

7.3 LOST TO FOLLOW-UP

A participant will be considered lost to follow-up if the participant repeatedly fails to return for scheduled visits and is unable to be contacted by the study site.

The following actions must be taken if a participant fails to return to the clinic for a required study visit:

- The site must attempt to contact the participant and reschedule the missed visit as soon as possible, counsel the participant on the importance of maintaining the assigned visit schedule and ascertain whether or not the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the Investigator or designee must make every effort to regain contact with the participant. These contact attempts should be documented in the participant’s medical record.
- Should the participant continue to be unreachable, he/she will be considered to have withdrawn from the study.

Details regarding the discontinuation of sites or of the study as a whole are outlined in Appendix 1.

8. STUDY ASSESSMENTS AND PROCEDURES

Study procedures and their time-points are summarized in the Schedules of Activities (SoA; Section 1.3). Protocol waivers or exemptions are not allowed.

Immediate safety concerns should be discussed with the Sponsor immediately upon occurrence or awareness to determine if the participant should continue or discontinue study treatment.

Procedures conducted as part of the participant’s routine clinical management (e.g., blood count) and obtained before signing of the Informed Consent Form (ICF) may

be utilized for screening or baseline purposes provided the procedure met the protocol-specified criteria and were performed within the time-frame defined in the SoA.

Samples for laboratory tests will be sent to one or several central laboratories or to the Sponsor for analysis. Instruction manuals and supply kits will be provided for all central laboratory assessments.

Based on continuous analysis of the data in this study, any *assessment,* sample type or biomarker evaluation not considered to be critical for safety may be stopped at any time if the data from the *assessments or* samples collected does not produce useful information.

8.1 EFFICACY ASSESSMENTS

Flexible sigmoidoscopy (or colonoscopy at screening if required by inclusion criteria) will be performed at the time-points specified in the SoA (see Section 1.3). The endoscopy will be centrally read and scored using the MCS-ES and UCEIS scoring systems. The Endoscopy subscore of the Mayo Score will be modified so that a value of 1 does not include friability. Sedation and preparation for colonoscopy or flexible sigmoidoscopy may follow local standards.

Mucosal biopsies will be obtained during flexible sigmoidoscopy at the most affected area 10 to 20 cm from the anal verge at screening/baseline and at Day 29 (Week 4) and Day 57 (Week 8) assessments. Biopsies will be assessed by standard histology for changes in inflammatory activity as measured by the Geboes Score (Geboes et al. 2000), Robarts Histology Index (Mosli et al. 2017) and the Nancy Histology Index (Marchal-Bressenot et al. 2017).

Additional details will be provided in the Procedural Manual.

8.2 SAFETY ASSESSMENTS

Planned time-points for all safety assessments are provided in the SoA (Section 1.3).

8.2.1 Physical Examinations

A complete physical examination will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, dermatological and neurological, musculoskeletal in addition to head, eyes, ears, nose, throat, neck and lymph nodes systems. Height and weight will also be measured and recorded. Further examination of other body systems may be performed in case of evocative symptoms at the Investigator's discretion.

The BMI will be calculated at Screening only. Height will be recorded at Screening only and body weight will be recorded at Screening, at all subsequent physical examinations and as clinically indicated.

Skinfold thickness at the intended site of injection of the abdomen, and outside the five centimeter area directly around the navel, will be measured once on Day –1, using a skinfold caliper. The result will be reported in the eCRF.

A brief physical examination will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, dermatological and neurological systems, and measurement of body weight.

A complete or abbreviated physical examination will be performed at the time-points specified in the SoA (Section 1.3) by trained medical personnel at the study center. Attention should be paid to signs and symptoms suggestive of type 3 hypersensitivity reactions.

Any abnormality identified at baseline should be recorded on the General Medical History and Baseline Conditions eCRF.

As clinically indicated, limited, symptom-directed physical examinations should be performed. Changes from baseline abnormalities should be recorded in participant's notes. New or worsened clinically significant abnormalities should be recorded as AEs on the Adverse Event eCRF.

8.2.2 Vital Signs

Blood pressure (BP), pulse rate, body temperature (tympanic or oral) and respiratory rate will be assessed at the time-points specified in the SoA (Section 1.3).

Blood pressure and pulse measurements will be assessed in a supine position with a completely automated device. Manual techniques will be used only if an automated device is not available. When possible, the same arm should be used for all blood pressure measurements.

Blood pressure and pulse measurements should be preceded by at least 5 minutes of rest for the participant in a quiet setting without distractions (e.g., television, cell phones).

Single measurements of vital signs (to be taken before blood collection for laboratory tests but after ECG collection when scheduled at the same time-point) will be performed in a supine position after 5 minutes rest and will include body temperature (tympanic), systolic and diastolic blood pressure, pulse rate and respiratory rate.

The timings of assessments may be amended or the number of assessments increased during study conduct on the basis of emerging data in order to allow for optimal characterization of the effect profile.

8.2.3 Electrocardiograms

Triplicate 12-lead ECG will be obtained as outlined in the SoA (see Section 1.3) using an ECG machine. Measurements of PR, QRS complex (QRS), QT, and QTc intervals may be performed by the instrument or calculated using an external tool. Refer to Section 4.1.3 for QTc withdrawal criteria and additional QTc readings that may be necessary.

At each time-point at which triplicate ECGs are required, three individual ECG tracings should be obtained as closely as possible in succession, but no more than 5 minutes apart. The full set of triplicates should be completed in less than 10 minutes.

To minimize variability, it is important that participants be in a resting position for ≥10 minutes prior to each ECG evaluation. Body position should be consistently maintained for each ECG evaluation to prevent changes in heart rate. Environmental distractions (e.g., television, radio, conversation) should be avoided during the pre-ECG resting period and during ECG recording. ECGs should be performed prior to any scheduled vital sign measurements and blood draws. In some cases, it may be appropriate to repeat abnormal ECGs to rule out improper lead placement as contributing to the ECG abnormality.

For safety monitoring purposes, the Investigator or designee must review, sign, and date all ECG tracings. Paper or electronic copies will be kept as part of the participant's permanent study file at the site. If considered appropriate by Roche, ECGs may be analyzed retrospectively at a central laboratory.

ECG characteristics, including heart rate, QRS duration, and PR, and QT intervals, will be recorded on the eCRF. QTcF (Fridericia's correction) and RR will be calculated and recorded on the eCRF. Changes in T-wave and U-wave morphology and overall ECG interpretation will be documented on the eCRF. T-wave information will be captured as normal or abnormal, U-wave information will be captured in two categories: absent/normal or abnormal.

8.2.4 Clinical Safety Laboratory Assessments

Planned time-points for all safety assessments are provided in the SoA (see Section 1.3).

Normal ranges for the study laboratory parameters must be supplied to the Sponsor before the study starts. A list of clinical laboratory tests to be performed is provided in Appendix 4 and these assessments must be conducted in accordance with the separate laboratory manual and the SoA (see Section 1.3).

The Investigator must review the laboratory report, document this review, and record any clinically relevant changes occurring during the study in the AE section of the CRF. The laboratory reports must be filed with the source documents. Clinically significant abnormal laboratory findings are those which are not associated with the underlying

disease, unless judged by the Investigator to be more severe than expected for the participant's condition.

In the event of unexplained abnormal clinically significant laboratory test values, the tests should be repeated immediately and followed up until they have returned to the normal range and/or an adequate explanation of the abnormality is found.

If such values do not return to normal/baseline within a period of time judged reasonable by the Investigator, the etiology should be identified and the Sponsor notified.

If laboratory values from non-protocol specified laboratory assessments performed at the local laboratory require a change in participant management or are considered clinically significant by the Investigator (e.g., SAE or AE or dose-modification) then, the results must be recorded in the CRF.

Results of clinical laboratory testing will be recorded on the eCRF or be received as electronically produced laboratory reports submitted directly from the local or central laboratory.

Additional blood or urine samples may be taken at the discretion of the Investigator if the results of any test fall outside the reference ranges, or clinical symptoms necessitate additional testing to monitor participant safety.

Where the clinical significance of abnormal lab results is considered uncertain, screening lab tests may be repeated before randomization to confirm eligibility.

If there is an alternative explanation for a positive urine or blood test for drugs of abuse, e.g., previous occasional intake of a medication or food containing for example, codeine, benzodiazepines or opiates, the test could be repeated to confirm washout.

Based on continuous analysis of the data in this study and other studies, any sample type not considered to be critical for safety may be stopped at any time if the data from the samples collected does not produce useful information.

8.2.5 Safety Biomarker Assessments

Quantitative changes in T_{reg}, T_{eff}, NK cells, and eosinophils will be evaluated as exploratory safety biomarkers. Cytokines IL-6, IL-8, TNF-α or IL-1β may be measured as exploratory safety biomarkers ad hoc in participants experiencing symptoms associated with injection-related reactions. These assessments are described in Sections 8.6 and 8.8.

IgE, tryptase, safety cytokines, and further exploratory safety biomarkers may be tested in case of participants experiencing signs and symptoms associated with IRRs. This includes hypersensitivity reactions.

8.2.6 Immunogenicity Assessments

Blood samples for ADA will be taken as described in the SoA (Section 1.3). The date and time of each sample will be recorded in the eCRF. Additional samples will be taken in participants with clinical signs of hypersensitivity reaction for ADA evaluation (see SoA Section 1.3). In each case, for each collected ADA sample, a corresponding PK sample will be collected at the same time-point for the determination of the RO7049665 concentration (See Section 8.5). Additional ADA sampling may be required after the safety and follow-up visit (Day 99) for participants who are ADA positive at the Day 99 visit. In this instance no corresponding PK sample is needed, however a sCD25 sample should be taken.

Validated ADA assays will be employed to detect ADAs against RO7049665. Samples which are positive for ADA will be further characterized and their neutralizing capability for recombinant IL-2 will be assessed using the neutralizing antibody (Nab) assay.

If required, remaining ADA samples may also be used for assay development/validation experiments. For exploratory purposes such as epitope characterization or IgG-IgM-isotyping, samples may be analyzed by assays which are not fully validated. Such samples will be kept for up to 6 months after the finalization of the bioanalytical report.

Samples from placebo-treated participants may not be analyzed in the first instance, but retained for subsequent analysis if appropriate.

Details on sampling procedures, sample storage and shipment are documented in the sample handling manual.

8.2.7 Local Pain and Skin Reactivity Assessments

Local pain and injection-site reactions (ISRs; e.g., burning, bleeding, itching, bruising, redness, hive formation, or other) will be assessed at the time-points indicated in the SoA (Section 1.3, Appendix 6). Photographs of skin may be taken on an ad hoc basis but are not required per protocol.

8.2.8 Clinical Outcome Assessment

Monitoring for signs and symptoms of worsening ulcerative colitis (e.g., rectal bleeding, stool frequency, abdominal pain, fever) will take place at every clinic visit. Elevation of the partial Mayo score (physician's global assessment, rectal bleeding, and stool frequency) by more than 2 points from baseline over two consecutive visits (scheduled or unscheduled) will meet disease exacerbation criteria. If disease worsening criteria meet individual stopping criteria (see Section 4.1.4) then participants will be treated as judged appropriate by their treating physician.

8.2.9 Medical History and Demographic Data

Medical history includes clinically significant diseases and all medications (e.g., prescription drugs, over-the-counter drugs, herbal or homeopathic remedies, nutritional supplements) used by the participant within 14 days prior to the screening visit.

Demographic data will include age, sex, and self-reported race/ethnicity.

8.3 ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

The definitions of an AE or SAE can be found in Appendix 2. The non-serious AEs of special interest and disease-related events and/or disease-related outcomes not qualifying as AEs or SAEs are discussed in Sections 8.3.6 and 8.3.7.

The Investigator and any qualified designees are responsible for ensuring that all AEs (including assessment of seriousness, severity and causality; see Appendix 2) are recorded on the Adverse Event eCRF and reported to the Sponsor in accordance with instructions provided in this section and in Appendix 2.

Procedures used for recording AEs are provided in Appendix 3:

- Diagnosis versus signs and symptoms:
  - Injection reactions
  - Other AEs
- AEs occurring secondary to other events
- Persistent or recurrent AEs
- Abnormal laboratory values
- Abnormal vital sign values
- Abnormal liver function tests
- Deaths
- Preexisting medical conditions
- Lack of efficacy or worsening of the condition being studied
- Hospitalization or prolonged hospitalization
- Patient-reported outcome data

8.3.1 Time Period and Frequency for Collecting Adverse Event and Serious Adverse Event Information

The method of recording, evaluating, and assessing causality of AEs and SAEs and the procedures for completing and transmitting SAE reports are provided in Appendix 2.

Investigators will seek information on adverse events at each participant's contact. All AEs, whether reported by the participant or noted by study personnel, will be recorded in the participant's medical record and on the Adverse Event eCRF as follows:

After informed consent has been obtained **but prior to initiation of study treatment**, only SAEs caused by a protocol-mandated intervention should be reported (e.g., SAEs related to invasive procedures such as biopsies). Any other adverse event should not be reported.

After initiation of study treatment, all AEs, regardless of relationship to study treatment, will be reported until the Day 99 safety and follow-up visit, or, 28 days after the last dose of study treatment in case of early termination.

Post-study adverse events and serious adverse events: The Investigator is not required to actively monitor participants for AEs after the final follow-up visit.

However, if the Investigator learns of any SAE (including a death) or other AEs of concern that are believed to be related to prior treatment with study treatment, at any time after a participant has been discharged from the study, and the Investigator considers the event to be reasonably related to the study treatment or study participation, the Investigator must promptly notify the Sponsor. For the procedure of reporting, see Appendix 2.

8.3.2 Method of Detecting Adverse Events and Serious Adverse Events

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the participant is the preferred method to inquire about AE occurrence.

A consistent methodology of non-directive questioning should be adopted for eliciting AE information at all participant evaluation time−points.

8.3.3 Follow-Up of Adverse Events and Serious Adverse Events

8.3.3.1 Investigator Follow-Up

The Investigator should follow each adverse event until the event has resolved to baseline grade or better, the event is assessed as stable by the Investigator, the event is otherwise explained, the participant is lost to follow-up (Section 7.3), or the participant withdraws consent. Every effort should be made to follow all SAEs considered to be related to study treatment or trial-related procedures until a final outcome can be reported.

During the study period, resolution of AEs (with dates) should be documented on the Adverse Event eCRF and in the participant's medical record to facilitate source data

verification. If, after follow-up, return to baseline status or stabilization cannot be established, an explanation should be recorded on the Adverse Event eCRF.

All pregnancies reported during the study should be followed until pregnancy outcome and reported according to the instructions provided in Section 8.3.5.

8.3.3.2 Sponsor Follow-Up

For SAEs, non-serious AEs of special interest, and pregnancies, the Sponsor or a designee may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details and outcome information (e.g., from hospital discharge summaries, consultant reports, autopsy reports) in order to perform an independent medical assessment of the reported case.

8.3.4 Regulatory Reporting Requirements for Serious Adverse Events

Prompt notification by the Investigator to the Sponsor of a SAE is essential so that legal obligations and ethical responsibilities towards the safety of participants and the safety of a study treatment under clinical investigation are met.

The Sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of a study treatment under clinical investigation. The Sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, IRB/ IEC, and investigators.

Investigator safety reports must be prepared for suspected unexpected serious adverse reactions (SUSAR) according to local regulatory requirements and Sponsor policy and forwarded to investigators as necessary.

An investigator who receives an investigator safety report describing a SAE or other specific safety information (e.g., summary or listing of SAEs) from the Sponsor will review and then, file it along with the Investigator's Brochure and will notify the IRB/IEC, if appropriate according to local requirements.

For immediate and expedited reporting requirements from Investigator to Sponsor and from Sponsor to Health Authority, investigators, IRB and EC, see Appendix 2.

8.3.4.1 Emergency Medical Contacts

To ensure the safety of study participants, access to the Medical monitors is available 24 hours a day 7 days a week. Details will be available on a separately.

8.3.5 Pregnancy

Female participants of childbearing potential will be instructed to immediately inform the Investigator if they become pregnant during the study or within 56 days after the last dose of study treatment.

Male participants will be instructed through the ICF to immediately inform the Investigator if their partner becomes pregnant during the study or within 56 days after the last dose of study drug.

If a pregnancy is reported, the Investigator should inform the Sponsor within 24 hours of learning of the pregnancy and should follow the pregnancy reporting process as detailed in Appendix 5.

Abnormal pregnancy outcomes (e.g., spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAEs (Appendix 5).

8.3.6 Non-Serious Adverse Events of Special Interest

Non-serious adverse events of special interest are required to be reported by the Investigator to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Appendix 2 for reporting instructions).

Non-serious adverse events of special interest for this study include the following:

- NCI CTCAE (v5.0) ≥ G2 hypersensitivity reaction/ *IRR*.
- Cases of an elevated alanine aminotransferase (ALT) or aspartate aminotransferase (AST) in combination with either an elevated bilirubin or clinical jaundice, as defined in Appendix 3.
- Suspected transmission of an infectious agent by the study treatment, as defined below:

  Any organism, virus, or infectious particle (e.g., prion protein transmitting transmissible spongiform encephalopathy), pathogenic or non-pathogenic, is considered an infectious agent. A transmission of an infectious agent may be suspected from clinical symptoms or laboratory findings that indicate an infection in a patient exposed to a medicinal product. This term applies only when a contamination of the study treatment is suspected.

8.3.7 Disease-Related Events and/or Disease-Related Outcomes Not Qualifying as AEs or SAEs

Not applicable for the participants in this study.

8.3.8 Management of Specific Adverse Events

Treatment of AEs will be considered on a case-by-case basis according to local standard of care.

For participants who report AEs of systemic IRRs or systemic hypersensitivities, following the first occurrence of an event, pre-medicate with paracetamol and antihistamine (as per local guidelines) prior to next administration of study treatment. At the reporting of an AE of systemic IRR or hypersensitivity, the Sponsor must be contacted.

If a subsequent systemic IRR or hypersensitivity AE occurs, stop dosing in this participant (see Section 4.1.4).

8.4 TREATMENT OF OVERDOSE

Study treatment overdose is the accidental administration of a drug in a quantity that is higher than the assigned dose. An overdose or incorrect administration of study treatment is not an AE unless it results in untoward medical effects (see Sections 5 and 5.2 of Appendix 2 for further details).

Any study drug overdose or incorrect administration of study drug should be noted on the Study Drug Administration eCRF.

All AEs associated with an overdose or incorrect administration of study drug should be recorded on the Adverse Event eCRF.

In the event of a dose administration error, the Investigator should:

1. Contact the Sponsor's Medical Monitor immediately.
2. Closely monitor the participant for AE/SAE and laboratory abnormalities until resolved.
3. Obtain a blood sample for PK analysis as soon as possible from the time of the last dose of study treatment, if requested by the Medical Monitor (determined on a case-by-case basis).
4. If known, document the variance from planned dose administration, in the CRF.

8.5 PHARMACOKINETICS

Blood samples will be collected to determine serum concentration of RO7049665. Samples will be collected at the timepoints specified in the Schedules of Activities (Section 1.3) and at any time point when an ADA sample is taken (Section 8.2.6).

The date and time of each sample collection will be recorded in the eCRF. Serum RO7049665 concentrations will be measured using validated specific enzyme-linked immunosorbent assay (ELISA). If deemed appropriate, samples may also be assayed with non-validated exploratory methods.

During the course of the study, PK sampling time-points may be modified and/or up to 3 additional PK samples may be collected per participant on the basis of emerging data in order to allow for optimal characterization of the PK profile of RO7049665, after agreement with the Sponsor's clinical pharmacologist and the investigator.

Additional PK samples will be taken at the time of treatment discontinuation, at time of ADA sample collection if additional ADA samples are collected, and if the participant experiences an infusion-related AE (such as an injection-related reaction [IRR]).

Samples collected from placebo-treated participants will not be analyzed in the first instance, but retained for subsequent analysis if appropriate.

Any volume of blood samples remaining after the specified analyses may also be used for additional assay development or study or compound-specific validation experiments (e.g., for PK Parallelism Assessment and ADAs).

Samples will be destroyed within 2 years after the date of final clinical study report (CSR).
Details on sampling procedures, sample storage and shipment are given in the Sample Handling Manual.

8.6 PHARMACODYNAMICS

Samples will be collected for PD assessments at the time points specified in the SOA (Section 1.3). The date and time of each sample will be recorded in the eCRF.

Blood samples

Blood samples will be collected for the following assessments:

- To quantify relative and absolute WBC counts (T cells, B cells, NK cells, and eosinophils) by flow cytometry as well as complete white blood count differential. Tests include (but are not limited to) markers for quantification and characterization of T_{regs} and T_{eff} cell subsets CD3, CD4, CD8, CD25, CD127 and FoxP3.
- Additional blood samples will be collected for potential evaluation of T cell subset specific methylation signature changes induced by drug. Decision to perform this exploratory assessment involving deoxyribonucleic acid (DNA) extraction will be made based on emerging PD data.

Tissue samples

Tissue colon biopsy samples will be collected for the following assessments:

- To characterize infiltrating WBCs by histology (hematoxylin and eosin staining) and immunohistochemistry (IHC). IHC markers tested may include but are not limited to CD3, CD4 and FoxP3.
- RNA extraction on tissue colon biopsy samples may be performed in order to undertake optional whole or targeted transcriptome gene expression analysis.
- Bacterial DNA or RNA may be extracted for subsequent mucosa-associated metagenomic analysis.
- Methylation signature changes induced by drug indicative of quantitative and specific changes of T cell subsets may be assessed. The decision to perform this exploratory assessment involving DNA extraction will be made based on emerging PD data.

Serum and plasma samples

Serum and plasma samples will be collected for the following assessments:

- To measure cytokines or cytokine receptors mechanistically linked with the drug's mode of action. Soluble biomarkers considered for testing include, but are not limited to, IL-5, IL-6, IL-8, IL-10 and sCD25.The final panel of soluble biomarkers will be informed by the outcome of the EIH study (WP39826) and the analysis of the resulting biomarker data, as well as by emerging data obtained in study WP40161.
- Cytokines IL-6, IL-8, TNF-α or IL‑1b may in addition be measured as exploratory safety biomarkers ad hoc in participants experiencing symptoms associated with injection‑related reactions.
- Exploratory disease biomarkers with potential for patient stratification or non-invasive response to treatment may be tested including, but not limited to perinuclear anti-neutrophil cytoplasmic antibody (pANCA) and Leucine-rich Alpha-2 Glycoprotein (LRG)
- Additional soluble biomarkers to obtain further confidence on mechanism of action of RO7049665 in humans, PD or disease markers may be tested.

Stool samples

Stool samples will be collected to evaluate exploratory disease biomarkers with potential for patient stratification or non-invasive response to treatment monitoring including but not limited to faecal Calprotectin.

Leftover biological samples.

Any remaining stool, serum, plasma, blood or colon biopsy sample after the specified analyses may also be used for additional method, study or compound specific (assay) validation experiments or biomarker measurements. The stool, serum, plasma samples, and tissue colon biopsy samples will be destroyed within 5 years after the date of final CSR. For participants who consent to RBR, leftover samples will be transferred to RBR (see Section 8.8.2.1).

8.7 GENETICS

8.7.1 Clinical Genotyping

A mandatory whole blood sample will be taken for DNA extraction from every participant in all countries where this assessment is permitted (See SoA Section 1.3). The DNA may be used for, but analysis is not limited to:

- Genotyping of alleles in genes encoding proteins involved in IL 2-mediated signal transduction
- Genotyping of alleles in genes, for which genetic polymorphisms have been previously associated with autoimmune diseases.

Data arising from all biosamples including samples for analyses of inherited DNA will be subject to the confidentiality standards described in Section 1.4 of Appendix 1.

Any remaining blood sample after the specified analyses may also be used for additional method, study or compound (assay) validation experiments.

The DNA or blood samples will be destroyed within 5 years after the date of final CSR. For participants who consent to RBR, leftover blood and DNA samples will be transferred to RBR (see Section 8.8.2.1).

Details on processes for collection and shipment of these samples can be found in Sample Handling Manual.

8.8 BIOMARKERS

Samples will be collected for biomarker assessments at the time points specified in the Schedules of Activities (Section 1.3). Biomarkers will be evaluated as described in Sections 8.6, and 8.7.

8.8.1 Exploratory Disease Biomarkers

Exploratory disease biomarkers will be tested for their ability to stratify participants or to evaluate their potential for non-invasive response to treatment monitoring. Markers to be tested may include, but are not limited to perinuclear anti-neutrophil cytoplasmic antibody (pANCA), LRG, and faecal Calprotectin.

8.8.2 Samples for Research Biosample Repository

8.8.2.1 Overview of the Research Biosample Repository

The Roche Research Biosample Repository (RBR) is a centrally administered group of facilities for the long-term storage of human biologic specimens, including body fluids, solid tissues, and derivatives thereof (e.g., DNA, RNA, proteins, peptides). The collection, storage and analysis of these specimens will facilitate the rational design of new pharmaceutical agents and the development of diagnostic tests, which may allow for individualized drug therapy for participants in the future.

For participants who consent to RBR, leftover samples will be transferred to RBR.

Leftover specimens will be used to achieve the following objectives:

- To study the association of biomarkers with efficacy, AEs, or progressive disease.
- To increase knowledge and understanding of disease biology.
- To study treatment response, including drug effects and the processes of drug absorption and disposition.
- To develop biomarker or diagnostic assays and establish the performance characteristics of these assays.

- To enable further exploratory biomarker research.

8.8.2.2 Sample Collection

The following samples will be stored in the RBR and used for research purposes, including, but not limited to, research on biomarkers related to study treatment, target and/or disease:

- Leftover plasma samples
- Leftover serum samples
- Leftover colon biopsy (and any derivatives) samples
- Leftover blood samples for DNA extraction
- Leftover extracted RNA and DNA samples
- Leftover stool samples

The samples collected for DNA extraction include, but are not limited to, genomic analysis and may be sent to one or more laboratories for analysis of germline or somatic mutations via whole genome sequencing (WGS), whole exome sequencing (WES), next-generation sequencing (NGS), or other genomic analysis methods.

Genomics is increasingly informing researcher's understanding of disease pathobiology. WGS provides a comprehensive characterization of the genome and, along with clinical data collected in this study, may increase the opportunity for developing new therapeutic approaches. Data will be analyzed in the context of this study but will also be explored in aggregate with data from other studies. The availability of a larger dataset will assist in identification of important pathways, guiding the development of new targeted agents.

Samples may be sent to one or more laboratories for analysis for WGS and other genomic analyses and associated clinical data may be shared with researchers who are not participating in the study or submitted to government or other health research databases for broad sharing with other researchers. Participant will not be identified by name or any other personally identifying information. WGS data will be analyzed in the context of this study but will also be explored in aggregate with data from other studies to increase researcher's understanding of disease pathobiology and guide the development of new therapeutic approaches. Given the complexity and exploratory nature of these analyses, WGS data and analyses will not be shared with investigators or study participants unless required by law.

For all samples, dates of consent and specimen collection should be recorded on the associated RBR page of the eCRF. For sampling procedures, storage conditions, and shipment instructions, see the separate Laboratory Manual.

RBR specimens will be stored and used until no longer needed or until they are exhausted. The Research Biosample Repository storage period will be in accordance

with the IRB/EC‑approved Informed Consent Form and applicable laws (e.g., health authority requirements).

The repository specimens will be subject to the confidentiality standards (as described under Confidentiality and in Appendix 1).

8.9 HEALTH ECONOMICS

Health Economics/Medical Resource Utilization and Health Economics parameters are not evaluated in this study.

8.10 TIMING OF STUDY ASSESSMENTS

8.10.1 Screening and Pre-treatment Assessments

Written informed consent for participation in the study must be obtained before performing any study‑specific screening tests or evaluations. Informed consent may be obtained up to 3 months prior to randomization. Screening start will be defined as the first assessment done after signature of the informed consent form. Informed Consent Forms (ICFs) for enrolled participant and for participants who are not subsequently enrolled will be maintained at the study site.

All screening and pre-treatment assessments must be completed and reviewed to confirm that participants meet all eligibility criteria. The Investigator will maintain a screening log to record details of all participants screened and to confirm eligibility or record reasons for screening failure.

An Eligibility Screening Form (ESF) documenting the Investigator's assessment of each screened participant with regard to the protocol's inclusion and exclusion criteria is to be completed by the Investigator and kept at the investigational site.

Screening and pre-treatment assessments will be performed within 35 days prior to Day 1 unless otherwise specified.

8.10.2 Assessments during Treatment

Under no circumstances will participants who enroll in this study and have completed treatment as specified, be permitted to be allocated a new randomization number and re-enroll in the study.

All assessments must be performed as per SoA (see Section 1.3). Assessments scheduled on the day of study treatment administration should be performed prior to administration of study treatment, unless otherwise noted in the schedule of activities. COA assessments (Section 8.2.7) should be performed prior to the completion of other study assessments.

8.10.3 Assessments at Study Completion/Early Termination Visit

Participants who complete the study (defined in Section 4.4) will be asked to return to the clinic 56 days after the last dose of study drug for a follow-up visit. Participants who discontinue from the study early will be asked to return to the clinic 28 days after the last dose of study drug for a follow-up visit.

8.10.4 Follow-Up Assessments

After the study completion/early termination visit, AEs should be followed as outlined in the SoA (Section 1.3) and in Sections 8.3.1 and 8.3.3.

Participants positive for ADAs at the study completion/early termination visit will be asked to return for further ADA assessments.

8.10.5 Assessments at Unscheduled Visits

Please see Section 1.3 for activities that are required to be performed in case of an unscheduled visit.

9. STATISTICAL CONSIDERATIONS

9.1 STATISTICAL HYPOTHESES

Not applicable.

9.2 SAMPLE SIZE DETERMINATION

The study will enroll up to *65* participants to assess the safety and tolerability of repeated dosing of RO7049665 (approximately 10 participants, randomized in the ratio of 4:1 RO7049665 to placebo, per cohort).

The sample size for this study was selected on the basis of clinical judgment and practicality. It was not determined on the basis of power against a formal statistical hypothesis. However, with a full cohort of 8 participants treated at a given dose level, there is an 83% chance to observe at least one AE that has a true incidence rate of 20% at that dose level in the population. After 4 participants have been treated at a given dose level, there is a 59% chance of observing such an AE.

9.2.1 Populations for Analyses

For purposes of analysis, the following populations are defined in Table 6.

Table 6 Analysis Populations

Population	Description
Intent-to-treat (ITT)	All randomized participants will be included in the intent-to-treat population.
Per-protocol (PP)	All randomized participants that received the expected number of doses will be included in the per-protocol population.
Safety	All participants randomized to study treatment and who received at least one dose of the study treatment, whether prematurely withdrawn from the study or not, will be included in the safety analysis.
Pharmacokinetic	Participants who have received active treatment will be included in the PK analysis population. Participants will be excluded from the PK analysis population if they significantly violate the inclusion or exclusion criteria, deviate significantly from the protocol, or if data are unavailable or incomplete which may influence the PK analysis. Excluded cases will be documented together with the reason for exclusion. All decisions on exclusions from the analysis will be made prior to database closure.
Efficacy	Participants who were included in the ITT population.
Pharmacodynamic	Participants who were included in the Safety analysis population.
Biomarker	Participants who were included in the Safety analysis population.
Immunogenicity	Participants who had at least one pre-dose and one post-dose ADA assessment will be included and analyzed according to the treatment they actually received. In first instance, only samples from RO7049665-treated participants will be analyzed. The relationship between ADA status and safety, efficacy, PK, and biomarker endpoints will be analyzed and reported descriptively.

9.3 STATISTICAL ANALYSES

9.3.1 Demographics and Baseline Characteristics

The following information collected at baseline will be summarized descriptively in the Safety analysis population to assess the comparability of treatment groups:

- Demographics
- Medical history
- Concomitant medications
- PD biomarkers

- Measures of endoscopic and histological appearance of mucosa of sigmoid colon

9.3.2 Safety Analyses

All safety analyses will be based on the Safety analysis population, with participants grouped according to the treatment received (dose level / dose frequency).

Table 7 Safety Statistical Analysis Methods

Endpoint	Statistical Analysis Methods
Adverse events	The original terms recorded on the eCRF by the Investigator for adverse events will be coded by the Sponsor. Adverse events will be summarized by mapped term and appropriate thesaurus level.
Clinical laboratory tests	All clinical laboratory data will be stored on the database in the units in which they were reported. Laboratory test values will be presented in International System of Units (SI units; Système International d'Unités) by individual listings with flagging of abnormal results. Summary tables of change from baseline over time will be displayed. See Appendix 4 for details on standard reference ranges and data transformation and the definition of laboratory abnormalities.
Vital signs	Vital signs data will be presented by individual listings with flagging of values outside the normal ranges and flagging of abnormalities. In addition, tabular summaries will be used, as appropriate.
ECG data analysis	ECG data will be presented by individual listings. In addition, tabular summaries will be used, as appropriate.
Concomitant medications	The original terms recorded on the participants' eCRF by the Investigator for concomitant medications will be standardized by the Sponsor by assigning preferred terms. Concomitant medications will be presented in summary tables and listings.
Other	Physical examination results, safety biomarkers and incidence of ADAs will be listed and summarized descriptively.

9.3.3 Pharmacokinetic Analyses

PK parameters will be read directly from the serum concentration-time profiles or calculated by using standard non-compartmental methods. The following PK parameters will be computed for RO7049665. However, other PK parameters might be computed in addition as appropriate:

- Time to maximum concentration (T_{max}) post the dose on Day 1, and on Day of last dose
- C_{max} post the dose on Day 1, and post the dose on Day of last dose
- Area under the serum concentration-time curve after the first dose administration extrapolated to infinity (AUC_{inf}) and over the dosing interval (AUC_{tau}) after the last dose administration (where tau=2 weeks). If deemed useful, a partial AUC_{0-t} may be used instead of AUC_{inf} or AUC_{tau} should extrapolation to infinity or up to tau not be considered reliable.

- Apparent clearance after the first dose administration (CL/F = Dose/AUC_{inf}) as well as after the last dose administration (CL_{ss}/F = Dose/AUC_{tau})
- Apparent volume of distribution (V/F) after the first dose administration
- Estimates of $T_{1/2}$ after the first dose (Day 1) as well as washout after last dose administration

The dose proportionality will be evaluated in an exploratory manner. C_{max} and AUC_{tau} (or AUC0-t if deemed useful) post the dose on Day 1 and C_{max} and AUC_{tau} post the dose on Day of last dose will be tested for dose proportionality. This will be done using one way Analysis of Variance (ANOVA) to test the dose effect using dose normalization and a natural log transform.

In addition, to evaluate drug accumulation, individual as well as mean (SD) of apparent peak (12 hour post dose) and trough (predose) after each dose administration will be plotted graphically over the course of assessments as per Table 2. In addition, AUC_{tau} post the dose on Day 1 will be tested statistically against AUC_{tau} post the dose on Day of last dose using an ANOVA with terms in the model for participants and day. A natural log transform of the data will be done prior to the analysis.

Individual and mean serum concentrations at each sampling time point for RO7049665 will be presented by listings and descriptive summary statistics including arithmetic means, geometric means, ranges, standard deviations, and coefficients of variation. Individual and mean concentration versus time will be plotted on linear or semi-logarithmic scales as appropriate.

All PK parameters will be presented by individual listings and summary statistics including arithmetic means, geometric means, medians, ranges, standard deviations, and coefficients of variation.

9.3.4 Efficacy Analyses

Preliminary efficacy will be evaluated as a secondary objective of the study by comparing between dose groups the following endoscopic and histologic measures of disease activity. Change from baseline to Day 57 will be analyzed and reported descriptively.

- Change from baseline to Day 57 (and descriptively at Day 29) in endoscopic indices of disease activity (Ulcerative Colitis Endoscopic Index of Severity [UCEIS] and MCS-ES
- Change from baseline to Day 57 (and descriptively at Day 29) in histology score of sigmoid colon biopsies (GS, RHI, and Nancy Histology Index [NHI])

In addition, preliminary efficacy will also be evaluated by comparing overall disease activity between dose groups using:

- Change from baseline to Day 57 (and descriptively at Day 29) in MCS

The main foreseen intercurrent event for this study is early treatment discontinuation. In the scenario that no intercurrent events occur (i.e., all participants complete the expected number of doses), the estimand is uncontroversial as simply the mean difference between treatment groups in change from baseline to Day 57 in efficacy scores. If however, intercurrent events are observed, three different strategies will be employed to handle early treatment discontinuation, each leading to their own respective estimands.

A treatment policy strategy (ITT population) will be used to estimate the difference between dose groups in the change from baseline in endoscopy, histology and overall disease activity scores, regardless of whether early treatment discontinuation occurred. In the absence of missing data, this can be achieved using Analysis of Covariance (ANCOVA) with dose group as a factor, otherwise an imputation method will be applied as appropriate.

A composite strategy (ITT population) will be used for endoscopy and overall disease activity scores to estimate the difference in the proportion of responders between dose groups, where responders are defined as those participants who reach a threshold of change from baseline in efficacy scores, in the absence of early treatment discontinuation. Endoscopic response is defined as at least a 1 point decrease in the MCS-ES and at least a 3 point decrease in the UCEIS. Clinical response (overall disease activity) is defined as at least a 3 point and 30% decrease from baseline in the MCS. MCS remission is a stool frequency score of 1 or 0 (with at least a 1 point decrease), a rectal bleeding score of 0, and an endoscopy score of 1 or 0 but with no friability. Endoscopic remission will be defined as a UCEIS score of 1 or 0. Treatment failure is defined as non-response or early treatment discontinuation. In the absence of missing data this can be analyzed using logistic regression with dose group as a factor, otherwise an imputation method will be applied as appropriate.

An on-treatment strategy (PP population) will be used to estimate the difference between dose groups in the change from baseline in endoscopy, histological and overall disease activity scores, restricted to those participants who completed the expected number of doses of their randomly assigned treatment. In the absence of missing data this can be achieved using ANCOVA with dose group as a factor, otherwise an imputation method will be applied as appropriate.

9.3.5 Immunogenicity Analyses

The immunogenicity analyses will include participants with at least one pre-dose and one post-dose ADA assessment.

The numbers and proportions of ADA-positive patients and ADA-negative participants during both the treatment and follow-up periods will be summarized.

- Participants are considered to be ADA positive if they are ADA negative at baseline but develop an ADA response following study drug administration (treatment-induced ADA response), or if they are ADA positive at baseline and the titer of one or more post-baseline samples is greater than the titer of the baseline sample by a scientifically reasonable margin such as at least 4-fold (treatment-enhanced ADA response).
- Patients are considered to be ADA negative if they are ADA negative at baseline and all post-baseline samples are negative, or if they are ADA positive at baseline but do not have any post-baseline samples with a titer that is greater than the titer of the baseline sample by a scientifically reasonable margin such as at least 4-fold (treatment unaffected).

The relationship between ADA status and safety, efficacy, PK, and biomarker endpoints may be analyzed and reported descriptively.

9.3.6 Pharmacodynamic Analyses

All PD analyses will be based on the Safety analysis population. Individual and mean PD data and parameters will be presented by listings and descriptive summary statistics including means, geometric means, medians, ranges, standard deviations, and coefficients of variation. Listings for the change from baseline (absolute and relative) and difference to placebo, as appropriate, and the corresponding summary statistics may also be presented. Graphical displays may be used, as appropriate.

9.3.7 Other Analyses

Exploratory biomarker analyses may be performed to investigate:

The association of biomarkers with improvements in endoscopic appearance of the mucosa, inflammatory infiltrate, or AEs.

Genotyping data may be analyzed for association with PD, efficacy, or safety of RO7049665.

The dose-exposure-response for pharmacological, clinical endpoints, and safety markers may be evaluated using graphical and pharmacometric analyses. Modeling and simulation may also be used for dose-escalation/decision purposes. If required, the population PK/PD analysis will be reported separately from the main CSR.

9.4 INTERIM ANALYSES

No formal interim analyses are planned.

9.5 SUMMARIES OF CONDUCT OF STUDY

All protocol deviations will be listed. Data for study drug administration and concomitant medication will be listed. The number of participants who were randomized, discontinued and completed the study will be summarized and listed.

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11. SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

The following section includes standard appendices such as Appendix 1 (for regulatory, ethical and study oversight considerations), Appendix 2 (for AE definitions, reporting) and Appendix 3 (procedures of recording), Appendix 4 (clinical laboratory tests), Appendix 5 (contraceptive guidance and collection of pregnancy information. Appendix 6 (pain and skin reactivity assessments), Appendix 7 (mayo clinical score [MCS]) and Appendix 8 (rationales for previous protocol amendments). Additional study-related appendices are in order of appearance in the protocol.

Appendix 1
Regulatory, Ethical, and Study Oversight Considerations

1. REGULATORY AND ETHICAL CONSIDERATIONS

1.1. COMPLIANCE WITH LAWS AND REGULATIONS

This study will be conducted in full conformance with the ICH E6 guideline for Good Clinical Practice and the principles of the Declaration of Helsinki, or the laws and regulations of the country in which the research is conducted, whichever affords the greater protection to the individual. The study will comply with the requirements of the ICH E2A guideline (Clinical Safety Data Management: Definitions and Standards for Expedited Reporting). Studies conducted in the United States or under a U.S. Investigational New Drug (IND) application will comply with U.S. FDA regulations and applicable local, state, and federal laws. Studies conducted in the European Union (EU)/ European Economic Area (EEA) will comply with the EU Clinical Trial Directive (2001/20/EC).

1.2. INSTITUTIONAL REVIEW BOARD OR ETHICS COMMITTEE

This protocol, the ICFs, any information to be given to the participant (e.g., advertisements, diaries etc), and relevant supporting information must be submitted to the IRB/EC by the Principal Investigator and reviewed and approved by the IRB/EC before the study is initiated. In addition, any participant recruitment materials must be approved by the IRB/EC.

The Principal Investigator is responsible for providing written summaries of the status of the study to the IRB/EC annually or more frequently in accordance with the requirements, policies, and procedures established by the IRB/EC. Investigators are also responsible for promptly informing the IRB/EC of any protocol amendments (Section 2.3.1 of this Appendix).

The Investigator should follow the requirements for reporting all adverse events to the Sponsor. Investigators may receive written IND safety reports or other safety-related communications from the Sponsor. Investigators are responsible for ensuring that such reports are reviewed and processed in accordance with health authority requirements and the policies and procedures established by their IRB/EC, and archived in the site's study file.

1.3. INFORMED CONSENT

The Sponsor's Master Informed Consent Form (and ancillary sample ICFs such as a Child's Assent or Caregiver's Informed Consent Form, if applicable) will be provided to each site. If applicable, it will be provided in a certified translation of the local language. Participants must be informed that their participation is voluntary. Participants will be required to sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act

(HIPAA) requirements, where applicable, and the IRB/IEC or study center. The Sponsor or its designee must review and approve any proposed deviations from the Sponsor's sample ICFs or any alternate consent forms proposed by the site (collectively, the "Consent Forms") before IRB/EC submission. The final IRB/EC‑approved Consent Forms must be provided to the Sponsor for health authority submission purposes according to local requirements. Participants must be re-consented to the most current version of the ICF(s) during their participation in the study. A copy of the ICF(s) signed by all parties must be provided to the participant.

The Consent Forms must be signed and dated by the participant before his or her participation in the study. The case history or clinical records for each participant shall document the informed consent process and that written informed consent was obtained prior to participation in the study.

The Consent Forms should be revised whenever there are changes to study procedures or when new information becomes available that may affect the willingness of the participant to take part. The final revised IRB/EC-approved Consent Forms must be provided to the Sponsor for health authority submission purposes if required as per local regulations.

Participants must be re-consented to the most current version of the Consent Forms (or to a significant new information/findings addendum in accordance with applicable laws and IRB/EC policy) during their participation in the study. For any updated or revised Consent Forms, the case history or clinical records for each participant shall document the informed consent process and that written informed consent was obtained using the updated/revised Consent Forms for continued participation in the study.

A copy of each signed Consent Form must be provided to the participant. All signed and dated Consent Forms must remain in each participant's study file or in the site file and must be available for verification by study monitors at any time.

A participant who is re-screened is not required to sign another ICF if the re-screening occurs within 30 days from the previous ICF signature date.

Consent to Participate in Exploratory Testing of Residual Material

The ICF will contain a separate section that addresses the use of remaining mandatory samples for optional exploratory research described in Section 8. A separate signature will be required to document a participant's agreement to allow any remaining specimens to be used for exploratory research.

Consent to Participate in the Research Biosample Repository

The Informed Consent Form will contain a separate section that addresses participation in the RBR. The investigator or authorized designee will explain to each participant the objectives, methods, and potential hazards of participation in the RBR. Participants will

be told that they are free to refuse to participate and may withdraw their specimens at any time and for any reason during the storage period. A separate, specific signature will be required to document a participant's agreement to provide optional RBR specimens. Participants who decline to participate will not provide a separate signature.

The Investigator should document whether or not the participant has given consent to participate by completing the RBR Sample Informed Consent eCRF.

In the event of death or loss of competence of a participant who is participating in the Research, the participant's specimens and data will continue to be used as part of the RBR.

For sites in the United States, each Consent Form may also include patient authorization to allow use and disclosure of personal health information in compliance with the U.S. Health Insurance Portability and Accountability Act of 1996 (HIPAA). If the site utilizes a separate Authorization Form for patient authorization for use and disclosure of personal health information under the HIPAA regulations, the review, approval, and other processes outlined above apply except that IRB review and approval may not be required per study site policies.

Approval by the Institutional Review Board or Ethics Committee

Sampling for the RBR is contingent upon the review and approval of the exploratory research and the RBR portion of the Informed Consent Form by each site's Institutional Review Board or Ethics Committee (IRB/EC) and, if applicable, an appropriate regulatory body. If a site has not been granted approval for RBR sampling, this section of the protocol will not be applicable at that site

Withdrawal from the Research Biosample Repository

Participants who give consent to provide specimens for the RBR have the right to withdraw their specimens at any time for any reason. If a participant wishes to withdraw consent to the testing of his or her specimens, the Investigator must inform the Medical Monitor in writing of the participant's wishes using the RBR Withdrawal Form and, if the trial is ongoing, must enter the date of withdrawal on the RBR Withdrawal of Informed Consent eCRF. The participant will be provided with instructions on how to withdraw consent after the trial is closed. A participant's withdrawal from Study WP40161 does not, by itself, constitute withdrawal of specimens from the RBR. Likewise, a participant's withdrawal from the RBR does not constitute withdrawal from Study WP40161. Data already generated before time of withdrawal of consent to RBR will still be used.

1.4. CONFIDENTIALITY

Participants will be assigned a unique identifier by the Sponsor. Any participant records or datasets that are transferred to the Sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.

The participant must be informed that his/her personal study-related data will be used by the Sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participant.

Medical information may be given to a participant's personal physician or other appropriate medical personnel responsible for the participant's welfare, for treatment purposes.

The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.

Confidentiality for Research Biosample Repository

Data generated from RBR specimens must be available for inspection upon request by representatives of national and local Health Authorities, and Roche monitors, representatives, and collaborators, as appropriate.

Participant medical information associated with RBR specimens is confidential and may only be disclosed to third parties as permitted by the Informed Consent Form (or separate authorization for use and disclosure of personal health information) signed by the participant, unless permitted or required by law.

Data derived from RBR specimen analysis on individual participants will generally not be provided to study investigators unless a request for research use is granted. The aggregate results of any conducted research will be available in accordance with the effective Roche policy on study data publication.

Genetic research data and associated clinical data may be shared with researchers who are not participating in the study or submitted to government or other health research databases for broad sharing with other researchers. Participants will not be identified by name or any other personally identifying information. Given the complexity and exploratory nature of these analyses, genetic data and analyses will not be shared with investigators or participants unless required by law.

Any inventions and resulting patents, improvements, and/or know-how originating from the use of the RBR specimen data will become and remain the exclusive and unburdened property of Roche, except where agreed otherwise.

Monitoring and Oversight Research Biosample Repository

Specimens collected for the RBR will be tracked in a manner consistent with Good Clinical Practice by a quality-controlled, auditable, and appropriately validated laboratory information management system, to ensure compliance with data confidentiality as well as adherence to authorized use of specimens as specified in this protocol and in the Informed Consent Form. Roche monitors and auditors will have direct access to appropriate parts of records relating to participant participation in RBR for the purposes

of verifying the data provided to Roche. The site will permit monitoring, audits, IRB/EC review, and health authority inspections by providing direct access to source data and documents related to the samples.

1.5. FINANCIAL DISCLOSURE

Investigators will provide the Sponsor with sufficient, accurate financial information in accordance with local regulations to allow the Sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate Health Authorities. Investigators are responsible for providing information on financial interests during the course of the study and for one year after completion of the study (i.e., LPLO).

2. DATA HANDLING AND RECORD

2.1. DATA COLLECTION AND MANAGEMENT RESPONSIBILITIES

2.1.1. Data Quality Assurance

All participant data relating to the study will be recorded on printed or electronic CRF unless transmitted to the Sponsor or designee electronically (e.g., laboratory data). The Investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the CRF.

The Investigator must maintain accurate documentation (source data) that supports the information entered in the CRF.

The Investigator must permit study-related monitoring, audits, IRB/IEC review, and regulatory agency inspections and provide direct access to source data documents.

The Sponsor or designee is responsible for the data management of this study including quality checking of the data.

Study monitors will perform ongoing source data verification to confirm that data entered into the CRF by authorized site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.

2.1.3. Source Data Records

Source documents (paper or electronic) are those in which participant data are recorded and documented for the first time. They include, but are not limited to, hospital records, clinical and office charts, laboratory notes, memoranda, COAs (paper or eCOA), evaluation checklists, pharmacy dispensing records, recorded data from automated instruments, copies of transcriptions that are certified after verification as being accurate and complete, microfiche, photographic negatives, microfilm or magnetic media, X-rays, patient files, and records kept at pharmacies, laboratories, and medico‑technical departments involved in a clinical trial.

Before study initiation, data to be entered directly into the eCRFs (i.e., no prior written or electronic record of the data) and considered source data must be defined in the Trial Monitoring Plan.

Source documents that are required to verify the validity and completeness of data entered into the eCRFs must not be obliterated or destroyed and must be retained per the policy for retention of records described below.

To facilitate source data verification, the Investigators and institutions must provide the Sponsor direct access to applicable source documents and reports for trial-related monitoring, Sponsor audits, and IRB/EC review. The investigational site must also allow inspection by applicable Health Authorities.

2.1.4. Use of Computerized Systems

When clinical observations are entered directly into an investigational site's computerized medical record system (i.e., in lieu of original hardcopy records), the electronic record can serve as the source document if the system has been validated in accordance with health authority requirements pertaining to computerized systems used in clinical research. An acceptable computerized data collection system allows preservation of the original entry of data. If original data are modified, the system should maintain a viewable audit trail that shows the original data as well as the reason for the change, name of the person making the change, and date of the change.

2.2. RETENTION OF RECORDS

Records and documents, including signed ICF, pertaining to the conduct of this study must be retained by the Investigator for at least 15 years after study completion or discontinuation of the study, or for the length of time required by relevant national or local health authorities, whichever is longer. After that period of time, the documents may be destroyed, subject to local regulations. No records may be destroyed during the retention period without the written approval of the Sponsor. No records may be transferred to another location or party without written notification to the Sponsor.

Roche will retain study data for 25 years after the final study results have been reported or for the length of time required by relevant national or local health authorities.

2.3. STUDY RECORDS

The Investigator must maintain adequate and accurate records to enable the conduct of the study to be fully reconstructed, including but not limited to the protocol, protocol amendments, ICFs, and documentation of IRB/EC and governmental approval.

Roche shall also submit an Annual Safety Report once a year to the IEC and CAs according to local regulatory requirements and timelines of each country participating in the study.

2.3.1. Protocol Amendments

Any substantial protocol amendments will be prepared by the Sponsor. Substantial protocol amendments will be submitted to the IRB/EC and to regulatory authorities in accordance with local regulatory requirements.

Approval must be obtained from the IRB/EC and regulatory authorities (as locally required) before implementation of any changes, except for changes necessary to eliminate an immediate hazard to participants or any non-substantial changes, as defined by regulatory requirements.

2.3.2. Publication Policy

The results of this study may be published or presented at scientific meetings. If this is foreseen, the Investigator agrees to submit all manuscripts or abstracts to the Sponsor for approval prior to submission. This allows the Sponsor to protect proprietary information and to provide comments based on information from other studies that may not yet be available to the Investigator.

The Sponsor will comply with the requirements for publication of study results. In accordance with standard editorial and ethical practice, the Sponsor will generally support publication of multicenter trials only in their entirety and not as individual center data. In this case, a coordinating Investigator will be designated by mutual agreement.

Any formal publication of the study in which contribution of Sponsor personnel exceeded that of conventional monitoring will be considered as a joint publication by the Investigator and the appropriate Sponsor personnel.

Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

Any inventions and resulting patents, improvements, and/or know-how originating from the use of data from this study will become and remain the exclusive and unburdened property of the Sponsor, except where agreed otherwise.

2.3.3. Dissemination of Clinical Study Data

A clinical study report containing the results of this trial will be made available to anyone who requests a copy.

2.3.4. Site Inspections

Site visits will be conducted by the Sponsor or an authorized representative for inspection of study data, participants' medical records, and eCRFs. The Investigator will permit national and local Health Authorities, Sponsor monitors, representatives, and collaborators, and the IRBs/ECs to inspect facilities and records relevant to this study.

3. ADMINISTRATIVE STRUCTURE

The Sponsor of this trial is F. Hoffmann-La Roche Lt. The Sponsor is responsible for the study management (monitoring will be outsourced to a CRO), data management, statistical analysis, and medical writing for the clinical study report (CSR). The CSR will be written and submitted to Health Authorities in the timeframe as required by applicable regulatory requirements. The protocol will be submitted to country's IRB/IEC.

3.1 INTERNAL MONITORING COMMITTEE (IMC)

A dedicated group of selected Sponsor representatives, which will be independent to the project, will be reviewing the safety data of the study. The roles, responsibilities, membership, scope of activities, time of meetings and communication plan for the IMC will be documented in an appropriate charter.

4. STUDY AND SITE CLOSURE

The Sponsor (or designee) has the right to close the study site or terminate this study at any time. Reasons for terminating the study may include, but are not limited to, the following:

- The incidence or severity of adverse events in this or other studies indicates a potential health hazard to participants
- Participant enrollment is unsatisfactory.

If general study stopping criteria are met, the study will be paused and the Sponsor will conduct a full review of all available safety data. The study will resume if the Sponsor and investigator agree and after informing and receiving agreement from relevant Health Authorities and Ethics Committees.

The Sponsor will notify the Investigator and Health Authorities if the study is placed on hold, or if the Sponsor decides to discontinue the study or development program.

Study sites will be closed upon study completion. A study site is considered closed when all required documents and study supplies have been collected and a study-site closure visit has been performed.

The Investigator may initiate study-site closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study site by the Sponsor or Investigator may include but are not limited to:

- Failure of the Investigator to comply with the protocol, the requirements of the IRB/IEC or local Health Authorities, the Sponsor's procedures, or GCP guidelines.
- Inadequate recruitment of participants by the Investigator.
- Discontinuation of further study treatment development.

Appendix 2
Adverse Events: Definitions and Procedures for Evaluating, Follow-up and Reporting

1. DEFINITION OF ADVERSE EVENTS

According to the E2A ICH guideline for Good Clinical Practice, an **adverse event** is any untoward medical occurrence in a patient or clinical investigation participant administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment.

An adverse event can therefore be:

- Any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Events Meeting the AE Definition:

- Any deterioration in a laboratory value (hematology, clinical chemistry, or urinalysis) or other clinical test (e.g., ECG, X−ray) that is associated with symptoms or leads to a change in study treatment or concomitant treatment or discontinuation from study treatment.
- Exacerbation of a chronic or intermittent pre-existing condition, including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study treatment administration even though it may have been present before the start of the study.
- Adverse events that are related to a protocol-mandated intervention, including those that occur prior to assignment of study treatment (e.g., screening invasive procedures such as biopsies).
- "Lack of efficacy" or "failure of expected pharmacological action" per se will not be reported as an AE or SAE unless the progression is unexpectedly accelerated and not in line with the natural history of the disease. If the “Lack of efficacy” would not require safety reporting such instances will be captured in the efficacy assessments. However, the signs, symptoms, and/or clinical sequelae resulting from lack of efficacy will be reported as AE or SAE if they fulfill the definition of an AE or SAE.

Events NOT Meeting the AE Definition:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the Investigator to be more severe than expected for the participant’s condition.
- The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the participant’s condition.

- Medical or surgical procedure (e.g., endoscopy, appendectomy): the condition that leads to the procedure is an AE.
- Situations where an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

2. DEFINITION OF SERIOUS ADVERSE EVENTS

If an event is not an AE per definition above, then it cannot be a serious adverse event (SAE) even if serious conditions are met (e.g., hospitalization for signs/symptoms of the disease under study, death due to progression of disease).

A serious adverse event is defined as any untoward medical occurrence that at any dose:

- **Results in death.**
- **Is life-threatening.**

  The term "life-threatening" in the definition of "serious" refers to an event in which the participant was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it was more severe.
- **Requires inpatient hospitalization or prolongation of existing hospitalization** (see Appendix 3).

  In general, hospitalization signifies that the participant has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred or was necessary, the AE should be considered serious.

  Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.
- **Results in persistent or significant disability/incapacity**

  Disability means substantial disruption of the participant's ability to conduct normal life functions.

  This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (e.g., sprained ankle) which may interfere with or prevent everyday life functions but do not constitute a substantial disruption.
- **Is a congenital anomaly/birth defect**.

- **Other significant events**:

  Medical or scientific judgment should be exercised in deciding whether SAE reporting is appropriate in other situations such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These events should usually be considered serious.

  Examples of such events include invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

  For **protocol-specific significant events**, see Sections 4.1.4 and 7.1 for consideration.

3. RECORDING OF ADVERSE EVENT AND/OR SERIOUS ADVERSE EVENT

When an AE/SAE occurs, it is the responsibility of the Investigator to review all documentation (e.g., hospital progress notes, laboratory reports, and diagnostics reports) related to the event.

The Investigator will then record all relevant AE/SAE information in the CRF.

It is **not** acceptable for the Investigator to send photocopies of the participant's medical records to Medical Monitor in lieu of completion of the eCRF.

There may be instances when copies of medical records for certain cases are requested by the Sponsor. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to the Sponsor.

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

3.1. ASSESSMENT OF SEVERITY

The terms "severe" and "serious" are not synonymous, i.e.:

- Severe refers to a category utilized for rating the intensity of an event; and both AEs and SAEs can be assessed as severe.
- Serious refers to an event that meets at least 1 of the predefined outcomes as described in the definition of an SAE, NOT when it is rated as severe

Severity and seriousness need to be independently assessed for each adverse event recorded on the eCRF.

The adverse event severity grading scale for the NCI CTCAE (v5.0) will be used for assessing adverse event severity. Table 1 will be used for assessing severity for adverse events that are not specifically listed in the NCI CTCAE.

Table 1 Adverse Event Severity Grading Scale

Grade	Severity
1	Mild; asymptomatic or mild symptoms; clinical or diagnostic observations only; or intervention not indicated
2	Moderate; minimal, local, or non-invasive intervention indicated; or limiting age-appropriate instrumental activities of daily living[a]
3	Severe or medically significant, but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; or limiting self-care activities of daily living[b,c]
4	Life-threatening consequences or urgent intervention indicated[d]
5	Death related to adverse event[d]

NCI CTCAE = National Cancer Institute Common Terminology Criteria for Adverse Events.

Note: Based on the NCI CTCAE (v5.0), which can be found at: https://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_50 and https://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/CTCAE_v5_Quick_Reference_8.5x11.pdf

[a] Instrumental activities of daily living refer to preparing meals, shopping for groceries or clothes, using the telephone, managing money, etc.

[b] Examples of self-care activities of daily living include bathing, dressing and undressing, feeding one's self, using the toilet, and taking medications, as performed by patients who are not bedridden.

[c] If an event is assessed as a "significant medical event," it must be reported as a serious adverse event (see Section 6 of this Appendix for reporting instructions), per the definition of serious adverse event in Section 2.

[d] Grade 4 and 5 events must be reported as serious adverse events (see Section 6 for reporting instructions), per the definition of serious adverse event in Section 2 with the exception of Grade 4 laboratory abnormality events which should only be reported as a SAE if it meets one or more of the serious conditions outlined in Section 2 (Definition of Serious Adverse Events) of Appendix 2.

Serious adverse events are required to be reported by the Investigator to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Appendix 3 for reporting instructions).

3.2. ASSESSMENT OF CAUSALITY

Investigators should use their knowledge of the participant, the circumstances surrounding the event, and an evaluation of any potential alternative causes to determine whether or not an adverse event is considered to be related to the study treatment, indicating "yes" or "no" accordingly. The following guidance should be taken into consideration:

- Temporal relationship of event onset to the initiation of study treatment.

- Course of the event, considering especially the effects of dose-reduction, discontinuation of study treatment, or reintroduction of study treatment.
- Known association of the event with the study treatment or with similar treatments.
- Known association of the event with the disease under study.
- Presence of risk factors in the participant or use of concomitant medications known to increase the occurrence of the event.
- Presence of non-treatment-related factors that are known to be associated with the occurrence of the event.

For participant receiving combination therapy, causality will be assessed individually for each protocol-mandated therapy.

4. FOLLOW-UP OF AES AND SAES

The Investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by the Sponsor to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.

If a participant dies during participation in the study or during a recognized follow-up period, the Investigator will provide the Sponsor with a copy of any post-mortem findings including histopathology.

New or updated information will be recorded in the originally completed eCRF.

The Investigator will submit any updated SAE data to the Sponsor within 24 hours of receipt of the information.

5. IMMEDIATE REPORTING REQUIREMENTS FROM INVESTIGATOR TO SPONSOR

Certain events require immediate reporting to allow the Sponsor to take appropriate measures to address potential new risks in a clinical trial. The Investigator must report such events to the Sponsor immediately; under no circumstances should reporting take place more than 24 hours after the Investigator learns of the event. The following is a list of events that the Investigator must report to the Sponsor within 24 hours after learning of the event, regardless of relationship to study treatment:

- Serious adverse events.
- Non-serious adverse events of special interest (NSAESI).
- Pregnancies (see Section 8.3.5).
- DLAEs (see Section 4.1.3; see the eCRF completion guideline for further guidance).

The Investigator must report new significant follow-up information for these events to the Sponsor immediately (i.e., no more than 24 hours after becoming aware of the information). New significant information includes the following:

- New signs or symptoms or a change in the diagnosis.
- Significant new diagnostic test results.
- Change in causality based on new information.
- Change in the event's outcome, including recovery.
- Additional narrative information on the clinical course of the event.

Investigators must also comply with local requirements for reporting serious adverse events to the local health authority and IRB/EC.

5.1 REPORTING REQUIREMENTS OF SERIOUS ADVERSE EVENTS, NON-SERIOUS ADVERSE EVENTS OF SPECIAL INTEREST AND DOSE-LIMITING ADVERSE EVENTS

Events that Occur prior to Study Treatment Initiation

After informed consent has been obtained but prior to initiation of study treatment, only serious adverse events caused by a protocol-mandated intervention should be reported. The Serious Adverse Event/Adverse Event of Special Interest Reporting Form provided to Investigators should be completed and submitted to the Serious Adverse Event Responsible immediately (i.e., no more than 24 hours after learning of the event).

Events that Occur after Study Treatment Initiation

For reports of serious adverse events and non-serious adverse events of special interest (Section 8.3.6) that occur after initiation of study treatment (Section 8.3.1), investigators should record all case details that can be gathered immediately (i.e., within 24 hours after learning of the event) on the appropriate Adverse Event of Special Interest/ Serious Adverse Event eCRF form and submit the report via the electronic data capture (EDC) system. A report will be generated and sent to the Sponsor's Safety Risk Management department.

In the event that the EDC system is unavailable, the Serious Adverse Event/Adverse Event of Special Interest Reporting Form provided to investigators should be completed and submitted to the Serious Adverse Event Responsible immediately (i.e., no more than 24 hours after learning of the event).

Once the EDC system is available, all information will need to be entered and submitted via the EDC system.

Reporting of Post-Study Adverse Events and Serious Adverse Events

If the Investigator becomes aware of any other serious adverse event occurring after the end of the AE reporting period, if the event is believed to be related to prior study treatment the event should be reported directly to the Sponsor or its designee, either by faxing or by scanning and emailing the SAE Reporting Form using the fax number or email address provided to investigators.

5.2 REPORTING REQUIREMENTS FOR CASES OF ACCIDENTAL OVERDOSE OR MEDICATION ERROR

Accidental overdose and medication error (hereafter collectively referred to as "special situations"), are defined as follows:

- Accidental overdose: accidental administration of a drug in a quantity that is higher than the assigned dose
- Medication error: accidental deviation in the administration of a drug

In some cases, a medication error may be intercepted prior to administration of the drug.

Special situations are not in themselves adverse events, but may result in adverse events. Each adverse event associated with a special situation should be recorded separately on the Adverse Event eCRF. If the associated adverse event fulfills seriousness criteria, the event should be reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event). For RO7049665 and placebo, adverse events associated with special situations should be recorded as described below for each situation:

- Accidental overdose: Enter the adverse event term. Check the "Accidental overdose" and "Medication error" boxes.
- Medication error that does not qualify as an overdose: Enter the adverse event term. Check the "Medication error" box.
- Medication error that qualifies as an overdose: Enter the adverse event term. Check the "Accidental overdose" and "Medication error" boxes.

In addition, all special situations associated with RO7049665 and placebo, regardless of whether they result in an adverse event, should be recorded on the Adverse Event eCRF and should be recorded as described below:

- Accidental overdose: Enter the drug name and "accidental overdose" as the event term. Check the "Accidental overdose" and "Medication error" boxes.
- Medication error that does not qualify as an overdose: Enter the name of the drug administered and a description of the error (e.g., wrong dose administered, wrong dosing schedule, incorrect route of administration, wrong drug, expired drug administered) as the event term. Check the "Medication error" box.
- Medication error that qualifies as an overdose: Enter the drug name and "accidental overdose" as the event term. Check the "Accidental overdose" and "Medication error" boxes. Enter a description of the error in the additional case details.

- Intercepted medication error: Enter the drug name and "intercepted medication error" as the event term. Check the "Medication error" box. Enter a description of the error in the additional case details.

As an example, an accidental overdose that resulted in a headache would require the completion of two Adverse Event eCRF pages, one to report the accidental overdose and one to report the headache. The "Accidental overdose" and "Medication error" boxes would need to be checked on both eCRF pages.

6. EXPEDITED REPORTING TO HEALTH AUTHORITIES, INVESTIGATORS, INSTITUTIONAL REVIEW BOARDS, AND ETHICS COMMITTEES

The Sponsor will promptly evaluate all serious adverse events and NSAESI against cumulative product experience to identify and expeditiously communicate possible new safety findings to investigators, IRBs, ECs, and applicable Health Authorities based on applicable legislation.

To determine reporting requirements for single adverse event cases, the Sponsor will assess the expectedness of these events using the following reference document{s}:

- RO7049665 Investigator's Brochure

The Sponsor will compare the severity of each event and the cumulative event frequency reported for the study with the severity and frequency reported in the applicable reference document.

Reporting requirements will also be based on the Investigator's assessment of causality and seriousness, with allowance for upgrading by the Sponsor as needed.

Appendix 3
Procedures for Recording Adverse Events

Investigators should use correct medical terminology/concepts when recording adverse events on the Adverse Event eCRF. Avoid colloquialisms and abbreviations.

Only one adverse event term should be recorded in the event field on the Adverse Event eCRF.

1. DIAGNOSIS VERSUS SIGNS AND SYMPTOMS

1.1. INJECTION REACTIONS

Adverse events that occur during or after study drug administration and are judged to be related to study treatment injection should be captured as a diagnosis (e.g., injection-related reaction, injection-site reaction or anaphylactic reaction) on the Adverse Event eCRF. If possible, avoid ambiguous terms such as “systemic reaction”. Associated signs and symptoms should be recorded on the dedicated Injection Reaction eCRF. If a participant experiences both a local and systemic reaction to the same dose of study drug, each reaction should be recorded separately on the Adverse Event eCRF, with signs and symptoms also recorded separately on the dedicated Injection Reaction eCRF.

1.2. OTHER ADVERSE EVENTS

For adverse events other than injection reactions, a diagnosis (if known) should be recorded on the Adverse Event eCRF rather than individual signs and symptoms (e.g., record only liver failure or hepatitis rather than jaundice, asterixis, and elevated transaminases). However, if a constellation of signs and/or symptoms cannot be medically characterized as a single diagnosis or syndrome at the time of reporting, each individual event should be recorded on the Adverse Event eCRF. If a diagnosis is subsequently established, all previously reported adverse events based on signs and symptoms should be nullified and replaced by one adverse event report based on the single diagnosis, with a starting date that corresponds to the starting date of the first symptom of the eventual diagnosis.

2. ADVERSE EVENTS OCCURRING SECONDARY TO OTHER EVENTS

In general, adverse events occurring secondary to other events (e.g., cascade events or clinical sequelae) should be identified by their primary cause, with the exception of severe or serious secondary events. However, medically significant adverse events occurring secondary to an initiating event that are separated in time should be recorded as independent events on the Adverse Event eCRF. For example:

- If vomiting results in mild dehydration with no additional treatment in a healthy adult, only vomiting should be reported on the eCRF.

- If vomiting results in severe dehydration, both events should be reported separately on the eCRF.
- If a severe gastrointestinal hemorrhage leads to renal failure, both events should be reported separately on the eCRF.
- If dizziness leads to a fall and subsequent fracture, all three events should be reported separately on the eCRF.

All adverse events should be recorded separately on the Adverse Event eCRF if it is unclear as to whether the events are associated.

3. PERSISTENT OR RECURRENT ADVERSE EVENTS

A persistent adverse event is one that extends continuously, without resolution, between participant evaluation time-points. Such events should only be recorded once on the Adverse Event eCRF. The initial severity of the event should be recorded, and the severity should be updated to reflect the most extreme severity any time the event worsens. If the event becomes serious, the Adverse Event eCRF should be updated to reflect this.

A recurrent adverse event is one that resolves between participant evaluation time-points and subsequently recurs. Each recurrence of an adverse event should be recorded separately on the Adverse Event eCRF.

4. ABNORMAL LABORATORY VALUES

Not every laboratory abnormality qualifies as an adverse event. A laboratory test result should be reported as an adverse event if it meets any of the following criteria:

- Accompanied by clinical symptoms.
- Results in a change in study treatment (e.g., dosage modification, treatment interruption, or treatment discontinuation).
- Results in a medical intervention (e.g., potassium supplementation for hypokalemia) or a change in concomitant therapy.
- Clinically significant in the Investigator's judgment.

It is the Investigator's responsibility to review all laboratory findings. Medical and scientific judgment should be exercised in deciding whether an isolated laboratory abnormality should be classified as an adverse event.

If a clinically significant laboratory abnormality is a sign of a disease or syndrome (e.g., alkaline phosphatase and bilirubin 5 times the upper limit of normal [ULN] associated with cholecystitis), only the diagnosis (i.e., cholecystitis) should be recorded on the Adverse Event eCRF.

If a clinically significant laboratory abnormality is not a sign of a disease or syndrome, the abnormality itself should be recorded on the Adverse Event eCRF, along with a descriptor indicating if the test result is above or below the normal range (e.g., "elevated potassium", as opposed to "abnormal potassium"). If the laboratory abnormality can be characterized by a precise clinical term per standard definitions, the clinical term should be recorded as the adverse event. For example, an elevated serum potassium level of 7.0 mEq/L should be recorded as “hyperkalemia”.

Observations of the same clinically significant laboratory abnormality from visit to visit should not be repeatedly recorded on the Adverse Event eCRF, unless the etiology changes. The initial severity of the event should be recorded, and the severity or seriousness should be updated any time the event worsens.

5. ABNORMAL VITAL SIGN VALUES

Not every vital sign abnormality qualifies as an adverse event. A vital sign result should be reported as an adverse event if it meets any of the following criteria:

- Accompanied by clinical symptoms.
- Results in a change in study treatment (e.g., dosage modification, treatment interruption, or treatment discontinuation).
- Results in a medical intervention or a change in concomitant therapy.
- Clinically significant in the Investigator’s judgment.

It is the Investigator’s responsibility to review all vital sign findings. Medical and scientific judgment should be exercised in deciding whether an isolated vital sign abnormality should be classified as an adverse event.

If a clinically significant vital sign abnormality is a sign of a disease or syndrome (e.g., high blood pressure), only the diagnosis (i.e., hypertension) should be recorded on the Adverse Event eCRF.

Observations of the same clinically significant vital sign abnormality from visit to visit should not be repeatedly recorded on the Adverse Event eCRF, unless the etiology changes. The initial severity of the event should be recorded, and the severity or seriousness should be updated any time the event worsens.

6. ABNORMAL LIVER FUNCTION TESTS

The finding of an elevated ALT or AST ($>3\times$ULN or >2 x baseline value if abnormal at baseline) in combination with either an elevated total bilirubin ($>2\times$ULN) or clinical jaundice in the absence of cholestasis or other causes of hyperbilirubinemia is considered to be an indicator of severe liver injury. Therefore, investigators must report as an adverse event the occurrence of either of the following:

- Treatment-emergent ALT or AST $>3\times$ULN (or >2 x baseline if value was abnormal at baseline) in combination with total bilirubin $>2\times$ULN.
- Treatment-emergent ALT or AST $>3\times$ULN (or >2 x baseline if value was abnormal at baseline) in combination with clinical jaundice.

The most appropriate diagnosis or (if a diagnosis cannot be established) the abnormal laboratory values should be recorded on the Adverse Event eCRF (see Appendix 2) and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event), either as a serious adverse event or a non-serious adverse event of special interest (see Section 8.3.6).

7. DEATHS

All deaths that occur during the protocol-specified adverse event reporting period (see Section 5 of Appendix 2), regardless of relationship to study treatment, must be recorded on the Adverse Event eCRF and immediately reported to the Sponsor (see Appendix 3).

Death should be considered an outcome and not a distinct event. The event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the Adverse Event eCRF. Generally, only one such event should be reported. If the cause of death is unknown and cannot be ascertained at the time of reporting, “unexplained death” should be recorded on the Adverse Event eCRF. If the cause of death later becomes available (e.g., after autopsy), “unexplained death” should be replaced by the established cause of death. The term "sudden death" should not be used unless combined with the presumed cause of death (e.g., "sudden cardiac death").

8. PREEXISTING MEDICAL CONDITIONS

A preexisting medical condition is one that is present at the screening visit for this study. Such conditions should be recorded on the General Medical History and Baseline Conditions eCRF.

A preexisting medical condition should be recorded as an adverse event only if the frequency, severity, or character of the condition worsens during the study. When recording such events on the Adverse Event eCRF, it is important to convey the concept that the preexisting condition has changed by including applicable descriptors (e.g., “more frequent headaches”).

9. LACK OF EFFICACY OR WORSENING OF ULCERATIVE COLITIS

Medical occurrences or symptoms of deterioration that are anticipated as part of ulcerative colitis should be recorded as an adverse event if judged by the Investigator to have unexpectedly worsened in severity or frequency or changed in nature at any time during the study. When recording an unanticipated worsening of ulcerative colitis on the Adverse Event eCRF, it is important to convey the concept that the condition has changed by including applicable descriptors (e.g., “accelerated ulcerative colitis”).

10. HOSPITALIZATION OR PROLONGED HOSPITALIZATION

Any adverse event that results in hospitalization or prolonged hospitalization should be documented and reported as a serious adverse event (per the definition of serious adverse event in Appendix 2), except as outlined below.

An event that leads to hospitalization under the following circumstances should not be reported as an adverse event or a serious adverse event:

- Hospitalization for respite care
- Planned hospitalization required by the protocol (e.g., for study treatment administration).
- Hospitalization for a preexisting condition, provided that all of the following criteria are met:

– The hospitalization was planned prior to the study or was scheduled during the study when elective surgery became necessary because of the expected normal progression of the disease.

– The participant has not suffered an adverse event.

An event that leads to hospitalization under the following circumstances is not considered to be a serious adverse event, but should be reported as an adverse event instead:

- Hospitalization for an adverse event that would ordinarily have been treated in an outpatient setting had an outpatient clinic been available.

Appendix 4
Clinical Laboratory Tests

The tests detailed in Table 1 will be performed by a local laboratory with the exception of Clostridium difficile*, which will be tested at a central laboratory.* Ova and parasite stool examination at screening *may be done locally at sites with such capabilities*. The results of the local laboratory assessments must be captured in source documentation and entered into the eCRF.

Any additional laboratory test (e.g., safety biomarkers listed in the Schedule of Activities but not listed in Table 1) will be performed by a central laboratory.

Protocol-specific requirements for inclusion or exclusion of participants are detailed in Sections 5.1 and 5.2, respectively, of the protocol.

Additional tests may be performed at any time during the study as determined necessary by the Investigator or required by local regulations.

Table 1 Protocol-Required Safety Laboratory Assessments

Laboratory Assessments	Parameters
Hematology	• Leucocytes, erythrocytes, hemoglobin, hematocrit, platelets, differential count (neutrophils, eosinophils, basophils, monocytes, lymphocytes).
Clinical Chemistry	• Sodium, potassium, chloride, bicarbonate, non-fasting glucose, urea, creatinine, creatine kinase (CK), protein, albumin, phosphate, calcium, total and direct bilirubin, alkaline phosphatase, ALT, AST, urate, hSCRP, GGT
Coagulation	• Prothrombin time (i.e., INR) and activated thromboplastin time (aPTT).
Viral Serology	• HIV (specific tests HIV-1 antibody, HIV-1/2 antibody, HIV-2 antibody), hepatitis B surface antigen (HBsAg), total hepatitis B core antibody (HBcAb), hepatitis C virus (HCV) by PCR.
Pregnancy Test	• All women of childbearing potential (including those who have had a tubal occlusion/ligation) will have a blood pregnancy test at screening. Urine pregnancy tests will be performed at specified subsequent visits. If a urine pregnancy test is positive, it must be confirmed by a blood pregnancy test.
Urinalysis	• Specific gravity • Dipstick: pH, glucose, protein, blood. • If there is a clinically significant positive result (confirmed by a positive repeated sample), urine will be sent to the laboratory for microscopy and culture. If there is an explanation for the positive dipstick results (e.g., menses), it should be recorded and there is no need to perform microscopy and culture. • Microscopic examination (red blood cells [RBCs], white blood cells [WBCs], casts, crystals, epithelial cells, bacteria), if blood or protein is abnormal.
Other Screening Tests	• Urine drug screen (to include at minimum: amphetamines, barbiturates, cocaine, opiates, and benzodiazepines). • Alcohol breath or blood test (as per local procedure). • *Clostridium difficile* (ELISA) • Ova and parasite stool examination
Other Tests	• IgE • Tryptase

The results of each test must be entered into the CRF.

Investigators must document their review of each laboratory safety report.

Laboratory results that could unblind the study that are performed centrally will not be reported to investigative sites or other blinded personnel until the study has been unblinded.

Additional Statistical Considerations for Clinical Laboratory Data

- Standard Reference Ranges and Transformation of Data

Roche standard reference ranges, rather than the reference ranges of the Investigator, will be used for all parameters. For most parameters, the measured laboratory test result will be assessed directly using the Roche standard reference range. Certain laboratory parameters will be transformed to Roche's standard reference ranges.

A transformation will be performed on certain laboratory tests that lack sufficiently common procedures and have a wide range of Investigator ranges, e.g., enzyme tests that include AST, ALT, and alkaline phosphatase and total bilirubin. Since the standard reference ranges for these parameters have a lower limit of zero, only the upper limits of the ranges will be used in transforming the data.

- Definition of Laboratory Abnormalities

For all laboratory parameters included, there exists a Roche predefined standard reference range. Laboratory values falling outside this standard reference range will be labeled "H" for high or "L" for low in participant listings of laboratory data.

In addition to the standard reference range, a marked reference range has been predefined by Roche for each laboratory parameter. The marked reference range is broader than the standard reference range. Values falling outside the marked reference range that also represent a defined change from baseline will be considered marked laboratory abnormalities (i.e., potentially clinically relevant). If a baseline value is not available for a participant, the midpoint of the standard reference range will be used as the participant's baseline value for the purposes of determining marked laboratory abnormalities. Marked laboratory abnormalities will be labeled in the participant listings as "HH" for very high or "LL" for very low.

Appendix 5
Contraceptive Guidance and Collection of Pregnancy Information

1. DEFINITIONS

- **Woman of Childbearing Potential (WOCBP)**

A woman is considered fertile following menarche and until becoming post-menopausal unless permanently sterile.

- **Women in the following categories are considered to be Woman of Non-Childbearing Potential (WONCBP)**

a. Pre-menarchal

b. Pre-menopausal female with one of the following:

– Documented hysterectomy.

– Documented bilateral salpingectomy.

– Documented bilateral oophorectomy.

Note: Documentation can come from the site personnel's: review of participant's medical records, medical examination, or medical history interview.

c. Post-menopausal female

– A post-menopausal state is defined as no menses for ≥ 12 months without an alternative medical cause other than menopause. A high follicle-stimulating hormone (FSH) level in the post-menopausal range may be used to confirm a post-menopausal state in women not using hormonal contraception or hormonal replacement therapy (HRT). However, in the absence of 12 months of amenorrhea, a single FSH measurement is insufficient.

– Females on HRT and whose menopausal status is in doubt will be required to use one of the non-hormonal highly effective contraception methods if they wish to continue their HRT during the study. Otherwise, they must discontinue HRT to allow confirmation of post-menopausal status before study enrollment.

2. CONTRACEPTION GUIDANCE

- **Female Participants**

Female participants of childbearing potential are eligible to participate if they agree to use a highly effective method of contraception consistently and correctly as described in Table 1 below.

Per ICH M3(R2), highly effective methods of birth control are defined as those, alone or in combination, that result in a low failure rate (i.e., less than 1% per year) when used consistently and correctly.

Table 1 Highly Effective Contraceptive Methods

Highly Effective Contraceptive Methods That Are User-Dependent[a] (Failure rate of < 1% per year when used consistently and correctly)
Combined (estrogen- and progestogen-containing) hormonal contraception associated with inhibition of ovulation • Oral • Intravaginal • Transdermal Progestogen-only hormonal contraception associated with inhibition of ovulation • Oral • Injectable
Highly Effective Methods That Are User-Independent[a]
• Implantable progestogen-only hormonal contraception associated with inhibition of ovulation • Intrauterine device (IUD) • Intrauterine hormone-releasing system (IUS) • Bilateral tubal occlusion/ligation **Vasectomized partner** A vasectomized partner is a highly effective contraception method provided that the partner is the sole male sexual partner of the WOCBP and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used. **Sexual abstinence** Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study treatment. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.

a) Typical use failure rates may differ from those when used consistently and correctly. Use should be consistent with local regulations regarding the use of contraceptive methods for participants participating in clinical studies.

3. PREGNANCY TESTING

For WOCBP enrolled in the study, blood sample and urine pregnancy tests will be performed according to Schedule of Activity tables (see Section 1.3). If a urine pregnancy test is positive, it must be confirmed by a blood pregnancy test.

Pregnancy testing will be performed whenever a menstrual cycle is missed or when pregnancy is otherwise suspected and according to local practice.

4. COLLECTION OF PREGNANCY INFORMATION

- **Male participants with partners who become pregnant**

The Investigator will attempt to collect pregnancy information on any male participant's female partner who becomes pregnant while the male participant is in this study (see Section 8.3.5 Pregnancy). This applies only to male participants who receive RO7049665.

Attempts should be made to collect and report details of the course and outcome of any pregnancy in the partner of a male participant exposed to study treatment. The Investigator will record pregnancy information on the Clinical Trial Pregnancy Reporting Form and submit it to the Sponsor within 24 hours of learning of the partner's pregnancy. When permitted by the site, the pregnant partner would need to sign an Authorization for Use and Disclosure of Pregnancy Health Information to allow for follow-up on her pregnancy. If the authorization has been signed, the Investigator should update the Clinical Trial Pregnancy Reporting Form with additional information on the course and outcome of the pregnancy when available. An Investigator who is contacted by the male participant or his pregnant partner may provide information on the risks of the pregnancy and the possible effects on the fetus, to support an informed decision in cooperation with the treating physician and/or obstetrician. The female partner will be followed to determine the outcome of the pregnancy. Information on the status of the mother and child will be forwarded to the Sponsor. Monitoring of the participant's partner should continue until conclusion of the pregnancy. Any termination of the pregnancy will be reported regardless of fetal status (presence or absence of anomalies) or indication for procedure.

- **Female participants who become pregnant**

The Investigator will collect pregnancy information on any female participant, who becomes pregnant while participating in this study (see Section 8.3.5 Pregnancy). Information will be recorded on the appropriate form and submitted to the Sponsor within 24 hours of learning of a participant's pregnancy. The participant will be followed to determine the outcome of the pregnancy. The Investigator will collect follow-up information on the participant and the neonate, which will be forwarded to the Sponsor. Monitoring of the participant should continue until conclusion of the pregnancy. Any

termination of pregnancy will be reported, regardless of fetal status (presence or absence of anomalies) or indication for procedure.

While pregnancy itself is not considered to be an AE or SAE, and should not be recorded on the AE eCRF, any pregnancy complication will be reported as an AE or SAE. A spontaneous abortion is always considered to be an SAE and will be reported as such. Any post-study pregnancy related SAE considered reasonably related to the study treatment by the Investigator, will be reported to the Sponsor as described in Appendix 2. While the Investigator is not obligated to actively seek this information in former study participants, he/she may learn of an SAE through spontaneous reporting.

Any female participant who becomes pregnant while participating in the study will be withdrawn from the study.

5 ABORTIONS

Any spontaneous abortion should be classified as a serious adverse event (as the Sponsor considers spontaneous abortions to be medically significant events), recorded on the Adverse Event eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5 of Appendix 2).

Any induced abortion due to maternal toxicity and/or embryo-fetal toxicity should also be classified as serious adverse event, recorded on the Adverse Event eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5 of Appendix 2).

Elective abortion not associated with toxicity (e.g., induced abortion for personal reasons) does not require expedited reporting but should be reported as outcome of pregnancy on the Clinical Trial Pregnancy Reporting Form.

6 CONGENITAL ANOMALIES/BIRTH DEFECTS

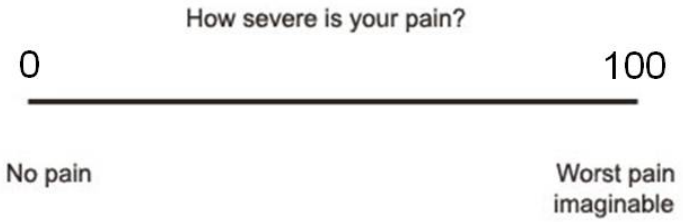
Any congenital anomaly/birth defect in a child born to a female participant or female partner of a male patient exposed to study treatment should be classified as a serious adverse event, recorded on the Adverse Event eCRF, and reported to the Sponsor immediately (i.e., no more than 24 hours after learning of the event; see Section 5 of Appendix 2).

Appendix 6 Pain and Skin Reactivity Assessments

Pain Assessment:

Pain will be assessed by the participant according to a 100-mm horizontal VAS:

- 0 = no pain,
- 100 = worst pain imaginable.



Skin Reactivity Assessment:

ISRs (i.e., burning, bleeding, itching, bruising, redness, hive formation, or other) will be assessed at the time-points indicated in the SoA. Assessment will be made according to a 0−3 scale:

- 0 = no skin reactivity
- 1 = mild skin reactivity (participant aware of the sign, but finds it easily tolerable)
- 2 = moderate skin reactivity (the participant has discomfort enough to cause interference with usual activities)
- 3 = severe skin reactivity (the participant is incapacitated and unable to work or participate in many or all usual activities).

The size of the injection site skin reactions will be captured according to a 0−4 scale:

- 0: unable to assess
- 1: ≤ 20 mm in diameter
- 2: ≤ 40 mm in diameter
- 3: ≤ 60 mm in diameter
- 4: > 60 mm in diameter

Appendix 7
Mayo Clinical Score (MCS)

Stool frequency*
- 0 = Normal no. of stools for this patient
- 1 = 1–2 stools more than normal
- 2 = 3–4 stools more than normal
- 3 = 5 or more stools more than normal

Rectal bleeding†
- 0 = No blood seen
- 1 = Streaks of blood with stool less than half the time
- 2 = Obvious blood with stool most of the time
- 3 = Blood alone passed

Findings of flexible proctosigmoidoscopy
- 0 = Normal or inactive disease
- 1 = Mild disease (erythema, decreased vascular pattern, mild friability)
- 2 = Moderate disease (marked erythema, absent vascular pattern, friability, erosions)
- 3 = Severe disease (spontaneous bleeding, ulceration)

Physician's global assessment‡
- 0 = Normal
- 1 = Mild disease
- 2 = Moderate disease
- 3 = Severe disease

*Each patient served as his or her own control to establish the degree of abnormality of the stool frequency.

†The daily bleeding score represented the most severe bleeding of the day.

‡The physician's global assessment acknowledged the three other criteria, the patient's daily record of abdominal discomfort and general sense of well-being, and other observations, such as physical findings and the patient's performance status.

Note: In accordance with the WP40161 protocol, the endoscopy subscore of the Mayo Clinic Score (MCS-ES) will be modified so that a value of 1 does not include friability.

Reference:

Schroeder et al. 1987

Appendix 8
Rationales for Previous Protocol Amendments

Protocol WP40161 Version 4

Protocol WP40161 Versions 2 and 3, and the local German Version 3 DEU-1, were updated in response to health authority feedback from the Paul-Ehrlich-Institute (PEI; the German Health Authority. Appendix 8 lists the rationales of protocol amendments Versions 2 and 3).

Protocol WP40161 Version 3 has been amended to incorporate the following changes into the global protocol:

- The stopping criterion relating to an increase in absolute regulatory T (T_{reg}) cell count of $>$ 450 cells/μL has been updated in response to the Paul-Ehrlich-Institute (PEI), the German Health Authority, to include a halt to the dosing if reached at any measurement following any dose administration, rather than at dose administration number 4, in 3 or more participants in a single dose level. The current cut-off of >450 cells/µL is not considered by the Sponsor to be a safety limit and is supported by recent data from the entry-into-human study WP39826 and data from a cynomolgus monkey 13-week GLP toxicity study, and to the Sponsor's knowledge there is no published threshold beyond which T_{reg} cell numbers in humans are considered a risk to human health. Therefore, the current cut-off will be maintained throughout the study treatment period.
- In order to resume dosing (or other dose adjustment) if a stopping criterion for the study is met, the requirement of health authority approval prior to resumption of dosing has been added for improved clarity as requested by the PEI.
- Further changes in response to the German ethics committee have been implemented, i.e., inclusion criterion 1 have been adjusted (minor change for clarity, that a written informed consent is available at the time of inclusion of the patient). The order of the statistical sections has been aligned with the order of objectives and endpoints and the efficacy assessments at Day 57 will be summarized descriptively.
- For the WP40161 study protocol, the dose unit has been adjusted from microgram (µg) to milligram (mg) in line with the RO7049665 dose vial (in mg/mL). Furthermore, a consistent use of dose level and cohort has been introduced throughout the protocol.
- The screening window has been increased by one week (i.e., to Day −35 prior to first dose instead of Day −28) in order to maximize the window of the patient's screening assessments. The study allows re-screening if specific criteria are fulfilled, among other for re-testing patients found to be positive to *Clostridium difficile* (where the time for treatment and re-testing may go beyond 28 days) and for the planning of the endoscopic screening assessment in relation to the ova and parasite stool examination that has a long turnaround time.

- The number of patients per cohort has been adjusted to approximately 10 participants.
- The collection timing of the fecal calprotectin stool sample has been changed to increase the flexibility of obtaining the sample from the patient and modified relative to the preparation requirements prior to endoscopic assessments on Days 29 and 57.
- Colon biopsies may be subjected to metagenomic DNA or RNA analysis to determine the composition of the mucosa-associated microbiome in patients. These data are of potential value for patient stratification purposes or for treatment monitoring.
- The skinfold thickness measurement is not required until the time of study treatment administration. In case a patient would be excluded after screening the measurement would not be required. Therefore, the time of measurement has been set to Day −1.
- Participants positive for anti-drug antibodies (ADA) will be requested to return and have additional ADA and soluble CD25 (sCD25) samples to monitor off-treatment ADA persistence. It has been clarified too that a corresponding pharmacokinetic (PK) sample does not have to be collected after the official study period and last planned safety follow-up visit (Day 99) in participants positive for ADA. As the ADA assay is sensitive for sCD25, time points have been moved and one new added to measure sCD25 concentrations toward the end of the study.
- The diagnostic term of "Injection-Related Reaction" (IRR) has been updated to the more general term of "Injection Reaction" (IR) with the options of either “Injection–Site Reaction” (ISR) to signify a local reaction or IRR to signify a systemic reaction as required for study WP40161. The reporting requirements for the eCRF have been updated accordingly.

Other:

Additional minor changes have been made to improve clarity and consistency between synopsis and protocol body, i.e.:

- The study figure and footnote have been aligned for consistency with the protocol.
- For the hematology laboratory assessments “other cells” have been removed from the differential count as this definition does not apply, “sediment” has been removed from the microscopic urine examination as the main components are listed, i.e., casts, crystals, epithelial cells, bacteria.
- Study discontinuation criteria have been introduced for clarity and to align with company requirements.
- The adverse event (AE) reporting time during the study treatment phase has been adjusted and aligned with given safety and follow-up visit (Day 99) and clarified in case of early termination.

- Insufficient response to standard of care with previous treatment with Janus kinase (JAK) inhibitors has been introduced to complete the list of inclusion criterion #6. 5- aminosalicylate (5-ASA) has been introduced to inclusion criterion #6 for clarity and consistency.
- Positivity for total hepatitis B core antibody (HBcAb) has been introduced to exclusion criterion 29 for clarity and to align with the clinical laboratory assessment (Appendix 4).
- The schedule of activities (SoA) tables and footnotes have been updated to ensure consistency with changes in the protocol body.
- Reporting of any medication used by the patient prior to screening has been aligned between the permitted and prohibited therapy sections (Sections 6.5.1. and Section 6.5.2.).
- For clarity the Mayo Clinical Score (MCS) table is shown (Appendix 7).
- The reporting requirements for special situations (i.e., accidental overdose and medication error) and dose-limiting adverse events (DLAEs) have been updated in line with global company requirements.
- Minor changes have been introduced to align with new updates to the Sponsor's protocol template.

Protocol WP40161 Version 3

Protocol WP40161 was previously amended (Version 2) in response to health authority feedback (United States Food and Drug Administration [US FDA]) pre-investigational new drug application [IND] and Paul-Ehrlich-Institut [PEI]; the German Health Authority). Protocol WP40161 V3 will be submitted globally for the purpose of study conduct.

Protocol WP40161 Version 2 has been amended to incorporate the following changes based on experience in the single ascending dose (SAD) entry-into-human (EIH) study of RO7049665, study WP39826:

- The starting dose and dose escalation structure have been included. The overall cohort structure has been simplified to include two doses (i.e., the same dose level may be administered in more than one cohort) with well understood safety, pharmacokinetics (PK), and pharmacodynamics (PD) in healthy volunteers. The WP39826 study has been summarized (including safety, PK, and PD). The number of expected participants has been modified and updated to reflect changes to the study design.
- In response to FDA feedback, the cohort that included RO7049665 as an adjunct to anti-tumor necrosis factor (TNF)-α treatment has been removed.
- Based on the identification of anti-drug antibodies (ADA) in study WP39826, the risk-benefit section has been updated, additional safety monitoring has been introduced (including management of potential hypersensitivity reactions), a

staggered enrollment plan has been implemented, and all doses will be administered with a 24 hour in-house stay for safety monitoring.

- The dose regimen of every 4-week (Q4W) has been removed based on safety, PK, and PD data from WP39826. The overall length of the study has been modified to reflect the timing of the every 2-week (Q2W) regimen, which will be maintained throughout the study.
- Based on the PK and PD findings from WP39826, the Schedule of Activities' (SoA) tables have been revised to optimize timing of PK and PD measurements. The SoAs have been updated to include injection-related reaction (IRR) sampling as needed, an early termination visit if required, *Clostridium difficile* enzyme-linked immunosorbent assay (ELISA) at screening, ova and parasite exam included at screening. Hepatitis C screening has been changed to from antibody screen to a polymerase chain reaction (PCR)-based assay. Follicle-stimulating hormone (FSH) sampling has been removed. Footnotes have been updated as applicable.
- The timing of key efficacy assessments (Ulcerative Colitis Endoscopic Index of Severity [UCEIS] and Mayo Clinic Score [MCS]) has been shifted from week 10 to week 8, and have been clarified as an assessment day rather than week.
- The blinding definition has been clarified.
- Most tertiary/exploratory objectives have been removed. These objectives included exploratory cytokines that are not fully identified. Objectives will now focus on safety, tolerability, PK, PD, and early efficacy in the primary and secondary endpoints.
- In response to FDA feedback, the definition of the patient population with moderately to severely active ulcerative colitis (UC) has been updated and clarified to include overall MCS, stool frequency subscore, and rectal bleeding subscore.
- A communication strategy has been introduced to detail management of staggered dosing, dose escalation, and safety risks.
- The requirement for genotype testing has been changed to clarify this is mandatory only in countries where this testing is permitted.
- The sections on overdose and medication error (i.e., accidental deviation in the administration of a drug) have been updated in line with new Sponsor template language relating to overdose, medication error, drug abuse, and drug misuse.
- General changes and updates to the protocol have been made to improve consistency and clarity and aligned with updated Sponsor and protocol template requirements.
- In Appendix 5 the contraceptive requirements and recommendations have been clarified.

Protocol WP40161 Version 2

Protocol WP40161 version 1 has been amended to incorporate the following changes in response to questions raised by Paul-Ehrlich-Institut (PEI; German Health Authority):

- The stopping criteria have been updated in order to allow administration of RO7049665 while maintaining participant's regulatory T (T_{reg}) cell levels within an upper limit of 450 cells/μl. In order to allow complete evaluation of all participants in a cohort and at a given dose (with possible variations in the participant's T_{reg} numbers after RO7049665 administration), the new stopping criterion will apply after four administrations given every second week (or after three administrations if given every 4 weeks) of RO7049665. The dose escalation criteria have also been updated to limit dosing beyond a level resulting in increases in off-target effects on eosinophils, above $5.0x10^9$ cells/L in 3 or more patients in a dose cohort.

- The Sponsor will implement an internal monitoring committee (IMC) for reviewing, specifically safety-related data on an ongoing basis for dose-escalation decision making.

- The individual participant stopping criterion with regards to other findings as a reason to stop dosing with RO7049665, has been modified to allow Investigator discretionary reasons only (instead of at the discretion of Investigator and Sponsor).

- The dose escalation criteria have been modified allowing dose-escalation between all non-biologic standard of care (SOC) cohorts based on the same criteria as for cohorts 1 and 2, i.e., safety and tolerability in at least 5 participants during the first 8 weeks.

- Inclusion criterion number 6 has been modified to clearly define insufficient treatment response and treatment intolerance to pharmacological treatment and failure based on defined previous doses and duration of treatment in accordance with CHMP/EWP/18463//2006 Rev.1 Guidelines on the development of new medicinal products 4 for the treatment of Ulcerative Colitis and the ECCO guidelines on ulcerative colitis.

- Inclusion criterion 9 will remain as is, i.e., with use of contraceptive methods resulting in a failure rate of <1% per year during the treatment period and for at least 28 days, as the Sponsor considers that this sufficiently covers the time prediction of 5 elimination half-lives.

Other:

- Additional minor changes have been made to improve clarity and consistency between synopsis and protocol body (i.e., the numbering of inclusion and exclusion criteria in the synopsis versus protocol body have been adjusted).