Protocol

Study ID: RVLO 121-04

Study Title: A Phase 2a, Double-blind, Placebo-Controlled, Multi-Center Study to Assess the Efficacy, Safety, and Tolerability of IRL201104 in Adult Participants with Active Eosinophilic Esophagitis (EoE).

NCT: NCT05084963

Document Date: 01 July 2021

A PHASE 2A, DOUBLE-BLIND, PLACEBO-CONTROLLED, MULTI-CENTER STUDY TO ASSESS THE EFFICACY, SAFETY, AND TOLERABILITY OF IRL201104 IN ADULT PARTICIPANTS WITH ACTIVE EOSINOPHILIC ESOPHAGITIS (EOE)

Protocol Number	RVLO 121-04
Compound Number	IRL201104
PIND Number	[redacted]
Sponsor:	Revolo Biotherapeutics 900 Camp Street New Orleans, LA 70130
Sponsor's Responsible Medical Officer:	[redacted] Chief Medical Officer Revolo Biotherapeutics
Protocol Version:	2.0
Date of Protocol:	01 July 2021

CONFIDENTIALITY STATEMENT

SIGNATURE PAGE

Sponsor's Approval

The protocol has been approved by Revolo Biotherapeutics.

Sponsor's Authorized Officer:

DocuSigned by:
[redacted]

07-Jul-2021 | 20:59 BST

Company/Sponsor signatory

Date

[redacted]
Chief Medical Officer
Revolo Biotherapeutics

INVESTIGATOR'S AGREEMENT

I have received and read the Investigator's Brochure for IRL201104. I have read the RVLO 121-04 protocol and agree to conduct the study as outlined.

I agree to comply with International Council for Harmonisation (ICH) Tripartite Guideline on Good Clinical Practice (GCP) and applicable Food and Drug Administration (FDA) regulations/ guidelines set forth in Code of Federal Regulations Title 21 (21 CFR) Parts 11, 50, 54, 56 and 312 and all locally applicable laws.

I agree to maintain the confidentiality of all information received or developed in connection with this protocol.

_____

Printed Name of Investigator

_____

Signature of Investigator

_____

Date

2. SYNOPSIS

<table>
<tr><td colspan="3">Name of Sponsor/Company: Revolo Biotherapeutics</td></tr>
<tr><td colspan="3">Name of Investigational Product: IRL201104</td></tr>
<tr><td colspan="3">Name of Active Ingredient: IRL201104</td></tr>
<tr><td>Protocol Number:
RVLO 121-04</td><td>Phase: 2a</td><td>Country: United States</td></tr>
<tr><td colspan="3">Title of Study: A Phase 2a, Double-blind, Placebo-Controlled, Multi-Center Study to Assess the Efficacy, Safety, and Tolerability of IRL201104 in Adult Participants with Active Eosinophilic Esophagitis (EoE)</td></tr>
<tr><td colspan="3">Study centers: Approximately 15 centers in the United States</td></tr>
<tr><td colspan="2">Studied period (years):
Estimated date first participant enrolled: Sep 2021
Estimated date last participant completed: Jun 2022</td><td>Phase of development: 2a</td></tr>
<tr><td colspan="3">Study Objectives
Primary Objective:
• To assess the efficacy of repeat intravenous (IV) doses of 4 mg and 8 mg IRL201104 compared to placebo in adult participants with EoE after 3 doses administered over 14 days as assessed by the change in peak esophageal intraepithelial eosinophilic count per high power field (eos/hpf) at Week 4.
Secondary Objectives:
• To explore the relationship between IRL201104 treatment and change in symptoms of dysphagia in adult participants with EoE, using the patient-reported outcomes (PRO) instrument Dysphagia Symptom Questionnaire (DSQ).
• To assess the efficacy of repeat IV doses of IRL201104 in adult participants with EoE after 3 doses administered over 14 days as assessed by thresholds of histologic response at Week 4.
• To evaluate the safety, tolerability, and immunogenicity of IRL201104 in adult participants with EoE.
Exploratory Objectives:
• To explore the relationship between IRL201104 and change in clinical outcomes assessments, to include the PROs Patient Global Impression of Severity (PGI-S) and Patient Global Impression of Change (PGI-C), and the clinician-reported outcomes EoE-Endoscopic Reference Score (EREFS) and EoE Histology Scoring System (EoEHSS].
• To explore the relationship between IRL201104 and the relative change in RNA gene expression to include EoE diagnostic panel (EDP) transcriptome signature.
• To explore the relationship between IRL201104 treatment on immune cells and markers of inflammation.</td></tr>
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Methodology:

This is a Phase 2a, double-blind, placebo-controlled, multi-center study to assess the efficacy and safety of a repeat-dose regimen of IRL201104 in participants with active EoE. A study diagram is provided in Figure 1.

Eligible participants will undergo an up to 6-week screening period to stabilize current therapy, confirm diagnosis by histologic assessment, and other eligibility criteria. Participant compliance to DSQ reporting will be assessed for at least 2 consecutive weeks prior to randomization.

At the end of the screening period, eligible participants will be randomized in a 1:1:1 ratio to one of 2 active arms or placebo, as displayed in Figure 1:

- Arm 1: IRL201104 4 mg IV with endoscopy and esophageal biopsy 2 weeks after last dose (Week 4).
- Arm 2: IRL201104 8 mg IV with endoscopy and esophageal biopsy 2 weeks after last dose (Week 4).
- Arm 3: Placebo IV with endoscopy and esophageal biopsy 2 weeks after last dose (Week 4)

Participants will receive IRL201104 (4 mg or 8 mg IV) or placebo on Days 0, 7, and 14 (3 total doses over 14 days). Participants will be evaluated for clinical and histologic responses on Week 4 . All participants will be assessed for safety and continued biomarker sampling through Week 8.

The Schedule of Events for the study is provided in Table 3.

Sample Size Justification:

Planned N = 36

Participants will be randomized 1:1:1to 4 mg or 8 mg IRL201104 or placebo. The sample size is based on clinical considerations of a reasonable number of participants for a preliminary investigation of this type and the opportunity to derive early estimates of histological response to treatment.

The proposed evaluation of IRL201104 will include, as the primary endpoint, the mean change (reduction) in eosinophil count from baseline for each treatment group and, as a secondary endpoint, the number and percentage of participants in each treatment group with a histologic eosinophil count below defined thresholds at Week 4. Other secondary and explorative objectives will be evaluated through the generation of summary statistics appropriate for categorical (eg, frequency counts) or continuous (eg, means) data. No formal statistical analysis of this result will be undertaken. Secondary endpoints will be similarly summarized by treatment group using measures appropriate for continuous data (eg, mean, standard deviation, median, minimum, maximum) or categorical data (eg, incidence rate of adverse events [AEs]).

Number of participants (planned): Approximately up to 50 participants are planned to be enrolled to yield36 participants who are randomized and complete the final endoscopy at Week 4.

Inclusion Criteria:

A participant must meet the following criteria to be eligible for inclusion in the study:

1. Age 18 to 75 years old, inclusive, at the time of signing the informed consent form.
2. Documented diagnosis of EoE by endoscopy prior to screening.

   Note: Must include a demonstration of intraepithelial eosinophilic infiltration (peak cell count ≥ 15 eos/hpf [400×]) from esophageal biopsy specimens from endoscopy.
3. History (by participant report) of on average at least 2 episodes of dysphagia (with intake of solids off anti-inflammatory therapy) per week in the 4 weeks prior to screening, and on average at least 2 episodes of documented dysphagia per week during any 2 consecutive weeks (qualifying period) between screening and baseline; dysphagia is defined as trouble swallowing

solid food, or having solid food stick, by participant report; and completed the DSQ on ≥ 70% of days during the qualifying period prior to baseline (Visit 1).

4. Must remain on a stabilized diet for at least 6 weeks prior to screening and during the course of the study; stable diet is defined as no initiation of single or multiple elimination diets or reintroduction of previously eliminated food groups.
5. Must be willing and able to continue any dietary therapy and/or medical regimens (including gastric acid suppression) in effect at the screening visit. There should be no change to these regimens during the study participation.
6. Willing and able to comply with all clinic visits and study-related procedures.
7. Able to understand and complete study-related questionnaires.
8. Provide signed informed consent.
9. Esophagogastroduodenoscopy (EGD) with photographs performed at screening (qualifying EGD), with a demonstration of intraepithelial eosinophilic infiltration (peak cell count ≥15 eos/hpf) in at least 2 of the 3 biopsied esophageal regions (proximal, mid, or distal).

Exclusion Criteria:

A participant who meets any of the following criteria will be ineligible to participate in this study:

1. Prior participation in an IRL201104 clinical study.
2. Has any current disease of the gastrointestinal tract (aside from EoE) that may impact, in the investigator's opinion, the patient's EoE disease status. This includes, but not limited to: eosinophilic gastritis, eosinophilic enteritis, eosinophilic duodenitis, eosinophilic colitis, or proctitis; inflammatory bowel disease; or celiac disease.
3. Has other causes of esophageal eosinophilia or the following diseases: hypereosinophilic syndromes, Churg-Strauss vasculitis (eosinophilic granulomatosis with polyangiitis), or peripheral blood absolute eosinophil count of > 1500 eosinophils/μL.
4. Has presence of oral or esophageal mucosal infection of any type.
5. Has any condition affecting the esophageal mucosa or altering esophageal motility other than EoE.
6. History of achalasia, active *Helicobacter pylori* infection, Crohn's disease, ulcerative colitis, celiac disease, and prior esophageal surgery (with the exception of a surgical repair of an EoE complication).
7. Any esophageal stricture unable to be passed with a standard, diagnostic, adult (9 to 10 mm) upper endoscope or any critical esophageal stricture that requires dilation at screening; or dilation within 2 months prior to screening.
8. On a pure liquid diet or any mouth or dental condition that prevents normal eating.
9. Has initiated, discontinued, or changed dosage regimen of PPIs within the 4 weeks prior to the qualifying EGD, between the qualifying EGD and baseline visit (Visit 1), or anticipates changes in the use of PPI during the study. PPI must remain constant throughout the study.
10. History of bleeding disorders or esophageal varices.
11. Use of anticoagulants within 2 weeks prior to screening. Participants should not stop these agents solely to become eligible for entry into this study.
12. Treatment with an investigational drug within 2 months or within 5 half-lives (if known), whichever is longer, prior to screening.
13. Use of systemic corticosteroids within 3 months or swallowed topical corticosteroids within 6 weeks prior to screening.
14. Treatment with oral immunotherapy (OIT) within 6 months prior to screening.

15. Allergen immunotherapy (sublingual immunotherapy [SLIT] and/or subcutaneous immunotherapy [SCIT]), unless on a stable dose for at least 1 year prior to screening.
16. The following treatments within 3 months before the screening visit, or any condition that, in the opinion of the investigator, is likely to require such treatment(s) during the study:

    Systemic immunosuppressive/immunomodulating drugs (eg, omalizumab, cyclosporine, mycophenolate-mofetil, interferon [IFN]γ, Janus kinase inhibitors, azathioprine, methotrexate, and other biologics that are ongoing [eg, dupilumab, benralizumab, mepolizumab, or vedolizumab]).
17. Diagnosed with active parasitic infection; or suspected parasitic infection, unless clinical and (if necessary) laboratory assessments have ruled out active infection before randomization.
18. Chronic or acute infection requiring treatment with systemic antibiotics, antivirals, or antifungals within 1 month prior to screening.
19. Use of oral antibiotics/anti-infectives within 2 weeks prior to screening.
20. Known or suspected immunosuppression, including history of invasive opportunistic infections (eg, tuberculosis, non-tuberculous mycobacterial infections, histoplasmosis, listeriosis, coccidioidomycosis, pneumocystosis, aspergillosis) despite infection resolution, or otherwise recurrent infections of abnormal frequency, or prolonged infections suggesting an immunocompromised status, as judged by the investigator.
21. Known history of human immunodeficiency virus (HIV) infection.
22. Positive or indeterminate hepatitis B surface antigen (HBsAg) or hepatitis C antibody at screening.
23. Moderate or severe renal impairment (eGFR <60 mL/min/1.73 m^2) or end stage renal disease.
24. Elevated transaminases (alanine aminotransferase [ALT] and/or aspartate aminotransferase [AST]) > 3 times the upper limit of normal (ULN) at screening.
25. History of malignancy within 5 years prior to screening, except completely treated in situ carcinoma of the cervix and completely treated and resolved nonmetastatic squamous or basal cell carcinoma of the skin.
26. Any other medical or psychological condition including relevant laboratory abnormalities at screening that, in the opinion of the investigator, suggest a new and/or insufficiently understood disease, may present an unreasonable risk to the participant as a result of his/her participation in this clinical study, may make the participant's participation unreliable, or may interfere with study assessments. The specific justification for participants excluded under this criterion will be noted in study documents (eg, chart notes, electronic case report form). These may include participant-reported alcohol or drug abuse and severe concomitant illness(es).
27. Planned or anticipated use of any prohibited medications and procedures (as described in the exclusion criteria) during study treatment.
28. Treatment with a live (attenuated) vaccine within 3 months prior to screening and/or treatment of a killed vaccine within 30 days prior to screening, until the end of the study with the exception of a coronavirus disease of 2019 (COVID-19) vaccine, as described in Section 9.2.1.
29. Pregnant or breastfeeding women, or women planning to become pregnant or breastfeed during the study.
30. Women unwilling to use adequate birth control, if of reproductive potential* and sexually active. Adequate birth control is defined as agreement to consistently practice an effective and accepted method of contraception throughout the duration of the study and for 30 days after the last dose of study treatment. These include: hormonal contraceptives, intrauterine device, or double barrier contraception (ie, condom and diaphragm), or male partner with documented vasectomy.

*For females, menopause is defined as at least 12 consecutive months without menses; to include laboratory confirmation of post-menopausal status (ie, a follicle stimulating hormone (FSH) of 2.25 U/mL must be documented). Hysterectomy, bilateral oophorectomy, or bilateral tubal ligation must be documented, as applicable; if documented, women with these conditions are not required to use additional contraception.
Investigational product, dosage, and mode of administration: IRL201104 and placebo is supplied as a sterile, lyophilized cake in a standard glass vial and is reconstituted with sterile water for injection. For both the 4 and 8 mg dose levels, the amount of drug substance in each 20 mL vial is 0.8 mg/ml, which is reconstituted with 10-20 mL water for injection (WFI). Investigational product will be administered in a volume of 10 mL. IRL201104 4 mg and 8 mg and placebo are administered as an IV bolus injection over 30 seconds (not to exceed 60 seconds) on Days 0, 7, and 14 (3 total doses over 14 days). Pharmacy staff will be unblinded for investigational product preparation. Investigators, study team members and study participants will be blinded to the dose level given (4 mg, 8 mg, or placebo) and will remain blinded throughout the study.
Duration of treatment: Screening period: up to 6 weeks Double-blind treatment and follow-up period: 8 weeks **Total study duration:** up to 14 weeks
Reference therapy, dosage, and mode of administration: None
Criteria for Evaluation **Primary Endpoints:** • Change from baseline in the peak esophageal intraepithelial eosinophil count at Week 4. **Secondary Endpoints:** • Absolute change in DSQ score from baseline. The DSQ is used to measure the frequency and intensity of dysphagia. DSQ scores can range from 0 to 84, with a lower score indicating less frequent or less severe dysphagia. • Proportion of participants achieving peak esophageal intraepithelial eosinophil count of < 15 eos/hpf (Week 4). • Percent reduction in peak esophageal intraepithelial eosinophil count (eos/hpf) (Week 4). **Exploratory Endpoints:** • Proportion of participants achieving peak esophageal intraepithelial eosinophil count of ≤ 6 eos/hpf (400×) at Week 4. • Proportion of participants achieving peak esophageal intraepithelial eosinophil count of ≤ 1 eos/hpf (400x) at Week 4. • Relative change in the EDP transcriptome signature (Week 4)

- Relative change in immune cells and markers of inflammation pre- and postdose administration at screening, baseline, and Weeks 1, 2, 4, 6, and 8. Immune cells and markers of inflammation include: immune cell phenotyping (T-subsets, B-subsets, and macrophage subsets); A20; serum cytokines; serum IgE/IgG4; and serum eosinophil derived neurotoxin (EDN) for eosinophil activation.
- Absolute change in EoE-EREFS (Week 4).

  EoE esophageal characteristics will be analyzed based on the EoE-EREFS, a scoring system for inflammatory and remodeling features of disease using both overall scores and scores for each individual characteristic. Proximal and distal esophageal regions will be scored separately; the score for each region ranges from 0 to 9 and the overall score ranges from 0 to 18.
- Absolute change in EoE grade score from the EoE Histology Scoring System (EoEHSS) (Week 4).

  EoE grade and stage scores evaluate 8 features: eosinophil density, basal zone hyperplasia, eosinophil abscesses, eosinophil surface layering, dilated intercellular spaces, surface epithelial alteration, dyskeratotic epithelial cells, and lamina propria fibrosis (absent/present). Severity (grade) and extent (stage) of abnormalities will be scored using a 4-point scale (0 normal; 3 maximum change).
- Absolute change in EoE stage score from the EoEHSS (Week 4).
- Absolute change in severity and/or frequency of EoE symptoms other than dysphagia (up to Week 8).
- Proportion of participants with use of rescue medications or procedures (up to Week 8).
- Concentration of functional IRL201104 (up to Week 8).
- Incidence of treatment-emergent antidrug antibody (ADA) responses (Week 8).
- Absolute days without dysphagia
- Change in PGI-S and PGI-C

Safety Endpoints:

Safety parameters will include monitoring of adverse events (AEs), physical examinations, vital signs, electrocardiograms (ECGs), clinical laboratory tests, and clinical evaluations. Participants will be asked to monitor all AEs experienced from the time of informed consent until their last study visit.

Statistical methods:

No formal statistical analysis is anticipated for this study. Change in histologic eosinophil count from baseline will be highlighted with summary statistics for continuous data (mean, standard deviation, median, minimum, maximum). The number and proportion of participants meeting the threshold eosinophil reduction criteria will also be summarized by treatment group. Changes in PRO measures from baseline and the incidence of AEs, vital signs, laboratory findings, and other safety evaluations will be similarly summarized using categorical or continuous data-derived summary statistics appropriate for the outcome under consideration.

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4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and specialist terms are used in this study protocol.

Table 1: Abbreviations and Specialist Terms

Abbreviation or Specialist Term	Explanation
ADA	antidrug antibody
AE	adverse event
AESI	adverse event of special interest
ALT	alanine aminotransferase
AST	aspartate aminotransferase
AUC_{last}	area under the plasma concentration-time curve from time = 0 to the last measurable concentration
CD	cluster of differentiation
C_{max}	maximum observed concentration
COVID-19	coronavirus disease of 2019
CPK	creatine phosphokinase
DSQ	Dysphagia Symptom Questionnaire
ECG	electrocardiogram
eCRF	electronic case report form
EDN	eosinophil derived neurotoxin
EDP	eosinophilic esophagitis diagnostic panel
EGD	esophagogastroduodenoscopy
EoE	eosinophilic esophagitis
EoEHSS	Eosinophilic Esophagitis-Histology Scoring System
eos/hpf	eosinophilic count per high-power field
EPIT	epicutaneous immunotherapy
EREFS	Eosinophilic Esophagitis-Endoscopic Reference Score
FAS	full analysis set
FDA	Food and Drug Administration
FFAR3	free fatty acid receptor 3
FIH	first-in-human
FOXP3	forkhead box P3
FSH	follicle stimulating hormone
GCP	Good Clinical Practice

Abbreviation or Specialist Term	Explanation
GERD	gastroesophageal reflux disease
HBsAg	hepatitis B surface antigen
HDM	house dust mite
HIV	human immunodeficiency virus
ICH	International Council for Harmonisation
IDMC	Independent Data Monitoring Committee
IEC	Independent Ethics Committee
IFN	interferon
Ig	immunoglobulin
IL	interleukin
IN	intranasal(ly)
IND	Investigational New Drug Application
IRB	Institutional Review Board
IV	intravenous
IWRS	interactive web response system
LDS	Loeys-Dietz syndrome
MAD	multiple ascending dose
mTB	mycobacterium tuberculosis
OIT	oral immunotherapy
PGI-C	Patient Global Impression of Change
PGI-S	Patient Global Impression of Severity
PK	pharmacokinetic(s)
PPI	proton pump inhibitors
PRO	patient-reported outcomes
QD	once daily
RNA	ribonucleic acid
SAE	serious adverse event
SAP	statistical analysis plan
SCIT	subcutaneous immunotherapy
SLIT	sublingual immunotherapy
$t_{1/2}$	terminal elimination half-life
TGFβ	transforming growth factor–β

Abbreviation or Specialist Term	Explanation
Th	T helper
Treg	regulatory T
UK	United Kingdom
ULN	upper limit of normal
US	United States
WFI	water for injection
WHO	World Health Organization

5. INTRODUCTION

5.1. Eosinophilic Esophagitis

Eosinophilic esophagitis (EoE) is a chronic and progressive disease of the immune system resulting in damage to the esophagus with subsequent loss of function resulting in difficulty swallowing and food impaction. If not adequately treated, symptoms and inflammation can progress, leading to scarring of the esophagus with irreversible damage. In the United States (US), there are approximately 160,000 patients with EoE who are currently treated, of which approximately 50,000 have failed multiple treatments. There are currently no therapies approved by the US Food and Drug Administration (FDA) for EoE.

Eosinophilic esophagitis is caused by the presence of large numbers of eosinophils in the esophagus. The production and accumulation of eosinophils may be caused by many factors, such as immune hypersensitivity responses to particular foods or environmental proteins (allergens) in affected individuals. Eosinophilic esophagitis is characterized by esophageal barrier defects and eosinophilic infiltrates. Therapeutic interventions that target reduction in esophageal eosinophils have been shown to correlate with symptom improvement (Hirano 2020).

Regulatory T (Treg) cells are a subset of T lymphocytes that play an important role in the prevention and control of many autoimmune and allergic diseases. Frischmeyer-Guerrerio, et al noted that transforming growth factor–β (TGFβ) regulates Treg maturation and demonstrated that patients with Loeys-Dietz syndrome (LDS), an autosomal dominant disorder caused by mutations in the genes encoding receptor subunits for TGFβ, TGFBR1 and TGFBR2, are strongly predisposed to develop allergic disease, including asthma, food allergy, eczema, allergic rhinitis, and eosinophilic gastrointestinal disease (Frischmeyer-Guerrerio 2013). Patients with LDS exhibited elevated immunoglobulin (Ig) E levels, eosinophil counts, and T helper (Th) 2 cytokines in their plasma. They had an increased frequency of cluster of differentiation (CD)4+ T-cells that expressed both forkhead box P3 (FOXP3) and interleukin (IL)-13, but retained the ability to suppress effector T-cell proliferation. T helper 2 cytokine–producing cells accumulated in cultures of naïve CD4+ T-cells from LDS subjects, but not controls, after stimulation with TGFβ, suggesting that LDS mutations support Th2 skewing in naïve lymphocytes in a cell-autonomous manner. The monogenic nature of LDS demonstrates that altered TGFβ signaling can predispose to allergic phenotypes in humans and underscores a prominent role for TGFβ in directing immune responses to antigens present in the environment and foods and may be relevant to nonsyndromic presentations of allergic diseases such as EoE.

Treg cells are reduced in the biopsies of adults with EoE as compared with control participants (Stuck 2011). In contrast, Treg cells seem to be increased in esophageal tissue of children with EoE compared with individuals with gastroesophageal reflux disease and healthy participants (Fuentebella 2010). However, Treg cell immunosuppressive function may be minimized in patients with EoE, similar to patients with allergic asthma. In animal models of EoE, epicutaneous immunotherapy (EPIT) induced Treg cells in the spleen and expression of forkhead box P3 (FOXP3) in the esophagus that correlated with reduced eosinophilic infiltration. Depletion of CD25+ cells abrogated Treg cell induction and resulted in increased esophageal inflammation. The transfer of Treg cells isolated from mice who had undergone EPIT prevented peanut-induced eosinophil infiltration and eotaxin expression in the esophagus of mice (Dioszeghy 2014). Interestingly, patients with the autosomal recessive form of hyper-IgE

syndrome caused by mutations in the dedicator of cytokinesis 8 gene have defective Treg cells and often develop EoE (Hsu 1993, Janssen 2014).

Finally, Wen, et al employed single-cell ribonucleic acid (RNA) sequencing to examine the heterogeneity of human tissue CD3+ T-cells during allergic inflammation, focusing on EoE. They investigated 1088 single T-cells derived from patients with a spectrum of disease activity with Treg (FOXP3+) enriched in the tissue. Prodigious levels of IL-5 and IL-13 were confined to HPGDS+ CRTH2+IL-17RB+FFAR3+CD4+ T8 effector Th2 cells. EoE severity closely paralleled a lipid/fatty acid–induced activation node highlighted by the expression of the short-chain free fatty acid receptor 3 (FFAR3). Ligands for FFAR3 induced Th2 cytokine production from human and murine T-cells, including in an in vivo allergy model. In summary, they elucidated the defining characteristics of tissue-residing CD3+ T-cells in EoE, a specific enrichment of CD4+ Treg and effector Th2 cells, and confinement of type 2 cytokine production to the CD4+ effector population (Wen 2019). Together, these findings show that Treg cells are dysregulated in patients with EoE and that they can protect against an EoE-like disease in some cases.

While the exact mechanism of the lack of suppressive Treg cell function in EoE isn't known, Treg cells appear to be important in the pathophysiology of EoE and their modulation may represent an opportunity for enduring therapy for patients with EoE.

5.2. Rationale for Treatment with IRL201104

Almost one third of the worlds' population is infected with mycobacterium tuberculosis (mTB) but less than 10% of those infected go on to develop the disease (World Health Organization [WHO] 2012), resulting in an estimated 2 million deaths each year (Miranda 2012). In the 2 billion people with 'latent' disease the bacteria survive in a dormant state entrapped in the lungs within a protective cellular structure known as the granuloma. The process of granuloma formation is initiated by the mTB bacteria being taken up by alveolar macrophages, which then release mediators to recruit inflammatory cells including more macrophages, dendritic cells and T-cells, which organize into the spherical structure of the granuloma. The bacteria can survive for decades inside the granuloma in a latent state. To further enhance its chances of survival the bacteria secrete proteins, known as chaperonins, which modulate the hosts' inflammatory response (Cehovin 2010) and enable the bacteria to remain undetected in the human host. The sponsor has explored the therapeutic potential of these proteins to produce drug-like peptides. One such peptide, IRL201104, formerly known as PIN201104, has been identified via phenotypic drug discovery to be a potential anti-inflammatory/immunomodulatory agent and is being developed as a potential treatment for a range of autoimmune and allergic diseases.

IRL201104 is a novel synthetic peptide derived from the active domain of the mTB-secreted protein chaperonin 60.1. It is being developed for the relief of symptoms in patients with active EoE. The peptide significantly decreases airway eosinophil infiltration in rodent models of allergic and nonallergic inflammation. IRL201104 appears to be effective in resetting the immune system from an inflammatory state and in inducing inflammatory disease remission in nonclinical models. Its short pharmacokinetic (PK) terminal elimination half-life ($t_{1/2}$) decreases the potential for the negative off-target effects associated with current therapies.

Importantly, in an assay using blood cells from healthy human participants, incubation with IRL201104 increased Treg cell concentrations in a dose-dependent manner, compared with the

control-treated cells. Treg cell levels were increased by more than 2-fold at the top concentration of IRL201104 (data on file).

Numerous nonclinical pharmacology studies performed suggest that IRL201104 has an anti-inflammatory and immunomodulatory mechanism of action. IRL201104 is effective in a range of models of inflammation and has also demonstrated efficacy in animal models of asthma, with prolonged duration of action.

5.3. Clinical Development Program

In non-clinical safety pharmacology and toxicology studies, IRL201104 was safe and well tolerated at exposures up to 11 times higher than the highest dose level planned for the current Phase 2a clinical study. In 14-day rat and dog Good Laboratory Practice toxicology studies, the no-observed-effect-level was the highest dose tested (0.5 mg/kg intravenous [IV] bolus).

IRL201104 was assessed in a first-in-human (FIH) Phase 1 study in 62 healthy participants and participants with mild asthma in the United Kingdom (UK) (Study C1104-001). The highest dose tested was a single 8 mg dose (IV bolus) in 4 healthy participants and 6 participants with mild asthma. Most participants in the study received a single dose by IV bolus; however, 1 group (4 participants) received 3 single IV bolus injections of 2 mg (2 hours apart) on a single day. In this study, IRL201104 was well tolerated and safe, with no significant adverse events (AEs) reported for single doses up to 8 mg IRL201104. After a single 2, 4, and 8 mg IV bolus, IRL201104 was detectable in blood up to 15, 30, and 60 minutes, respectively, with exposure increasing in a dose-dependent manner. The mean $t_{1/2}$ of the 8 mg dose was approximately 16 minutes.

IRL201104 is currently being assessed for safety, tolerability, and PK in an ongoing Phase 1 multiple ascending dose (MAD) study in 18 healthy participants in the UK (Study C1104-003). The IRL201104 IV dose levels were 4 mg once daily (QD) for 5 days and 8 mg QD for 7 days. At each dose level, up to 8 participants received IRL201104 and 2 received placebo. Following a blinded review of safety data, study treatment was well tolerated in the groups receiving either IRL201104 4 mg (6 participants), 8 mg (8 participants) or placebo (4 participants) for 5 days and followed through Day 21.

In combination, these nonclinical data and a good clinical safety/tolerability profile show that IRL201104 is well suited for further development as a treatment for EoE.

This Phase 2a clinical study is a double-blind, placebo-controlled study designed to investigate the efficacy, safety, and tolerability of IRL201104 in adult participants with active EoE. Participants will receive a total of 3 doses of IRL201104 4 mg or 8 mg IV or placebo over a 14-day period. Study treatment will be delivered by IV bolus injection via cannula.

The justification for the dose levels selected in the current study is presented in Section 7.8.

The study will be conducted in compliance with the protocol, Good Clinical Practice (GCP), and all applicable regulatory requirements.

6. STUDY OBJECTIVES AND PURPOSE

6.1. Primary Objective

The primary objective is to assess the efficacy of repeat IV doses of 4 mg and 8 mg IRL201104 or placebo in adult participants with EoE after 3 doses administered over 14 days as assessed by the change in peak esophageal intraepithelial eosinophilic count per high-power field (eos/hpf) at Week 4.

6.2. Secondary Objectives

The secondary objectives are:

- To explore the relationship between IRL201104 treatment and change in symptoms of dysphagia in adult participants with EoE, using the patient-reported outcomes (PRO) instrument Dysphagia Symptom Questionnaire (DSQ).
- To assess the efficacy of repeat IV doses of IRL201104 in adult participants with EoE after 3 doses administered over 14 days as assessed by thresholds of histologic response at Week 4.
- To evaluate the safety, tolerability, and immunogenicity of IRL201104 in adult participants with EoE.

6.3. Exploratory Objectives

The exploratory objectives are:

- To explore the relationship between IRL201104 and change in clinical outcomes assessments, to include the PROs PGI-S and PGI-C, and the clinician-reported outcomes EoE-Endoscopic Reference Score (EREFS) and EoE Histology Scoring System (EoEHSS).
- To explore the relationship between IRL201104 and the relative change in RNA gene expression to include EoE diagnostic panel (EDP) transcriptome signature.
- To explore the relationship between IRL201104 treatment on immune cells and markers of inflammation.

6.4. Endpoints

6.4.1. Primary Endpoint

The primary endpoint is:

- Change from baseline in the peak esophageal intraepithelial eosinophil count at Week 4.

6.4.2. Secondary Endpoints

The secondary endpoints are:

- Absolute change in DSQ score from baseline.

- Proportion of participants achieving peak esophageal intraepithelial eosinophil count of < 15 eos/hpf (Week 4).
- Percent reduction in peak esophageal intraepithelial eosinophil count (eos/hpf) (Week 4).

The safety endpoints are:

- Monitoring of AEs, physical examinations, vital signs, electrocardiograms (ECGs), clinical laboratory tests, and clinical evaluations. Participants will be asked to monitor all AEs experienced from the time of informed consent until their last study visit.

6.4.3. Exploratory Endpoints

Exploratory endpoints include:

- Proportion of participants achieving peak esophageal intraepithelial eosinophil count of ≤ 6 eos/hpf (400×) at Week 4.
- Proportion of participants achieving peak esophageal intraepithelial eosinophil count of ≤ 1 eos/hpf (400x) at Week 4.
- Relative change in the EDP transcriptome signature (Week 4).
- Relative change in immune cells and markers of inflammation pre- and postdose administration at screening, baseline, and Weeks 1, 2, 4, 6, and 8. Immune cells and markers of inflammation include: immune cell phenotyping (T-subsets, B-subsets, and macrophage subsets); A20; serum cytokines; serum IgE/IgG4; and serum eosinophil derived neurotoxin (EDN) for eosinophil activation.
- Absolute change in EoE-EREFS (Week 4).
- Absolute change in EoE grade score from the EoE Histology Scoring System (EoEHSS) (Week 4).
- Absolute change in EoE stage score from the EoEHSS (Week 4).
- Absolute change in severity and/or frequency of EoE symptoms other than dysphagia (up to Week 8).
- Proportion of participants with use of rescue medications or procedures (up to Week 8).
- Concentration of functional IRL201104 (up to Week 8).
- Incidence of treatment-emergent antidrug antibody (ADA) responses (Week 8).
- Absolute days without dysphagia
- Change in PGI-S and PGI-C

7. INVESTIGATIONAL PLAN

7.1. Overall Study Design and Flow Chart

This is a Phase 2a, double-blind, placebo-controlled, multi-center study to assess the efficacy and safety of a repeat-dose regimen of IRL201104 in participants with active EoE. A study diagram is provided in Figure 1.

Eligible participants will undergo an up-to-6-week screening period to stabilize current therapy, confirm diagnosis by histologic assessment, and other eligibility criteria. Participant compliance to DSQ reporting will be assessed for at least 2 consecutive weeks prior to randomization.

At the end of the screening period, eligible participants will be randomized in a 1:1:1 ratio to one of 3 arms, as displayed in Figure 1:

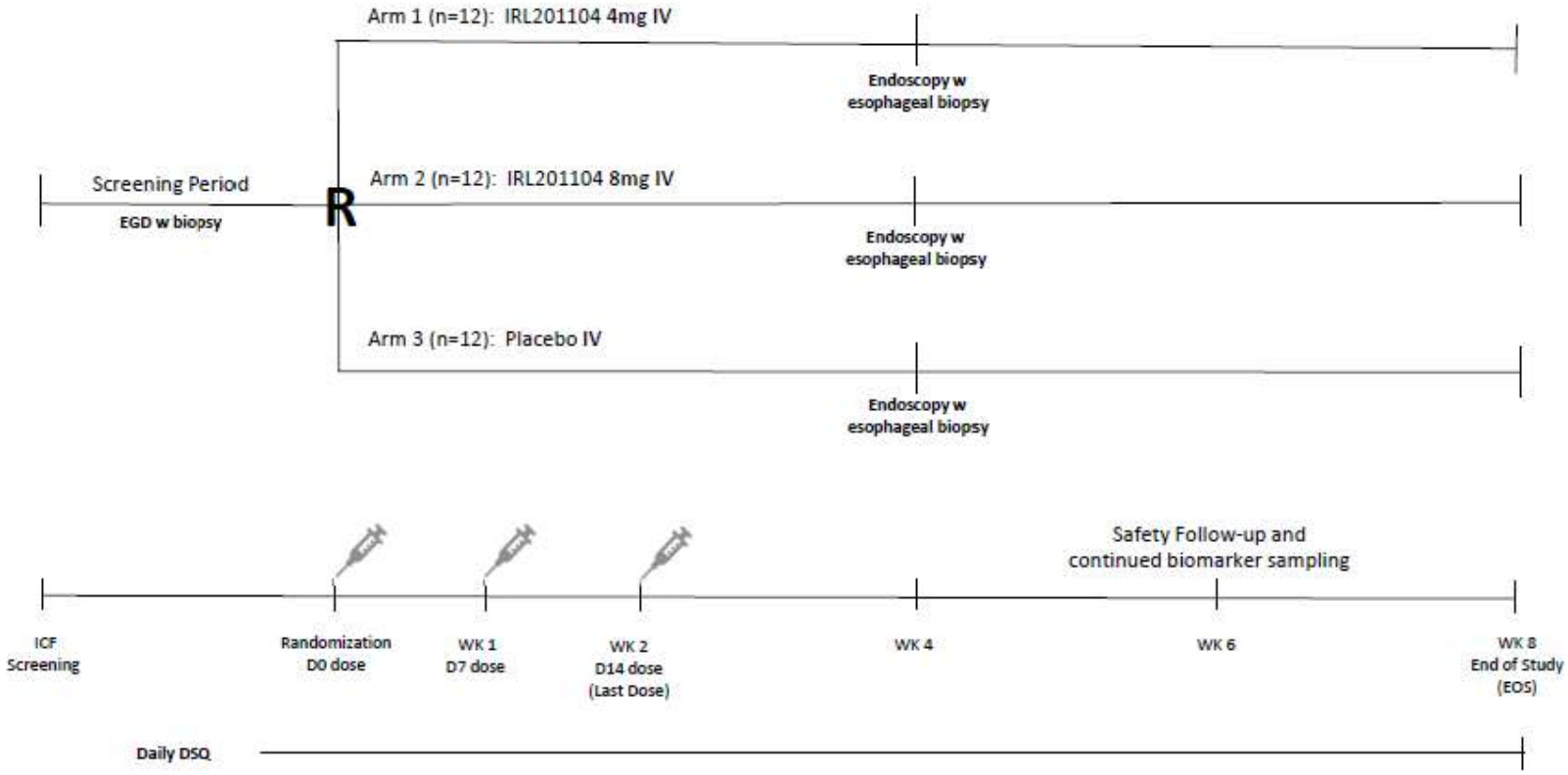
- Arm 1: IRL201104 4 mg IV with endoscopy and esophageal biopsy 2 weeks after last dose (Week 4).
- Arm2: IRL201104 8 mg IV with endoscopy and esophageal biopsy 2 weeks after last dose (Week 4).
- Arm 3: Placebo IV with endoscopy and esophageal biopsy 2 weeks after last dose (Week 4).

Participants will receive IRL201104 (4 mg or 8 mg IV) or placebo on Days 0, 7, and 14 (3 total doses over 14 days). Participants will be evaluated for clinical and histologic responses at Week 4. All participants will be assessed for safety and continued biomarker sampling through Week 8.

The Schedule of Events for the study is provided in Table 3.

The total duration of the study for a given participant is up to 14 weeks.

Figure 1: Study RVLO 121-04 Diagram



D = day; EGD = esophagogastroduodenoscopy; EOS = End of Study; ICF = informed consent form; IV = intravenous; R = randomization; WK = week

7.2. Number of Participants

Approximately up to 50 participants are planned to be enrolled to yield 36 participants who are randomized and complete the final endoscopy at Week 4. The sample size determination is provided in Section 15.4.

7.3. Treatment Assignment

Eligible participants will be randomized in a 1:1:1 ratio to receive IRL201104 4 mg or 8 mg or placebo and to complete the final endoscopy with esophageal biopsy 2 weeks after last dose (Week 4).

7.4. Definition of the End of the Study

The end of the study is defined as the final follow-up visit by the last participant. If the study is terminated prematurely, the study ends when the sponsor notifies the investigator in writing that the study has finished, or when the last participant attends the final follow-up visit, whichever is later.

7.5. Dose Modification Criteria

Dose modification for an individual participant is not allowed.

7.6. Planned Interim Analysis

No interim analysis is planned.

7.7. Independent Data Monitoring Committee

An independent data monitoring committee (IDMC), composed of members who are independent from the sponsor and the study investigators, will monitor participant safety by conducting formal reviews of accumulated safety data.

The IDMC will provide the sponsor with appropriate recommendations on the conduct of the clinical study to ensure the protection and safety of the participants enrolled in the study. The IDMC may also institute measures required for ensuring the integrity of the study results during the study execution.

All activities and responsibilities of the IDMC are described in the IDMC charter.

7.8. Rationale for Dose Selection

A total of 3 doses of 4 mg or 8 mg (IV bolus) administered once on Days 0, 7, and 14 will be tested for efficacy in participants with active EoE in this study.

In safety pharmacology and toxicology studies, IRL201104 was safe and well tolerated at exposures at least 3-fold and up to 11 times higher than the dose level planned for the current Phase 2a clinical study. In 14-day rat and dog Good Laboratory Practice toxicology studies, the no-observed-effect-level was the highest dose tested (0.5 mg/kg IV bolus). Safety margins are summarized in Table 2.

Table 2: Safety Margins for the IRL201104 8 mg Clinical Dose

Species	C_{max} (ng/mL)	C_{max} Fold over Human Dose	AUC (ng.h/mL)	AUC Fold over Human Dose
Human 8 mg single dose (N=10) (Study C1104-001)	545.99 (C_{max})	-	145.1 (AUC_{0-t})	-
Dog NOEL (0.5 mg/kg [1]) 14-day GLP toxicity (Day 1) (Study CGH0005)	2020 (C_{1min})	3.7	456.5 (AUC_{0-24})	3.1
Rat NOEL (0.5 mg/kg [1]) 14-day GLP toxicity (Study CGH0004) (Estimated Day 1)	5940 (C_{max}) [2]	10.9	674 (AUC_{last}) [2]	4.6

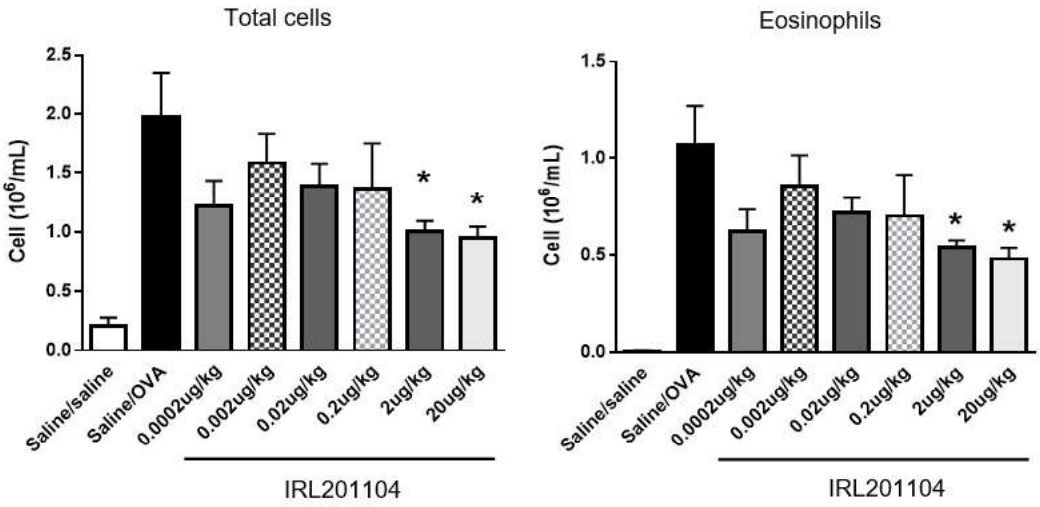
AUC = area under the concentration-time curve; AUC_{0-24} = area under the steady-state blood concentration-time curve over 1 dose interval (ie, from hour 0 to 24 for q24h administration); AUC_{0-t} = area under the blood concentration-time curve from time = 0 to the last measurable concentration at time = t; AUC_{last} = area under the plasma concentration-time curve from time = 0 to the last measurable concentration; C_{1min} = exposure at 1 minute post first dose that day; C_{max} = maximum observed concentration; GLP = Good Laboratory Practice; NOEL = no-observed-effect-level; q24h = once every 24 hours.
[1] Highest dose tested.
[2] Data extrapolated from Study PSN15-318.
Note: The sample matrix was plasma for rat PK studies and whole blood for toxicology and clinical studies.

The dose and dosing regimen have been selected from nonclinical models of allergic inflammation, which assess impact on eosinophils. A number of models assessing eosinophilic inflammation have been conducted showing significant reduction in eosinophils.

7.8.1. Selection of Highest Dose Level

In the ovalbumin challenge model, single IV doses of 2 and 20 µg/kg significantly inhibited eosinophil recruitment into the lung (Study D0028 GSK; refer to the Investigator's Brochure), with a greater effect being seen with 20 µg/kg (Figure 2). Pharmacokinetics were not measured in this study but were measured in a lipopolysaccharide acute inflammation mouse model, where an IV dose of 20 µg/kg (which was maximally efficacious) resulted in a maximum observed concentration (C_{max}) of 152 ng/mL and an area under the plasma concentration-time curve from time = 0 to the last measurable concentration (AUC_{last}) of 18 ng.h/mL (Studies D0055 PNE and PSN18-0258; refer to the Investigator's Brochure). This exposure is consistent with other mouse studies where PK has been measured. A clinical dose of 8 mg (C1104-001) gives an exposure approximately 4- to 8-fold higher than the maximum efficacious dose in mouse model (20 µg/kg).

Figure 2: Effect of IRL201104 (IV) on Ovalbumin-Induced Eosinophil Influx into Mouse Lung



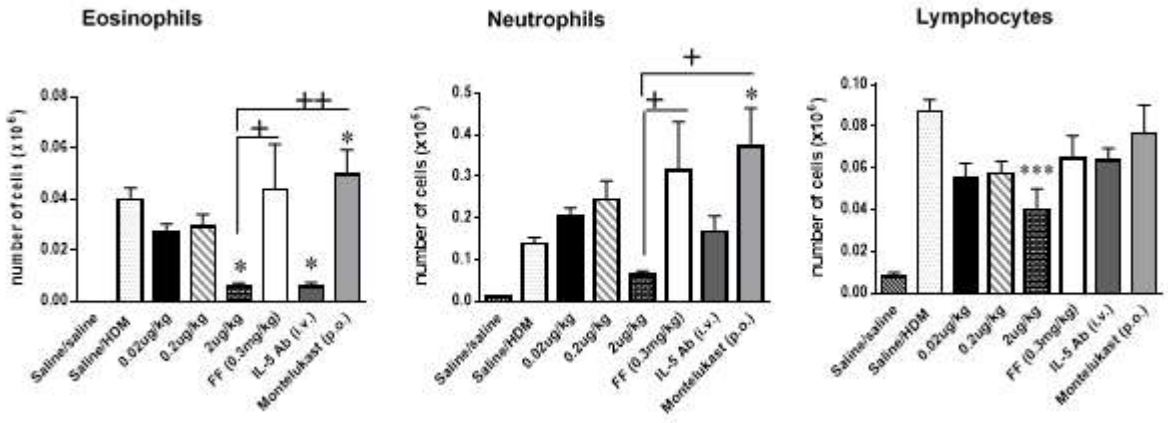
Data represent mean ± standard error of the mean of n= 6 animals; *p<0.05 vs ovalbumin, T- test

7.8.2. Selection of Dose Interval

In addition, an allergic house dust mite (HDM) model has been conducted. Briefly, mice underwent 3 weeks of sensitization to HDM, rested for a period of 2 weeks, and then were challenged intranasally (IN) with HDM. Animals were treated with vehicle, IRL201104, or comparator drugs 15 minutes prior to HDM challenge as a single administration. Lung inflammatory infiltration was measured by analyzing the bronchoalveolar lavage fluid at 4 hours after HDM challenge (acute time point) or after 7 days (chronic time point). A third group of animals was rechallenged with HDM 14 days later and cellular infiltration was evaluated 4 hours later.

IRL201104, given as a single IN administration, dose-dependently reduced eosinophil, neutrophil, and lymphocyte lung influx following both the HDM challenge and rechallenge (Figure 3), indicating that the effect lasted for at least 14 days. Eosinophil recruitment was significantly inhibited by IRL201104 and achieved the same degree of eosinophil inhibition as anti-interleukin (IL)-5 at the maximum dose tested (2 µg/kg).

Figure 3: Effects of IRL201104 IN on Cellular Recruitment at the 14-Day Rechallenge Time Point in the Murine HDM Model



Ab = antibody; ANOVA=analysis of variance; FF = fluticasone furoate; HDM = house dust mite; IL = interleukin; IN=intranasal; IV = intravenous; p.o. = oral; SEM = standard error of the mean
Data represent mean ± SEM of n=6 animals.
*p<0.05, ***p<0.001 vs HDM, ANOVA-Tukey's post-test, +p<0.05, ++p<0.01 vs IRL201104 2 µg/kg

Therefore, 3 doses administered over a 2-week period has been selected as the dosing regimen based on the nonclinical evidence that, despite having a short PK $t_{1/2}$, IRL201104 IV bolus is maximally efficacious in the ovalbumin mouse model (Studies D0028 and D0024) and that the effect lasts for 14 days after a single dose in the HDM mouse model (Study D0024 GSK; refer to the Investigator's Brochure) but the effects appeared to subside at 20 days postdose (ovalbumin mouse model; results published in Riffo-Vasquez 2020). This long PD effect is hypothesized to be because of an effect of IRL201104 on Treg cells; IRL201104 was shown to increase the number of Treg cells in human in vitro assays (Study D0066 KCL; data on file).

Two dose strengths were selected to be tested in the current Phase 2a clinical study: 4 mg and 8 mg. These dose levels have been well tolerated in healthy participant studies to date, the maximum doses studied to date being 8 mg on 7 consecutive days. Experience with anti-IL-5 compounds would indicate that marked and sustained suppression of eosinophils has not been associated with adverse effects for example increased risk of parasitic infection.

7.9. Schedule of Events

The visit schedule is presented in Table 3.

Table 3: Study RVLO 121-04 Schedule of Events

Procedures	Screening Period	Prior to Rand	Rand/ Baseline	Treatment Period				End of Study/ Final Safety	Notes
Visit	**V -1**	**V0**	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	
Week	**-6**	**-**	**0**	**1**	**2**	**4**	**6**	**8**	
Day	**-**	**-**	**0**	**7**	**14**	**28**	**42**	**56**	
Window	**≤ 6 weeks**	**-**	**-**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	
Screening/ Baseline									
Informed consent	X	-	-	-	-	-	-	-	
Demographics/ Medical history review	X	-	-	-	-	-	-	-	
Inclusion/exclusion criteria review	X	X	X	-	-	-	-	-	
Treatment									
Randomization IWRS	-	-	X	-	-	-	-	-	
Administer study treatment	-	-	DBL	DBL	DBL	-	-	-	Pharmacy site staff will be unblinded to prepare the treatment. Investigators, study staff members, and study participants will be blinded to the treatment and dose level (4 mg or 8 mg or placebo) and will remain blinded throughout the study.
Efficacy									
DSQ training and issue handset	X	X	-	-	-	-	-	-	Daily completion of the DSQ starts on the first day it is issued to the participant.

Procedures	Screening Period	Prior to Rand	Rand/ Baseline	Treatment Period				End of Study/ Final Safety	Notes
Visit	**V -1**	**V0**	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	
Week	**-6**	**-**	**0**	**1**	**2**	**4**	**6**	**8**	
Day	**-**	**-**	**0**	**7**	**14**	**28**	**42**	**56**	
Window	**≤ 6 weeks**	**-**	**-**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	
DSQ compliance assessment	-	-	X	X	X	X	X	X	
Retrieval of DSQ handset	-	-	-	-	-	-	-	X	
Esophageal endoscopy and biopsy with EoE-EREFS	EGD	-	-	-	-	X		-	
EoEHSS - histology scoring	X	-	-	-	-	X		-	
PGI-S	-	-	X	-	X	X	X	X	
PGI-C	-	-	-	-	X	X	X	X	
Safety									
Physical examination	X	-	X	X	X	X	X	X	Predose on Day 0.
ECG	X	-	X	X	X	X	X	X	
Height	X	-	-	-	-	-	-	-	
Weight	X	-	X	X	X	X	X	X	
Vitals	X	-	X	X	X	X	X	X	Should be measured within 15 minutes prior to dosing, and +15 minutes (+/- 5 minutes) postdose on Days 0, 7, and 14.
Diet history	X	X	X	X	X	X	X	X	
Concomitant medications and procedures	X	X	X	X	X	X	X	X	
Review of adverse events	-	X	X	X	X	X	X	X	

Procedures	Screening Period	Prior to Rand	Rand/ Baseline	Treatment Period				End of Study/ Final Safety	Notes
Visit	**V -1**	**V0**	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	
Week	**-6**	**-**	**0**	**1**	**2**	**4**	**6**	**8**	
Day	**-**	**-**	**0**	**7**	**14**	**28**	**42**	**56**	
Window	**≤ 6 weeks**	**-**	**-**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	
Local laboratory									
Hematology and serum chemistry	X	-	X	X	X	X	X	X	
Coagulation	X	-	X	X	X	X	X	X	Should be performed before the procedure of endoscopy with esophageal biopsies on days the procedure is performed.
Urinalysis	X	-	X	X	X	X	X	X	
Serology	X	-	-	-	-	-	-	-	
Pregnancy testing	X	-	X	-	-	-	-	X	Serum pregnancy test at screening; urine pregnancy test at all subseqeuent time points.
FSH testing	X	-	-	-	-	-	-	-	FSH of 2.25 U/mL must be documented to confirm post-menopausal status in female participants who have had at least 12 months without menses.
Research samples/ Central laboratory									
Blood PK sampling	-	-	X	X	X	X	X	X	Blood sample for PK concentrations collected predose on Day 0 only; 4min (+/- 1 min) postdose on Days 0, 7, and 14.
Antidrug antibodies	-	-	X	-	-	-	-	X	Predose on Day 0.

Procedures	Screening Period	Prior to Rand	Rand/ Baseline	Treatment Period				End of Study/ Final Safety	Notes
Visit	**V -1**	**V0**	**V1**	**V2**	**V3**	**V4**	**V5**	**V6**	
Week	**-6**	**-**	**0**	**1**	**2**	**4**	**6**	**8**	
Day	**-**	**-**	**0**	**7**	**14**	**28**	**42**	**56**	
Window	**≤ 6 weeks**	**-**	**-**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	**± 3 days**	
Biopsies for central lab	X	-	-	-	-	X		-	Screening endoscopy with esophageal biopsies and photographs are to be performed during screening period to allow results to be available prior to Visit 1.
Central histology result received prior to visit	-	X	-	-	-	-	-	-	
Whole blood for PBMC: Immune cell phenotyping (T, B, & Mo-subsets) and A20	X	-	X	X	X	X	X	X	Predose and 2 hours (+/- 10 minutes) postdose on Days 0, 7, and 14.
Serum cytokines	X	-	X	X	X	X	X	X	Predose and 2 hours (+/- 10 minutes) postdose on Days 0, 7, and 14.
Serum IgE/IgG4 (total)	X	-	X	X	X	X	X	X	Predose and 2 hours (+/- 10 minutes) postdose on Days 0, 7, and 14.
Serum EDN	X	-	X	X	X	X	X	X	Predose and 2 hours (+/- 10 minutes) postdose on Days 0, 7, and 14.

Base = baseline; DSQ = Dysphagia Symptom Questionnaire; ECG = electrocardiogram; EDN = eosinophil derived neurotoxin; EDP = EoE diagnostic panel; EGD = esophagogastroduodenoscopy; EoE = eosinophilic esophagitis; EoEHSS = EoE Histology Scoring System; EREFS = Endoscopic Reference Score; d = days; FSH = follicle stimulating hormone; FU = follow-up; Ig = immunoglobulin; IWRS = Interactive Web Response System; IV = intravenous; Mo = macrophage; q2wk = once every 2 weeks; PBMC = peripheral blood mononuclear cell; PGI-C = Patient Global Impression of Change; PGI-S = Patient Global Impression of Severity; PK = pharmacokinetic; QoL = quality of life; Rand = randomization; DBL = double-blind; Screen = screening; TBD = to be determined; V = visit

8. SELECTION AND WITHDRAWAL OF PARTICIPANTS

8.1. Participant Inclusion Criteria

A participant must meet the following criteria to be eligible for inclusion in the study:

1. Age 18 to 75 years old, inclusive, at the time of signing the informed consent form.
2. Documented diagnosis of EoE by endoscopy prior to screening.

   Note: Must include a demonstration of intraepithelial eosinophilic infiltration (peak cell count ≥ 15 eos/hpf [400×]) from esophageal biopsy specimens from endoscopy.
3. History (by participant report) of on average at least 2 episodes of dysphagia (with intake of solids off anti-inflammatory therapy) per week in the 4 weeks prior to screening, and on average at least 2 episodes of documented dysphagia per week during any 2 consecutive weeks (qualifying period) between screening and baseline; dysphagia is defined as trouble swallowing solid food, or having solid food stick, by participant report; and completed the DSQ on ≥ 70% of days during the qualifying period prior to baseline (Visit 1).
4. Must remain on a stabilized diet for at least 6 weeks prior to screening and during the course of the study; stable diet is defined as no initiation of single or multiple elimination diets or reintroduction of previously eliminated food groups.
5. Must be willing and able to continue any dietary therapy and/or medical regimens (including gastric acid suppression) in effect at the screening visit. There should be no change to these regimens during the study participation.
6. Willing and able to comply with all clinic visits and study-related procedures.
7. Able to understand and complete study-related questionnaires.
8. Provide signed informed consent.
9. Esophagogastroduodenoscopy (EGD) with photographs performed at screening (qualifying EGD), with a demonstration of intraepithelial eosinophilic infiltration (peak cell count ≥15 eos/hpf) in at least 2 of the 3 biopsied esophageal regions (proximal, mid, or distal).

8.2. Participant Exclusion Criteria

A participant who meets any of the following criteria will be ineligible to participate in this study:

1. Prior participation in an IRL201104 clinical study.
2. Has any current disease of the gastrointestinal tract (aside from EoE) that may impact, in the investigator's opinion, the patient's EoE disease status. This includes, but not limited to: eosinophilic gastritis, eosinophilic enteritis, eosinophilic duodenitis, eosinophilic colitis, or proctitis; inflammatory bowel disease; or celiac disease.

3. Has other causes of esophageal eosinophilia or the following diseases: hypereosinophilic syndromes, Churg-Strauss vasculitis (eosinophilic granulomatosis with polyangiitis), or peripheral blood absolute eosinophil count of > 1500 eosinophils/μL.
4. Has presence of oral or esophageal mucosal infection of any type.
5. Has any condition affecting the esophageal mucosa or altering esophageal motility other than EoE.
6. History of achalasia, active *Helicobacter pylori* infection, Crohn's disease, ulcerative colitis, celiac disease, and prior esophageal surgery (with the exception of a surgical repair of an EoE complication).
7. Any esophageal stricture unable to be passed with a standard, diagnostic, adult (9 to 10 mm) upper endoscope or any critical esophageal stricture that requires dilation at screening; or dilation within 2 months prior to screening.
8. On a pure liquid diet or any mouth or dental condition that prevents normal eating.
9. Has initiated, discontinued, or changed dosage regimen of PPIs within the 4 weeks prior to the qualifying EGD, between the qualifying EGD and baseline visit (Visit 1), or anticipates changes in the use of PPI during the study. PPI must remain constant throughout the study.
10. History of bleeding disorders or esophageal varices.
11. Use of anticoagulants within 2 weeks prior to screening. Participants should not stop these agents solely to become eligible for entry into this study.
12. Treatment with an investigational drug within 2 months or within 5 half-lives (if known), whichever is longer, prior to screening.
13. Use of systemic corticosteroids within 3 months or swallowed topical corticosteroids within 6 weeks prior to screening.
14. Treatment with oral immunotherapy (OIT) within 6 months prior to screening.
15. Allergen immunotherapy (sublingual immunotherapy [SLIT] and/or subcutaneous immunotherapy [SCIT]), unless on a stable dose for at least 1 year prior to screening.
16. The following treatments within 3 months before the screening visit, or any condition that, in the opinion of the investigator, is likely to require such treatment(s) during the study:

    Systemic immunosuppressive/immunomodulating drugs (eg, omalizumab, cyclosporine, mycophenolate-mofetil, interferon [IFN]γ, Janus kinase inhibitors, azathioprine, methotrexate, and other biologics that are ongoing [eg, dupilumab, benralizumab, mepolizumab, or vedolizumab]).
17. Diagnosed with active parasitic infection; or suspected parasitic infection, unless clinical and (if necessary) laboratory assessments have ruled out active infection before randomization.
18. Chronic or acute infection requiring treatment with systemic antibiotics, antivirals, or antifungals within 1 month prior to screening.

19. Use of oral antibiotics/anti-infectives within 2 weeks prior to screening.
20. Known or suspected immunosuppression, including history of invasive opportunistic infections (eg, tuberculosis, non-tuberculous mycobacterial infections, histoplasmosis, listeriosis, coccidioidomycosis, pneumocystosis, aspergillosis) despite infection resolution, or otherwise recurrent infections of abnormal frequency, or prolonged infections suggesting an immunocompromised status, as judged by the investigator.
21. Known history of human immunodeficiency virus (HIV) infection.
22. Positive or indeterminate hepatitis B surface antigen (HBsAg) or hepatitis C antibody at screening.
23. Moderate or severe renal impairment (eGFR <60 mL/min/1.73 m^2) or end stage renal disease.
24. Elevated transaminases (alanine aminotransferase [ALT] and/or aspartate aminotransferase [AST]) > 3 times the upper limit of normal (ULN) at screening.
25. History of malignancy within 5 years prior to screening, except completely treated in situ carcinoma of the cervix and completely treated and resolved nonmetastatic squamous or basal cell carcinoma of the skin.
26. Any other medical or psychological condition including relevant laboratory abnormalities at screening that, in the opinion of the investigator, suggest a new and/or insufficiently understood disease, may present an unreasonable risk to the participant as a result of his/her participation in this clinical study, may make the participant's participation unreliable, or may interfere with study assessments. The specific justification for participants excluded under this criterion will be noted in study documents (eg, chart notes, electronic case report form). These may include participant-reported alcohol or drug abuse and severe concomitant illness(es).
27. Planned or anticipated use of any prohibited medications and procedures (as described in the exclusion criteria) during study treatment.
28. Treatment with a live (attenuated) vaccine within 3 months prior to screening and/or treatment of a killed vaccine within 30 days prior to screening, until the end of the study with the exception of a coronavirus disease of 2019 (COVID-19) vaccine, as described in Section 9.2.1.
29. Pregnant or breastfeeding women, or women planning to become pregnant or breastfeed during the study.
30. Women unwilling to use adequate birth control, if of reproductive potential* and sexually active. Adequate birth control is defined as agreement to consistently practice an effective and accepted method of contraception throughout the duration of the study and for 30 days after the last dose of study treatment. These include: hormonal contraceptives, intrauterine device, or double barrier contraception (ie, condom and diaphragm), or male partner with documented vasectomy.

    *For females, menopause is defined as at least 12 consecutive months without menses; to include laboratory confirmation of post-menopausal status (ie, a follicle stimulating hormone (FSH) of 2.25 U/mL must be documented). Hysterectomy, bilateral

oophorectomy, or bilateral tubal ligation must be documented, as applicable; if documented, women with these conditions are not required to use additional contraception.

8.3. Participant Withdrawal Criteria

8.3.1. Withdrawal of Participants

Participants are free to withdraw from the study at any time without giving reasons. Furthermore, the investigator may withdraw a participant for reasons such as intolerance to study treatment, intercurrent illness, need for medication that is contraindicated, significant noncompliance with the requirements of the study, or withdrawal of consent. The investigator will assess the reasons for withdrawal as far as possible and will fully record the circumstances and medical details.

Participants who permanently discontinue study treatment and who do not withdraw from the study will be asked to return to the clinic for all remaining study treatment visits and participate in all follow-up assessments according to the visit schedule.

8.3.2. Individual Stopping Criteria

Participants will be permanently discontinued from study treatment in the event of:

- Anaphylactic reaction to study treatment
- Treatment with any prohibited concomitant medication or procedure (Section 9.2.2)
- Diagnosis of a malignancy during study, excluding carcinoma in situ of the cervix or squamous or basal cell carcinoma of the skin
- Evidence of pregnancy
- Any infection that:
  - Requires parenteral treatment with antibiotic, antifungal, antiviral, anti-parasitic, or anti-protozoal agent
  - Requires oral treatment with such agents for longer than 2 weeks
  - Is opportunistic, such as tuberculosis and other infections whose nature or course may suggest an immunocompromised status
- Severe laboratory abnormalities, such as:
  - Neutrophil count $<0.5 \times 10^3/\mu L$
  - Platelet count $<50 \times 10^3/\mu L$
  - ALT and/or AST values >3 x ULN with total bilirubin >2 x ULN, excluding confirmed Gilbert's Syndrome
- Confirmed AST and/or ALT >5 x ULN (for more than 2 weeks)
- Other reasons that may lead to the permanent discontinuation of study treatment include:

- Certain AEs deemed related to the study treatment (eg, severe and prolonged injection site reactions) or study procedures

8.3.3. Allowable Deviations in Timing of Study Treatment

Study treatment dosing may be delayed around the allowable 3-day windows as specified in the Schedule for Events, for any reason, such as:

- Clinically important laboratory abnormalities, such as:
  - Neutrophil count $<1.0 \times 10^3/\mu L$ but $>0.5 \times 10^3/\mu L$
  - Platelet count $<100 \times 10^3/\mu L$ but $>50 \times 10^3/\mu L$
  - Creatine phosphokinase (CPK) >10 ULN
- Short-term, self-limiting conditions (eg, infections resolving spontaneously or requiring <2 weeks of oral anti-infective treatment), with the exception of upper respiratory infections
- Other intercurrent illnesses

A decision to delay study treatment should be discussed with the medical monitor. The investigator may delay study treatment at any time, even without consultation with the medical monitor if the urgency of the situation requires immediate action and if this is determined to be in the participant's best interest. However, the medical monitor should be contacted as soon as possible in any case of study treatment delay.

A participant is not allowed to skip any doses of study treatment during the study.

Dose reductions are not allowed.

8.3.4. Replacement of Participants

Participants who discontinue from the study will be replaced if they did not receive 3 doses of study treatment and the final endoscopy as randomized.

9. TREATMENT OF PARTICIPANTS

9.1. Description of Study Treatment

Study treatment will be administered as a repeat-dose regimen of IRL201104 4 mg or 8 mg or placebo on Days 0, 7, and 14 (3 total doses over 14 days).

Study treatment will be delivered by IV bolus via a cannula over 30 seconds, and not to exceed 60 seconds.

An overview of the investigational product used in the study is provided in Table 4.

Table 4: Investigational Product

	Investigational Product		
Product Name:	**IRL201104**		**Placebo**
Dosage Form:	Lyophilized cake reconstituted with sterile water for injection	Lyophilized cake reconstituted with sterile water for injection	Lyophilized cake reconstituted with sterile water for injection
Unit Dose	4 mg (0.8 mg/ml per 20 mL vial reconstituted with 20 mL)	8 mg (0.8 mg/ml per 20 mL vial reconstituted with 10 mL)	N/A
Volume Required for Reconstitution	20 mL	10 mL	10 mL
Volume for Administration	10 mL	10 mL	10 mL
Route of Administration	IV injection	IV injection	IV injection

9.2. Concomitant Medications

Any treatment (including nutritional supplements) or procedure administered from the time of informed consent to the end of the final study visit is considered concomitant medication/procedures. This includes medications that were started before the study and are ongoing during the study. All concomitant treatments will be reported in the eCRF along with their daily dosage, duration, and reasons for administration. Participants who have received any concomitant treatment may be withdrawn from the study at the discretion of an investigator.

9.2.1. Permitted Concomitant Medications and Procedures

Other than the prohibited medications listed in Section 9.2.2, treatment with concomitant medications are permitted during the study.

The dosage regimen of PPIs for any condition (such as gastroesophageal reflux disease, asthma, or allergic rhinitis) must remain constant throughout the study.

Participants in this study may be offered COVID-19 vaccines during this study. Where possible, vaccinations should not occur within 7 days of a clinical visit (7 days either before or after the visit). The date of vaccination should be accurately recorded in the CRF. Study treatment should not be administered within 48 hours (48 hours before or after) of the COVID-19 vaccine.

Other vaccines are prohibited as specified in Section 9.2.2.

9.2.2. Prohibited Concomitant Medications and Procedures

Treatment with the following concomitant medications is prohibited. Study treatment will be permanently discontinued if any of the following are used:

- Medications used for the treatment of EoE (these are considered rescue medications):
  - Swallowed topical corticosteroids
  - Systemic corticosteroids
  - Dose change of systemic leukotriene inhibitors, topical, nasal, and/or inhaled corticosteroids
  - Systemic treatment for EoE with an immunosuppressive/immunomodulating substance (including, but not limited to, omalizumab, cyclosporine, mycophenolate-mofetil, IFNγ, Janus kinase inhibitors, azathioprine, methotrexate, leukotriene inhibitors and other biologics [eg, dupilumab, benralizumab, mepolizumab, vedolizumab])
- Allergen immunotherapy (SCIT and SLIT are allowed if dose is stable for 1 year or more; however, OIT is prohibited)
- Participants who are not using PPI in the 8 weeks prior to screening cannot start PPI therapy prior to End of Study visit
- Treatment with a live attenuated vaccine
- Treatment with a killed vaccine, with the exception of a COVID-19 vaccine (Section 9.2.1)
- Treatment with an investigational drug (other than IRL201104)
- The following concomitant procedures are prohibited during the study:
  - Major elective surgical procedures
  - Esophageal dilation (considered rescue procedure)
  - Diet change (participants should remain on stable diet for at least 6 weeks prior to screening and during the course of the study; stable diet is defined as no initiation of single or multiple elimination diets or reintroduction of previously eliminated food groups)

9.2.3. Prohibited Concomitant Medications or Procedures as Rescue

If medically necessary (eg, for treatment of intolerable EoE symptoms), participants may be rescued with a prohibited medication or procedure (as defined in Section 9.2.2) at the discretion of the investigator. Participants who receive rescue treatment with a prohibited medication or procedure will be permanently discontinued from study treatment. These participants will be asked to return to the clinic for all remaining study treatment visits and participate in all follow-up assessments according to the visit schedule. Investigators should make every attempt to obtain efficacy measurements before initiation of rescue treatment.

9.3. Method of Treatment Assignment

Approximately 36 participants will be randomized in a 1:1:1ratio to receive IRL201104 4 mg or 8 mg or placebo according to a central randomization scheme provided by an interactive web response system (IWRS).

9.3.1. Blinding

This is a double-blind study using open-label investigational product. Pharmacy staff will be unblinded for investigational product preparation. Investigators, study team members and study participants will be blinded to the dose level given (4 mg, 8 mg, or placebo) and will remain blinded throughout the study.

10. STUDY TREATMENT MATERIALS AND MANAGEMENT

10.1. Study Treatment

IRL201104 drug product and placebo is packaged as a sterile, lyophilized cake in a standard glass vial and is reconstituted with sterile water for injection (WFI) (Table 4). The WFI used to reconstitute the drug product will be an authorized product supplied by the clinical study center.

For both the 4 and 8 mg doses, the amount of drug substance in each 20 mL vial is 0.8 mg/ml, which is reconstituted with 10 or 20 mL WFI. Investigational product will be administered in a volume of 10 mL.

The process for reconstitution will be detailed in the sponsor pharmacy manual.

10.2. Study Treatment Packaging and Labeling

The study treatment will be packaged and labeled according to Good Manufacturing Practice (GMP) guidelines and applicable laws and regulations.

10.3. Study Treatment Storage

Investigational product will be stored at –20°C ± 5°C.

If an out-of-range temperature is noted, the procedures will be followed as set forth in the sponsor pharmacy manual.

10.4. Study Treatment Preparation

Instructions for the preparation and reconstitution of study treatment will be provided in the sponsor pharmacy manual.

10.5. Administration

IRL201104 is administered by IV injection over 30 seconds (not to exceed 60 seconds) via a cannula. Study dosing is considered completed when the study treatment and subsequent flushing of the line has been fully administered. Additional dosing details will be provided in the Pharmacy Manual.

10.6. Study Treatment Accountability

Accountability is ensured by administering the study treatment as an IV injection by designated personnel at the study center.

All study treatment accountability records must be kept current.

The investigator is accountable for proper storage temperature and conditions of study treatment. At the end of the study, all unused study treatment supplies will be returned to the sponsor or destroyed in accordance with the sponsor's instructions.

The investigator must be able to account for all opened and unopened study treatment. These records should contain the dates, quantity, and study treatment dispensed to each participant and disposed of at the study center.

All accountability records must be made available for inspection by the sponsor and regulatory agency inspectors; photocopies must be provided to the sponsor at the conclusion of the study.

10.7. Study Treatment Handling and Disposal

Procedures for study treatment handling and disposal are provided in the study manuals.

11. ASSESSMENT OF EFFICACY

All efficacy assessments will be collected at the time points specified in Table 3.

11.1. Histologic Response

Esophageal biopsies will be obtained by endoscopy. The peak eos/hpf is the primary measure of efficacy.

The screening endoscopy should be performed during the screening period to allow results to be available prior to Visit 1 for assessment of eligibility. A total of 9 mucosal pinch biopsies will be collected at each time point from across 3 esophageal regions: proximal, mid-esophagus, and distal. Three samples from each region will be used for the histology (needed for study inclusion criteria, as well as efficacy endpoints). Biopsy tissue will be placed in level-specific vials and sent to the central pathology laboratory for processing of tissue into slides. Esophageal biopsies will be used to assess histologic changes and to examine EoE gross esophageal mucosal features based on a validated scoring system for inflammatory and remodeling aspects of the disease. To be randomized in the study, participants must have a peak intraepithelial eosinophil count of at least 15 eos/hpf [400×]) from esophageal biopsy specimens in at least 2 of the 3 esophageal regions sampled.

Eosinophil counts, histopathologic features, and gross endoscopic findings will be evaluated and scored for each endoscopy. Eight histopathologic epithelial features (basal layer hyperplasia, eosinophil density, eosinophil microabscesses, eosinophil surface layering, dilated intercellular spaces, surface epithelial alteration, dyskeratotic epithelial cells, lamina propria fibrosis) will be scored on a 4-point scale (0 = normal, 3 = worst) for both the severity of the abnormality (ie, grade) and the amount of tissue affected by the abnormality (ie, stage).

Endoscopic findings with separate evaluations of the proximal and distal esophagus will be recorded with respect to 5 categories by the endoscopist: 1) exudates or plaques (grade 0–2); 2) fixed esophageal rings (grade 0–3); 3) edema (grade 0–1); 4) furrows (grade 0–1); and 5) strictures (grade 0–1). An endoscopy score for each category will be calculated and summed for each anatomic location (proximal and distal). The maximum endoscopy score is 10 points for

each location, and a Total Endoscopy Score is the sum of the scores for the proximal and distal locations.

In addition, the general appearance of the stomach and duodenum will be assessed by the endoscopist. Biopsies will be taken from the stomach and duodenum for the screening EGD as follows: gastric body (greater curvature): 2 specimens, gastric antrum: 2 specimens, and duodenum (third part or distal): 2 specimens. Biopsies from the stomach should be submitted in one vial; biopsies from the duodenum should be submitted in a separate vial to the central pathology laboratory for processing of tissue into slides. If the pre-treatment biopsy identifies eosinophilia in the stomach and/or duodenum, the participant will be excluded from the study.

As a secondary endpoint, histologic response will be defined as eosinophil count < 15 eos/hpf (400×) in all available esophageal levels following 14 days of treatment with IRL201104 based on anatomical and histological findings from endoscopy with esophageal biopsies. Change in peak esophageal eos/hpf from baseline will be determined by counting eosinophils in the most inflamed areas of each esophageal region sampled at each time point and calculating the change in the peak count at each location obtained at baseline compared with the count obtained at subsequent time points.

Histology results will be interpreted by a pathologist at a central pathology reading center. Gastric and duodenal biopsies may be repeated at the discretion of the investigator, but are not required. Detailed instructions for biopsy sample collection and handling will be provided in the study manuals.

Tissue blocks remaining after the histological assessment will be banked for exploratory research. Remaining samples from each region will be processed to bank tissue RNA for EDP transcriptome signature (Section 11.7) and other EoE related gene expression exploratory research (Section 11.8). Biopsy samples will be sent to a central pathology laboratory for processing and analysis. If required by the investigator institution, biopsy samples will be processed and analyzed by the local laboratory, and the processed specimen will be sent to the central pathology laboratory for central reading.

In addition to the samples to assess histological response, the next set of biopsies will be taken from the most inflamed area of the esophagus. A total of 4 biopsies will be obtained and stored in RNA*later*™ (1 fragment), flash frozen (1 fragment), and formalin-fixed (2 fragments). The banked samples will be used for exploratory analysis which could include, but are not limited to, gene expression, epigenetics, cell culture, and microbiome, etc. Detailed instructions for biopsy sample collection and handling will be provided in the study manuals.

The number of and locations of required biopsy sampling is summarized in Table 5.

Table 5: Biopsy Sampling

	Screening: # Biopsy Samples				Postdose: # Biopsy Samples			
	Histologic	Exploratory			Histologic	Exploratory		
		RNAseq	Flash Frozen	Formalin-fixed		RNAseq	Flash Frozen	Formalin-fixed
Esophagus								
Proximal	3				3			
Mid	3				3			
Distal	3				3			
Most Inflamed Area		1	1	2		1	1	2
Stomach and Duodenum								
Gastric body	2							
Gastric antrum	2							
Duodenum	2							

11.2. Dysphagia Symptom Questionnaire

The DSQ is used to measure the frequency and intensity of dysphagia. The DSQ scores can range from 0 to 84, with a lower score indicating less frequent or less severe dysphagia (Dellon 2013).

The questionnaire will be completed by participants daily. Each evening before bedtime, participants will be asked to indicate if they experienced dysphagia symptoms (eg, food passing slowly or food sticking) during that day. Participants must have experienced dysphagia (response of "yes" to Question 2 on DSQ) on a minimum of 2 days total and completed the DSQ on ≥ 70% of days in at least 2 consecutive weeks (qualifying period) prior to the baseline visit (Visit 1). Participants must fill out the DSQ at least 5 or more days during a given week in order to be compliant.

Calculations will be performed on daily ePRO entries from the qualifying period prior to baseline, and prior to each study visit during the treatment and follow-up periods. The DSQ score for the secondary endpoints will be calculated by summing the scores of responses to Questions 2 and 3 only. Questions 1 and 4 will be excluded from the DSQ score.

DSQ score for 14-day period =

$$\frac{\text{(Sum of points from questions 2+3 in the daily DSQ with non-missing data over the interval being reported)} \times 14}{\text{Number of diaries reported with non-missing data over the interval being reported}}$$

For each DSQ calculation, the sum of the specified question(s) from each reported daily diary with non-missing data over the interval being reported will be obtained, dividing this by the number of reported daily diaries with non-missing data in that same interval. This result will then be multiplied by the interval. This normalizes each DSQ sum over the interval. For the DSQ score, if a response of "No" is recorded for Question 2, the DSQ score will be set to zero for that day. The DSQ calculations during the interval being reported prior to the baseline visit will be defined as the baseline DSQ evaluation. For each 14-day period, at least 8 reported diaries are needed to effectuate a DSQ calculation for that period. If at least 8 diaries are not available, the DSQ result for that 14-day period will be set to missing.

11.3. Eosinophilic Esophagitis-Endoscopic Reference Score

The EoE-EREFS will be used to measure the endoscopically identified EoE esophageal mucosal inflammatory and remodeling features. This instrument includes a total of 17 items related to the presence and severity of esophageal features. The specific esophageal features include: rings (absent, mild, moderate, severe, not applicable); stricture (yes, no, not applicable); diameter of the stricture (if applicable); exudates (absent, mild, severe); furrows (absent, present); edema (absent, present); crepe paper esophagus (absent, present); overall general appearance incorporating all endoscopically identified EoE findings (ie, fixed rings, strictures, whitish exudates, furrowing, edema, and crepe paper mucosa). In addition, mucosal changes associated with gastroesophageal reflux disease (GERD) will also be recorded using the Los Angeles classification system for erosions (No Erosions or LA Classification A, B, C, D). The EoE esophageal characteristics will be analyzed based on the EoE-EREFS, a validated scoring system for inflammatory and remodeling features of disease using both overall scores and scores for each individual characteristic (Hirano 2013). The EoE-EREFS should be performed by the physician who performs the endoscopy procedure at the time of the endoscopy. In addition to the local read, images will be submitted to the central imaging laboratory for central reading.

11.4. Eosinophilic Esophagitis-Histology Scoring System

The EoEHSS is a validated histology scoring system for esophageal biopsies that evaluates 8 features: eosinophil density, basal zone hyperplasia, eosinophil abscesses, eosinophil surface layering, dilated intercellular spaces, surface epithelial alteration, dyskeratotic epithelial cells, and lamina propria fibrosis (absent/present) (Collins 2017). Severity (grade) and extent (stage) of abnormalities will be scored using a 4-point scale (0 normal; 3 maximum change).

11.5. Patient Global Impression of Severity

The PGI-S is a PRO measure that uses a verbal rating scale of severity. The PGI-S uses a recall period that aligns with the assessment period used for calculating DSQ endpoint scores (ie, DSQ endpoints are based on 14-day scores, and the PGI-S is administered on the last day of the 14-day assessment period. The PGI-S instructs participants to recall over the past 14 days, such that the anchor covers the same 14-day period as the 14-day DSQ endpoint score).

11.6. Patient Global Impression of Change

The PGI-C is a PRO measure that assesses change compared with prior to starting treatment and reflects the participant's belief about the efficacy of treatment. The PGI-C is a scale depicting a participant's rating of overall improvement.

11.7. RNA Sequencing and EDP Transcriptome

RNA sequencing and EDP transcriptome will be evaluated.

A molecular EDP has been identified that is composed of 94 EoE genes and distinguishes EoE from control individuals without esophagitis or with GERD (Wen 2013).

The EDP signature will be evaluated in biopsy samples. Using a method reported by Wen (2015), the EoE transcriptome will be determined using the EDP from extracted RNA.

Further details will be provided in the study manuals.

11.8. Other Biomarkers of Eosinophilic Esophagitis

Other biomarkers of disease will be explored and may include, but are not limited to: immunohistochemistry and immunofluorescence, DNA analysis, microdissection further immunophenotyping, gene expression, epigenetics, cell culture, and microbiome, etc.

12. ASSESSMENT OF SAFETY

All safety assessments will be collected at the time points specified in the Schedule of Events (Table 3).

12.1. Screening and Baseline Assessments

A screening assessment to determine study eligibility will be performed after the participant has provided signed informed consent. The participant will be assigned a unique participant number once the ICF has been signed.

Demographic characteristics will include standard demography (eg, age, race, and weight), medical history, medication history, and diet history for each participant. Characteristics of disease, including duration, and disease symptoms, will be collected. Viral serology, demographic characteristics, medical and medication history, and height are collected at screening only.

12.2. Vital Signs

Vital sign measurements include systolic and diastolic blood pressure, heart rate, temperature, and respiratory rate and will be assessed within 15 minutes predose and at 15 minutes (+/-5 minutes) postdose.

Measurements should be made with participants in a sitting position, after resting for at least 5 minutes.

Height will be measured at screening only.

12.3. Physical Examination

A complete physical examination comprises a routine medical examination, including gross neurological assessments. The following body systems will be examined: general appearance, neurological, eyes, ear/nose/throat, cardiovascular, respiratory, abdominal, hepatic, gastrointestinal, musculoskeletal, and dermatological.

For each body system the result will be recorded as "normal", "abnormal not clinically significant", or "abnormal clinically significant".

12.4. Electrocardiogram

A standard 12-lead ECG will be performed before blood is drawn during visits requiring blood draws. The ECG strips or reports will be retained with the source documentation, and the results will be documented in the eCRF. Electrocardiogram results will be locally interpreted.

12.5. Laboratory Assessments

Detailed instructions for blood sample collection are in the study manuals provided to study centers.

Local Laboratory Assessments:

The following clinical laboratory parameters will be assessed:

Blood Chemistry

Sodium	Total protein, serum	Total and indirect bilirubin
Potassium	Creatinine	Total cholesterol
Chloride	Blood urea nitrogen	Low-density lipoprotein
Carbon dioxide	AST	High-density lipoprotein
Calcium	ALT	Triglycerides
Glucose	Alkaline phosphatase	Uric acid
Albumin	Lactate dehydrogenase	CPK

Hematology

Hemoglobin	Differential:
Hematocrit	Neutrophils
Red blood cells	Lymphocytes
White blood cells	Monocytes
Red cell indices	Basophils
Platelet count	Eosinophils

Urinalysis

Microscopic analysis will only be done in the event of abnormal dipstick results.

Color	Glucose	Red blood cells
Clarity	Blood	Hyaline and other casts

pH	Bilirubin	Bacteria
Specific gravity	Leukocyte esterase	Epithelial cells
Ketones	Nitrite	Crystals
Protein	White blood cells	Yeast

Serological markers

HIV-1/2 antibodies	HBsAg	Hepatitis C virus antibodies

Other laboratory tests:

- Serum pregnancy tests will be performed for females of reproductive potential at screening, after which urine pregnancy tests will be performed.
- FSH testing will be performed at screening to confirm post-menopausal status in female participants who have had at least 12 months without menses.
- Coagulation testing (prothrombin time and partial thromboplastin time) will be performed as indicated in the Schedule of Events (Table 3). Samples for coagulation tests should be obtained before the procedures of endoscopy with esophageal biopsies are performed.

Abnormal laboratory values and laboratory AEs:

- All laboratory values must be reviewed by the investigator or authorized designee.
- Significantly abnormal tests must be repeated to confirm the nature and degree of the abnormality. When necessary, appropriate ancillary investigations should be initiated. If the abnormality fails to resolve or cannot be explained by events or conditions unrelated to the study treatment or its administration, the medical monitor must be consulted.
- The clinical significance of an abnormal test value, within the context of the disease under study, must be determined by the investigator.

Criteria for reporting laboratory values as an AE are provided in Section 12.6.1.4.

Central laboratory assessments for the assessment of PK and immunogenicity are described in Section 13.

12.6. Adverse and Serious Adverse Events

12.6.1. Definition of Adverse Events

12.6.1.1. Adverse Event

An AE is the development of an undesirable medical condition or the deterioration of a pre-existing medical condition following or during exposure to a pharmaceutical product, whether or not considered causally related to the product. In clinical studies, an AE can include an undesirable medical condition occurring at any time, including baseline or washout periods, even if no study treatment has been administered.

All AEs that occur after any participant has been enrolled, before treatment, during treatment, or within 30 days following the cessation of treatment, whether or not they are related to the study, must be recorded on forms provided by the sponsor.

12.6.1.2. Serious Adverse Event

A serious adverse event (SAE) is an AE occurring during any study phase (ie, baseline, treatment, washout, or follow-up), and at any dose of the study treatment, that fulfils one or more of the following:

- Results in death
- It is immediately life-threatening
- It requires in-patient hospitalization or prolongation of existing hospitalization
- It results in persistent or significant disability or incapacity
- Results in a congenital abnormality or birth defect
- It is an important medical event that may jeopardize the participant or may require medical intervention to prevent one of the outcomes listed above.

All SAEs that occur after any participant has been enrolled, before treatment, during treatment, or within 30 days following the cessation of treatment, whether or not they are related to the study, must be recorded on forms provided by the sponsor.

12.6.1.3. Other Adverse Events

The following events require reporting to the sponsor (or designee) within 24 hours of learning of the event:

Pregnancy

Should a pregnancy occur, it must be reported and recorded on the sponsor's pregnancy form. Pregnancy in itself is not regarded as an AE unless there is a suspicion that an investigational product may have interfered with the effectiveness of a contraceptive medication.

Although pregnancy is not considered an AE, it is the responsibility of the investigator to report to the sponsor (or designee) by telephone within 24 hours of identification, any pregnancy occurring in a female participant or female partner of a male participant, during the study or within 120 days of the last dose of study treatment. Any complication of pregnancy affecting a female study participant or female partner of a male study participant, and/or fetus, and/or newborn must be reported as an SAE.

The outcome of all pregnancies (spontaneous miscarriage, elective termination, normal birth, or congenital abnormality) must be followed up and documented even if the participant was discontinued from the study.

Overdose of Study Treatment

Any accidental or intentional overdose over the intended dose of study treatment within the intended therapeutic window, if associated with an AE, should be reported within 24 hours.

Adverse Events of Special Interest

Adverse events of special interest (AESIs) must be reported within 24 hours of identification. Adverse events of special interest in this study include:

- Anaphylactic reactions or acute allergic reactions that require immediate treatment
- Severe injection site reactions that last longer than 24 hours
- Any severe infection, any bacterial infection requiring treatment with parenteral antibiotics or treatment with oral antibiotics for longer than 2 weeks, any clinical endoparasitosis, any opportunistic infection, any viral infection requiring antiviral treatment
- Note: Generally, all uncommon, atypical, peculiar, or unusually persistent infections, especially viral infections, should be reported as AESI.

Refer to the study manuals for the procedures to be followed.

12.6.1.4. Abnormal Laboratory, Vital Signs, or Electrocardiogram Results

The criteria for determining whether an abnormal objective test finding should be reported as an AE include:

- the test result is associated with accompanying symptoms; and/or
- the test result requires additional diagnostic testing or medical/surgical intervention; and/or
- the test result leads to discontinuation from the study, significant additional concomitant drug treatment, or other therapy.

Contact the medical monitor in the event the investigator feels that an abnormal test finding should be reported as an AE, although it does not meet any of the above criteria.

Repeating an abnormal test, in the absence of any of the above conditions, does not constitute an AE. Any abnormal test result that is determined to be an error does not require reporting as an AE.

12.6.2. Relationship to Study Drug

An investigator who is qualified in medicine must make the determination of relationship to the investigational product for each AE (Unrelated, Possibly Related or Probably Related). The investigator should decide whether, in his or her medical judgment, there is a reasonable possibility that the event may have been caused by the investigational product. If no valid reason exists for suggesting a relationship, then the AE should be classified as “unrelated.” If there is any valid reason, even if undetermined, for suspecting a possible cause-and-effect relationship between the investigational product and the occurrence of the AE, then the AE should be considered “related.”

If the relationship between the AE/SAE and the investigational product is determined to be “possible” or “probable” the event will be considered to be related to the investigational product for the purposes of expedited regulatory reporting.

12.6.3. Recording Adverse Events

Adverse events spontaneously reported by the participant and/or in response to an open question from the study personnel or revealed by observation will be recorded during the study at the

investigational study center. Clinically significant changes in laboratory values, blood pressure, and pulse need not be reported as AEs. However, abnormal values that constitute an SAE or lead to discontinuation of administration of study treatment must be reported and recorded as an AE. Information about AEs will be collected from the signing of the consent form until 30 days following the last dose of study treatment. Serious AE information will be collected from signing of the consent form until 30 days following the last dose of study treatment. The AE term should be reported in standard medical terminology when possible. For each AE, the investigator will evaluate and report the onset (date and time), resolution (date and time), intensity, causality, action taken, serious outcome (if applicable), and whether or not it caused the participant to discontinue the study.

Intensity will be assessed according to the following scale:

- Mild (awareness of sign or symptom, but easily tolerated)
- Moderate (discomfort sufficient to cause interference with normal activities)
- Severe (incapacitating, with inability to perform normal activities)

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity whereas seriousness is defined by the criteria under Section 12.6.1.2. An AE of severe intensity may not be considered serious.

12.6.4. Reporting Adverse Events

All SAEs (related and unrelated) will be recorded from the signing of consent form until 30 days following the end of treatment exposure. Any SAEs considered possibly or probably related to the investigational product and discovered by the investigator at any time after the study should be reported. All SAEs must be reported to the sponsor within 24 hours of the first awareness of the event. The investigator must complete, sign, and date the SAE pages, verify the accuracy of the information recorded on the SAE pages with the corresponding source documents, and send to the sponsor.

Additional follow-up information, if required or available, should all be reported to the sponsor within 24 hours of receipt and this should be completed on a follow-up SAE form and placed with the original SAE information and kept with the appropriate section of the eCRF and/or study file.

The sponsor is responsible for notifying the relevant regulatory authorities of certain events. It is the Principal Investigator's responsibility to notify the Institutional Review Board (IRB) or Independent Ethics Committee (IEC) of all SAEs that occur at his or her study center. Investigators will also be notified of all unexpected, serious, drug-related events (7/15 Day Safety Reports) that occur during the clinical study. Each study center is responsible for notifying its IRB or IEC of these additional SAEs.

All AEs that lead to a participant's withdrawal from the study must be reported to the sponsor's medical monitor within 30 days.

Refer to the study manuals for procedures to be followed.

13. ASSESSMENT OF PHARMACOKINETICS AND IMMUNOGENICITY

Samples will be collected pre- and postdose at the time points listed in the Schedule of Events (Table 3) for the following assessments:

- IRL201104 PK concentrations
- Antidrug antibodies (ADA)
- Immune cell phenotyping (T-subsets, B-subsets, and macrophage subsets)
- A20
- Serum cytokines
- Serum IgE/IgG4
- Serum EDN

Immunogenicity variables include ADA status. Results are reported as an optical density (OD) unit with a cut-off value. If the OD value is lower than the cut-off value, it is reported as negative, and if OD value is higher than the cut-off value it is positive. Participants who are assessed as ADA positive at their last study visit (early termination or end of study) will be considered for follow-up based on the overall clinical presentation at that time. Participants who are identified for follow-up may be asked to return to the clinic to have additional ADA samples collected for analysis.

The PK and immunogenicity assessments will be analyzed by a central laboratory.

14. RESEARCH SAMPLES

Research samples (blood/esophageal biopsies) will be collected at time points listed in the Schedule of Events (Table 3).

Use and Storage of Research Samples (Serum/Plasma/Esophageal Biopsies)

Research blood samples and esophageal biopsy samples will be banked and may be used to study the effects of the study treatment on target pathway modulation and EoE pathophysiology. Stored samples may also be used to discover markers predictive of IRL201104 response as well as identify markers associated with toxicity. Details for banking samples will be provided in the study manuals.

Technologies that may be employed include circulating protein marker analyses (eg, enzyme-linked-immunosorbent serologic assay, electrochemiluminescence), immunohistochemistry and/or histology of biopsies, and/or transcriptome analyses of blood and/or biopsy samples (eg, microarray, transcriptome sequencing, and/or quantitative reverse transcriptase-polymerase chain reaction).

Detailed instructions for sample collection and handling will be provided in the study manuals.

15. STATISTICS

15.1. General Considerations for Data Analysis

No formal statistical analysis will be conducted for this study. Summary statistics appropriate for categorical and continuous variables will be presented by treatment group and all inferences will be based on clinical evaluation of those results. A detailed statistical analysis plan (SAP) will be prepared that describes the method and style of the data summaries and listings for each endpoint.

15.2. Analysis Populations

15.2.1. Full Analysis Set

The full analysis set (FAS) will consist of all participants randomized at baseline who receive at least one dose of treatment.

This is the principal data set for the formal evaluation of the primary and secondary efficacy endpoints at Week 4. Participants will be evaluated based on the treatment group to which they were randomized.

15.2.2. Safety Analysis Set

The safety analysis set will include all participants who were documented to have taken at least 1 dose of study treatment. Safety evaluations will consider participants according to the actual treatment they received.

15.3. Planned Analysis

15.3.1. Study Population Analysis

Demographics and baseline disease characteristics and medical history will be summarized descriptively by treatment group. Participant disposition will be summarized, including the reasons for discontinuation. The number of participants in each analysis population will be displayed and an accounting of exclusions from each study population will be provided.

15.3.2. Efficacy Analysis

For the primary efficacy endpoint, the change from baseline in histologic eosinophil count in each treatment group will be summarized as the mean, standard deviation, median, minimum, and maximum, appropriately presented as defined by the treatment group of interest.

As a secondary endpoint of interest, number and the proportion of participants with a histologic eosinophil count of < 15 eos/hpf will also be summarized for each treatment group. The secondary endpoint, DSQ change from baseline, will be summarized in the same manner as the primary endpoint.

For the exploratory EoE-EREFS and EoEHSS endpoints, a categorical summary of the scores obtained will be highlighted along with the mean, standard deviation, median, minimum, and maximum change from baseline. The PGI-S and PGI-C will be summarized categorically and as summarized continuous data. Post-baseline evaluation for each PRO measure will be undertaken

as described in the Schedule of Events (Table 3). The EREFS data will be summarized as the frequency and percentage distribution of observed scores within each EREFS domain.

15.3.3. Safety Analysis

All on-treatment safety data will be assessed descriptively for AEs, SAEs, clinical laboratory measurements, viral serology, vital signs, physical examinations, and ECGs. No formal statistical analysis will be performed on safety outcomes; inferences, if any, will be derived through clinical review and interpretation.

All TEAEs will be summarized overall and for each body system and preferred term by treatment group, relationship to investigational product, and severity. For tabulations by severity, only a participant's most severe event within the category (eg, overall, body system, or preferred term) will be counted. Adverse events will be dichotomized into "related" (definitely, probably, and possibly) and "unrelated" (unlikely and not related). "Treatment-emergent" will be defined as starting or worsening after the first dose of investigational product. If the start date is missing, the event is assumed to be treatment emergent. All SAEs will be tabulated.

Vital signs, including blood pressure, heart rate, body temperature, respiration rate, and weight will be summarized by treatment group at baseline and at each scheduled visit.

Clinical safety laboratory data will be summarized descriptively by treatment group at baseline and at subsequent scheduled visits. Summaries of safety laboratory parameters will include the first measurement of each scheduled assessment but repeat assessments done at the same study time point will not be included in summary calculations. Laboratory data will also be listed by treatment, participant, and visit. Listings will include scheduled, unscheduled, and repeat evaluations. A listing of markedly abnormal values, as defined in the SAP, will additionally be generated.

Abnormal physical examination findings that suggest a clinically significant worsening from baseline will be reported as AEs and analyzed as such. Clinically significant findings noted prior to start of investigational product treatment will be recorded as medical history and analyzed as such.

Concomitant medications will be tabulated by preferred term, and treatment group. For the purposes of data collection, a medication's usage will be considered concomitant if it was started within 14 days prior to informed consent and continued after administration of the investigational product. If the start date is missing, it will be assumed that the medication was used concomitantly.

15.3.4. Pharmacokinetic and Immunogenicity Analysis

Concentrations of IRL201104 will be listed by participant and summarized by treatment group and time point from predose to Week 8 based on the safety set.

Antidrug antibody status (negative or positive based on whether results are lower or higher, respectively, than the OD cut-off value) will be listed and summarized by treatment group and time point from predose to Week 8based on the safety set.

Details will be provided in the SAP.

15.4. Determination of Sample Size

The chosen sample size for this study reflects reasonable clinical expectations of the number of participants who could be evaluated in this observational investigation for the purpose of obtaining preliminary estimates of histological response to treatment. It also provides for some level of accounting of the variability of estimates and the repeatability of results among a small group of participants treated in each proposed arm.

16. DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

16.1. Study Monitoring

Before an investigational study center can enter a participant into the study, a representative of the sponsor will assess the investigational study center to:

- Determine the adequacy of the facilities
- Discuss with the investigator(s) and other personnel their responsibilities with regard to protocol adherence, and the responsibilities of the sponsor or its representatives. This will be documented in a Clinical Study Agreement between the sponsor and the investigator.

During the study, a monitor from the sponsor or representative will have regular contacts with the investigational study center, for the following:

- Provide information and support to the investigator(s).
- Confirm that facilities remain acceptable.
- Confirm that the investigational team is adhering to the protocol, that data are being accurately recorded in the eCRFs, and that investigational product accountability checks are being performed.
- Perform source data verification. This includes a comparison of the data in the eCRFs with the participant's medical records at the hospital or practice, and other records relevant to the study. This will require direct access to all original records or certified copies for each participant (eg, clinic charts).
- Record and report any protocol deviations not previously sent to the sponsor.
- Confirm AEs and SAEs have been properly documented on eCRFs and confirm any SAEs have been forwarded to the sponsor and those SAEs that met criteria for reporting have been forwarded to the IRB.

The monitor will be available between visits if the investigator(s) or other staff needs information or advice.

16.2. Audits and Inspections

Authorized representatives of the sponsor, a regulatory authority, IEC, or an IRB may visit the study center to perform audits or inspections, including source data verification. The purpose of a sponsor audit or inspection is to systematically and independently examine all study-related activities and documents to determine whether these activities were conducted, and data were

recorded, analyzed, and accurately reported according to the protocol, GCP guidelines of the International Council for Harmonisation (ICH), and any applicable regulatory requirements. The investigator should contact the sponsor immediately if contacted by a regulatory agency about an inspection.

16.3. Institutional Review Board

The Principal Investigator must obtain IRB approval for the investigation. Initial IRB approval, and all materials approved by the IRB for this study including the ICF and recruitment materials must be maintained by the investigator and made available for inspection.

17. QUALITY CONTROL AND QUALITY ASSURANCE

To ensure compliance with GCP and all applicable regulatory requirements, the sponsor may conduct a quality assurance audit. Please see Section 16.2 for more details regarding the audit process.

18. ETHICS

18.1. Ethics Review

The final study protocol, including the final version of the ICF, must be approved or given a favorable opinion in writing by an IRB or IEC as appropriate. The investigator must submit written approval to the sponsor before he or she can enroll any participant into the study.

The Principal Investigator is responsible for informing the IRB or IEC of any amendment to the protocol in accordance with local requirements. In addition, the IRB or IEC must approve all advertising used to recruit participants for the study. The protocol must be re-approved by the IRB or IEC upon receipt of amendments and annually, as local regulations require.

The Principal Investigator is also responsible for providing the IRB with reports of any reportable serious adverse drug reactions from any other study conducted with the investigational product. To the sponsor will provide this information to the Principal Investigator.

Progress reports and notifications of serious adverse drug reactions will be provided to the IRB or IEC according to local regulations and guidelines.

18.2. Ethical Conduct of the Study

The study will be performed in accordance with ethical principles that have their origin in the Declaration of Helsinki and are consistent with the ICH/GCP, applicable regulatory requirements and the sponsor's policy on Bioethics.

18.3. Written Informed Consent

The Principal Investigator at each center will ensure that the participant is given full and adequate oral and written information about the nature, purpose, possible risk and benefit of the study. Participants must also be notified that they are free to discontinue from the study at any

time. The participant should be given the opportunity to ask questions and allowed time to consider the information provided.

The participant's signed and dated informed consent must be obtained before conducting any study procedures.

The Principal Investigator(s) must maintain the original, signed ICF. A copy of the signed ICF must be given to the participant.

19. DATA HANDLING AND RECORDKEEPING

19.1. Inspection of Records

The sponsor will be allowed to conduct study center visits to the investigational facilities for the purpose of monitoring any aspect of the study. The investigator agrees to allow the monitor to inspect the drug storage area, study treatment stocks, drug accountability records, participant charts and study source documents, and other records relative to study conduct.

19.2. Retention of Records

The Principal Investigator must maintain all documentation relating to the study for a period of 2 years after the last marketing application approval, or if not approved 2 years following the discontinuance of the test article for investigation. If it becomes necessary for the sponsor or the Regulatory Authority to review any documentation relating to the study, the investigator must permit access to such records.

20. PUBLICATION POLICY

If the data merit, the investigator and the sponsor will discuss the preparation of a manuscript for publication in a peer-reviewed professional journal or an abstract for presentation, oral or written, to a learned society or symposium. Either party may undertake the task, but both must agree to the strategy before the work is started. Each party will allow the other 30 days to comment before any results are submitted for publication or presentation. Authorship should reflect work done by the investigators and personnel of the sponsor, in accordance with generally recognized principles of scientific collaboration.

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22. APPENDIX

The following scales/assessments will be utilized in this study:

Full Title of Scale/Assessment	Completed By
DSQ	Participant
EoE-EREFS	Endoscopist / Central reader
EoEHSS	Central reader
PGI-S	Participant
PGI-C	Participant

DSQ = Dysphagia Symptom Questionnaire; EREFS = Eosinophilic Esophagitis-Endoscopic Reference Score; EoEHSS = Eosinophilic Esophagitis-Histology Scoring System; PGI-C = Patient Global Impression of Change; PGI-S = Patient Global Impression of Severity

A separate master file containing each scale/assessment listed above will be provided to the study centers. Updates to scales/assessments during the study (if applicable) will be documented in the table above, and a new master file containing the revised scale/assessment will be provided to the study centers.