CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

217759Orig1s000

MULTI-DISCIPLINE REVIEW

Summary Review
Office Director
Cross Discipline Team Leader Review
Clinical Review
Non-Clinical Review
Statistical Review
Clinical Pharmacology Review

NDA Multi-Disciplinary Review and Evaluation

Application Type	NDA
Application Number(s)	217759
Priority or Standard	Priority
Submit Date(s)	July 29, 2022
Received Date(s)	July 29, 2022
PDUFA Goal Date	March 29, 2023
Division/Office	Division of Pulmonology, Allergy, and Critical Care (DPACC), Office of Immunology and Inflammation (OII)
Review Completion Date	March 22, 2023
Established/Proper Name	Leniolisib phosphate
(Proposed) Trade Name	Joenja
Pharmacologic Class	Phosphoinositide 3-kinase inhibitor
Code name	CDZ173
Applicant	Pharming Technologies B.V.
Doseage form	Oral tablet
Applicant proposed Dosing Regimen	70 mg twice daily
Applicant Proposed Indication(s)/Population(s)	Treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in patients age ≥ 12 years
Applicant Proposed SNOMED CT Indication Disease Term for each Proposed Indication	Activated PI3K-delta syndrome
Recommendation on Regulatory Action	Approval
Recommended Indication(s)/Population(s) (if applicable)	Treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adult and pediatric patients 12 years of age and older 45 kg or greater
Recommended SNOMED CT Indication Disease Term for each Indication (if applicable)	Activated PI3K-delta syndrome
Recommended Dosing Regimen	70 mg administered orally twice daily approximately 12 hours apart, with or without food

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Abbreviations: DIIP, Division of Inflammation and Immune Pharmacology; DPACC, Division of Pulmonology, Allergy, and Critical Care; OB, Office of Biostatistics; PBPK, physiologically based pharmacokinetic modelling.

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Abbreviations: DEPI, Division of Epidemiology; DMEPA, Division of Medication Error Prevention and Analysis; DRISK=Division of Risk Management; OPDP, Office of Prescription Drug Promotion; OPQ, Office of Pharmaceutical Quality; OSE, Office of Surveillance and Epidemiology; OSI=Office of Scientific Investigations.

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Glossary

AC	advisory committee
ADME	absorption, distribution, metabolism, excretion
AE	adverse event
AR	adverse reaction
BLA	biologics license application
BPCA	beats per minute
BPM	Best Pharmaceuticals for Children Act
BRF	Benefit Risk Framework
CBER	Center for Biologics Evaluation and Research
CDER	Center for Drug Evaluation and Research
CDRH	Center for Devices and Radiological Health
CDTL	Cross-Discipline Team Leader
CFR	Code of Federal Regulations
CMC	chemistry, manufacturing, and controls
COSTART	Coding Symbols for Thesaurus of Adverse Reaction Terms
CRF	case report form
CRO	contract research organization
CRT	clinical review template
CSR	clinical study report
CSS	Controlled Substance Staff
DHOT	Division of Hematology Oncology Toxicology
DMC	data monitoring committee
ECG	electrocardiogram
eCTD	electronic common technical document
ETASU	elements to assure safe use
FDA	Food and Drug Administration
FDAAA	Food and Drug Administration Amendments Act of 2007
FDASIA	Food and Drug Administration Safety and Innovation Act
GCP	good clinical practice
GRMP	good review management practice
ICH	International Conference on Harmonisation
IND	Investigational New Drug
IRT	Immunoglobulin Replacement Therapy
ISE	integrated summary of effectiveness
ISS	integrated summary of safety
ITT	intent to treat
MedDRA	Medical Dictionary for Regulatory Activities

mITT	modified intent to treat
NCI-CTCAE	National Cancer Institute-Common Terminology Criteria for Adverse Event
NDA	new drug application
NME	new molecular entity
OCS	Office of Computational Science
OLE	Open Label Extension
OPQ	Office of Pharmaceutical Quality
OSE	Office of Surveillance and Epidemiology
OSI	Office of Scientific Investigation
PBRER	Periodic Benefit-Risk Evaluation Report
PD	pharmacodynamics
PI	prescribing information
PK	pharmacokinetics
PMC	postmarketing commitment
PMR	postmarketing requirement
PP	per protocol
PPI	patient package insert (also known as Patient Information)
PREA	Pediatric Research Equity Act
PRO	patient reported outcome
PSUR	Periodic Safety Update report
REMS	risk evaluation and mitigation strategy
SAE	serious adverse event
SAP	statistical analysis plan
SGE	special government employee
SOC	standard of care
TEAE	treatment emergent adverse event

1. Executive Summary

1.1. Product Introduction

Pharming Technologies B.V. submitted new drug application (NDA) 217759 to support the approval of leniolisib for the "treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adult and pediatric patients 12 years of age and older." APDS is caused by gain-of-function mutations in the *PIK3CD* gene and loss-of-function mutations in the *PIK3R1* gene, encoding the p110δ catalytic subunit and the p85a regulatory subunits of phosphoinositide 3-kinase delta (PI3Kδ), respectively; PI3Kδ is expressed primarily in hematopoietic cells. These mutations result in activation of PI3Kδ with resulting increased phosphatidylinositol-3,4,5-trisphosphate (PIP3) production, hyperactivity of the downstream mTOR/AKT pathway, and dysregulation of T cells and B cells. Leniolisib (CDZ173) is a small molecule inhibitor of the p110δ subunit of PI3Kδ that reduces signaling through this pathway. It is a new molecular entity (NME) that is not approved for any indication in the U.S. or any other country. The proposed dosing regimen is 70 mg orally twice daily.

1.2. Conclusions on the Substantial Evidence of Effectiveness

The recommended regulatory action for this NDA is **approval** of leniolisib for the treatment of APDS in adult and pediatric patients 12 years of age and older. This will be the first product approved for the treatment of APDS.

Substantial evidence of effectiveness for leniolisib in subjects with APDS was established with one adequate and well-controlled trial, with confirmatory evidence. The 12-week, pivotal phase 3 trial (Study CCDZ173X2201 Part 2) conducted in 31 adults and adolescents 12 years of age and older demonstrated a clinically meaningful and statistically significant improvement in the percentage of naïve B cells and lymphadenopathy for subjects randomized to leniolisib compared to those treated with placebo. The low proportions of naïve B cells are characteristic in APDS and a direct result of PI3K pathway dysregulation that blocks B cell development at the transitional B cell stage. Dysregulation in the PI3K pathway manifests clinically as a primary immunodeficiency with autoimmune and lymphoproliferative features. Correction of the abnormal immunophenotype associated with APDS, specifically an increase in the percentage of naïve B cells out of total B cells, is expected to lead to normalization of immune function and improvement in clinical sequelae such as fewer infections, autoimmune manifestations, and lymphoproliferative disease.

Benign lymphoproliferation caused by PI3K pathway dysregulation typically manifests as chronic or reactive lymphadenopathy, splenomegaly, hepatomegaly, or gut infiltration and is another hallmark of APDS. Lymphoproliferative disease does not spontaneously regress; consequently, APDS patients are at increased risk of mechanical obstruction and malignancy

such as lymphoma over time as the disease progresses. As such, decreased lymphadenopathy, as measured by the change from baseline in the log10 transformed sum of product of diameters (SPD) in the index lesions selected on MRI/CT imaging, is an endpoint that reflects improvement in the underlying immune dysregulation and is expected to predict clinical benefit.

Confirmatory evidence providing strong mechanistic support includes the well-established etiology of the disease, the mechanism of action of leniolisib, and pharmacodynamic biomarker data showing reductions in pAKT, as well as normalization of other immunologic parameters.

1.3. Benefit-Risk Assessment

Benefit-Risk Summary and Assessment

Activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) is a primary immunodeficiency disease caused by autosomal dominant gain-of-function mutations in the catalytic p110δ (*PIK3CD*) or loss-of-function mutations in the regulatory p85α (*PIK3R1*) subunits of PI3Kδ, leading to APDS1 and APDS2, respectively, both of which result in hyperactive PI3Kδ signaling. Normal lymphocyte development is dependent upon a balanced PI3Kδ pathway. In APDS, however, PI3Kδ is constitutively active, resulting in dysregulated B and T cell development with features of both underactive (immunodeficiency) and overactive or inappropriate (autoimmunity/lymphoproliferation) immune responses (Preite et al. 2018; Tangye et al. 2019). Inhibition of the p110δ subunit of PI3Kδ prevents the intracellular conversion of phosphatidylinositol 4,5-bisphosphate (PIP2) to phosphatidylinositol 3,4,5-trisphosphate (PIP3) downstream of receptor tyrosine kinases that is important for immune function and is thought to be beneficial for the treatment of hematological malignancies or gain-of-function mutations in the PI3K pathway that lead to hyperactive lymphoproliferation, such as APDS.

The prevalence of PI3Kδ-related germline mutations is unknown, but APDS prevalence is estimated to be between 1-2 cases per million, globally; more subjects may be identified as genetic testing becomes more widely available (Michalovich and Nejentsev 2018; Nunes-Santos et al. 2019).

Genetic defects in PI3Kδ affect B-cell differentiation and lead to intrinsic B-cell dysregulation, hypogammaglobulinemia, and recurrent sinopulmonary bacterial infections. Clinical manifestations of APDS typically progress from recurrent infections and lymphoproliferation in early childhood, to autoimmunity in mid-childhood, and malignancy in late childhood-adulthood. Benign lymphoproliferation, driven by the proliferation of abnormal B and T cell subsets, is a clinical manifestation of the immune dysregulation component of APDS and presents as lymphoid hyperplasia, lymphadenopathy, splenomegaly, or hepatomegaly. Autoimmune phenomena commonly present after the first decade of life (Thouenon et al. 2021). Treating the underlying etiology of this condition early may prevent downstream sequelae of lymphoproliferation (i.e., malignancy, persistent lymphadenopathy) and recurrent infections (i.e. bronchiectasis).

Currently, there are no approved therapies targeted specifically to offset the hyperactive PI3Kδ signaling of APDS. Available treatments are supportive and aimed at disease symptoms and sequelae, such as prophylactic antibiotics, immunoglobulin replacement, immunosuppressants

(e.g., systemic corticosteroids, off-label p110/mTOR inhibitors), chemotherapy/radiation for lymphoma, and splenectomy. Hematopoietic stem cell transplant (HSCT) has also been performed with variable results.

The Applicant has submitted an NDA for leniolisib, a small molecule inhibitor of p110δ, for the treatment of APDS in patients age ≥ 12 years. Efficacy and safety were supported by one adequate and well controlled 12-week, pivotal phase 3 trial (Trial 2201, Part 2), in a total of 31 adults and adolescents. The demonstration of substantial evidence of effectiveness is based on clinically meaningful and statistically significant improvements in co-primary endpoints measuring the percentage of naïve B cells and lymphadenopathy in subjects randomized to leniolisib compared to those treated with placebo, along with confirmatory evidence from mechanistic data.

No safety concerns were identified in this review that would preclude approval. The risks of leniolisib can be adequately addressed through labeling.

Overall, the benefit/risk assessment for leniolisib for the treatment of APDS in adults and adolescents aged 12 or older is favorable. Efficacy and safety findings support approval of leniolisib for the indication of APDS.

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Analysis of Condition	• APDS is an ultra-rare genetic condition with an estimated prevalence of <1000 individuals worldwide. • APDS1 is caused by gain-of-function (GOF) mutations in the *PI3KCD* gene and APDS2 is caused by loss-of-function (LOF) mutations in the *PIK3R1* gene, both of which lead to constitutive PI3Kδ signaling that is responsible for the lymphoproliferation-associated primary combined immunodeficiency syndrome. • Dysregulated activation of PI3Kδ signaling leads to characteristic immunophenotypic abnormalities consistent with disrupted maturation beyond the transitional B cell stage, including elevated transitional B cells and reduced naïve and memory B cells; disruption of B cell development is central to the increased susceptibility to recurrent sinopulmonary infections • Although a heterogenous disorder, typical clinical features of APDS include early-onset, recurrent and severe sinopulmonary infections, as well as persistent viral infections; lymphoproliferative disease leading to mechanical obstructions, hepatosplenomegaly, and/or malignancy; and autoimmunity. Available natural history data suggest complications are common (Coulter et al. 2017; Oh et al. 2021). • Along with significant disease morbidity, APDS patients have decreased life-expectancy (Elkaim et al. 2016; Okano et al. 2019).	APDS is a serious, progressive, ultra-rare genetic disease caused by dysregulated activation of the PI3K pathway that manifests as a primary immunodeficiency with increased infections, along with autoimmunity and lymphoproliferative disease. APDS is associated with significant morbidity and shortened life-expectancy.

Dimension	Evidence and Uncertainties	Conclusions and Reasons
Current Treatment Options	• There are no FDA-approved drugs for the treatment of APDS. • Current therapies target symptoms and sequelae of disease (e.g., antibiotics, immunoglobulin replacement, splenectomy). • Sirolimus, an mTOR inhibitor, has been used off-label for lymphoproliferation with variable results. • HSCT has the potential to be curative, but it is associated with high rates of complications and mortality.	There is an unmet need for the treatment of APDS as there are no approved therapies.
Benefit	• Leniolisib acts directly on the affected PI3K pathway in APDS by inhibiting the p110δ subunit of PI3Kδ. • The efficacy of leniolisib was evaluated in an adequate and well-controlled trial (Trial 2201, Part 2) in 31 subjects (19 adults and 12 adolescents) with APDS1 and APDS2 (21 leniolisib, 10 placebo). The changes in percent of naïve B cells and index lymph node size, the co-primary efficacy measures, were compared between subjects after 12 weeks of treatment. • Leniolisib demonstrated statistically significant improvements over placebo for both co-primary endpoints. The LS mean treatment difference estimate was -0.25 [95% CI: -0.38, -0.12; p-value = 0.0006] in change from baseline in the log10 transformed sum of product of diameters (SPD) in the index lesions at Day 85. The LS mean treatment difference estimate was 37.30 [95% CI: 24.06, 50.54; p-value = 0.0002] in change from baseline at Day 85 in percentage of naïve B cells out of total B cells. • The treatment effect was preserved across sensitivity and subgroup analyses, including by age.	The co-primary endpoints for percentage of naïve B cells and index lesion size showed a statistically significant advantage for subjects treated with leniolisib over placebo. Correction of the underlying immune abnormality, which is characterized by a low percentage of naïve B cells, is expected to result in normalization of immune function and improvement in clinical sequelae (e.g., fewer infections, decreased lymphoproliferative disease). The pathophysiology of disease is the same across phenotypes and age groups. Mechanistically, leniolisib is anticipated to have

Dimension	Evidence and Uncertainties	Conclusions and Reasons
	• Other pharmacodynamic endpoints (pAKT, spleen size/volume, other immunologic parameters) provided numerically favorable improvements with leniolisib treatment compared to placebo. • Data beyond 12 weeks of treatment is uncontrolled.	a similar effect in patients regardless of the disease phenotype or age.
Risk and Risk Management	• The safety of leniolisib was based on 38 adult and pediatric patients 12 years of age and older with APDS in Trial 2201, Part 1 and 2, as well as the OLE. • There were two deaths reported in the clinical development program, one in the placebo group and one in the leniolisib treatment group, both of which appeared related to the underlying disease. • Nonfatal SAEs were similar between treatment groups in Trial 2201 Part 2 with 3 (14%) subjects experiencing an SAE in the leniolisib group versus 2 (20%) in the placebo group. • There were no dropouts or discontinuations due to adverse events in Trial 2201, Part 1 or 2, and few in the OLE. • The overall rate of severe TEAEs was similar between treatment and placebo group. Three individuals in both the leniolisib and placebo treatment groups experienced an adverse event of grade 3 or higher • Neutrophil counts were lower in the leniolisib treatment group compared to placebo but were not associated with adverse events or infections. • Although limited by size, subgroup analyses revealed no imbalances in the safety profile.	The safety database was adequate for the safety assessment of leniolisib for the proposed indication, patient population, dosage regimen, and duration. Potential adverse drug reactions may be mitigated through product labeling and routine pharmacovigilance. No REMS is needed.

1.4. Patient Experience Data

Patient Experience Data Relevant to this Application (check all that apply)

☐	The patient experience data that were submitted as part of the application include:			Section of review where discussed, if applicable
	☑	Clinical outcome assessment (COA) data, such as		
		☑	Patient reported outcome (PRO)	8.1.1, 8.1.5
		☐	Observer reported outcome (ObsRO)	
		☑	Clinician reported outcome (ClinRO)	8.1.1, 8.1.5
		☐	Performance outcome (PerfO)	
	☐	Qualitative studies (e.g., individual patient/caregiver interviews, focus group interviews, expert interviews, Delphi Panel, etc.)		
	☐	Patient-focused drug development or other stakeholder meeting summary reports		
	☐	Observational survey studies designed to capture patient experience data		
	☑	Natural history studies		2.1
	☐	Patient preference studies (e.g., submitted studies or scientific publications)		
	☐	Other: (Please specify): patient-generated narratives		
☑	Patient experience data that were not submitted in the application, but were considered in this review:			
	☐	Input informed from participation in meetings with patient stakeholders		
	☐	Patient-focused drug development or other stakeholder meeting summary reports		
	☐	Observational survey studies designed to capture patient experience data		
	☑	Other: (Please specify): natural history studies from literature		N/A
☐	Patient experience data was not submitted as part of this application.			

2. Therapeutic Context

2.1. Analysis of Condition

Activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) is a primary immunodeficiency caused by autosomal dominant gain of function mutations in the *PIK3CD* gene encoding the p110δ catalytic subunit or loss-of-function mutations in the *PIK3R1* gene encoding the regulatory p85α (*PIK3R1*) subunit leading to APDS1 and APDS2, respectively. APDS is also known as p110δ-activating mutation causing senescent T cells, lymphadenopathy, and immunodeficiency (PASLI). The subunit p110δ is expressed primarily in hematopoietic cells whereas p85α expression is thought to be ubiquitous. Activation of PI3Kδ by antigen receptors, co-receptors, growth receptors and cytokine receptors leads to phosphorylation of phosphatidylinositol 3,4-bisphosphate (PIP2) to generate phosphatidylinositol 3,4,5-triphosphate (PIP3) resulting in cell activation, growth, metabolism and inhibition of apoptosis via the AKT/mTOR/S6K signaling pathways (Redenbaugh and Coulter 2021).

APDS1 and APDS2 are classified predominantly as antibody deficiencies as PI3Kδ signaling pathways are integral to immunoglobulin class switching. However, there is substantial phenotypic overlap with other combined immunodeficiencies as made evident by the additional susceptibility to severe viral infections seen in many individuals suffering from APDS (Jamee et al. 2020). Individuals with mutations in PI3Kδ have been shown to have intrinsic B-cell defects that lead to reduced differentiation beyond the transitional stage, increased AKT phosphorylation and increased apoptosis of which the resultant B-cell phenotype contributes to the clinical phenotype. Emerging analyses of the B cell phenotype pathognomonic to those suffering from APDS show they have decreased naïve B cell numbers with an expanded double negative B cell population; a B cell population that lacks expression of immunoglobulin D and CD27 (a memory marker) that prematurely accumulates in autoimmune diseases, infectious diseases, and immunodeficiencies. Additionally, the B cells in patients with APDS exhibit impaired class switch recombination, but normal plasmablast differentiation that manifests as predominantly IgM-secreting cells and detected as the typically elevated IgM levels in patients (Wang et al. 2022). This constellation of immunophenotypic findings is characteristic for APDS and is thought to directly reflect the underlying immune dysregulation that results in the clinical features.

Although the underlying epidemiology is evolving given the recent discovery of the mutation that leads to APDS, in several families, APDS mutations were shown to appear de novo among children and an analysis of families with the E1021K mutation showed no founder effect (Angulo et al. 2013). These findings indicate that APDS mutations appear recurrently in human populations.

Clinical manifestations of APDS typically progress from recurrent infections and lymphoproliferation in early childhood, to autoimmunity in mid-childhood, and malignancy in

late childhood-adulthood. While clinical manifestations are heterogenous, recurrent respiratory infections and lymphoproliferation occur in the overwhelming majority of those diagnosed with the disease. Patients with APDS suffer from sinopulmonary tract infections and have a concomitant predisposition to herpesviruses and bacterial infections.

Persistent benign lymphoproliferation, a hallmark of the disease, may present as lymphoid hyperplasia, lymphadenopathy, splenomegaly, or hepatomegaly, and may potentially involve the airways and gastrointestinal tract, with increased susceptibility to malignancy. The molecular defect responsible for the constitutive activity of PI3K and subsequent immune dysregulation with decreased naïve B cell and expanded transitional B cell populations leads to the lymphoproliferation and increased risk for the development of autoimmunity. A substantial proportion of patients also experience autoimmune hematologic disease including hemolytic anemia, pancytopenia, and thrombocytopenia. Available natural history data suggest complications such as bronchiectasis occurs in 50-60% of patients suffering from APDS, persistent viremia in 38-49%, splenomegaly in 55%, hepatomegaly in 28-45%, autoimmune phenomena in 42%, and malignant lymphoma in 13% (Coulter et al. 2017; Oh et al. 2021). Despite available treatments, survival for individuals with APDS appears to be shortened from the average lifespan (Elkaim et al. 2016; Okano et al. 2019). Preliminary data show the survival rates to 20, 30, and 40 years of age are approximately 87%, 74%, and 68%, respectively (Hanson and Bonnen 2022). Patients with APDS have a high incidence of lymphoma compared to the general population with cases starting in early childhood. The most common malignancy is B cell lymphoma and is the most common cause of death, followed by complications from HSCT. Early recognition and diagnosis are essential to prevent long term sequelae, such as bronchiectasis and malignancy, which are often fatal.

2.2. Analysis of Current Treatment Options

There are currently no approved pharmacological treatments for the disease. As such, current management focuses on supportive care and treatment of disease sequelae such as antibiotics for prevention and treatment of infections and immunoglobulin replacement therapy. Immunosuppressive agents including systemic corticosteroids and rituximab have been used to manage autoimmune phenomena and benign lymphoproliferation. Rapamycin and sirolimus have also been used off-label to inhibit mTOR located downstream of PIK3. In case reports, sirolimus treatment has been associated with a decrease in hepatosplenomegaly, restoration of T cell proliferation and decrease in non-neoplastic lymphoproliferation, but has been less effective for the treatment of autoimmune phenomena (Lucas et al. 2014; Maccari et al. 2018). Hematopoietic stem cell transplantation (HSCT) has been performed in some patients with severe infection or lymphoma; however, clinical outcomes have been variable as engraftment failure and adverse complications, including increased mortality are common.

3. Regulatory Background

3.1. U.S. Regulatory Actions and Marketing History

Leniolisib is not approved or marketed in the United States or any foreign country for any indication.

3.2. Summary of Presubmission/Submission Regulatory Activity

Leniolisib (CDZ173) was developed under investigational new drug (IND) application 124045, which was opened on January 20, 2015. Leniolisib was granted orphan therapy designation for APDS on January 30, 2018, and rare pediatric disease designation on August 26, 2020. Relevant interactions between the Agency and Applicant to discuss the clinical development program are summarized below.

Type B Pre-IND Meeting: July 1, 2014:

- Recommended further dose exploration and justification
- Recommended a randomized, placebo-controlled trial design
- Encouraged the collection of relevant biomarkers and data on functional impairment
- The Division noted that a reduction in lymph node size alone is unlikely to be sufficient to support approval in the absence of supportive efficacy data for other clinically meaningful endpoints
- Recommended efficacy endpoints that capture symptomatic benefit
- Agreed mechanistic data on the effects of leniolisib may be used as supportive of clinical efficacy

Type C Guidance Meeting: June 8, 2017

- Need a randomized placebo-controlled study to demonstrate efficacy
- Endpoints do not directly reflect a clinically meaningful outcome and the totality of data may take precedence over a statistical “win” on the co-primary endpoints
- Recommended inclusion of physician or patient global assessment scores as primary objective
- Recommended assessment of the correction of immune defects and a decrease in serious infections

Type C Guidance Meeting: November 17, 2020

- Agreement on Regulatory Starting Material with justification in NDA
- Advice provided on the proposed stability package
- Agreement on post-approval submission of 2-year rat carcinogenicity data and pre-post-natal development study

Type B Pre-NDA Meeting: April, 29, 2022

- The leniolisib APDS clinical and nonclinical programs appear adequate to characterize the safety and efficacy of leniolisib
- Additional analyses and assessments were requested (e.g., analysis of supportive literature and assessment of concomitant therapies)
- Additional background information was requested (e.g., relevant contextual information for endpoint analysis and patient reported outcome instruments and predicted immunologic and clinical phenotype information for each subject).
- Clarifications on clinical pharmacology data were also requested

4. Significant Issues From Other Review Disciplines Pertinent to Clinical Conclusions on Efficacy and Safety

4.1. Office of Scientific Investigations (OSI)

The OSI conducted clinical inspections of 2 study sites from Trial 2201, Part 2. Sites were selected for GCP inspections using a risk-based approach that considered numbers of enrolled subjects, treatment effect, and prior inspectional history. Source documents were reviewed for all randomized subjects. Records reviewed included, but were not limited to, protocol versions, IRB submissions, for FDA 1572s, financial disclosures, eligibility records, informed consent forms, source data evaluation, adverse event reports, laboratory reports, clinical source data, concomitant medications, questionnaires, paper case report forms, investigational product accountability, staff training, and Applicant/monitor correspondence, and co-primary efficacy endpoint source records. At site #1001, no discrepancies were noted. There was no evidence of underreporting of adverse events. However, for 11 of the 16 randomized subjects, hsCRP lab results were missing for Baseline (Day of Randomization) and Visit 101 (Day 1 Dosing Day). These missing values did not impact the review, but a corrective and preventative action (CAPA) plan to institute a check list to ensure all tests results are completed per protocol was provided by the clinical investigator. At site #3001, 5 subjects were screened and 3 were randomized. All 3 randomized subjects completed Trial 2201, Part 2. No discrepancies were noted. There was no evidence of underreporting of adverse events or of unreported protocol deviations.

For additional information, refer to the Clinical Inspection Summary dated January 11, 2023.

4.2. Product Quality

The recommended regulatory action from the Office of Pharmaceutical Quality (OPQ) is Approval. Assessment of the drug substance, drug product, labeling, manufacturing, and biopharmaceutics were all considered adequate.

Leniolisib tablets (70 mg) are for oral administration and immediate release. The drug is a free base that has low solubility and high permeability (BCS Class II). The drug substance has one chiral center (b) (4) All regulatory starting materials satisfy the ICH Q11 guidelines, and the Applicant has performed adequate evaluations of the potential impurities in the drug substance based on the manufacturing process. The drug substance container closure system components are typical of what the Agency accepts, including the (b) (4) as the primary packaging material.

The drug product is formulated with compendial grade excipients and is manufactured (b) (4)

(b) (4). It contains the active ingredient in its phosphate salt form, leniolisib phosphate, which is a new molecular entity. The Applicant provided up to 12 months of long-term stability studies and 6 months of accelerated stability studies for three registration batches with supporting stability results of development and scale-up batches. There are no trends observed for any of the stability parameters under both accelerated and long-term conditions for studied periods and the Applicant committed to continue the stability studies for primary stability batches. The proposed shelf-life of 24 months for the leniolisib film-coated tablets with the container closure system of HDPE bottles with an aluminum induction seal and (b) (4) screw cap closures with a storage condition of do not store above 25°C, do not refrigerate is acceptable.

For additional information, refer to the Integrated Quality Assessment dated December 14, 2022.

4.3. Clinical Microbiology

Not applicable.

4.4. Devices and Companion Diagnostic Issues

Not applicable.

5. Nonclinical Pharmacology/Toxicology

5.1. Executive Summary

The Applicant has conducted a comprehensive program of pharmacology, pharmacokinetic (PK) and toxicology studies to support clinical development and the NDA submission of leniolisib (CDZ173) for APDS. All pivotal toxicity studies and juvenile toxicology studies have been submitted and reviewed previously under IND 124045 and IND (b) (4) The pharmacological, pharmacokinetic, and impurities studies have been submitted and reviewed previously under NDA 217759. The key findings from the nonclinical studies are cited and summarized in this review to support the approval of NDA 217759.

Pharmacological studies demonstrated that leniolisib binds to the active site of PI3Kδ. In cell-free isolated enzyme assays, leniolisib was selective for PI3Kδ over PI3Kα (28-fold), PI3Kβ (43-fold), and PI3Kγ (257-fold), as well as the broader kinome. In cell-based assays, leniolisib reduced pAKT pathway activity and inhibited proliferation and activation of B and T cell subsets.

The Applicant has conducted subchronic and chronic toxicology studies with leniolisib in both rats and cynomolgus monkeys. Leniolisib-related toxicities included immune suppression (rats & monkeys), skin lesions (rats & monkeys), germ cell depletion (rats), GI tract inflammation (monkeys), and QTc prolongation (monkeys). These toxicities were generally pharmacodynamic-related and/or clinically monitorable and not considered dose-limiting. These findings were observed at plasma exposures approximately 3 times or equivalent to the human exposure at the maximum recommended human dose (MRHD) in rats and monkeys, respectively. In animal reproduction studies, oral administration of leniolisib to pregnant rats and rabbits during the period of organogenesis at exposures approximately 2-6 times the MRHD on an AUC basis, produced embryofetal toxicity including malformations. Leniolisib was not mutagenic or clastogenic in in vitro and in vivo assays. The pre- and postnatal developmental study in rats as well as the 6-month Tg-RasH2 mouse and 2-year rat carcinogenicity studies will be completed as post-marketing requirements. To conclude, NDA 217759 is recommended for approval from the nonclinical perspective.

5.2. Referenced NDAs, BLAs, DMFs

None.

5.3. Pharmacology

Primary Pharmacology

In biochemical assays, leniolisib inhibited human recombinant enzymes PI3Kα and PI3Kβ with IC_{50}s of 0.28 and 0.48 µM, respectively. Leniolisib selectively inhibits the lipid kinase PI3Kδ at least 28-fold over other Class I PI3K isoforms.

In human cell-based assays, leniolisib inhibited PI3Kδ, PI3Kα, and PI3Kβ with IC_{50}s of 0.056, 1.62, and 2.37 µM, respectively. Leniolisib inhibited PI3Kγ with IC_{50}s of 7.76 or >7.42 µM, respectively. In the Rat-1 cell line expressing mutant PI3Kδ (i.e., N334K, C416R, E525K, or E1021K) with elevated levels of pAkt, leniolisib inhibited enhanced activity of the mutant enzymes with IC_{50} values between 0.205 µM for the wt and 0.05 µM for the C416R mutant. In whole blood assays from rats, monkeys, and humans and mouse splenocytes, leniolisib inhibited both activation (i.e., phosphorylation of Akt, expression of CD69 and CD86) and proliferation of B cells at nanomolar to sub-micromolar concentration range. In mouse splenocytes and human peripheral blood mononuclear cells (PBMCs), leniolisib inhibited T cell proliferation and differentiation in the sub-micromolar range. In human PBMC and whole blood and mouse bone marrow-derived mast cells, leniolisib inhibited the oxidative burst of human neutrophils and monocytes and activation of mouse mast cells in the sub-micromolar range.

Currently, there is no validated APDS disease model in animals. The Sponsor explored antibody response to sheep red blood cells (SRBC) in rats and ozone-induced neutrophil infiltration in mouse bronchoalveolar lavage (BAL). In a 5-day antibody response to SRBC model, leniolisib inhibited T-cell dependent specific antibody response (i.e., anti-SRBC producing plaque-forming cells) in a dose-dependent manner. In a C57BL/6 mouse model of ozone-induced acute lung inflammation, leniolisib inhibited the ozone-induced increase in BAL neutrophil and macrophage numbers in a dose-dependent manner.

Secondary Pharmacology

Leniolisib did not show inhibitory activity on a panel of 4 other enzymes in the PI3K family (Vps34, mTOR, DNA-PK, PI4K). In KINOMEScan against a total of 442 serine/threonine, tyrosine and lipid kinases and a Novartis Protease panel (a panel of proteases and 47 serine/threonine and tyrosine kinases), 10 µM leniolisib did not inhibit any kinases or proteases other than the expected Class I PI3K isoforms except for RPS6KA5 (76% inhibition). In additional receptor and ion channel screens, 10 µM leniolisib inhibited the G-protein coupled receptor GPR8 (72% inhibition). In the Novartis P5 receptor profile, leniolisib had an IC_{50} of 4.7 µM for the hPDE4D receptor and 7.7 µM for the 5HT2B receptor.

Safety Pharmacology

Leniolisib was found to inhibit hERG channel currents in hERG-transfected HEK cells with an IC_{50} of 11.9 µM. Consistent with this finding, single dose leniolisib (150 mg/kg, oral) administration induced QTcI (QT interval, individual correction) prolongation in two separate monkey telemetry studies. Prolongation was observed between 3 – 14 hours post dose. An additional single dose CNS and respiratory safety pharmacology study in monkeys (n=10) found no effects of 300 mg/kg leniolisib in the functional observational battery (FOB) or on tidal volume, respiratory rate, or minute volume.

5.4. ADME/PK

Leniolisib was rapidly absorbed after oral gavage with a mean T_{max} of 0.25-0.5 hour in mice, 0.25 hour in rats and 2.6 hours in monkeys. In rats, terminal plasma $T_{1/2}$ of total radioactivity was approximately 33.4 hours after oral gavage dosing. The plasma clearance was approximately 15 mL/min/kg in rats and monkeys, while the blood clearance was moderate to high compared to the hepatic blood flow (i.e., 23%-44%). Leniolisib has a low first-pass effect and a moderate to strong binding to plasma proteins (fraction unbound: 12.4% in mice, 8.2% in rats, 3.8% in dogs, 9.9% in monkeys and 5.5% in humans) in a concentration independent manner. The oral bioavailability was 59% in rats and ~100% in monkeys.

In radioactive studies, leniolisib was moderately distributed into tissues (Vss of 1.06 L/kg in rats and 0.657 L/kg in monkeys) with a high skin affinity. After a single oral dose of 10 mg/kg to male rats, the highest exposures were found in the bile, hair follicle, liver, intestinal wall, kidney as well as in melanin-containing structures.

In mouse, rat, dog, monkey, and human hepatocytes, the biotransformation of leniolisib occurred mainly via phase I metabolism (i.e., at the tetrahydropyridopyrimidine and the pyrrolidine parts of the molecule). Characterization and structural elucidation of metabolites in plasma, urine or bile from nonclinical species have demonstrated the similarities in the metabolic profiles between these species and humans. It is noted that no major human metabolites were identified in completed clinical studies.

In rats and monkeys, the major route of excretion of [^{14}C] leniolisib was via the feces (approximately 91% and 51-58% of the radioactive dose by oral gavage).

In in vitro assays, leniolisib inhibited BCRP with an IC_{50} of 18.9 µM. Leniolisib did not inhibit efflux transporter P-gp. Leniolisib inhibited OATPB1 (IC_{50}=3.0 µM), OATPB3 (IC_{50}=13.4 µM), OAT3 (IC_{50}=15.2 µM), and OCT2 (IC_{50}=3.4 µM). Leniolisib did not inhibit OCT1 and OAT1. Leniolisib inhibited the human multidrug and toxin extrusion protein family MATE1 and MATE2K (MATE1 IC_{50} =6.70 µM and MATE2K IC_{50} =0.85 µM).

5.5. Toxicology

5.5.1. General Toxicology

As listed in the following table, general toxicity studies with leniolisib were conducted in rats and monkeys with dosing durations up to 6 months in rodents and 9 months in non-rodents. All of the pivotal studies have been reviewed previously under IND 124045 and IND (b) (4) *(DARRTS Reference ID: 3162851, 3624707, 3666991, 3952011, 4173292, and 5056867)*. Key findings discussed in the previous reviews are summarized below.

Table 1. Leniolisib General Toxicology Studies in Rats and Monkeys

General Toxicology Study	GLP	Report Number	Review Reference ID
Rats			
2-Week oral (gavage) exploratory study in rats Doses: 0, 10, 50 or 300 mg/kg/day, 10/sex/group	Yes	1170360	3162851
13-Week oral (gavage) administration toxicity study in the rat followed by a 4-week treatment-free period Doses: 0, 10, 35, or 90/120[a] mg/kg/day, 10/sex/group	Yes	1270409	3624707, 3666991, 3952011
26-Week oral (gavage) administration toxicity study in the rat followed by a 4-week treatment-free period Doses: 0, 15, 40, or 120 mg/kg/day, 20/sex/group	Yes	1470444	4173292
Monkeys			
2-Week oral gavage administration toxicity study in the cynomolgus monkey including jacket telemetry and a 4-week recovery period Doses: 0, 30, 100, or 300 mg/kg/day; 3/sex/group	Yes	1170359	3162851
13-Week oral gavage administration toxicity study in the cynomolgus monkey with a 4-week recovery phase Doses: 0, 20, 40, or 60 mg/kg/day; 3/sex/group	Yes	1270410	3624707, 3666991, 3952011
39-Week oral (gavage) administration toxicity study in the monkey followed by a 4-week treatment-free period Doses: 0, 20, 40, or 60 mg/kg/day, 4/sex/group	Yes	1470445	5056867

Source: Nonclinical Reviewer generated
[a] Dose level increased from 90 mg/kg to 120 mg/kg from the start of Week 6.
Abbreviations: GLP, good laboratory practice.

5.5.1.1. Repeat Dosing Toxicities With Leniolisib in Rats

2-Week Rat Study (Study# 1170360)

Rats (n=10/sex/group) were dosed once daily at 0, 10, 50, or 300 mg/kg/day (oral) for 15 days (HD animals did not receive treatment for full duration, see below). An additional 5 animals/sex/group in the control and HD groups were included in the 28-day recovery study. Premature deaths were observed in 2/10 HD males and 3/10 HD females between Days 2 and 7. The remaining animals in the HD group were euthanized on days 6 and 7. The microscopic findings in GI tract (minimal to moderate inflammation) along with the clinical signs of severe

diarrhea in HD animales indicated significant GI toxicity. Target organs of toxicity in the rat included adrenal gland, kidney, liver, spleen, thymus, pancreas, lymph nodes, GI tract, thyroid, and femur (marrow). The no observed adverse effect level (NOAEL) was judged to be the LD (10 mg/kg) based on dose limiting toxicities at the MD in thymus, pancreas, and GI tract (stomach, ileum, jejunum, duodenum, cecum) and mortality at the HD.

13-Week Rat Study (Study# 1270409)

Rats (n=10/sex/group) were dosed by oral gavage once per day with 0, 10, 30, or 120 mg/kg/day leniolisib. Spleen, lymph nodes (mesenteric and mandibular), thymus and GI tract (acute/subacute inflammation in ileum, cecum) were identified as target organs of toxicity. These findings provided evidence of immune suppressive effects of the test article. These effects are considered to be pharmacodynamic effects of leniolisib that are monitorable in a clinical setting. Therefore, the high dose of 120 mg/kg/day was used in assessment of safety.

39-Week Rat Study (Study# 1470444)

Rats (20/sex/group) were exposed to 0, 15, 40, and 120 mg/kg/day of leniolisib for 26 weeks via oral gavage with additional animals included in the control and HD groups for a 4-week recovery period. Toxicity was observed in MD and HD groups. The target organs were identified to be the testis, prostate, spleen, thymus, lymph nodes, skin, GALT/Peyer patches, bone marrow, and tongue. The HD induced severe toxicity, particularly for female rats. All HD females and 1 MD female had to be removed from the study early (9/20 HD between weeks 7-21, remaining 11/20 HD at week 24; the 1 MD at week 22) due to drug-related skin lesions secondary to the immunosuppressive properties of leniolisib. The sex difference in toxicity may be related to increased leniolisib exposure in females (female $AUC_{0\text{-}24hr}$ is 1.50- 2.68-fold higher than males). Toxicity to the germinal epithelium of testes (i.e., disrupted germinal epithelium, multinucleated germ cells, and tubule vacuoles) was observed at the MD and HD and it did not reverse following a 4-week recovery period. However, these findings were consistent with germ cell depletion and not considered indicative of damage to the static cellular mass of the testes. Minimal to moderate subacute inflammation was observed in the skin at all doses, and the tongue in the HD group for both males and females. Treatment-related atrophy of the mesenteric lymph node, thymus, and GALT/Peyer's patches were observed in male and female rats at all doses and attributed to immune suppression based on the pharmacology of leniolisib. Based on the germ cell findings in the MD and HD groups, the NOAEL was determined to be 15 mg/kg; however, these findings were not considered dose-limiting. The high dose of 120 mg/kg/day (AUC: 142000 (M) and 311000 (F) ng*hr/ml) was considered to be the limit dose with respect to observed germ cell depletion and findings attributed to immunosuppression. The findings attributed to immune suppression including skin lesions/infections were considered clinically monitorable findings that were consequences of the pharmacologic activity of leniolisib.

5.5.1.2. Repeat Dosing Toxicities With Leniolisib in Monkeys

2-Week Monkey Study (Study# 1170359)

Monkeys (n=3/sex/group) were dosed once daily at 0, 30, 100, or 300 mg/kg/day via oral gavage for 14 days. An additional 2 animals/sex/group in the control and HD groups were included in the 28-day recovery study. 3/5 males and 3/5 females in the HD group were sacrificed on Day 7 or Day 8/9. Dosing was stopped on Day 7 for the remaining animals in the HD group. The QTc prolongation was observed at the HD, which was consistent with findings from 2 single dose safety pharmacology studies in which QTc prolongation was observed in monkeys after a single 150 mg/kg dose. Target organs included adrenal gland, GI tract (stomach, cecum, colon, ileum, jejunum), pancreas, thyroid, lymphoid tissue, spleen, thymus, liver, and kidney. The NOAEL is judged to be the MD (100 mg/kg) based on premature deaths in the HD group as well as drug-related toxicities present in HD animals.

13-Week Monkey Study (Study# 1270410)

Monkeys (3/sex/group) were dosed once per day with 0, 20, 40, or 60 mg/kg/day leniolisib by oral gavage. These doses were reduced from 0, 40, 80, and 160 mg/kg/day starting at day 30 due to emesis, diarrhea, dehydration, and body weight loss. QTc prolongation was observed in the HD group at the 13-week time point (maximum increase = +15% vs. control at t = 5 hr post-dose) in male monkeys. Toxicity was observed in organs of the GI tract including ileum, stomach, and rectum in the HD group. Specific findings included acute/subacute inflammation, degeneration, and erosion. These findings are considered to be clinically monitorable. Therefore, the high dose of 60 mg/kg/day was used in the assessment of safety.

39-Week Monkey Study (Study# 1470445)

Monkeys (4/sex/group) were dosed once per day at oral doses of 0, 20, 40, and 60 mg/kg/day. QTc(B) prolongation was observed in the MD and HD groups. The finding is consistent with the 13-week oral toxicology study with monkeys. Prolongation did not increase (maximum increase = +15%) in 39-week study compared with 13-week study. The finding is considered adverse but clinically monitorable. The target organs of toxicity were identified as colon, caecum, rectum, spleen, GALT/Peyer's patch, mandibular lymph node, and thymus. Erosion/ulcer formation was observed in the caecum and rectum in a single male in the HD group. The finding of erosion/ulcer is considered adverse. In the colon, caecum, and rectum, crypt microabscess formation was observed in MD and HD dose groups. Additionally, lymphoid depletion was observed in the spleen, GALT/Peyer's patch, mandibular lymph node, and thymus. These findings were attributed to the pharmacological action of leniolisib. The NOAEL was identified as the low dose of 20 mg/kg/day due to the QTc(B) prolongation in the MD and HD groups and the erosion/ulcer formation in both the caecum and rectum in the single HD male. The mid

dose of 40 mg/kg/day (AUC: 48500 ng*h/mL) was used for safety margin calculations as the QTc(B) prolongation is considered clinically monitorable.

5.5.1.3. Leniolisib Toxicokinetics (TK) and Exposure Margins

Adequate exposure margins of leniolisib were obtained supporting the corresponding clinical studies during the development. Toxicities were generally PD-related and/or clinically monitorable, not considered dose-limiting.

Table 2. Summary of Leniolisib Exposures in Repeat Dose Toxicity Studies

Species	Study Duration	Dose (mg/kg)	$AUC_{0\text{-}24h}$, Male/Female (ng·h/mL)	Exposure Margin (M/F)
Rat	2-weeks	10[a]	6720/14700	0.2/0.4[a]
		50	40600/86800	1.0/2.1
		300	878000/1170000	21.5/28.7
	13-weeks	10	8420/13500	0.2/0.3
		30	44900/68800	1.1/1.7
		90/120[a,b]	273000/288000	6.7/7.1[a]
	26-weeks	15	13600/25900	0.3/0.6
		40	55800/83600	1.4/2.0
		120[a]	142000/311000	3.5/7.6[a]
Monkey	2-weeks	30[a]	21000/14100	0.5/0.3[a]
		100	13200/78200	0.3/1.9
		300	78200/42900	1.9/1.1
	13-weeks	40/20	1130/6970	0.03/0.2
		80/40	78200/42900	1.9/1.1
		60[a]	147000/73700	3.6/1.8[a]
	39-weeks	20	25900/14700	0.6/0.4
		40[a]	50500/46500	1.2/1.1[a]
		60	71500/79400	1.8/1.9

Source: Nonclinical Reviewer generated, See Table 1 for study report and review references

[a] Doses used for exposure margin calculations.

[b] Dose level increased from 90 to 120 mg/kg from the start of Week 6.

Note: Clinical exposure of leniolisib in healthy subjects at an oral dose of 70 mg BID at the steady state on Day 15 (Study 2101 CSR, part 3: AUC_{tau},ss = 20400 ng·h/mL, $AUC_{0\text{-}24h}$= 2 x 20400 ng·h/mL = 40800 ng·h/mL,).

Abbreviations: $AUC_{0\text{-}24h}$, area under the curve from 0 to 24 hours; AUC_{tau},ss, area under the curve during a dosing interval at steady state; BID, twice daily; CSR, clinical study report; F, female; M, male.

5.5.2. Genetic Toxicology

A complete battery of genetic toxicology studies was conducted. In the bacterial reverse mutation assay, there was no evidence of mutagenicity of leniolisib at up to 5,000 µg/plate. In an in vitro micronucleus assay in TK6 human lymphoblastoid cells, leniolisib at 343 µg/mL did not induce micronuclei after 3-hour incubation in the presence or absence of S9 or after 20-hour incubation in the absence of S9. In a chromosomal aberration assay in human peripheral blood lymphocytes, leniolisib at 325 µg/ml or at 350 µg/ml + S9 increased the number of structural chromosomal aberrations observed. The 4% incidence of structural aberrations (excluding gaps) in 1/2 assays in experiment 1 and 2/2 assays in experiment 2 are within the

historical control range (0-4%). The increase was observed at high cytotoxicity (50% and 46% mitosis inhibition in experiment 1 and 2, respectively) and no significant increases were observed at lower, more moderate toxicity. These observations lessen the concern of the findings at the HD. In an in vivo micronucleus assay with rats, there were no statistically significant effects of leniolisib treatment on the number of micronucleated polychromatic erythrocytes at doses up to 700 mg/kg/day. The totality of the evidence indicates leniolisib is negative for genotoxicity.

5.5.3. Carcinogenicity

Carcinogenicity studies with leniolisib have not been conducted. The 6-month transgenic (Tg-RasH2) mouse carcinogenicity study and 2-year carcinogenicity study with rats are ongoing under Special Protocol Assessments with ECAC concurrence. The final study reports will be submitted as post-marketing requirements.

5.5.4. Reproductive and Developmental Toxicology

The following GLP studies were submitted and reviewed previously *(DARRTS Reference ID 3951976)*. A pre- and postnatal development study with leniolisib has not been conducted. A pre- and postnatal development study with rats will be submitted as a post-marketing requirement.

Table 3. Leniolisib Reproductive and Developmental Toxicology Studies in Rats and Rabbits

Reproductive and Developmental Toxicology Studies	Report Number	Review Reference ID
CDZ173: 10-week oral (gavage) administration toxicity study in the juvenile rat followed by a 7-week treatment-free period with a fertility and early embryonic development assessment Doses: 0, 10, 30, or 90 mg/kg/day; 20/sex/group	1470452	3951976
CDZ173: Oral (gavage) study of embryo-fetal development in the rat Doses: 0, 10, 30, and 120 mg/kg/day; 20 females/group	1470306	3951976
CDZ173: Oral (gavage) study of embryo-fetal development in the rabbit Doses: 0, 10, 30, and 100 mg/kg/day ; 22 females/group	1470305	3951976

Source: Nonclinical Reviewer generated

Fertility and Early Embryonic Development

The Fertility and Early Embryonic Development (FEED) study was conducted in a subset (20/sex/group) of rats immediately after the conclusion of a juvenile toxicology study in which rats were treated with 0, 10, 30, or 90 mg/kg/day leniolisib once daily by oral gavage from PND 7 – 77. The study was conducted in accordance with methods outlined in the ICH S5(R2) Guidance to evaluate potential toxic effects resulting from leniolisib treatment from before mating through mating and implantation. For males, treatment related adverse findings in the male reproductive tract included decreased round spermatids (minimal) and decreased

spermatocytes (minimal) in the testis in HD group animals. Furthermore, mean total sperm count was decreased in males in the MD and HD groups. The mean sperm count values in both groups were within the historical control range. Decreased round spermatids and decreased spermatocytes in the testis in the HD group had no impact on mating and fertility indices. Therefore, the NOAEL with respect to fertility parameters in males is the HD at 90 mg/kg/day. The $AUC_{0\text{-}24h}$ at the 90 mg/kg/day dose is 90,900 ng·hr/ml. This value results in an exposure margin of 2.2 relative to the predicted maximum clinical exposure at 70 mg BID.

There was no evidence for leniolisib effects on female reproductive performance. Furthermore, there was no evidence for an effect of leniolisib on the number of corpora lutea, implantation sites, pre-implantation loss, viable embryos, and post-implantation loss. The NOAEL with respect to fertility parameters in females is the HD at 90 mg/kg/day, the highest dose tested. $AUC_{0\text{-}24h}$ at the 90 mg/kg/day dose is 143,000 ng·hr/ml. This value results in an exposure margin of 3.5 relative to the predicted maximum clinical exposure at 70 mg BID.

Embryo-Fetal Development (EFD) Study in Rats

Pregnant rats were treated with 0, 10, 30, or 120 mg/kg/day leniolisib by oral gavage from gestational day (GD) 6 – 17 and euthanized on GD 21.

A modest, statistically significant decrease in mean body weight gain was observed in HD animals over the course of the treatment period. Body weight gain returned to control levels in all dose groups during the post-treatment period. The NOAEL with respect to maternal toxicity was the MD at 30 mg/kg/day. The mean $AUC_{0\text{-}24h}$ at the 30 mg/kg dose on GD 17 = 65,000 ng·hr/ml. This provides a systemic exposure margin of 1.6 relative to the predicted maximum clinical exposure at 70 mg BID.

Mean fetal body weight was reduced in male and female offspring in the HD group. The magnitude of the body weight decreases in female fetuses (-7.8%) achieved statistical significance. Leniolisib-related external, visceral, and skeletal malformations were observed in fetuses in the HD group. Most notably, malformations of the eye were observed in fetuses from 6/20 litters. These findings included eye bulge, microphthalmia, anophthalmia, and small orbital socket. All findings exceeded the incidence range of historical controls. Additionally, treatment-related visceral and skeletal variations were noted in fetuses from HD animals. These included dilated ureter as well as multiple skeletal variations related to ossification or alignment.

The NOAEL with respect to teratogenicity is defined as the MD at 30 mg/kg/day based on decreased fetal body weights and malformations of the eye in offspring of 120 mg/kg treated females. The mean $AUC_{0\text{-}24h}$ at the 30 mg/kg dose on GD 17 = 65,000 ng·hr/ml. This provides a systemic exposure margin of 1.6 relative to the predicted maximum clinical exposure at 70 mg BID.

EFD Study in Rabbits

Pregnant rabbits were treated with 0, 10, 30, or 100 mg/kg/day leniolisib by oral gavage from gestational day (GD) 7 – 20 and euthanized on GD 29.

Females in the HD group were observed to have thin appearance as well as marked decreases in mean body weight gain (-63%) and food consumption (-55%). All of the above parameters returned to control levels after the cessation of leniolisib treatment. The NOAEL with respect to maternal toxicity was defined as the MD at 30 mg/kg/day. This dose was associated with a mean $AUC_{0\text{-}24h}$ = 13,200 ng·hr/ml. The exposure margin relative to the clinical $AUC_{0\text{-}24h}$ at the 70 mg BID dose is 0.3.

Leniolisib-related malformations were observed in fetuses in the HD group, in the presence of marked maternal toxicity. Consistent with fetal observations in rats, 2 fetuses from 1 female were found with eye malformations. Both animals were found with microphthalmia and small orbital socket. Such findings were not detected in the historical control database. Additionally, there were low incidences of skeletal malformations (2 – 3 fetuses/1 -2 litters) in fetuses in the HD group that exceeded the historical control range in the sternebra, vertebra, and ribs. Treatment related skeletal variations were noted in fetuses in the HD group. These included bent hyoid bone (skull), misshapen squamosal bone (skull), and additional ossification site of the cervical centrum (vertebra) in the HD group.

The NOAEL with respect to teratogenic effects is defined as the MD at 30 mg/kg/day based on eye malformations as well as additional skeletal malformations in fetuses in the HD group. The mean maternal $AUC_{0\text{-}24h}$ at the 30 mg/kg dose on GD 21 = 13,200 ng·hr/ml, resulting in an exposure ratio of 0.3 relative to the maximum clinical exposure.

Other Development Studies

Juvenile Animal Toxicology Study

The Applicant conducted a 10-week GLP toxicology study (1470452), in which juvenile rats (10/sex/group) received leniolisib at oral doses of 0, 10, 30, and 90 mg/kg/day for 10 weeks from PND 7-77. Mortality was observed for both males and females in the HD group. The age of balanopreputial separation for males in the MD and HD groups increased one day. The age of vaginal opening for females in the MD and HD groups decreased one day. These findings had no adverse impact on fertility or reproductive performance as assessed in the fertility and early embryonic study conducted at the conclusion of the juvenile toxicity portion of the study. Decreased mean total activity during the 30-minute test was observed for males in all treated groups and females in the HD group. Spleen (marginal zone depletion), GALT/Peyer's patch (decreased germinal centers), mesenteric lymph node, mandibular lymph node, and popliteal lymph node (decreased germinal centers), and testes (decreased round spermatids and/or decreased spermatocytes) were identified as potentially related to the pharmacodynamics of

leniolisib and not considered adverse. The MD of 30 mg/kg/day (AUC: 21600/40200 ng·h/mL, Male/Female) was used for exposure margin calculations due to mortality in the HD group.

Table 4. Leniolisib Toxicokinetics (TK) and Exposure Margins in Developmental and Reproductive Studies

Study	Species	Dose (mg/kg)	$AUC_{0\text{-}24h}$, Male/Female or Average (ng·h/mL)	Exposure Margin
Fertility and early embryonic development (FEED) study[a]	Rat	10	5270/8470	0.1/0.2
		30	21600/40200	0.5/1.0
		90[b]	90900/14300	2.2/3.5[b]
Embryofetal development (EFD) studies	Rat	10	12800	0.3
		30[b]	65000	1.6[b]
		120	248000	6.1
	Rabbit	10	5210	0.1
		30[b]	13200	0.3[b]
		100	87300	2.1
Juvenile toxicology study	Rat	10	5270/8470	0.1/0.2
		30[b]	21600/40200	0.5/1.0[b]
		90	90900/14300	2.2/3.5

Source: Nonclinical Reviewer generated, See Table 3 for study report and review references
[a] $AUC_{0\text{-}24h}$ of leniolis b in the rat FEED study are assessed on PND 77 in the juvenile toxicology study.
[b] NOAELs
Note: Clinical exposure of leniolisib in healthy subjects at an oral dose of 70 mg BID at the steady state on Day 15 (Study 2101 CSR, part 3: AUC_{tau},ss = 20400 ng·h/mL, $AUC_{0\text{-}24h}$ = 2 x 20400 ng·h/mL = 40800 ng·h/mL).
Abbreviations: $AUC_{0\text{-}24h}$, area under the curve from 0 to 24 hours; AUC_{tau},ss, area under the curve during a dosing interval at steady state; BID, twice daily; CSR, clinical study report; EFD, embryo fetal development; FEED, fertility and early embryonic development; NOAELs, no observed adverse effect levels; PND, postnatal day; TK, toxicokinetic.

5.5.5. Other Toxicology Studies

Phototoxicity

In an in vitro melanin binding assay, [^{3}H]leniolisib bound to synthetic melanin at two binding sites with Kd1 of 36.6 µM and Kd2 of 1.8 µM, respectively, suggesting prolonged retention in the posterior eye cup is to be expected. In a one-day ocular tolerability study, male rabbits (3/group) received ophthalmology suspension of leniolisib at concentrations of 0.5%, 2% or 5% by dropper into the right eye. One animal in the 2% group and two animals in the 5% group showed a minimal and transient conjunctival hyperemia which resolved by 24 hours post last dose. All fluorescein staining results at the 24-hour examination were negative. In addition, there were no test article-related ophthalmic or histopathologic findings in the eyes of treated animals in the 39-week repeat dose toxicity study in monkeys or in the 26-week repeat dose toxicity study in rats.

The UV spectrum of leniolisib shows absorption at a 290 nm wavelength and a secondary absorption maximum at a 330 nm wavelength. The molar extinction coefficients at these wavelengths were 2,261 and 2,042 L mol^{-1} cm^{-1}, respectively. In the follow-up GLP compliant in

vitro Balb/c 3T3 neutral red uptake phototoxicity assay, leniolisib was not phototoxic at up to 100 µg/mL. The risk of phototoxicity of leniolisib is considered minimal according to the guidance *S10 Photosafety Evaluation of Pharmaceuticals* (January 2015).

5.5.6. Conclusion

The Applicant has characterized leniolisib in safety pharmacology, genetic toxicology, general toxicology, and developmental/reproductive toxicology studies. Adequate exposure margins for leniolisib were obtained in all species that exceed the predicted human exposure at the MRHD of 70 mg BID. Leniolisib administration to pregnant rats and rabbits was associated with embryofetal toxicity including malformations at exposures approximately 2-6 times the MRHD on an AUC basis. Patients of childbearing potential should be advised of the risks to a fetus and recommend use of highly effective contraception during leniolisib treatment.

No major human metabolites were identified in completed clinical trials. Dose comparisons in the product labeling are based on the AUC of leniolisib. This approach is consistent with the labeling language describing nonclinical studies conducted with other kinase inhibitors. Based on the unmet medical need for the APDS indication, the carcinogenicity and pre/ postnatal development assessments will be completed as PMRs. In conclusion, there are no outstanding pharmacology-toxicology issues. The NDA is recommended for approval from the nonclinical perspective.

6. Clinical Pharmacology

6.1. Executive Summary

Leniolisib is a small molecule selective inhibitor of phosphoinositide 3-kinase delta (PI3Kδ) catalytic subunit p110δ that converts phosphatidylinositol (4,5)-bisphosphate (PI(4,5)P2) into phosphatidylinositol-3,4,5- trisphosphate (PIP3), an important phospholipid that anchors and activates many downstream kinases. p110δ is principally expressed in leukocytes. Overactive PI3Kδ caused by certain gain-of-function mutations in eith catalytical subunit p110δ (encoded by PIK3CD gene) or regulatory subunit p85 (encoded by PIK3R1 gene) interferes with immune cells maturation and differentiation, which results in the pathogenesis of activated PI3Kδ syndrome (APDS).

Leniolisib phosphate has pH-dependent solubility, high permeability, and falls under BCS class 2. The Applicant has submitted this NDA to support the use of leniolisib 70 mg twice daily orally for the treatment of APDS in adults and adolescents aged 12 years or older.

The clinical pharmacology program for leniolisib includes 8 clinical studies (Studies 2101, 2102, 2104, 2203, 2201, 2201E1, LE1101, and LE2101) (Table 5). Of the clinical studies in the program, 5 were conducted in healthy participants (Studies LE1101, LE2101, 2101, 2102, and 2104), 1 was conducted in subjects with Sjögren's syndrome (Study 2203), and 2 were conducted in subjects with APDS (Studies 2201 and 2201E1). Phase 1 clinical studies provided plasma and urine leniolisib pharmacokinetic (PK) information following the single- and multiple-dose in healthy participants; assessed absorption, distribution, metabolism, excretion (ADME) of leniolisib; and evaluated the drug-drug interaction (DDI) potential with cytochrome P450 (CYP)3A4/CYP2D6/P-glycoprotein (P-gp) inhibitors (itraconazole and quinidine) and CYP3A/UDP-glucuronosyl transferase (UGT)1A1/sulfotransferase (SULT)1E1 substrates (oral contraceptives ethinylestradiol and levonorgestrel). Phase 2 and 3 clinical studies provided single- and multiple-dose PK data on plasma leniolisib as well as pharmacodynamic (PD) data on % phosphorylated protein kinase B (pAkt)-positive B cells in subjects with Sjögren's syndrome and subjects with APDS.

The clinical pharmacology review is focused on the appropriateness of the proposed dosing regimen of leniolisib in patients with APDS, food effect, DDIs, and effect of various factors on the PK of leniolisib.

Recommendation

The Office of Clinical Pharmacology, Division of Inflammation and Immune Pharmacology (DIIP) have reviewed the clinical pharmacology data relevant to this application and recommend approval from a Clinical Pharmacology Perspective.

Post Marketing Requirement (PMR)

- Conduct a hepatic impairment clinical study to assess the impact of moderate and severe hepatic impairment on the pharmacokinetics of leniolisib.
- Conduct a cocktail drug-drug interaction (DDI) clinical study to assess the effect of leniolisib on the pharmacokinetics of CYP1A2 sustrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and MATE/ OCT2 substrate (metformin).

Table 5. Overview of Clinical Studies in the Clinical Pharmacology Program

Study Number; Summary; Phase	Key Clinical Pharmacology Objectives	Study Design (Number of Participants[a])	Treatments[b]
Studies in Healthy Participants			
CCDZ173X2101 (CSR Study 2101); Section 2.2.1; Phase 1	• Assess PK of ascending single and multiple oral doses of leniolisib • Investigate PK of leniolisib under fed and fasted conditions • Investigate PD effect of leniolisib on pAkt in B cells after *ex vivo* stimulation of peripheral blood • Explore the PK/PD relationship of different single and multiple oral doses of leniolisib • Explorative ADME and metabolite profiling and identification (using ^{19}F-NMR)	Multicenter, randomized, double-blind, placebo-controlled, ascending single- and multiple-dose study in healthy participants Part 1: Randomized, double-blind, placebo-controlled, single ascending dose study (N=64) Part 2: Randomized, open-label, 2-way crossover, food effect study (N=12) Part 3: Randomized, double-blind, placebo-controlled, multiple ascending dose study (N=42)	Part 1: Single oral dose of placebo or leniolisib in the fasted state at ascending dose levels of 10, 20, 40, 80, 110, 140, 200, 300, and 400 mg Part 2: Single oral dose of leniolisib 70 mg (1 × 70 mg capsule) given in the fasted and fed state Part 3: Oral placebo or leniolisib bid in the fasted state at ascending dose levels of 20, 40, 70, and 140 mg for 15 days Parts 1 and 3 used 10, 70 (Part 1, Cohort 20 only), and 100 mg capsules to make the required dose. The 10 and 100 mg capsules were (b) (4) formulation, while the 70 mg capsules were (b) (4) formulation.
LE1101 (CSR Study LE1101); Section 2.2.2; Phase 1	• Determine if (Novartis)70 mg leniolisib hard gelatin capsules are bioequivalent to (Pharming) 70 mg leniolisib film-coated tablets	Single-center, randomized, open-label, 2-way crossover study in healthy participants (N=18)	Single oral dose of leniolisib 70 mg in the fasted state as Novartis capsule (1 × 70 mg capsule) and as Pharming tablet (1 × 70 mg tablet) formulations
LE2101 (CSR Study LE2101); Section 2.2.3; Phase 1	• Determine total recovery and relative excretion of radioactivity in urine and feces after a single dose of 70 mg ^{14}C-leniolisib, containing 40 µCi of ^{14}C-radioactivity • Determine plasma PK parameters of total ^{14}C-radioactivity and of leniolisib	Single-center, open-label, ADME study in healthy male participants (N=6)	Single oral dose of 70 mg ^{14}C-leniolisib (40 µCi) as oral solution in the fasted state

CCDZ173X2102 (CSR Study 2102); Section 2.2.4; Phase 1	• Evaluate effect of itraconazole, a strong dual CYP3A/P-gp inhibitor, on single-dose PK of oral leniolisib • Evaluate effect of quinidine, a strong P-gp inhibitor, on single-dose PK of oral leniolisib	Single-center, open-label, single-sequence, 3-period crossover DDI study in healthy male participants (N=20)	Single oral dose of leniolisib 10 mg (1 × 10 mg capsule) on Day 1 of Period 1, on Day 5 of Period 2 (approximately 3 h after the itraconazole dose), and on Day 1 of Period 3 (approximately 1 h after the initial quinidine dose); Single oral dose of itraconazole 200 mg (2 × 100 mg capsule) on Days 1 through 9 of Period 2; Two oral doses of quinidine 300 mg (1 × 300 mg tablet) 4 h apart on Day 1 of Period 3 All morning doses were administered after an overnight fast, a standard light breakfast, and another fasting period. Leniolisib was administered at least 3.5 h after the start of the breakfast.
CCDZ173X2104 (CSR Study 2104); Section 2.2.5; Phase 1	• Assess effect of multiple oral doses of leniolisib on the PK of a single dose of a monophasic oral contraceptive containing ethinylestradiol and levonorgestrel in healthy female participants • Evaluate PK of oral leniolisib	Single-center, open-label, fixed-sequence, 2-period DDI study in healthy female participants (N=30)	Single dose of oral contraceptive tablet containing 30 µg ethinylestradiol and 150 µg levonorgestrel on Day 1 of Period 1 and on Day 15 of Period 2 (coadministered with leniolisib dose); Oral leniolisib 70 mg (1 × 70 mg capsule) bid on Days 1 through 17 of Period 2 All morning doses were administered after an overnight fast. Evening doses of leniolisib were given at least 1 h after dinner was consumed.
Studies in Patients with Sjögren's Syndrome			
CCDZ173X2203 (CSR Study 2203); Section 2.3.1; Phase 2	• Explore relationship between leniolisib PK, clinical efficacy outcomes, PD markers, and potential protein soluble serum and salivary biomarkers • Assess PK of leniolisib in patients with primary Sjögren's syndrome	Multicenter, multinational, double-blind, randomized, placebo-controlled, parallel-design study in patients with primary Sjögren's syndrome (N=20)	Oral placebo or leniolisib 70 mg (1 × 70 mg capsule) bid for 12 weeks Morning doses on Days 1, 8, 15, 29, 57, and 85 were given following an overnight fast; all other doses were given irrespective of food.
Study Number; Summary; Phase	**Key Clinical Pharmacology Objectives**	**Study Design (Number of Participants[a])**	**Treatments[b]**
Studies in Patients with APDS			
CCDZ173X2201 (CSR Study 2201 Part 1 and CSR Study 2201 Part 2); Section 2.4.1; Phase 2/3	• Assess dose-PD and PK/PD relationship of leniolisib in patients with APDS for dose selection in Part 2 • Assess PK of leniolisib in patients with APDS	Part 1: Multicenter, multinational, open-label, nonrandomized, dose-finding study in patients with APDS (N=6) Part 2: Multicenter, multinational, participant-, investigator-, and sponsor-blinded, randomized, placebo-controlled, fixed-dose study in patients with APDS (N=19)	Part 1: Oral leniolisib bid at sequentially increasing dose levels every 4 weeks from 10 mg (1 × 10 mg capsule), to 30 mg (3 × 10 mg capsule), to 70 mg (1 × 70 mg capsule). Evening doses of 10 and 30 mg on Days 28 and 56, respectively, were not taken to allow for pAkt washout Part 2: Oral placebo or leniolisib 70 mg (1 × 70 mg capsule) bid for 12 weeks
CCDZ173X2201E1[c] (Interim CSR Study 2201E1); Section 2.4.2; Phase 2/3	• Characterize PK of leniolisib in patients with APDS • Evaluate PK and relative bioavailability of (Novartis) leniolisib film-coated tablets compared to (Novartis) leniolisib hard gelatin capsules[d]	Multicenter, multinational, open-label, nonrandomized, active-treatment extension study in patients with APDS (N=35)	Oral leniolisib 70 mg bid initially as Novartis capsule (1 × 70 mg capsule) then as Novartis tablet (1 × 70 mg tablet) formulation[e] through the end of treatment period, up to a maximum of 6 years Treatment was given in fasted state or 0.5 h after a light breakfast for morning doses when serial PK sampling was performed and irrespective of food for all other doses.

Source: Summary of Clinical Pharmacology Studies, Table 2.

aNumber of participants who received leniolisib and contributed PK data.

b Treatment was given without regard to food unless otherwise stated.

c Study CCDZ173X2201E1 is an ongoing study.

d Statistical analysis for relative bioavailability assessment was no longer performed following reclassification of leniolisib as a BCS class 2 (low solubility, high permeability) compound.

Abbreviations: ADME, absorption, distr bution, metabolism, excretion; APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; (b) (4) CSR, clinical study report; CYP3A, cytochrome P450 3A; DDI, drug-drug interaction; N, total number of subjects; pAkt, phosphorylated protein kinase B; PD, pharmacodynamic; P-gp, P-glycoprotein; PK, pharmacokinetic; ^{19}F-NMR, fluorine-19 nuclear magnetic resonance.

6.2. Summary of Clinical Pharmacology Assessment

6.2.1. Pharmacology and Clinical Pharmacokinetics

The PK and PD properties of leniolisib following single-(10 to 400 mg) and multiple-dose administration (20 to 140 mg twice daily) have been characterized in 5 completed studies in healthy participants, 1 completed study in subjects with primary Sjögren's syndrome, and the completed and ongoing extension studies in subjects with APDS (10 to 70 mg twice daily) (Table 5). No dedicated clinical studies evaluating leniolisib PK in the hepatically or renally impaired population have been conducted.

Dosage Form/Formulation Development

Leniolisib is supplied as 70 mg tablets for oral administration. Leniolisib initially entered clinical development as a capsule formulation (Novartis hard gelatin capsule [HGC]) before a tablet formulation was developed by Novartis (Novartis file-coated tablet [FCT]). Two formulations of Novartis HGC were used in clinical studies, (b) (4) and (b) (4) All Novartis HGC at the 70 mg leniolisib dosage strength were of the (b) (4) formulation. Following acquisition by Pharming, the manufacturing process for the tablet formulation was transferred from Novartis Pharma AG to Pharming's intended manufacturer of the commercial product, Skyepharma (Pharming FCT). Healthy participants who received (b) (4) and (b) (4) formulations of Novartis HGC in Study 2101 showed similar dose-normalized leniolisib exposures, suggesting no clinically relevant differences between the 2 capsule formulations. Study LE1101 assessed the bioequivalence between leniolisib 70 mg Novartis HGC (b) (4) and Pharming FCT in healthy participants and found the formulations bioequivalent.

Pharmacokinetics

The systemic drug exposure (AUC and C_{max}) of leniolisib increased dose proportionally within the studied range of doses (20 to 140 mg twice a day dosing and single doses of 10 to 400 mg). During twice daily dosing approximately 12 hours apart, leniolisib accumulates approximately 1.4-fold (range of 1.0 to 2.2) in achieving steady-state, consistent with an effective half-life ($t_{1/2}$) of approximately 7 hours. Steady state drug concentrations were reached after approximately 2 to 3 days following the twice daily dosing regimen. The pharmacokinetics of leniolisib are similar between healthy participants and APDS subjects.

Absorption

In a placebo controlled, single and multiple ascending dose study in healthy participants, leniolisib median time to maximum plasma concentration (T_{max}) occurred at about 1-hour postdose. T_{max} appeared independent of dose and was not altered after multiple oral doses.

Food is unlikely to have a clinically meaningful effect on the systemic exposure of leniolisib for the FCT formulation.

Distribution

The systemic decay in leniolisib plasma concentration over time is bi exponential, indicating a distribution delay towards peripheral tissues. The apparent terminal elimination $t_{1/2}$ is approximately 10 hours. The volume of distribution of leniolisib is estimated to be 28.5 L in pateints with APDS. Leniolisib was highly bound (94.5%) to plasma proteins.

Metabolism

Leniolisib was 60% metabolized by the liver, with CYP3A4 being the most predominant enzyme involved (94.5%) in the primary oxidative metabolism of leniolisib with minor contribution from other enzymes (3.5% CYP3A5, 0.7% CYP1A2 and 0.4% CYP2D6). Intestinal secretion by BCRP as well as extrahepatic CYP1A1 cannot be excluded as excretion routes.

Elimination and Excretion

The mean recovery of total ^{14}C-radioactivity following a single oral dose of 70 mg ^{14}C leniolisib was 92.5% (67.0% and 25.5% recovered via feces and urine, respectively) 168 hours postdose. Unchanged leniolisib (6.32%) was the predominant drug-related material recovered in urine.

Pharmacodynamics

Ex vivo pharmacodynamics of leniolisib [proportion of phosphorylated Akt (pAkt)-positive B cells] were assessed intra-individually at 10, 30, and 70 mg twice daily for 4 weeks at each dose level in subjects with APDS. Within the explored dose range, higher leniolisib plasma concentrations were generally associated with higher reduction of pAkt-positive B cells and higher doses were associated with a slightly higher peak reduction as well as more sustained reduction. Treatment with leniolisib 70 mg twice a day at steady-state is estimated to produce time-averaged reduction of pAkt-positive B cells by approximately 80%.

6.2.2. General Dosing and Therapeutic Individualization

General Dosing

The proposed dosing regimen of leniolisib in pediatric patients 12 years of age and older and adults weighing more than 45 kg is 70 mg administered orally twice daily approximately 12 hours apart, with or without food. Clinical pharmacology review team agrees with the proposed dosing regimen.

6.2.2.1. Therapeutic Individualization

Drug-Drug Interactions

Drug-Drug Interactions as Victim

In a clinical DDI Study 2102, co-administration with itraconazole (a strong dual inhibitor of CYP3A4 and P-gp) increased leniolisib AUC_{inf} by approximately 2-fold and C_{max} by 25%. Hence, concomitant use of leniolisib with strong CYP3A4 inhibitors should be avoided. PBPK modeling and simulation predicted increased leniolisib AUC_{0-12} and C_{max} in the presence of erythromycin, a moderate CYP3A4 inhibitor, (approximately 60% and 30% increase, respectively) and decreased leniolisib AUC_{0-12} and C_{max} in the presence of rifampicin, a strong CYP3A4 inducer, (approximately 80% and 50% decrease, respectively) and efavirenz, a moderate CYP3A4 inducer (approximately 60% and 30% decrease, respectively). Hence, concomitant use with moderate CYP3A4 inhibitors may be allowed since doses up to 140 mg BID were well tolerated in the phase 1 MAD study in healthy subjects. Based on the PBPK results, co-administration of strong or moderate CYP3A4 inducers should be avoided.

Leniolisib exhibits pH-dependent solubility, with lower solubility at higher pH-values. No dedicated studies evaluating the effects of gastric acid reducing agents on leniolisib PK have been conducted. However, a post hoc analysis of interim data from Study 2201E1 showed nearly comparable plasma leniolisib concentration-time profiles between subjects with APDS who reported taking proton pump inhibitors (PPIs) and those who did not. Thus, leniolisib may be used concomitantly with PPIs or histamine-2 receptor antagonists with no dose adjustment. For details, refer to Section 6.3.2.

Drug-Drug Interactions as Perpetrator

Based on in vitro results, clinically relevant interactions at the therapeutic dose of leniolisib could not be excluded for inhibition of CYP1A2, UGT1A1, BCRP, OATP1B1, OATP1B3, OCT2, and MATE2K. While little to no reversible inhibition of CYP1A2 by leniolisib was observed in vitro, time-dependent inhibition was observed, indicating potential for clinically relevant DDI with CYP1A2 substrates. Because no dedicated clinical studies have been conducted to assess the effects of leniolisib on the PK of CYP1A2 substrates, it is recommended to avoid concomitant use of leniolisib with CYP1A2 substrates with narrow therapeutic indices. No dedicated clinical studies have been conducted to evaluate the effects of concomitant use of leniolisib on the PK of BCRP, OATP1B1, and OATP1B3 substrates (e.g., statins). Hence, concomitant use of these substrates with leniolisib should be avoided. The Applicant states that clinically relevant DDIs with leniolisib are not anticipated for substrates of the renal transporters, OCT2 and MATE2K, because leniolisib is only marginally (6%) renally excreted. This justification is not valid as limited renal elimination of leniosilib does not necessarily mean it cannot inhibit renal

transporters. In PBPK model-based simulations, co-administration with steady-state leniolisib led to decreased exposure of midazolam (a prototypical CYP3A4 substrate).

Based on these results, a PMR is issued to the Applicant to conduct a cocktail DDI clinical study to assess the effect of leniolisib on the pharmacokinetics of CYP1A2 sustrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and metformin (MATE/OCT2 substrate). For details, refer to Section 6.3.2.

Hepatic and Renal Impairment

No dedicated clinical studies to assess the effects of hepatic or renal impairment on leniolisib PK have been conducted. Considering that metabolism is the predominant pathway for leniolisib elimination, hepatic impairment may lead to significant changes in leniolisib exposure. Even though the current label states that leniolisib is not recommended in the moderate-to-severe hepatically impaired population, data on prevalence of hepatic impairment in the APDS patient population from published literature suggests that hepatomegaly is commonly reported as one of the noninfectious phenotypic features of APDS in 28.8% to 45% of patients, and, therefore, a proportion of APDS patients may experience hepatic impairment. In addition, in the absence of clinical PK data in subjects with hepatic impairment for model verification, PBPK modeling cannot provide the exposure estimation of leniolisib in subjects with hepatic impairment with high confidence. Therefore, a clinical PK study in subjects with hepatic impairment to inform labeling is required (see PMR in Section 13.1.5).

Renal clearance is a minor elimination pathway for leniolisib, with approximately 6% of elimination occurring renally. Therefore, the Applicant's justification to not conduct a renal impairment study and not excluding subjects with renal impairment in the label appears reasonable. For details, refer to Section 6.3.2.

Outstanding Issues

Based on the evaluation of the DDI results and potential risk in patients with hepatic impairment, the Clinical Pharmacology review team proposed the following two PMRs:

- Conduct a hepatic impairment clinical study to assess the impact of moderate and severe hepatic impairment on the pharmacokinetics of leniolisib.
- Conduct a cocktail drug-drug interaction (DDI) clinical study to assess the effect of leniolisib on the pharmacokinetics of CYP1A2 sustrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and MATE/ OCT2 substrate (metformin).

6.3. Comprehensive Clinical Pharmacology Review

6.3.1. General Pharmacology and Pharmacokinetic Characteristics

The clinical pharmacology program for leniolisib included 8 clinical studies (2101, 2102, 2104, 2203, 2201, 2201E1, LE1101, and LE2101). Of the clinical studies in the program, 5 were conducted in healthy participants (Studies LE1101, LE2101, 2101, 2102, and 2104), 1 was conducted in subjects with Sjögren's syndrome (Study 2203), and 2 were conducted in subjects with APDS (Studies 2201 and 2201E1). An overview of the studies included in the Clinical Pharmacology program is shown in Table 5. Phase 1 clinical studies provided plasma and urine leniolisib PK information following the single- and multiple-dose in healthy participants; assessed absorption, distribution, metabolism, excretion (ADME) of leniolisib, and evaluated the DDI potential with CYP3A4/CYP2D6/P-gp inhibitors (itraconazole and quinidine) and CYP3A/UGT1A1/ SULT1E1 substrates (oral contraceptives ethinylestradiol and levonorgestrel). In addition, the effect of race on PK was evaluated in a healthy Japanese participant cohort. Phase 2 and 3 clinical studies provided single- and multiple-dose PK data on plasma leniolisib as well as PD results on %pAkt-positive B cells in subjects with Sjögren's syndrome and subjects with APDS. A modeling and simulation approach was used to support the clinical pharmacology of leniolisib. A plasma leniolisib PopPK model was established from the healthy participant data in Study 2101 and used to generate individual post hoc PK parameter estimates for exposure-corrected QT interval (QTc) analysis. A subsequent reanalysis for QTc prolongation through modeling of change from baseline QTc (ΔQTc) and placebo-corrected ΔQTc was also performed. The QT-IRT review team concludes that leniolisib did not prolong the QtcF interval in a review dated January 26, 2023. In addition, PK data from subjects with APDS in Study 2201 were fitted to a 1-compartment PopPK model, and the estimated PK parameters were used for PK/PD analysis of pAkt inhibition in B cells (Study 2201 Part 1). PBPK analyses were also performed using data from Studies 2101 and 2102 to support evaluation of the DDI potential with leniolisib.

Formulation Development

Leniolisib was initially developed by Novartis as (b) (4) hard gelatin capsules (Novartis HGC) for its clinical development program. The Novartis HGC formulation at 10 and 100 mg dosage strengths (b) (4) used in early development (b) (4) the Novartis HGC formulation at 70 mg dosage strength (b) (4) (b) (4) used in the pivotal Phase 2/3 study in subjects with APDS (Study 2201). Subsequently, film-coated tablets (Novartis FCT) were developed to support further clinical development in the open-label extension study (Study 2201E1), and subjects were transitioned from Novartis HGC to Novartis FCT.

Novartis FCT was the intended formulation for commercial use in the Novartis APDS development program. Following acquisition of exclusive rights under certain Novartis intellectual property to research, develop, manufacture, and commercialize leniolisib, Pharming Group N.V. (Pharming) transferred the manufacturing process of Novartis FCT to its intended manufacturer of the final commercial presentation (Pharming FCT), Skyepharma. (b) (4) Leniolisib formulations used in clinical development are presented in Table 6.

Table 6. Leniolisib Formulations Used in Clinical Development

Ingredient	Amount per Dosage Unit (mg)			
	Hard Gelatin Capsule[a]			Film-Coated Tablet
	10 mg	70 mg	100 mg	70 mg
Leniolisib phosphate	(b) (4)			
Lactose monohydrate				
Microcrystalline cellulose				
(b) (4)				
Sodium starch glycolate				
(b) (4)				
Hydroxypropyl methylcellulose				
(b) (4)				
Colloidal silicon dioxide				
Magnesium stearate				
Coating[b]				
(b) (4)				

Source: Summary of Biopharmaceutical Studies, Table 4.

a (b) (4)

b Coating is composed of (b) (4)

Yellow for Skyepharma-manufactured tablets.

(b) (4)

In vitro dissolution profiles showed (b) (4) However, in vitro differences in dissolution at pH ≥4.0 did not translate to in vivo differences in leniolisib exposure among formulations in clinical studies.

Healthy participants who received (b) (4) and (b) (4) formulations of Novartis HGC showed similar dose-normalized leniolisib exposures in Study 2101 (Table 63), suggesting no clinically relevant differences between the 2 formulations. The geometric mean dose-normalized AUC_{inf} and C_{max} following a single dose of (b) (4) in the fasted state were 276 (19300/70) ng*h/mL/mg and 45.6 (3190/70) ng/mL/mg, respectively, comparable to those observed with (b) (4) in the fasted state (215 to 297 ng*h/mL/mg and 32.6 to 42.4 ng/mL/mg, respectively).

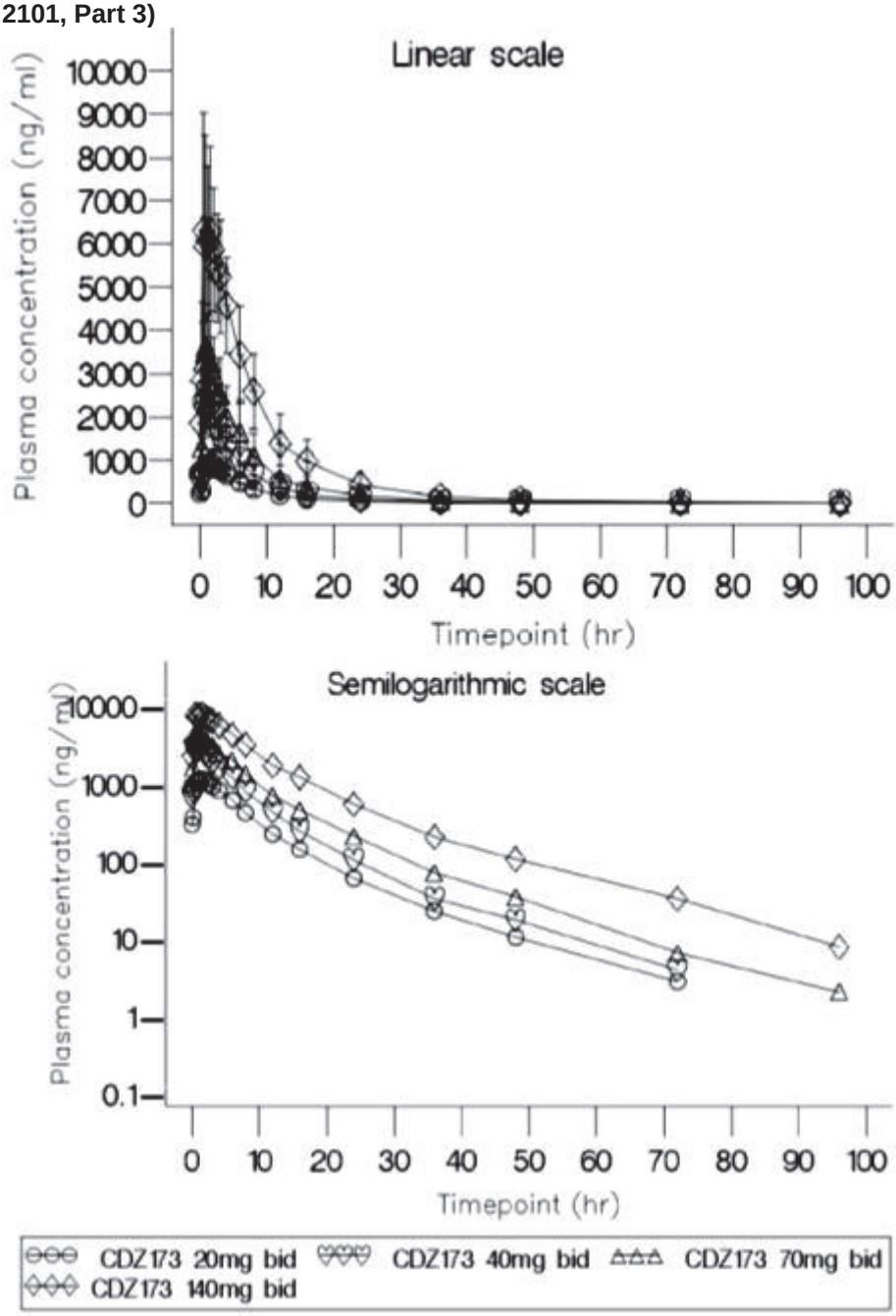
Study LE1101 assessed the bioequivalence between leniolisib 70 mg Novartis HGC (b) (4) and Pharming FCT in healthy participants and found the formulations bioequivalent (the GMR 90% CI comparing Pharming FCT to Novartis HGC was well contained within 0.80 to 1.25 C_{max} and AUC_{0-t}, and AUC_{inf}) (Table 65). Hence, no clinically relevant difference in leniolisib exposure is expected between the proposed commercial product (Pharming FCT) and drug product used in the pivotal clinical study (Novartis HGC).

Reviewer Comment: *The results of Study LE1101 demonstrated that the Novartis HGC (b) (4) (the drug product used in pivotal Study 2201) and Pharming FCT (the to-be-marketed drug product) are bioequivalent and therefore the PK, PD, and efficacy results from Study 2201 in APDS subjects conducted with the capsule formulation can also be applied to the to-be-marketed tablet formulation.*

Absorption

Following a single oral dose from 10 to 400 mg in healthy participants, leniolisib was rapidly absorbed with the median T_{max} ranging from 1 to 2 hours postdose. With twice daily dosing from 20 to 140 mg, median T_{max} was mostly unchanged at approximately 1-hour post-dose (Figure 1). Leniolisib exposure increased proportional to dose across the dose ranges evaluated (20 to 140 mg twice a day dosing and single doses of 10 to 400 mg). In subjects with APDS, the median T_{max} following a single dose of 10 to 70 mg leniolisib ranged from approximately 2 to 3 hours postdose. With twice daily dosing, there was low accumulation (accumulation ratio 1.32 to 1.57) in healthy participants, and steady state was achieved within 2 to 3 days from onset of therapy. PK of leniolisib were comparable between healthy participants and APDS subjects. PK parameters of leniolisib following single ascending doses and multiple ascending doses in healthy volunteers are shown in Table 7 and Table 8, respectively. PK parameters of leniolisib in subjects with APDS are shown in Table 9.

Figure 1. Mean (SD) Leniolisib Plasma Concentration-Time Profiles on the Final Dose on Day 15 Following Twice Daily Dosing of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 3)



Source: Study 2101 CSR, Figure 14.2-2.1c.
Abbreviations: BID, twice daily; CSR, clinical study report; SD, standard deviation.

Table 7. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following Single-Dose Administration of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 1)

		Geometric Mean (Geometric CV%)				
Leniolisib Dose (mg)	n	C_{max} (ng/mL)	T_{max} (h)	AUC_{inf} (ng*h/mL)	$t_{1/2}$ (h)	CL_R (L/h)
10	8	402 (35.5)	1.00 (0.5, 2.5)	2380 (43.5)	5.32 (37.1)	0.159 (51.7)
20	8	790 (20.1)	1.00 (1.00, 2.00)	5200 (30.9)	6.49 (29.6)	0.144 (51.6)
40	6	1470 (33.0)	0.875 (0.5, 1.50)	8590 (39.2)	6.53 (24.4)	0.173 (13.4)
80	6	3400 (42.4)	1.25 (0.5, 2.50)	23800 (44.5)	9.15 (50.5)	0.146 (17.5)
110	6	4010 (33.9)	1.25 (0.5, 6.00)	28300 (29.6)	8.83 (38.5)	0.147 (36.7)
140	6	5980 (27.4)	1.00 (0.517, 3.00)	28400 (20.2)	5.94 (71.7)	0.178 (36.7)
200	12	7110 (22.6)	1.31 (0.5, 2.17)	45700 (33.2)	8.40 (37.7)	0.206 (19.6)
300	6	9780 (17.6)	1.52 (1.50, 2.05)	70400 (20.9)	8.61 (26.0)	0.153 (38.6)
400	6	13800 (29.3)	2.00 (1.50, 3.00)	111000 (21.9)	10.4 (18.8)	0.187 (55.0)

Source: Study 2101 CSR, Figure 14.2-2.1a and Table 14.2-2.2a.
Abbreviations: $AUC_{0-\infty}$, area under the curve to infinity; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; CL_R, renal clearance; L/h, liter per hour; n, number of subjects; T_{max}, time to maximum observed plasma concentration; $t_{1/2}$, half-life.

Table 8. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following Twice Daily Dosing of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 3)

		Geometric Mean (Geometric CV%)						
Dose Regimen	n	C_{max} (ng/mL)	T_{max} (h)	AUC_{0-12} (ng*h/mL)	CL/F (L/h)	$t_{1/2}$ (h)	R_{acc}	CL_R (L/h)
20 mg bid								
Day 1	8	811 (21.9)	1.05 (0.75, 2.00)	4470 (17.1)	-	-	-	-
Day 15	6	1030 (24.4)	1.00 (0.583, 2.50)	5890 (28.7)	3.40 (28.7)	9.85 (46.7)	1.32 (16.1)	0.0861 (24.9)
40 mg bid								
Day 1	6	1910 (21.7)	0.875 (0.5, 1.50)	9030 (14.9)	-	-	-	-
Day 15	6	2680 (25.3)	1.00 (0.6, 1.00)	12500 (30.8)	3.20 (30.8)	9.66 (47.5)	1.38 (21.4)	0.193 (53.5)
70 mg bid								
Day 1	22	3020 (22.5)	1.00 (0.5, 4.00)	13900 (28.0)	-	-	-	-
Day 15	16	3650 (29.6)	1.00 (0.5, 2.00)	19000 (43.5)	3.69 (43.5)	9.38 (43.6)	1.45 (18.8)	0.179 (55.5)
140 mg bid								
Day 1	8	5620 (10.4)	0.875 (0.5, 2.50)	27400 (28.6)	-	-	-	-
Day 15	7	7640 (21.6)	0.75 (0.5, 2.55)	42000 (27.8)	3.33 (27.8)	10.9 (29.8)	1.57 (12.4)	0.251 (31.1)

Source: Study 2101 CSR, Figure 14.2-2.1c and Table 14.2-2.2c.
Abbreviations: AUC_{0-12}, area under the curve from 0 to 12 hours; BID, twice daily; CLF, apparent clearance; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; CL_R, renal clearance; L/h, liter per hour; n, number of subjects; R_{acc} accumulation ratio; T_{max}, time to maximum observed plasma concentration; $t_{1/2}$, half-life.

Table 9. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following the First Dose of 70 mg Leniolisib Capsule in Subjects With APDS – Stratified by Age Group (Study 2201, Part 2)

Age Group Visit	n	Geometric Mean (Geometric CV%)[a]			
		C_{max} (ng/mL)	T_{max} (h)	AUC_{0-t} (ng*h/mL)	AUC_{0-8} (ng*h/mL)
Overall					
Day 1	19[b]	2080 (25.4)	2.87 [0.92, 7.78]	10100 (27.6)	10400 (26.3)
<18-year-old					
Day 1	7	2070 (25.4)	3.00 [1.00, 5.05]	9890 (31.1)	10200 (28.4)
≥18-year-old					
Day 1	12[c]	2090 (26.6)	1.16 [0.92, 7.78]	10100 (26.9)	10600 (26.3)

Source: Study 2201 Part 2 CSR, Table 14.2-1.2.1.1b.

a Median [minimum, maximum] presented for Tmax.

b n=18 for AUC0-8.

c n=11 for AUC0-8.

Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{0-8}, area under the curve from 0 to 8 hours; AUC_{0-t}, area under the curve to the last quantifiable time point; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects; T_{max}, time to maximum observed plasma concentration.

Distribution

Leniolisib is highly bound (94.5%) to plasma proteins and primarily associated with plasma in vitro (mean blood-to-plasma ratio of 0.643). Systemic decline in leniolisib plasma concentration over time appeared biexponential, indicating a distribution delay toward peripheral tissues. Leniolisib has a low-to-moderate volume of distribution. In healthy participants, the geometric mean apparent volume of distribution ranged from 32.3 to 54.3 L following a single dose from 10 to 400 mg. In subjects with APDS, the mean typical volume of distribution was estimated as 28.5 L.

Metabolism

In vitro, phase 1 oxidative metabolism (mainly hydroxylation and dealkylation) was identified as the major metabolic pathway of leniolisib, and no phase 2 metabolism was observed. Leniolisib was 60% metabolized by the liver, with CYP3A4 being the most predominant enzyme involved (94.5%) in the primary oxidative metabolism of leniolisib with minor contribution from other enzymes (3.5% CYP3A5, 0.7% CYP1A2 and 0.4% CYP2D6).

Elimination

The mean recovery of total ^{14}C-radoactivity following a single oral dose of 70 mg ^{14}C-leniolisib was 92.5% (67.0% and 25.5% recovered via feces and urine, respectively) 168 hours postdose. Based on renal clearance estimated from 48-hour urine excretion in healthy participants,

approximately 6% of oral dose was recovered as unchanged leniolisib in the urine. The terminal elimination $t_{1/2}$ of leniolisib is approximately 5-10 hours in healthy volunteers.

Population Pharmacokinetic Analysis

PopPK analysis for leniolisib was performed on data from healthy participants in Study 2101. The PopPK model was built using a total of 2803 leniolisib concentration measurements from 118 participants with median [range] body weight of 76.3 [50.0, 105] kg who received leniolisib as single doses from 10 to 400 mg and under a twice daily regimen from 20 to 140 mg for 14 days. Leniolisib PK was well-described by a 2-compartment model with absorption lag time (ALAG), first-order absorption (Ka), and first-order elimination. The model included a body weight effect on the apparent volume of distribution in the central (V_c/F) and peripheral (V_p/F) compartments, a Japanese race effect on apparent clearance (CL/F), an evening dose time (light-fed condition) effect on Ka, and estimated different Ka and ALAG for fasted- and fed-state administration.

Parameter estimates of the healthy participant population PK model appear in line with those estimated from 1-compartment PopPK modeling of APDS subject data in Study 2201, suggesting that PK of leniolisib were similar between healthy participants and APDS subjects. Leniolisib V_c/F and V_p/F increased with weight relative to a 70-kg individual according to a power coefficient of 0.662 (i.e., [body weight in kg/70]$^{0.662}$). Based on the model, V_c/F and V_p/F were 20% lower relative to a 70-kg individual for participants weighing 50 kg and 31% higher relative to a 70-kg individual for participants weighing 105 kg. PopPK model did not determine age or sex to be a significant predictor of leniolisib PK and did not include it as a covariate effect in the final model. Leniolisib CL/F was 37% higher for Japanese participants relative to non-Japanese participants. However, the PopPK analysis dataset only has 6 Japanese participants who were enrolled in Study 2101. Caution should be taken when compare the PK between Japanese and non-Japanese subjects. In addition, the estimated 95% CI of the effect of formulation ((b) (4) versus (b) (4) on Ka included 1 and its inclusion did not result in improved predictive performance. Thus, formulation was not included as a covariate in the final model.

6.3.2. Clinical Pharmacology Questions

Does the Clinical Pharmacology Program Provide Supportive Evidence of Effectiveness?

The PK and PD properties of leniolisib following single-(10 to 400 mg) and multiple-dose administration (20 to 140 mg twice daily) have been characterized in 5 completed studies in healthy participants, and the completed and ongoing extension studies in subjects with APDS (10 to 70 mg twice daily). In Part 1 of Study 2201, a clear and apparent reduction in pAkt-positive B cells and pAkt reduction was observed in subjects with APDS (N=-6) following all three investigated doses (10, 30, and 70 mg). The maximum reduction was observed as early as 1-hour post-dose. In Part 2 of Study 2201 in APDS subjects (N=31) on the proposed dosing

regimen (i.e., 70 mg BID), the co-primary efficacy endpoints (change from baseline in the log10 transformed SPD in index lesions and change from baseline in percentage of naïve B cells) were met. Refer to clinical/statistical Section 8 for the efficacy and safety results from Study 2101 in subjects with APDS.

Is the Proposed Dosing Regimen Appropriate for the General Patient Population for Which the Indication is Being Sought?

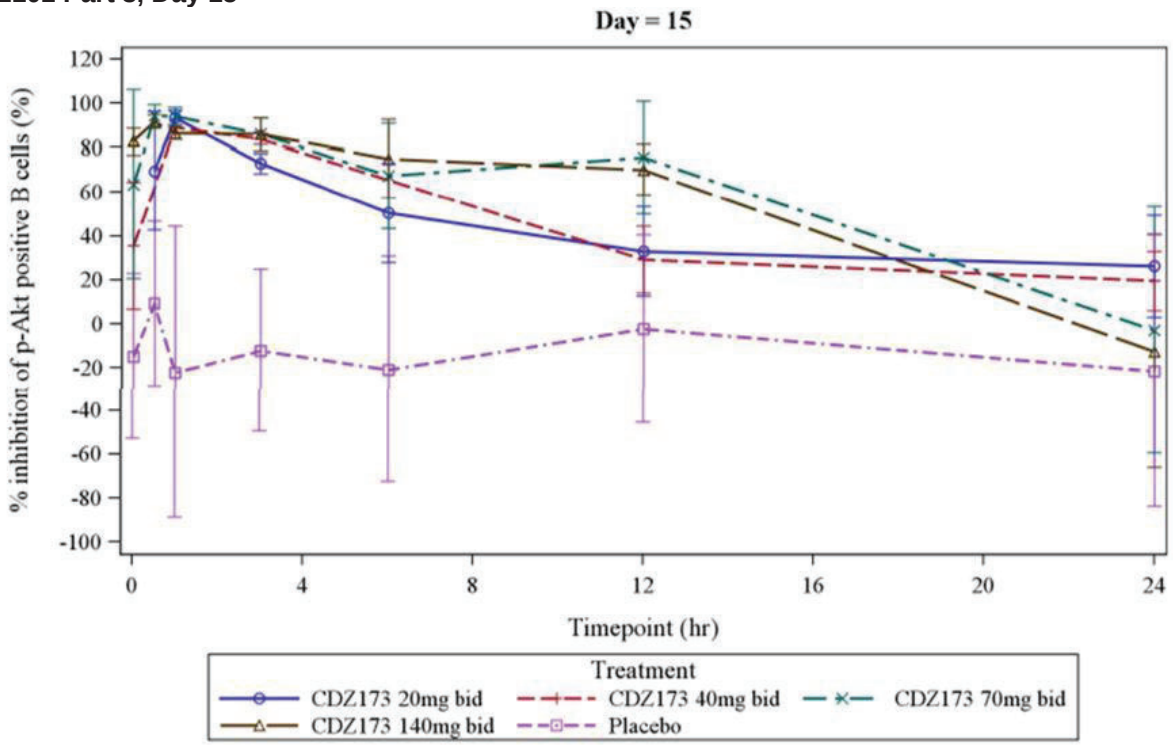
Exposure-Response Relationship

Percent Change From Baseline in %pAkt-Positive B Cells

The PD endpoint following leniolisib treatment was measured ex vivo %pAkt-positive B cells, as Akt is a direct downstream target of activated PI3Kδ. Inhibition of pAkt in ex vivo non-stimulated and stimulated B cells was observed shortly after dosing with leniolisib in a dose- and concentration-dependent manner in both the first-in-human study at leniolisib doses from 10 to 400 mg (Study 2101 Parts 1 and 3) and the dose-finding part of the pivotal study in subjects with APDS evaluating leniolisib at 10, 30, and 70 mg twice daily (Study 2201 Part 1).

In healthy participants, inhibition of ex vivo stimulated pAkt-positive B cells was observed shortly after dosing with leniolisib, while negligible reduction was observed with placebo. Percent pAkt inhibition was highest in the 70 and 140 mg twice daily dose groups and peaked at approximately 1-hour post-dose (Figure 2). Percent pAkt-positive B cells gradually returned close to baseline levels in approximately 24 hours from the last dose. The maximal pAkt inhibition appeared to reach at doses of 70 mg and 140 mg BID (approximately 90%). Therefore, doses up to 70 mg BID were evaluated in Study 2201 (Part 1) in APDS subjects.

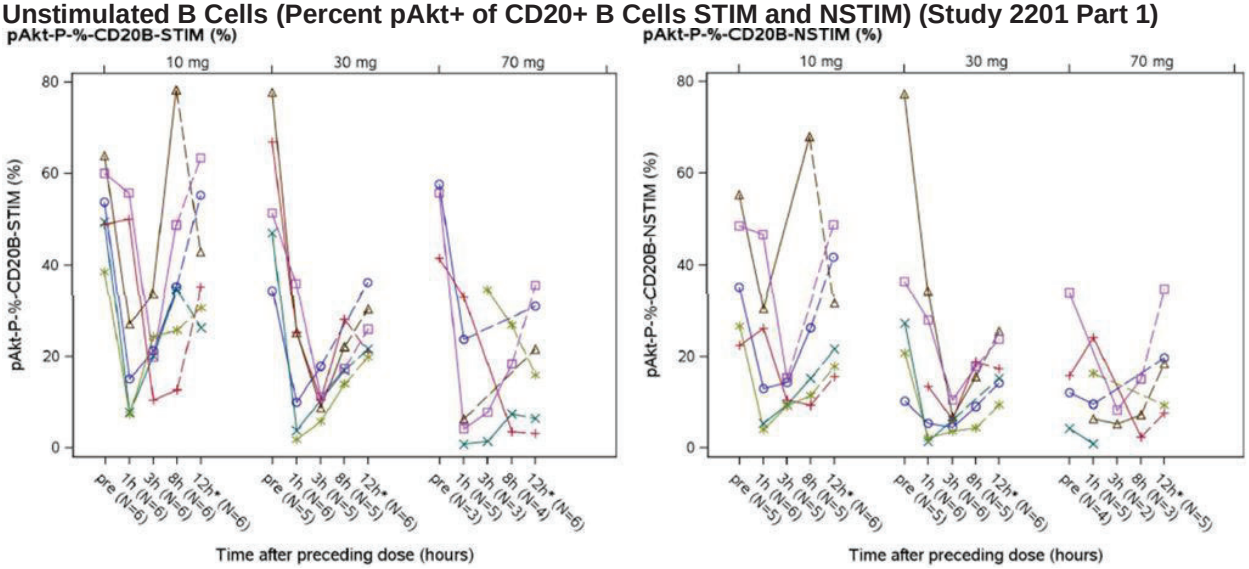
Figure 2. Arithmetic Mean (SD) Inhibition of Phospho-Akt Positive B Cells Profiles (N=35, Study 2101 Part 3, Day 15



Source: Study 2101 CSR, Figure 14.2-10c
Abbreviations: BID, twice daily; N, total number of subjects; CSR, clinical study report; pAkt, phosphorylated protein kinase B; SD, standard deviation.

In subjects with APDS from Part 1 of Study 2201, a clear and apparent maximum reduction in both stimulated and non-stimulated pAkt-positive B cells was observed as early as 1-hour post-dose (Figure 3). The early onset of pAkt inhibition occurred at approximately the same time as plasma leniolisib T_{max}. The pAkt inhibition started to rebound after T_{max} (1 hr) and returned to baseline level after 12 hours thereby providing support for the proposed BID dosing regimen. Although a single dose of 10 mg generally reduced %pAkt-positive B cells to less than 20%, the inhibition generally waned over the ensuing 12 hours. While the 30- and 70-mg dose levels did not differ in their maximum effect, inhibition was more sustained over time at 70 mg, which may therefore be more optimal in terms of time-averaged inhibition and inhibition at trough (12 hours post-dose).

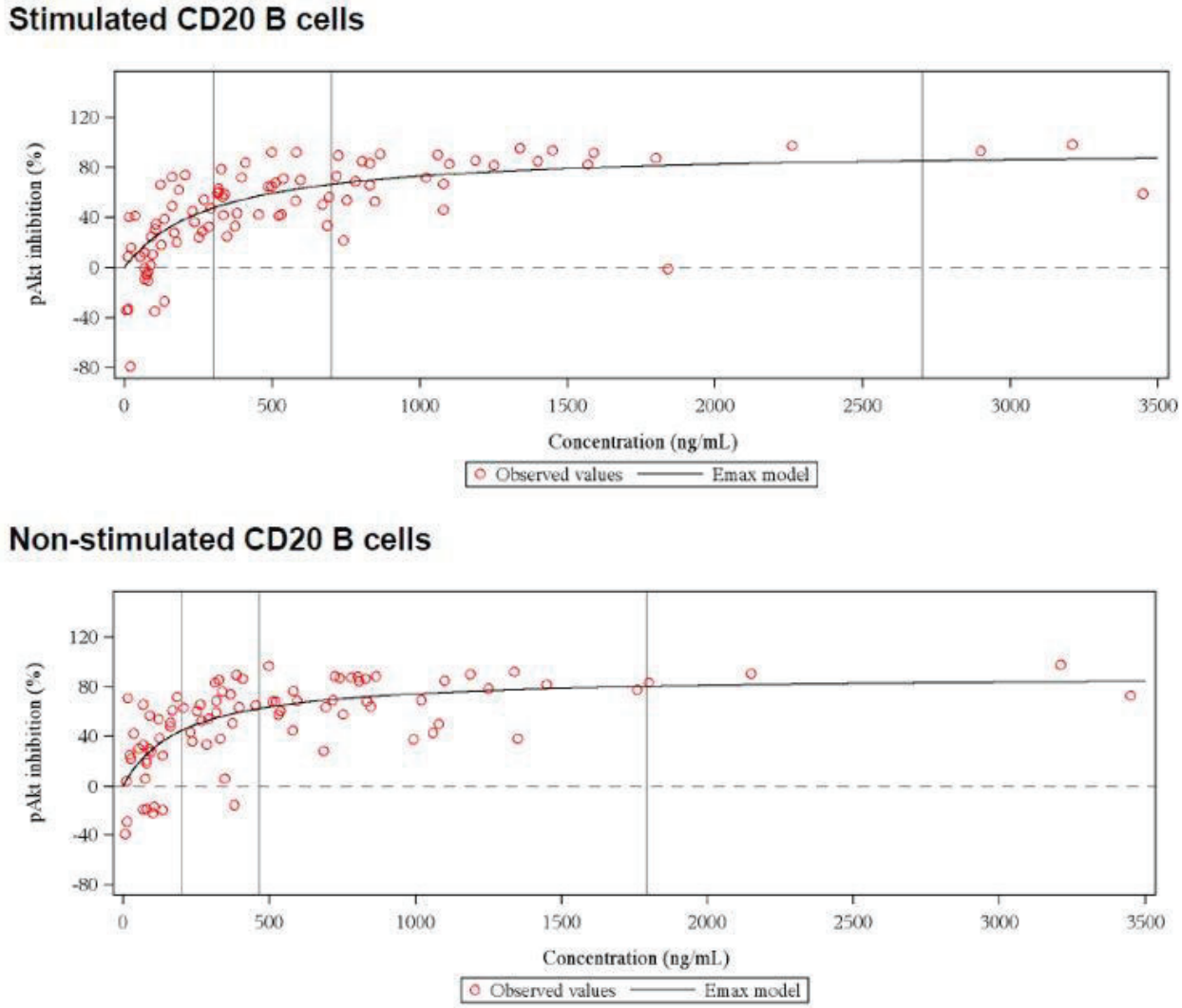
Figure 3. Individual Subject Profiles (N=6) of Phosphorylated Akt in Ex Vivo Stimulated and Unstimulated B Cells (Percent pAkt+ of CD20+ B Cells STIM and NSTIM) (Study 2201 Part 1)



Source: Figure 11-1 on page 59 from Study 2201 Part 1 CSR
Abbreviations: CSR, clinical study report; N, total number of subjects; NSTIM, unstimulated; pAkt, phosphorylated protein kinase B; STIM, stimulated.

Based on the E_{max} model (Figure 4), treatment with leniolisib 10, 30, and 70 mg twice daily is expected to result in time-averaged inhibition of stimulated pAkt in B cells of approximately 38%, 63%, and 78%, respectively, with an EC_{50} of 300 ng/mL, and time-averaged pAkt inhibition at 50% (ED_{50}), 70% (ED_{70}), and 90% (ED_{90}) of E_{max} are expected with leniolisib doses of 15, 35, and 136 mg twice daily, respectively, in ex vivo stimulated B cells (Table 10). The treatment with leniolisib 10, 30, and 70 mg twice daily is expected to result in time-averaged inhibition of non-stimulated pAkt in B cells of approximately 44%, 77%, and 78%, respectively, with an EC_{50} of 200 ng/mL, and time-averaged pAkt inhibition at 50% (ED_{50}), 70% (ED_{70}), and 90% (ED_{90}) of E_{max} are expected with leniolisib doses of 10, 23, and 90 mg twice daily, respectively, in ex vivo non-stimulated B cells (Table 10).

Figure 4. E_{max} Concentration-Response of Phosphorylated Akt in B Cells (Study 2201 Part 1)



Source: Figure 11-2 on page 60 from Study 2201 Part 1 CSR
Abbreviations: CSR, clinical study report; E_{max}, maximal effect; pAkt, phosphorylated protein kinase B.

Table 10. E_{max} Model of Concentration-Response of Phosphorylated Akt in B Cells – Study 2201 Part 1

Biomarker	Parameter	Estimate	Standard Error	Two-sided 95% Confidence interval
pAkt-P-%-CD20B-STIM (%)	Emax	94.60	5.80	(78.49, 110.72)
	EC50	300.32	55.68	(145.72, 454.92)
	EC70	700.75	129.93	(340.02, 1061.49)
	EC90	2702.91	501.14	(1311.51, 4094.30)
	ED50	15.06	2.79	(7.31, 22.82)
	ED70	35.15	6.52	(17.06, 53.24)
	ED90	135.58	25.14	(65.79, 205.37)
	Time averaged pAkt inhibition (%) at 10 mg	37.74	2.23	(31.57, 43.92)
	Time averaged pAkt inhibition (%) at 30 mg	62.98	1.46	(58.92, 67.04)
	Time averaged pAkt inhibition (%) at 70 mg	77.85	2.58	(70.69, 85.01)
pAkt-P-%-CD20B-NSTIM (%)	Emax	88.93	7.57	(69.47, 108.39)
	EC50	199.19	68.36	(23.45, 374.92)
	EC70	464.77	159.51	(54.72, 874.81)
	EC90	1792.67	615.27	(211.08, 3374.26)
	ED50	9.99	3.43	(1.18, 18.81)
	ED70	23.31	8.00	(2.75, 43.88)
	ED90	89.92	30.86	(10.59, 169.25)
	Time averaged pAkt inhibition (%) at 10 mg	44.48	5.50	(30.35, 58.62)
	Time averaged pAkt inhibition (%) at 30 mg	66.71	4.17	(55.99, 77.43)
	Time averaged pAkt inhibition (%) at 70 mg	77.82	4.76	(65.59, 90.05)

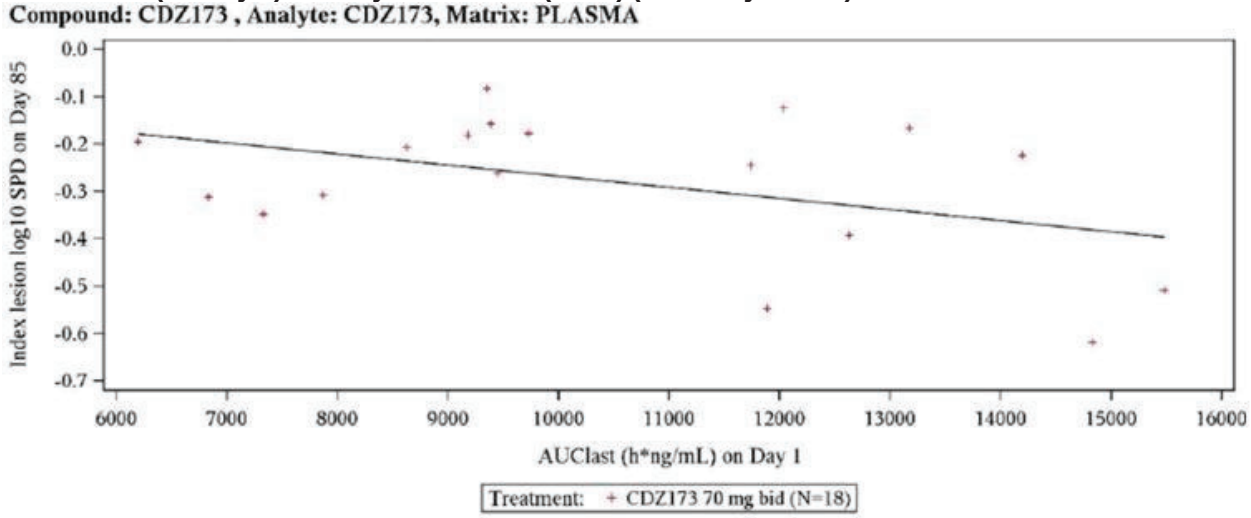
Source: Table 11-3 on page 61 from Study 2201 Part 1 CSR.
Abbreviations: CSR, clinical study report; EC50, effect concentration 50; EC70, effect concentration 70; EC90, effect concentration 90; ED50, effective dose 50; ED70, effective dose 70; ED90; effective dose 90; E_{max}, maximal effect; NSTIM, unstimulated; pAkt, phosphorylated protein kinase B; STIM, stimulated

Effect on Lymphoproliferation and Immunophenotype Normalization

In Part 2 of the study, the effect of 70 mg leniolisib twice daily on lymphoproliferation and immunophenotype normalization in subjects with APDS was assessed by the primary efficacy endpoints, log10 SPD of index lesions and % naive B cells. Scatterplots of the change from baseline in primary efficacy endpoints at Day 85 of treatment against leniolisib plasma AUC_{0-t}

(on Day 1) showed slight trends for greater reduction in log10 SPD (on Day 85) of index lesions and increase in % naive B cells at higher AUC_{0-t} values (Figure 5).

Figure 5. Scatterplot of Change From Baseline in log10 Transformed SPD (on Day 85) Versus PK Parameters (on Day 1) – Study 2201 Part 2 (N=18) (PD Analysis Set)



Source: Figure 11-11 on page 80 from Study 2201 Part 2 CSR
Abbreviations: AUC_{last}, area under the curve to the last quantifiable time point; BID, twice daily; CSR, clinical study report; N, total number of subjects; PD, pharmacodynamic; PK, pharmacokinetic; SPD, sum of product of diameters.

Overall, based on the PK/PD relationship and on the E_{max} concentration-response model, while both 30 and 70 mg may achieve near complete pathway inhibition shortly post dose, 70 mg BID is expected to confer a more sustained and higher time-averaged inhibition relative to lower doses which provides support for the recommended dose of 70 mg BID of leniolisib.

Is an Alternative Dosing Regimen or Management Strategy Required for Subpopulations Based on Intrinsic Patient Factors?

An alternative dosing regimen is not recommended for subpopulations based on intrinsic patient factors.

Subjects With Hepatic and/or Renal Impairment

No dedicated clinical studies to assess the effects of hepatic or renal impairment on leniolisib PK have been conducted. Renal clearance is a minor elimination pathway for leniolisib, with approximately 6% of elimination occurring renally. Therefore, the Applicant's justification to not conduct a renal impairment study and not excluding subjects with renal impairment in the label appears reasonable based on the draft guidance *Pharmacokinetics in Patients With Impaired Renal Function — Study Design, Data Analysis, and Impact on Dosing and Labeling* (September 2020).

The Applicant proposes in the current label that leniolisib is not recommended in the moderate-to-severe hepatically impaired population. In vitro and in vivo results indicate that leniolisib is approximately 90% eliminated through metabolism with 60% metabolized by the liver. Therefore, leniolisib plasma concentrations in APDS subjects with moderate-to-severe hepatic impairment can be expected to exceed those observed with dosing with 140 mg twice daily (highest dose assessed in leniolisib clinical program). To determine the need to conduct a hepatic impairment study to evaluate the impact of moderate and severe hepatic impairment on the PK of leniolisib and understand the prevalence of hepatic impairment in the APDS patient population, the following IR was sent to the Applicant on October 4, 2022:

> *In the proposed label, you indicate that the use of leniolisib in patients with moderate to severe hepatic impairment is not recommended. Based on the exclusion criteria for your Study 2201 (Parts 1 and 2), we note that subjects with liver disease or liver injury as indicated by clinically significant abnormal liver function tests (ALT and AST greater than 2.5 times upper limit of normal) were excluded from the study. Provide information regarding how many subjects were excluded from the Study 2201 due to the presence of hepatic impairment based on your exclusion criteria. In addition, provide epidemiology evidence to estimate the possible prevalence of concomitant hepatic disease and hepatic impairment (by both your Study 2201 hepatic impairment criteria and Child-Pugh Scoring system) in the target patient population.*

The Applicant responded that there were no subjects in the clinical database excluded from Study 2201 due to the presence of hepatic impairment based on the exclusion criteria. However, screen failures were not recorded in the CRF and therefore not captured in the clinical database. The Applicant also stated that upon completing an extensive literature search, there are currently no comprehensive sources for international longitudinal natural history data for APDS. Data has been reported in the published literature in both Europe (Elgizouli et al. 2016; Coulter et al. 2017; Maccari et al. 2018) and the U.S. (Oh et al. 2021), and a global systematic review was conducted in the APDS population (Jamee et al. 2020). None of the currently available published literature characterizes potential hepatic impairment by AST/ALT levels or the Child-Pugh criterion.

An extensive systematic review of all published literature in 2019 identified 243 APDS subjects (Jamee et al. 2020). Regarding potential hepatic impairment, the review reported liver or kidney transplants in 4/243 (1.6%) cases without mentioning the underlying reason for the solid organ transplant or the exact number of liver versus kidney transplants. The same review mentions hepatomegaly in 70/243 (28.8%) and hepatobiliary disorders in 17/243 (7%) of APDS subjects. In a report of 38 APDS subjects from the United States Immunodeficiency Network (USIDNET) Registry and 2 additional U.S. APDS subjects from Mt Sinai Hospital, one subject with a history of ulcerative colitis and sclerosing cholangitis was reported to require a liver transplant. One subject in the USIDNET Registry was reported to have developed EBV hepatitis,

which was not previously noted in the literature. Hepatomegaly was reported in 11/40 (27.5%) of these APDS subjects.

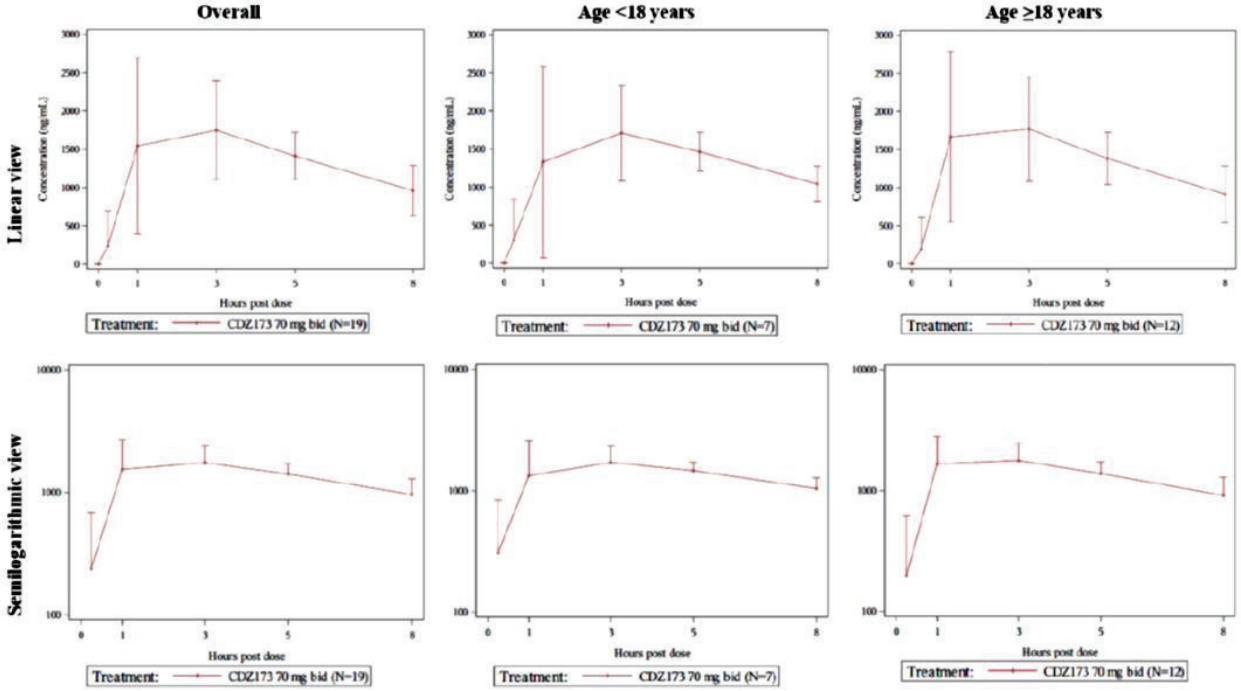
A few articles have been published on the European APDS population, mainly from the subjects captured in the European Society for Immunodeficiencies (ESID) Registry (Elgizouli et al. 2016; Coulter et al. 2017; Maccari et al. 2018). No hepatic pathology was reported in a 2015 review of APDS subjects from the ESID Registry that included 5 APDS subjects in the population of 669 subjects with immunodeficiency (Elgizouli et al. 2016). Another publication in 2018 of APDS subjects exposed to rapamycin in the ESID Registry identified one subject that had rapamycin therapy interrupted due to liver toxicity and two subjects developed autoimmune hepatitis out of 77 APDS subjects enrolled in the registry with clinical and immunological information available in 68 of those subjects (Maccari et al. 2018). Another publication from Europe in 2017 of 53 APDS subjects, including 25 subjects previously published and 28 subjects published for the first time (Coulter et al. 2017), reported hepatomegaly in 24/53 (45%) with one case of liver disease with cholangitis with associated cryptosporidium infection.

Reviewer Comment: *Considering that metabolism is the predominant pathway for leniolisib elimination, the Applicant has proposed in the label that leniolisib is not recommended in the moderate-to-severe hepatically impaired population. However, the data on prevalence of hepatic impairment provided by the Applicant suggests that hepatomegaly is commonly reported as one of the noninfectious phenotypic features of APDS in 28.8% to 45% of patients, therefore the possibility of occurrence of some degree of hepatic impairment in patients with APDS cannot be completely ruled out. To address this, a PMR will be issued to the Applicant to conduct a hepatic impairment clinical study to assess the impact of moderate and severe hepatic impairment on the pharmacokinetics of leniolisib. Renal clearance is a minor elimination pathway for leniolisib, with only approximately 6% of elimination occurring renally. Therefore, the Applicant's justification to not conduct a renal impairment study and not excluding subjects with renal impairment in the label appears reasonable.*

Adolescents

In Part 2 of Study 2201, leniolisib plasma PK profiles appeared comparable between age groups <18 years and ≥18 years (Figure 6 and Table 9).

Figure 6. Mean (SD) Leniolisib Plasma Concentration-Time Profile on Day 1 of Twice Daily Dosing of 70 mg Leniolisib Capsule in Subjects With APDS - Stratified by Age (Study 2201, Part 2)

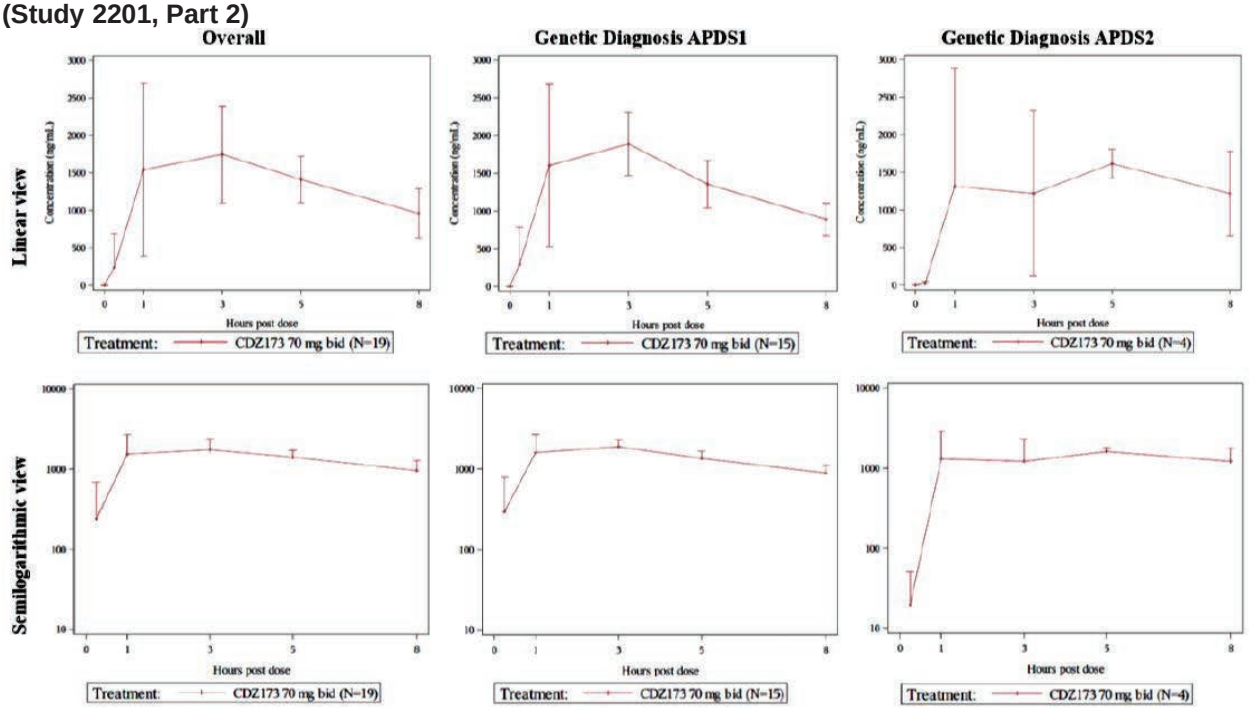


Source: CSR Study 2201 Part 2, Figure 14.2-1.1.1b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Other Intrinsic Demographic Factors

PopPK analysis of data from healthy participants in Study 2101 did not determine age or sex to be a significant predictor of leniolisib PK. In subjects with APDS, leniolisib plasma PK characteristics appeared comparable between subjects aged <18 years and ≥18 years receiving leniolisib 70 mg twice daily. Based on the Pop PK results, V_c/F and V_p/F were 20% lower relative to a 70 kg individual for participants weighing 50 kg and 31% higher relative to a 70-kg individual for participants weighing 105 kg. The same popPK model indicated that the CL/F of Japanese descents was approximately 37% higher than non-Japanese descents. However, the results should not be over-interpretated by considering there were only 6 Japanese descents enrolled in Study 2011 and a different formulation ((b) (4) was used in Japanese descents compared to other subjects in Part 1. Leniolisib plasma PK characteristics appeared comparable between subjects with genetic diagnosis of APDS1 and APDS2 receiving leniolisib 70 mg twice daily (Figure 7).

Figure 7. Mean (SD) Leniolisib Plasma Concentration-Time Profile on Day 1 of Twice Daily Dosing With 70 mg Leniolisib Capsule in Subjects With APDS - Stratified by Genetic Diagnosis (Study 2201, Part 2)



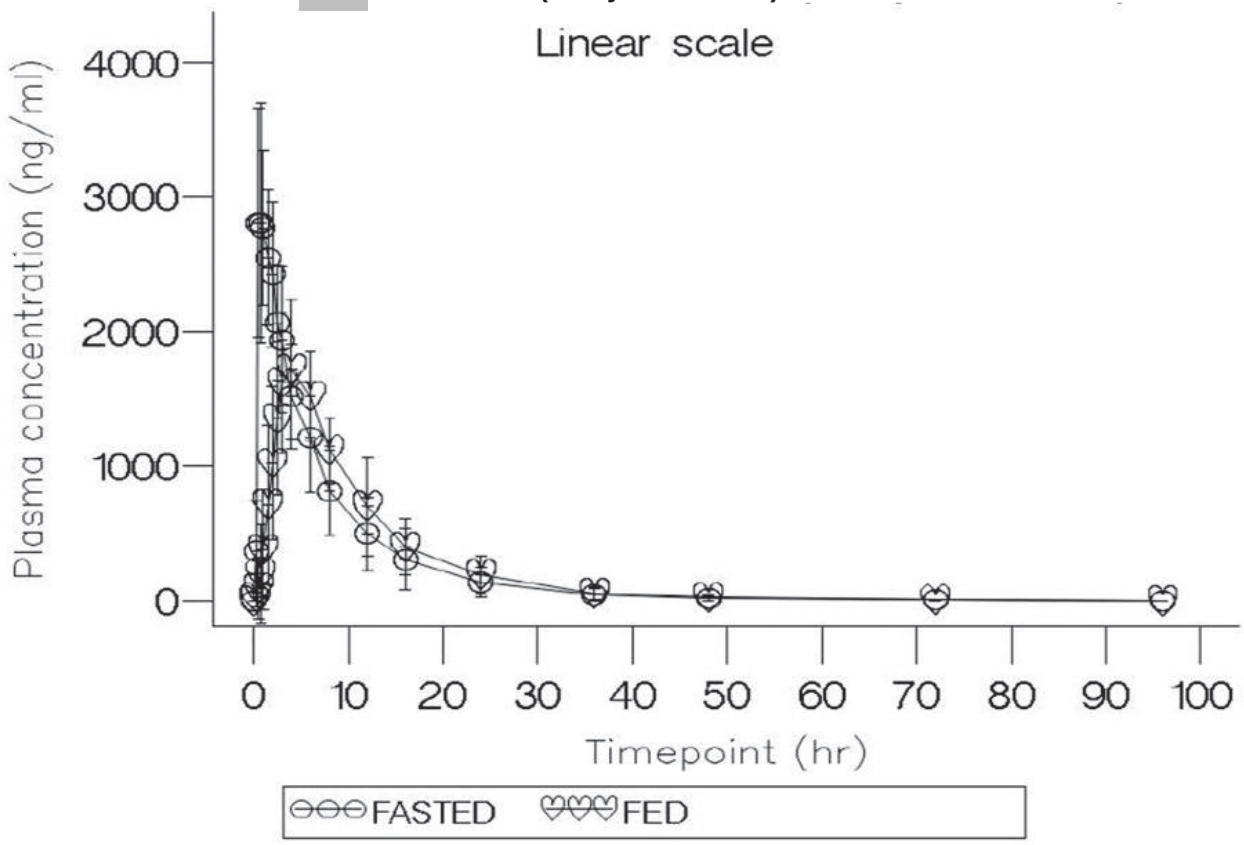
Source: CSR Study 2201 Part 2, Figure 14.2-1.1.2b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CSR, clinical study report; N, total number of subjects; SD, standard deviation

Are There Clinically Relevant Food-Drug or Drug-Drug Interactions, and What is the Appropriate Management Strategy?

Food Effect

The effect of food on leniolisib was assessed in Study 2101 Part 2 where healthy participants received a single dose of leniolisib 70 mg capsule ([(b) (4)] under fasted and fed conditions. Co-administration with a high-fat meal decreased the absorption rate, as indicated by a delayed time to maximum concentrations (T_{max}) from approximately 1 to 4 hours postdose. High-fat meal reduces C_{max} by approximately 40% but with little effect on AUC_{inf} (fed/fasted geometric mean ratio [90% CI] of 0.59 [0.53, 0.66] and 1.00 [0.93, 1.07], respectively) (Figure 8 and Table 11).

Figure 8. Arithmetic Mean (SD) Leniolisib Plasma Concentration-Time Plot (N=12) Under Fed and Fasted Conditions With (b) (4) Formulation (Study 2101 Part 2)



Source: Figure 11-3 on page 91 from Study 2101 CSR
Abbreviations: (b) (4); CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Table 11. Assessment of Food Effect on Leniolisib PK Parameters With (b) (4) Formulation (Study 2101 Part 2)

	CDZ173 70mg Fed		CDZ173 70mg Fasted			
Parameter	N	Geometric least squares mean[1]	N	Geometric least squares mean[1]	Ratio (Fed to Fasted)[2]	90% confidence interval for ratio[2]
AUC0-12h (hr*ng/mL)	12	13083.12	12	15097.80	86.66	(79.33, 94.66)
AUClast (hr*ng/mL)	12	19105.35	12	19214.41	99.43	(92.40, 107.00)
AUCinf (hr*ng/mL)	12	19223.28	12	19288.30	99.66	(92.52, 107.35)
Cmax (ng/mL)	12	1886.32	12	3186.31	59.20	(52.92, 66.23)

Source: Table 11-10 on page 93 from Study 2101 CSR
Abbreviations: $AUC_{0\text{-}12h}$, area under the curve from 0 to 12 hours; AUC_{inf}, area under the curve to infinity; AUC , area under the curve to the last quantifiable time point; C_{max}, maximum plasma concentration; (b) (4); CSR, clinical study report; PK, pharmacokinetic; N, total number of subjects.

The Applicant states that that C_{max} achieved at the fed state (1890 ng/mL) following the therapeutic dose (70 mg) remains well above the plasma concentration that is required for 50%

of maximal effect (EC_{50}) for ex vivo pAkt inhibition in B cells (~200 to 300 ng/mL, see Table 10). the food effect on leniolisib exposure is not expected to compromise on-target pathway inhibition, and leniolisib may be taken with or without food.

However, it should be noted that food effect study was conducted with the capsule dosage form (i.e., (b) (4) and the food effect on the to-be-marketed tablet dosage form (i.e., FCT) has not been evaluated in leniolisib clinical program. Therefore, the Applicant provided following justifications:

- The to-be-marketed formulation (i.e., FCT) is not significantly different from the clinical trial formulation (i.e, HGC (b) (4)). The major difference is (b) (4) (Table 6).
- (b) (4) dissolution profile of HGC dosage form compared to FCT dosage at pH 4.0 was not translated to in vivo differences in leniolisib pharmacokinetics (AUC and C_{max}) as demonstrated in the bioequivalence Study LE1101.
- Bioequivalence was established between the capsule and tablet dosage forms in Study LE1101.
- Furthermore, leniolisib was administered without regard to food in the pivotal Study 2201. In the extension study 2201E, both capsule and tablet dosage forms were taken irrespective of food as indicated by a food intake survey conducted by the Applicant. The survey shows that the majority of the subjects in Study 2201E1 took leniolisib with food.

The review team considers the above justifications are acceptable. Further details regarding the applicability of the results of food effect with the capsule formulation to the to-be-marketed tablet formulation can be found in Section 6.3.1.

Drug-Drug Interaction

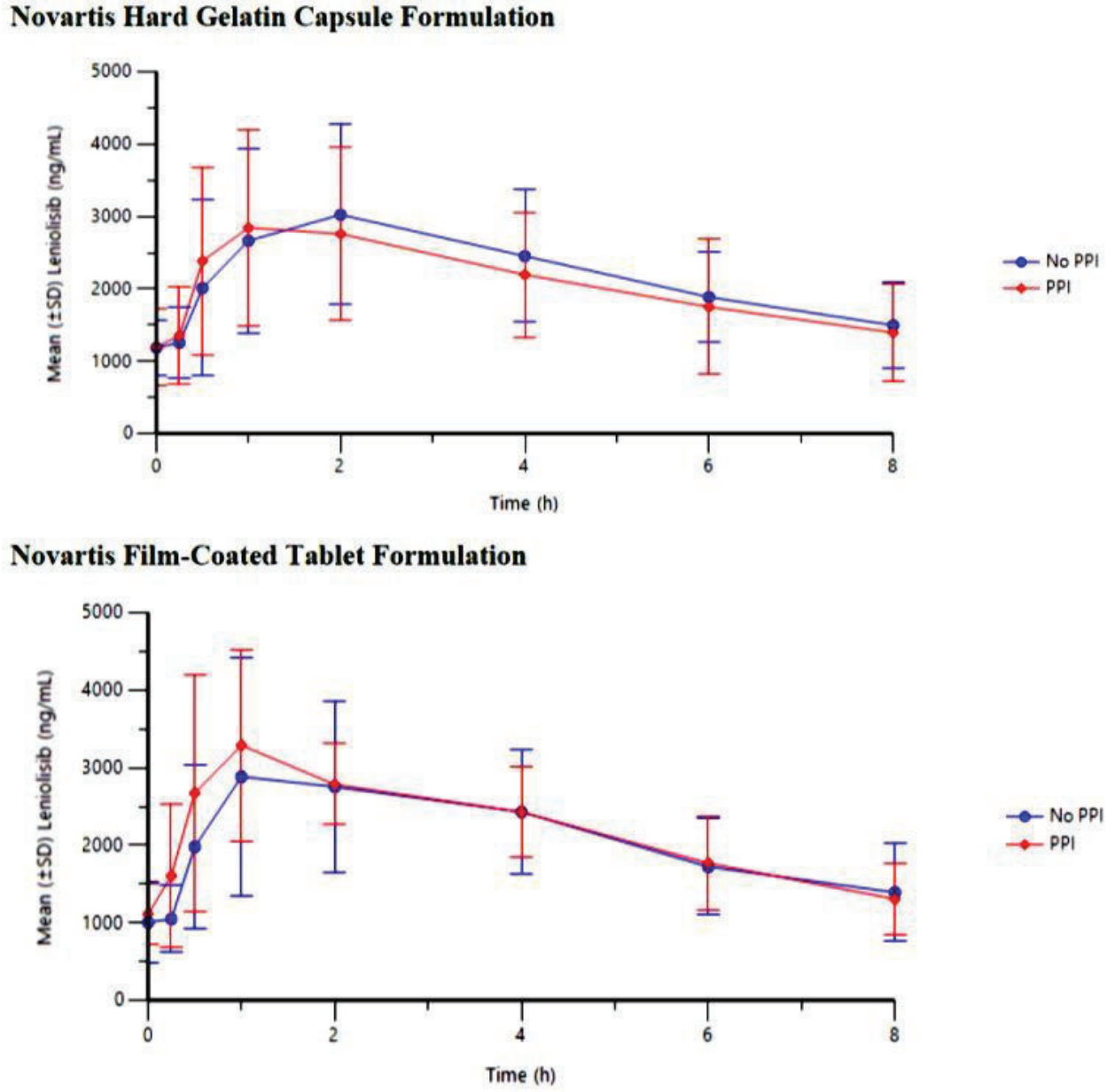
Drug-Drug Interactions as Victim

In a clinical DDI Study 2102, co-administration with itraconazole (a strong dual inhibitor of CYP3A4 and P-gp) increased leniolisib AUC_{inf} by approximately 2-fold and C_{max} by 25%, while co-administration with quinidine (a strong dual inhibitor of CYP2D6 and P-gp) did not affect leniolisib exposure. Hence, leniolisib may be co-administered with CYP2D6 and P-gp inhibitors without dose adjustment, but concomitant use with strong CYP3A4 inhibitors should be avoided. PBPK simulations were used to assess how leniolisib PK will be affected by co-administration with erythromycin (a moderate CYP3A4 inhibitor), rifampicin (a strong CYP3A4 inducer), and efavirenz (a moderate CYP3A4 inducer). The model predicted increased leniolisib AUC_{0-12} and C_{max} in the presence of erythromycin (approximately 60% and 30% increase, respectively) and decreased leniolisib AUC_{0-12} and C_{max} in the presence of rifampicin

(approximately 80% and 50% decrease, respectively) and efavirenz (approximately 60% and 30% decrease, respectively). Hence, concomitant use with moderate CYP3A4 inhibitors may be allowed since doses upto 140 mg BID were well tolerated in the phase 1 MAD study in healthy subjects. Based on the PBPK results, co-administration of strong or moderate CYP3A4 inducers should be avoided.

Leniolisib exhibits pH-dependent solubility, with lower solubility at higher pH-values. Thus, gastric acid-reducing agents may impair the absorption of leniolisib. No dedicated studies evaluating the effects of gastric acid reducing agents on leniolisib PK have been conducted. However, a post hoc analysis of interim data from Study 2201E1 showed nearly comparable plasma leniolisib concentration-time profiles between subjects with APDS who reported taking proton pump inhibitors (PPIs) and those who did not (Figure 9**,** Table 12). Thus, leniolisib may be used concomitantly with PPIs or histamine-2 receptor antagonists with no dose adjustment.

Figure 9. Mean (SD) Leniolisib Plasma Concentration-Time Profile Following Twice Daily Dosing of 70 mg Leniolisib in Subjects With APDS, Stratified by Formulation and Concomitant Use of PPI (Study 2201E1)



Source: Summary of Clinical Pharmacology Studies, Figure 24
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; CSR, clinical study report; PPI, protein pump inh bitor; SD, standard deviation.

Table 12. Summary of Statistical Assessment for Effect of PPI Use on Leniolisib Exposure Following Twice Daily Dosing of 70 mg Leniolisib in Subjects With APDS, Stratified by Formulation (Study 2201E1)

Formulation Parameter	Treatment	n	LS Mean	Treatment Comparison	
				LS Mean Ratio (%)	90% Confidence Interval
Novartis hard gelatin capsule formulation					
C_{max} (ng/mL)	PPI use reported (test)	11	3260	101.93	80.54, 129.00
	No PPI use reported (reference)	19	3198		
AUC_{0-8} (ng*h/mL)	PPI use reported (test)	10	16478	98.25	77.22, 125.00
	No PPI use reported (reference)	19	16945		
Novartis film-coated tablet formulation					
C_{max} (ng/mL)	PPI use reported (test)	9	3628	109.07	88.61, 134.25
	No PPI use reported (reference)	18	3327		
AUC_{0-8} (ng*h/mL)	PPI use reported (test)	9	17033	104.03	84.59, 127.93
	No PPI use reported (reference)	17	16373		

Source: Summary of Clinical Pharmacology Studies, Table 16.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{0-8}, area under the curve from 0 to 8 hours; C_{max}, maximum plasma concentration; CSR, clinical study report; LS, least squares; n, number of subjects in category; PPI, protein pump inhibitor.

Drug-Drug Interactions as Perpetrator

In vitro, leniolisib was found to potentially inhibit CYP1A2, CYP2C8, CYP2C9, CYP2C19, CYP2D6, UGT1A1, OATP1B1, OATP1B3, OCT1, OCT2, OAT1, OAT3, MATE1, MATE2K, BCRP, and BSEP and potentially induce CYP2B6, CYP2C9, and CYP3A4. Clinically relevant interactions at the therapeutic dose of leniolisib (70 mg twice daily) could not be excluded for inhibition of CYP1A2, UGT1A1, BCRP, OATP1B1, OATP1B3, OCT2, and MATE2K based on the anticipated $I_{max,ss,u}$; the observed IC_{50} and K_i values; and the calculated R1, R2, R3, and R values.

While little to no reversible inhibition of CYP1A2 by leniolisib was observed in vitro, time-dependent inhibition was observed and the calculated R_2 value was ≥1.25, indicating potential for clinically relevant DDI with CYP1A2 substrates. Because no dedicated clinical studies have been conducted to assess the effects of leniolisib on the PK of CYP1A2 substrates, it is recommended in the current label to avoid concomitant use of leniolisib with CYP1A2 substrates with narrow therapeutic indices. The use of in vitro methods to predict the effects of time-dependent inhibition of CYP1A2 may often over-predict the in vivo situation. Therefore, the Applicant should conduct a clinical DDI study to characterize the effect of leniolisib on CYP1A2 substrates in humans.

No dedicated clinical studies have been conducted to evaluate the effects of concomitant use of leniolisib on the PK of BCRP, OATP1B1, and OATP1B3 substrates (e.g., statins). Given the in vitro results that showed that leniolisib can inhibit BCRP and OATP1B1/B3 transporters, the

Applicant is proposing to avoid the use of leniolisib with substrates of BCRP or OATP1B1/3 in the current label. However, this may lead to exclusion of multiple drugs (e.g., all the statin drugs are substrates of OATP1B). Therefore, the Applicant should conduct a clinical DDI study to assess the effect of leniolisib on BCRP and OATP1B1/3substrates in humans to fully characterize this DDI potential.

The in vitro results showed that the leniolisib can inhibit OCT2 and MATE2K transporters and lead to potential clinically relevant DDIs when co-administered with OCT2 and MATE2K substrates. Limited renal elimination of leniolisib does not necessarily mean it cannot inhibit renal transporters. Further, PBPK modeling cannot be used for DDI risk evaluation because the current metformin substrate model (PBPK software library) has not been adequately validated regarding the contributions of the transporters MATEs and OCTs to metformin elimination. Therefore, the Applicant should assess the effect of leniolisib on OCT2 and MATE substrates in a clinical DDI study.

Based on PBPK model-based simulations, co-administration with steady-state leniolisib is expected to result in decreased exposure of midazolam (a prototypical CYP3A4 substrate) (see Appendix 16.3.5.1 for details on PBPK modeling results). The fifth percentile of the simulated ratios for midazolam exposure (AUC_{inf} and C_{max}) with/without leniolisib fell below 0.8 and ranged from 0.32 to 0.77, indicating that leniolisib is a weak-to-moderate inducer of CYP3A4. However, the model could not be used to accurately predict the magnitude of the effect of leniolisib on the PK of midazolam due to the uncertainty associated with the in vitro-to-in vivo extrapolation of the CYP3A4 induction parameter values. No substantial CYP3A4 auto-induction was observed in healthy participants receiving leniolisib at doses up to 140 mg twice daily for 14 days (Study 2101). However, upon a longer dosing period in subjects with primary Sjögren's syndrome (Study 2203) and subjects with APDS (Study 2201, Part 2) at 70 mg twice daily, the mean C_{trough} was lower at later time points (Days 57 and 85) compared to earlier time points (Day 29). Hence, CYP3A4 autoinduction with prolonged use of leniolisib cannot be excluded. Therefore, the Applicant should assess the effect of leniolisib on sensitive CYP3A4 substrates in a clinical DDI study.

Therefore, to address the concerns mentioned above, a PMR will be issued to the Applicant to conduct a cocktail DDI clinical study to assess the effect of leniolisib on the pharmacokinetics of CYP1A2 substrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and MATE/ OCT2 substrate (metformin).

In a clinical DDI Study 2104, co-administration of leniolisib at steady state with CYP3A4, SULT1E1, and UGT1A1 substrate oral contraceptives (ethinylestradiol + levonorgestrel) increased ethinylestradiol exposure by approximately 30% and had no effect on levonorgestrel exposure. The increase in ethinylestradiol exposure is unlikely to compromise the efficacy of a combined oral contraceptive composed of ethinylestradiol and levonorgestrel. Hence, leniolisib may be co-administered with oral contraceptives without dose adjustment.

7. Sources of Clinical Data and Review Strategy

7.1. Table of Clinical Studies

To support the proposed indication, the Applicant submitted clinical data from study 2201, which consisted of two parts, and study 2201E1, an open label extension (OLE) (Table 13).

Table 13. Summary of Clinical Studies Included in This Submission

Trial Identity	NCT no.	Trial Design	Regimen/ Schedule/Route	Study Endpoints	Treatment Duration/ Follow Up	No. of Subjects Enrolled	Study Population	No. of Centers and Countries
Controlled studies to support efficacy and safety								
CCDZ173X2201 Part 2	02435173	R, DB, PC, MC	Leniolisib 70 mg PO BID Placebo PO BID	Change from baseline in the log10 transformed SPD in the index lesions selected as per the Cheson methodology from MRI/CT imaging Change from baseline in naïve B cells out of total B cells	12 weeks	Leniolisib 21 Placebo 10	Adults and adolescents with APDS	10 centers 9 countries
Studies to support safety								
CCDZ173X2201E1 (Long-term extension)	02859727	OLE, NR, MC	Leniolisib 70 mg PO BID	AEs, physical examination, vital signs, ECG, safety laboratory	6 years	37	Adults and adolescents with APDS from study CCDZ173X2201 or who received PI3Kδ inhibitors	8 centers 7 countries
Other studies pertinent to the review of efficacy or safety								
CCDZ173X2201 Part 1	02435173	OL, single arm, MAD, DRF, MC	Leniolisib 10 mg PO BID x 4W Leniolisib 30 mg PO BID x 4W Leniolisib 70 mg PO BID x 4W	PK/PD/safety	12 weeks	6	Adults and adolescents with APDS	3 centers 4 countries

Source: Clinical Reviewer generated.
Abbreviations: AEs, adverse events; : APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CT, computed tomography; DB, double-blind; DRF, dose range finding; ECG, electrocardiogram; MAD, multiple ascending dose; MC, multicenter; MRI, magnetic resonance imaging; NCT, national clinical trial; NR, non-randomized; OL, open label; OLE, open label extension; PC, placebo controlled; PD, pharmacodynamic; PK, pharmacokinetic; PO, orally; PI3Kδ; phosphoinositide 3-kinase-δ; R, randomized; SPD, sum of product of diameters.

7.2. Review Strategy

The Applicant submitted clinical data from Studies 2201, Parts 1 and 2, and 2201E1 to support the efficacy and safety of leniolisib in the proposed indication of treatment of APDS in patients age ≥ 12 years. Part 1 included a 12-week single-arm, open-label, multiple ascending dose, study with 4 week treatment periods to evaluate safety/tolerability and PK of leniolisib at three different dosing regimens. Part 2 consisted of a 12-week multicenter, randomized, double-blind, placebo-controlled study and is the focus of the efficacy and safety review. Trial 2201E1 was an OLE to Part 2 where long-term safety was further assessed. The protocol design and study results for each trial are outlined in Sections 8.1.1, 8.1.3, and 8.1.5. The safety data are reviewed in Section 8.2. A detailed review of the clinical pharmacology program for leniolisib is located in Section 6 by Dr. Nisha Kwatra.

Data from Trial 2201, Part 2 provide the primary evidence evaluating the efficacy of leniolisib for the treatment of APDS. These data are presented in Sections 8.1.2, 8.1.4, and 8.1.6 by FDA biostatistician, Dong-Hyun Ahn, Ph.D., who confirmed the Applicant's efficacy analyses and generated tables and figures for this review.

For the evaluation of safety, FDA medical officer, Katherine Clarridge, M.D., analyzed data from Trials 2201, Parts 1 and 2, and 2201E1 using JMP, JMP Clinical, and the Office of Computational Science Analysis Toolbox. The safety results presented in 8.2 represent the medical officer reviewer's own analyses.

8. Statistical and Clinical and Evaluation

8.1. Review of Relevant Individual Trials Used to Support Efficacy

8.1.1. CCDZ173X2201, Part 1

Title and Administrative Information

Trial CCDZ173X2201, referred to as 2201, Part 1 was the first part of the trial entitled "An open-label, non-randomized, within-subject dose-finding study followed by a randomized, subject, investigator and sponsor-blinded placebo controlled study to assess the efficacy and safety of CDZ173 (Leniolisib) in subjects with APDS (Activated phosphoinositide 3-kinase delta syndrome/ p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency)." The trial was conducted at three centers, in the US, Czech Republic, and the Netherlands, from December 24, 2015 to October 17, 2016.

Trial Objective

The primary objective was to assess the dose-PD and PK/PD relationships of leniolisib in subjects with APDS.

Trial Design

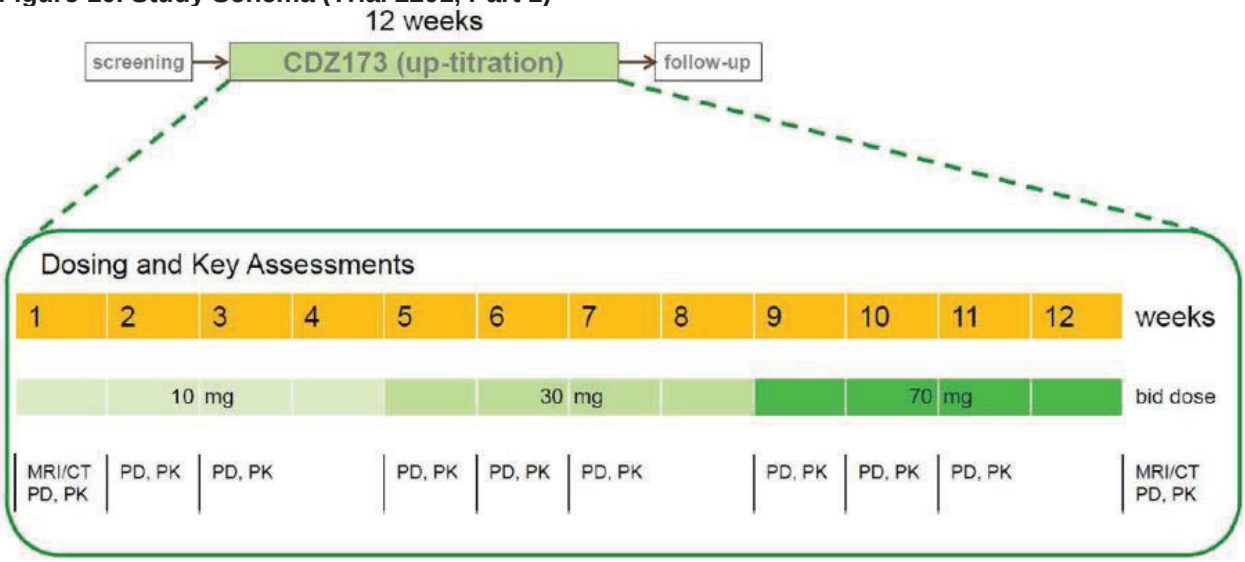
Trial 2201 Part 1 was a phase 2/3, 12-week, single-arm, open-label, within-subject, up-titration, dose-finding study evaluating the safety, tolerability, and PK/PD of leniolisib 10 mg, 30 mg, and 70 mg delivered orally twice daily in subjects ≥ 12 years of age diagnosed with APDS. Each subject received the starting dose of leniolisib 10 mg, followed by 30 mg and 70 mg, twice daily for 4 weeks at each dose level, respectively.

Details of each study visit are summarized below, and the study design is presented in Figure 10.

- During the Screening period (Day -50 to Day -2), informed consent and demographic data were collected. Eligibility was assessed and reviewed, physical exam was performed, vital signs, laboratory evaluations, ECGs were obtained, and subjects underwent an MRI or CT scan.
- At the Baseline Visit (Day -1), eligibility was reviewed. The baseline visit period was extended by three days if required for logistical purposes to complete any necessary screening assessments that were not performed during the Screening period such as the MRI or CT scan.

- The first day of dosing started on Day 1 of the Treatment Period (Day 1 to Day 84). Subjects were started on leniolisib 10 mg taken orally twice daily until Day 28. Prior to escalation to the next dose level a safety review and review of the PK/PD data up to seven days after the first dose were assessed. Subjects followed with leniolisib 30 mg taken orally twice daily starting on Day 29 and continued to Day 56. After a safety review and PK/PD data review up to Day 36, subjects were started on leniolisib 70 mg taken orally twice daily from Day 57 to Day 84. Again, safety and PK/PD data reviews were performed at the end of the third dosing period. Assessments completed on Day 84 included the end of treatment MRI or CT scan.
- The End of Study Visit occurred on Day 112, four weeks after the last day of dosing, at which time final safety assessments were performed and subjects were offered participation in the OLE.

Figure 10. Study Schema (Trial 2201, Part 1)



Source: Applicant's Clinical Trial Report for Study 2201 Part 1, Section 9.1.
Abbreviations: BID, twice daily; CT, computed tomography; MRI, magnetic resonance imaging; PD, pharmacodynamic; PK, pharmacokinetic.

Study Population

Subjects between the ages of 12 and 75 who had a documented APDS-associated genetic PI3K delta mutation were enrolled in the trial. Subject eligibility was established at screening and reviewed at the Baseline Visit. The key inclusion and exclusion criteria were the same as those described in Trial CCDZ173X2201, Part 2, described in Section 8.1.1.

Concomitant Therapy

Prohibited therapies and drugs to be used with caution during the course of the study were similar to CCDZ173X2201, Part 2, with the exception that systemic glucocorticoids above 10 mg rather than 25 mg prednisone or equivalent per day were prohibited.

Study Assessments

Study assessments were performed as shown in Table 14 with treatment divided into three periods where subjects received consecutive treatments of 10 mg BID for four weeks, 30 mg BID for four weeks, followed by 70 mg BID for four weeks. Imaging, immunophenotyping, and patient reported outcome assessments were performed as described in Section 8.1.4.

Table 14. Schedule of Assessments for Trial 2201, Part 1

Study Phase	SCR	BL	Dose 1 (10 mg)				Dose 2 (30 mg)				Dose 3 (70mg)				EOS
Visit numbers[a]	1	2	101	102	103	199	201	202	203	299	301	302	303	304	499
Days	-50 to -2	-1	1	8	15	28	29	36	43	56	57	64	71	84	112
Visit windows		-3 days*		+/- 1 day	+/- 1 day		+/- 1 day	+/- 1 day	+/- 1 day		+/- 1 day	+/- 1 day	+/- 1 day	+ 3 days	+/- 2 days
Written informed consent	X														
Inclusion/exclusion criteria	X	X[b]													
Relevant medical history/current medical conditions	X	X[b]													
Demography (incl. smoking status)	X														
Physical examination	X	X					X				X			X	X
Hepatitis and HIV screen	X														
Pregnancy test[c]	X[c]	X					X				X			X	X
Vital signs and body measurements															
Body height	X														
Body weight	X	X													X
Body temperature	X	X	X	X	X		X	X	X		X	X	X	X	X
Blood pressure / pulse rate[d]	X	X	X[e]	X	X		X[e]	X	X		X[e]	X	X	X	X
ECG evaluation (triplicates)	X	X	X[e]	X	X		X[e]	X	X		X[e]	X	X	X	X
Hematology, blood chemistry, urinalysis	X	X	X	X	X		X	X	X		X	X	X	X	X
Tuberculosis test	X														
Serum immunoglobulins		X	X				X				X			X	
EBV/CMV test		X	X				X				X			X	
DNA blood collection (consent optional)							X[f]								
PK blood collection			X[g]	X	X		X[g]	X	X		X[g]	X	X	X	
PD blood collection		X	X[h]	X	X		X[h]	X	X		X[h]	X	X	X	

Study Phase	SCR	BL	Dose 1 (10 mg)				Dose 2 (30 mg)				Dose 3 (70mg)				EOS
Immunophenotyping[i]		X	X				X				X			X	
Soluble biomarker/miRNA[j]		X	X				X				X			X	
PBMC for functional T cell assay		X												X	
RNA blood collection		X												X	
Drug administration record[k]			X	X	X	X	X	X	X	X	X	X	X	X	
MRI or CT scan	X[l]													X[m]	
SF-36/WPAI-CIQ questionnaires		X					X				X			X	
Physician/patients global assessment		X					X				X			X	
Patient narratives														X	
Phase/study completion		X				X				X				X[n]	X
Comments	As required														
Adverse events (comment)	As required														
Concomitant meds/therapies	As required														

Source: Modified from Applicant's Clinical Protocol for Study 2201 Part 1, Section 8.
Note: X means any blood collection unless otherwise mentioned pre-dose is meant
[a] Visit structure given for internal programming purpose only.
[b] Review of inclusion and exclusion criteria and current medical conditions is required at baseline evaluation.
[c] Serum pregnancy test required screening, urine pregnancy test at baseline and further time points.
[d] Following 3 minutes of sitting rest, sitting blood pressure and pulse measurements will be taken.
[e] Blood pressure/pulse rate and ECG measurement at pre-dose, 1h, 3h and 5 h post-dose.
[f] DNA blood collection informed consent (optional) can be granted during the whole study participation except screening and the sample should be collected accordingly.
[g] PK samples to be taken at pre-dose, 0.25h, 1h, 3h, 5h and 8h post-dose.
[h] PD samples to be taken at -1h, 1h, 3h and 8h post-dose.
[i] This sample to be taken at the -1h timepoint with the PD blood collection except at BL and Day 84 where it should be taken pre-dose.
[j] This sample to be taken at the -1h timepoint except at BL and Day 84 where it should be taken pre-dose.
[k] Twice daily dosing leniolisib except on Day 28 and 56 when the evening dose was to be omitted.
[l] The duration between the scan and first dosing was less than 21 days; the scan may be performed during the screening or baseline period..
[m] The Day 84 scan can be taken up to 21 days post Day 84 visit.
[n] Phase completion page on Day 84 will be captured under Visit number 399.
[o] Baseline visit may be extended up to D-4 if required for logistical reasons. All screening assessments must be completed prior to this visit.
Abbreviations: BL, baseline; CIQ, classroom impairment questions; CT, computed tomography; CMV, cytomegalovirus; DNA, deoxyribonucleic acid; EBV, Epstein-barr virus; ECG, electrocardiogram; EOS, end of study; HIV, human immunodeficiency virus; MRI, magnetic resonance imaging; miRNA, micro ribonucleic acid; PD, pharmacodynamic; PK, pharmacokinetic; PMBC, peripheral blood mononuclear cell; RNA, ribonucleic acid; SCR, screening; SF-36; short form health survey; WPAI, work productivity and activity questionnaire.

Trial Endpoints

The primary endpoints for Part 1 included:

- Number of subjects with AEs and SAEs, including significant changes from baseline in physical findings, vital signs, electrocardiograms, and laboratory values qualifying and reported as AEs. The number of subjects in each category (AEs and SAEs) were reported per dose level: leniolisib 10 mg from Day 1 to Day 28, leniolisib 30 mg from day 29 to day 56 and leniolisib 70 mg from day 57 to day 84
- Single and multiple dose concentrations of leniolisib at days 1, 29, 57, and 84
- Percentage of inhibition of unstimulated and stimulated pAkt Levels in B cell inhibition in unstimulated and stimulated whole blood in order to determine the appropriate dose for Part 2 at days 1, 29, 57, and 84

Secondary endpoints consisted of changes from baseline in SF-36 and WPAI-CIQ scores, and of visual analogue scale scores for PGA and PtGA as described in Section 8.1.3 at Week 12. Additionally, summary statistics were provided for the change from baseline in the log 10 transformed SPD of index lesions, spleen, and liver as well B cell and T cell immunophenotype, serum immunoglobulins, soluble biomarkers, high sensitivity C-reactive protein (hsCRP), and lactate dehydrogenase (LDH) also at Week 12.

8.1.2. Study Results

Compliance With Good Clinical Practices

Trial 2201, Part 1 was conducted in accordance with the International Council for Harmonisation (ICH) Harmonised Tripartite Guideline for Good Clinical Practice (GCP) E6. A statement of compliance with Good Clinical Practices is located in the clinical trial reports.

Financial Disclosure

Details of financial disclosure are presented in Section 16.2.

Subject Disposition

A total of six subjects diagnosed with APDS were enrolled in Trial 2201, Part 1. All six subjects completed the trial according to the study protocol and received leniolisib 10 mg, followed by 30 mg and 70 mg, twice daily for 4 weeks at each dose level, respectively.

Protocol Violations/Deviations

One subject reported eight protocol deviations during the trial due to enrollment prior to the availability of the full set of required screening lab results and loss of one set of blood samples at a subsequent visit.

Demographic Characteristics

Table 15 **provides baseline characteristics for the 6 subjects enrolled in Trial 2201 Part 1.**

Table 15. Baseline Demographic and Clinical Characteristics, Trial 2201, Part 1

Subgroup	Leniolisib (N = 6) n (%)
Sex	
Female	2 (33)
Male	4 (67)
Age (years), n (%)	
Mean (SD)	22.2 (10)
Median	22
Range	16, 31
Age group, n (%)	
< 18	2 (33)
≥ 18	4 (67)
Race	
Caucasian	6 (100)

Subgroup	Leniolisib (N = 6) n (%)
Ethnicity	
Hispanic or Latino	1 (17)
Not Hispanic or Latino	3 (50)
Not reported	2 (33)
Baseline medications	
Glucocorticoids	0
Immunoglobulin replacement	6 (100)
Previous rapamycin use	1 (17)

Source: Clinical Reviewer Generated using OCS Analysis Studio, Custom Table Tool.
Note: Dataset: Demographics; Filter: SAFFL = 'Y', TRT01A = 'CDZ173 10 mg bid / 30 mg bid / 70 mg bid' or 'Placebo'.
Abbreviations: N, total number of subjects; n, number of subjects in category; SD, standard deviation

Other Baseline Characteristics (e.g., Disease Characteristics, Important Concomitant Drugs)

Of the six subjects enrolled in Trial 2201, Part 1, all were diagnosed with APDS1.

Treatment Compliance, Concomitant Medications, and Rescue Medication Use

Subjects were instructed to take the study drug exactly as prescribed and to contact the investigator if unable to take the study drug for any reason. Subjects were provided individual diary cards to record each administration of study drug and were advised to return unused medication at specific time points for drug accountability and verification. Furthermore, sequential blood samples were collected in all subjects up to 8 hours after the first dose administration (Day 1) and after the first dose following each escalation to the next dose level (Day 29 and 57) with results consistent with appropriate administration. There were no recorded study drug dose adjustments other than the planned increase per protocol in Part 1.

Primary Endpoints

The primary objectives of Part 1 were to evaluate the safety/tolerability and PK/PD of leniolisib in subjects with APDS to inform dose selection for Part 2. The endpoint used to make this assessment was the single and multiple dose concentrations of leniolisib required to inhibit pAkt in unstimulated and stimulated whole blood the results for which are described in Section 6.3.1. The proposed BID dosing regimen was chosen based on a maximum reduction in both stimulated and non-stimulated pAkt-positive B cells observed one-hour post-dose that returned to baseline level after 12 hours. The 30- and 70-mg dose levels had similar maximum effect, but inhibition was better sustained over time with the 70 mg dose. Relevant findings related to safety/tolerability are included in Section 8.2.3.

Secondary and Exploratory Endpoints

Imaging for Lymphoproliferation

All subjects enrolled in Trial 2201, Part 1 experienced a reduction in spleen volume and SPD of index lesions. Four out of the six subjects experienced a decrease in liver volume as outlined in Table 16.

Table 16. Reduction in Spleen Volume, Sizes of Index Lesions, and Liver Volume, Trial 2201, Part 1

Subject	Spleen Volume (% Change From Baseline)	Index Lesions SPD (% Change From Baseline)	Liver Voume (% Change From Baseline)
(b) (6)	-26	-51	-11
	-39	-13	-2
	-57	-65	7
	-36	-33	-5
	-39	-48	-3
	-37	-31	5

Source: Applicant's Clinical Trial Report for Study 2201 Part 1, Section 11.4.
Abbreviations: SPD, sum of product of diameters

B cell Immunophenotype

Transitional and naïve B cells were the focus of the B cell subset analysis. All subjects except one experienced a decrease in transitional B cells out of total B cells over the course of the treatment period. All subjects experienced an increase in percentage of naïve B cells out of total B cells during the course of treatment.

T cell Immunophenotype

Senescent T cells, as represented by CD57+CD8+ and PD-1+CD4+ T cells, decreased gradually in all subjects over the course of treatment.

Serum Immunoglobulins

Consistent with the laboratory findings of APDS, four of the six subjects presented with elevated serum IgM at screening. Serum IgM decreased for all subjects over the course of treatment. There were no clear trends with any of the other Ig classes.

Soluble Biomarkers

The serum levels of CXCL13/BLC, CXCL10/IP-10, CCL22/MDC, CCL4/MIP-1β, IFNγ, TNFα, IL-1β, IL-2, IL-4, GM-CSF, CXCL1/GRO-alpha, CXCL11/ITAC, CCL26/Eotaxin-3 and CL3/MIP-1α were obtained regularly throughout the treatment period. Mean levels of CXCL13/BLC, CXCL10/IP-10, CCL22/MDC, CCL4/MIP-1β, IFNγ and TNFα decreased during the treatment period. Levels of IL-

1β, IL-2, IL-4, GM-CSF, CXCL1/GRO-alpha, CXCL11/ITAC, CCL26/Eotaxin-3 and CL3/MIP-1α were below limits of quantification for most samples analyzed.

Clinical Chemistry

There was no apparent pattern in serum levels of hsCRP and LDH, evaluated to assess inflammation, over the course of treatment in Trial 2201, Part 1.

SF-36, WPAI-CIQ

There were no conclusive trends in these patient-reported outcomes.

PtGA and PGA

PtGA questionnaire in the form of 100 mm visual analogue scales (VAS) showed a mean±SD increase in wellbeing of 10.5±10.7 mm (range -3 to 22 mm) and PGA questionnaire in the form of 100 mm VAS demonstrated less disease activity following 12 weeks of treatment (mean±SD reduction of 25.8±15.98 mm, ranging from 12-51 mm reduction).

Reviewer Comment: *While there were consistent positive trends observed in several of the parameters used to assess effect of leniolisib on APDS, results were based solely on a descriptive analysis and were not used to make any conclusions on efficacy. The results from Part 1, however, were the basis for dose selection in Part 2 as the 70-mg dose resulted in sustained inhibition of pAKT compared to the lower doses. Overall, the results supported the continued evaluation of leniolisib for the treatment of APDS in Part 2 of the trial.*

8.1.3. CCDZ173X2201, Part 2

Title and Administrative Information

Trial 2201 Part 2 was the second part of the trial entitled “An open-label, non-randomized, within-subject dose-finding study followed by a randomized, subject, investigator and sponsor-blinded placebo-controlled study to assess the efficacy and safety of CDZ173 (Leniolisib) in subjects with APDS (Activated phosphoinositide 3-kinase delta syndrome/ p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency).” The trial was conducted at ten centers: 2 in Italy and 1 each in United States, United Kingdom, Czech Republic, Netherlands, Ireland, Belarus, Russia and Germany from December 5, 2017 to August 16, 2021.

Trial Objective

The primary objective was to assess the clinical efficacy (lymphadenopathy and immunophenotype normalization) of leniolisib in subjects with APDS.

Trial Design

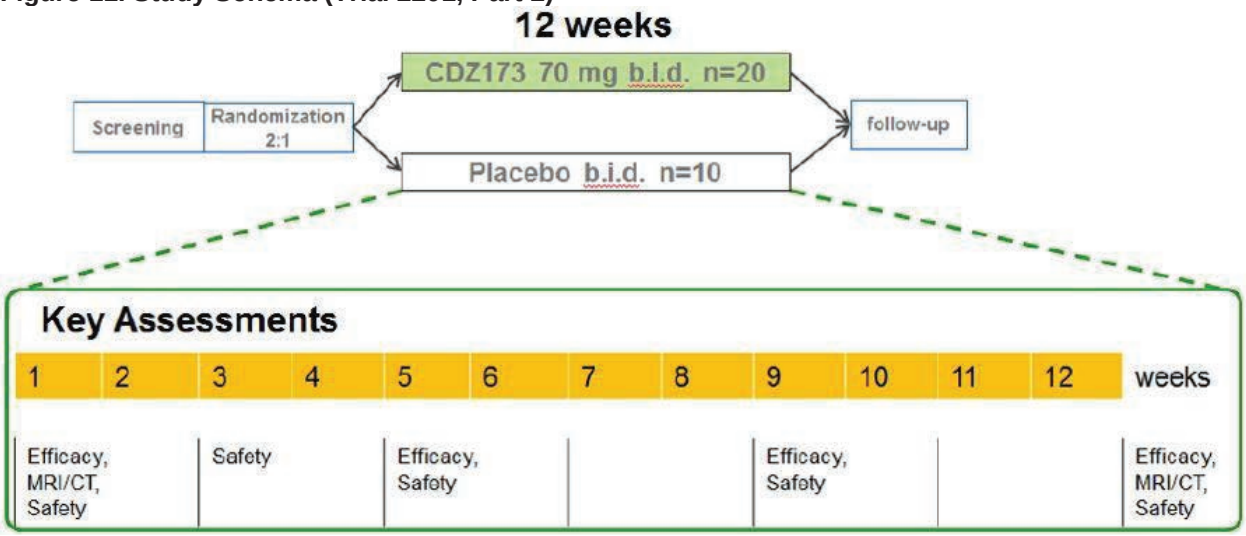
Trial 2201, Part 2 was the pivotal 12-week, randomized, double-blind, placebo-controlled, multi-center study to assess the efficacy and safety of leniolisib in subjects ≥ 12 years of age diagnosed with APDS. Subjects were randomized 2:1 to leniolisib 70 mg or placebo taken orally twice daily for twelve weeks.

Subjects enrolled in Part 1 of the study did not participate in Part 2. Subjects were permitted to directly roll over from Part 2 to treatment in OLE study, CCDZ173X2201E1. The subjects who did not directly roll over from Part 2 into the OLE were followed for safety four weeks after the last day of dosing. On Day 112 / Week 16, these subjects participated in the end of study visit.

Details of each study visit are summarized below, and the study design is presented in Figure 11.

- During the Screening period (Day -50 to Day -2), informed consent and demographic data were collected. Eligibility was assessed and reviewed, physical exam was performed, vital signs, laboratory evaluations, ECGs were obtained, and subjects underwent an MRI or CT scan.
- At the Baseline Visit (Day -1), eligibility was assessed and reviewed, physical exam was performed, and vital signs, laboratory evaluations, and ECGs were obtained. Additional laboratory evaluations were obtained including PD blood collection. Subjects were provided the SF-36/WPAI-CIQ questionnaires and the patient/physician global assessment.
- At the first Treatment Phase Visit (Day 1), subjects were randomized 2:1 to either leniolisib 70 mg twice daily or placebo for 12 weeks.
- At subsequent treatment visits (Days 15, 29, 57, and 85), efficacy and safety measurements were evaluated, and subjects received either study drug or placebo.
- On the last visit of the Treatment Phase (Day 105), neither study drug nor placebo was dispensed at this visit. However, efficacy and safety evaluations were performed.
- Subjects who did not directly roll over from Part 2 to treatment in the open label extension, 2201E1, were followed for safety and participated in the End of Study Visit (Day 112).

Figure 11. Study Schema (Trial 2201, Part 2)



Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Section 9.1.
Abbreviations: BID, twice daily; CT, computed tomography; MRI, magnetic resonance imaging, n, number of subjects in category.

Study Population

Subjects were assessed at screening and baseline for eligibility. Enrollment criteria are outlined below.

Key Inclusion Criteria

- Male and female subjects 12 to 75 years of age (inclusive), who had a documented APDS-associated genetic PI3K delta mutation in either PIK3CD or PIK3R1.
- Nodal and/or extranodal lymphoproliferation and clinical findings and manifestations compatible with APDS such as a history of repeated oto-sino-pulmonary infections and/or organ dysfunction (e.g., lung, liver).
- At least one measurable nodal lesion on a CT or MRI scan.
- Sitting vital signs (i.e., obtained after resting in a seated position for at least 3 minutes) at Screening within the following ranges:
  - Systolic blood pressure, 90-139 mm Hg.
  - Diastolic blood pressure, 50-89 mm Hg.
  - Pulse rate, 50 - 100 bpm; up to 110 bpm in adolescents.
- Minimum body weight of 45 kg.
- Legal representatives (for subjects under the age of 18 years) able to communicate well with the Investigator, to understand and comply with the requirements of the study.

Key Exclusion Criteria

- Use of other investigational drugs within 5 half-lives of enrollment, or within 30 days, whichever is longer.
- History of hypersensitivity to any of the study drugs or to drugs of similar chemical classes.
- Previous or concurrent use of immunosuppressive medication such as:
  - mTOR inhibitor or a PI3Kδ inhibitor within 6 weeks prior to first dosing, however short-term use for up to a total of 5 days was allowed but only up to 1 month prior to enrollment in the study.
  - B cell depleters within 6 months prior to first dosing of study medication; if subjects have received prior treatment with a B cell depleter, absolute B lymphocyte counts in the blood must have regained normal values.
  - Belimumab or cyclophosphamide within 6 months prior to first dosing of study medication.
  - Cyclosporine A, mycophenolate, 6-mercaptopurine, azathioprine or methotrexate within 3 months prior to first dosing of study medication.
  - Glucocorticoids above 25 mg prednisone or equivalent per day within 2 weeks prior to first dosing of study medication.
  - Other immunosuppressive medication where effects are expected to persist at start of dosing of study medication.
- History or current diagnosis of ECG abnormalities indicating significant risk of safety for subjects participating in the study.
- Current use of medication known to be strong inhibitors, or moderate or strong inducers of isoenzyme CYP3A and the treatment cannot be discontinued or switched to a different medication prior to starting study treatment.
- Current use of medication that are metabolized by isoenzyme CYP1A2 and have a narrow therapeutic index.
- Any surgical or medical condition which might significantly alter the absorption, distribution, metabolism, or excretion of drugs, or which may jeopardize the subject in case of participation in the study.
- History of acquired immunodeficiency diseases.
- A positive Hepatitis B surface antigen or Hepatitis C test (by PCR) result at screening.
- History of malignancy of any organ system (other than localized basal cell carcinoma of the skin), treated or untreated, within the past 5 years, regardless of whether there is evidence of local recurrence or metastases.

- Uncontrolled chronic or recurrent infectious disease or evidence of tuberculosis infection as defined by a positive QuantiFERON TB-Gold test at Screening.
- Donation or loss of 400 mL or more of blood within 8 weeks before randomization
- Administration of live vaccines starting from 6 weeks before study entry, during the study and up to 7 days after the last dose of CDZ173.
- Pregnant or nursing (lactating) women, where pregnancy is defined as the state of a female after conception and until the termination of gestation, confirmed by a positive hCG laboratory test.
- Women of child-bearing potential, defined as all women physiologically capable of becoming pregnant, unless they are using highly effective methods of contraception during dosing of study medication and for 2 days after stopping study treatment.

Concomitant Therapy

Prohibited therapies during the course of the study included other investigational therapies, live or attenuated vaccines, strong CYP3A inhibitors and moderate to strong CYP3A inducers, and drugs that are metabolized by CYP1A2. Co-administration of the immunosuppressive medications described in the exclusion criteria were prohibited during trial participation.

In addition to the aforementioned prohibited therapies, drugs that are moderate inhibitors of CYP3A, have a risk of Torsades de Pointes, are weak inducers CYP3A, are substrates or inhibitors/inducers of BRCP and OATP1B1/B3, and certain food and herbal medications were to be administered with caution.

Study Assessments

Study assessments were performed according to the schedule shown in Table 17. Descriptions of the individual assessments and techniques used to obtain data are described below.

Table 17. Study Assessments in Trial 2201, Part 2

Epoch	Screening		Treatment Period					Follow-up
Visit Name	SCR	BL	Treatment				EOT	EOS
Visit numbers, core period[a]	1	2	101	102	103	104	105	199
Days	-50 to -7	-1	1	15	29	57	85	112
Visit windows				+/- 3 day	+/- 3 day	+/- 3 day	+/- 3 day	+/- 5 day
Written informed consent	X							
Inclusion/exclusion criteria	X	X[b]						
Relevant medical history/current medical conditions	X	X[b]						
Demography (incl. smoking status)	X							
Physical examination[c]	X	X	X[d]	X	X	X	X	X
Tanner staging[c,e]	X						X	
Hepatitis and HIV test	X							
Pregnancy test [c,f]	X[f]	X			X	X	X	X
COVID-19 test	X							
Randomization			X					

Epoch	Screening		Treatment Period					Follow-up
Visit Name	SCR	BL	Treatment				EOT	EOS
Vital signs and body measurements								
Body height[c]	X						X	
Body weight[c]	X	X			X	X	X	X
Body temperature[c]	X	X	X	X	X	X	X	X
Blood pressure/pulse rate[c,g]	X	X	X[h]	X	X	X	X	X
ECG evaluation[c]	X	X	X[h]	X	X	X	X	X
Hematology, blood chemistry, urinalysis[c,i]	X	X	X	X	X	X	X	X
Tuberculosis test	X							
Serum Immunoglobulins[c]		X	X		X	X	X	
EBV/CMV test[c,j]		X	X		X	X	X	
DNA blood collection (consent required)								X[k]
PK blood collection[c]			X[l]		X	X	X	
Immunophenotyping[c]		X	X		X	X	X	
Soluble biomarker/miRNA[c,m]		X	X		X	X	X	
PBMC for functional T cell assay[c]		X					X	
RNA blood collection[c]		X					X	
Drug administration record[c,n]			X	X	X	X	X[o]	
MRI or CT scan[c]	X[p]						X[p]	
SF-36/WPAI-CIQ questionnaires[c]		X			X	X	X	
Activity tool training	X							
Provide activity measuring tool[q]	X					X		
Return activity measuring tool[c]		X					X	
Patient/physician global assessment[c]		X			X	X	X	
Subject narratives							X	
Study/epoque completion information		X						X[r]

Epoch Visit Name	Screening SCR BL	Treatment Period Treatment EOT	Follow-up EOS
Comments	As required		
Adverse events (comment)	As required		
Concomitant meds/therapies	As required		

Source: Adapted from Applicant's CSR

Note: X means any blood collection unless otherwise mentioned pre-dose is meant.

[a] Visit structure given for internal programming purpose only.

[b] Review of inclusion and exclusion criteria and current medical conditions is required at baseline evaluation. The position of the PI3KCD mutation and the date of first diagnosis of COVID-19 need to be recorded in the eCRF.

c Assessment to be performed in an early treatment discontinuation visit (named V104.1) in case of a subject discontinuing study treatment before schedule. If the subject discontinues treatment prior to V104, all the marked assessments should be performed at the time of treatment discontinuation and in addition the subject should come in for the same assessments, except imaging, at the scheduled V105, Day 85, which will be their last visit. For subjects that discontinue treatment at or after the V104 the marked assessments will be done at the time of discontinuation in an early treatment discontinuation visit (named V104.1) and the subject will in addition be asked to come back for an EOS visit 28 +/- 5 days later.

[d] If assessment of the subject's general clinical status shows no significant change between baseline (visit 2) and the first treatment visit (visit 101), the results of the physical examination at baseline (visit 2) can be used for first treatment visit (visit 101) data entry as well.

[e] Only for subjects below 18 years of age.

[f] Serum pregnancy test required at screening, urine pregnancy test at baseline and further time points.

[g] Following 3 minutes of sitting rest, sitting blood pressure and pulse measurements will be taken.

[h] Blood pressure/pulse rate and ECG measurement at pre-dose, 1h, 3h and 5 h post-dose.

[i] Safety lab including hsCRP, LDH, beta2 microglobulin, ferritin, fibrinogen, and erythrocyte sedimentation rate.

[j] Samples for EBV/CMV test will be collected as EBV/CMV viremia (Visits 2, 101, 103-105) and EBV/CMV latent (Visits 2, 101, and 105 only).

[k] DNA samples can be collected anytime from BL till EOS, but before sample collection the specific optional genetic ICF must be obtained.

[l] PK samples only on day 1 to be taken at pre-dose, 0.25h, 1h, 3h, 5h and 8h post-dose (samples only taken predose for day 29, 57 and 85).

[m] This sample to be taken pre-dose.

[n] BID daily dosing CDZ173.

[o] Note that no IMP from the core study will be dispensed on that visit. Please refer to protocol CCDZ173X2201E1 for guidance regarding the transition to the extension study.

[p] In case a subject has a historical scan this can be used for verifying eligibility. However, the baseline scan should still be performed in the period of -30 to -1 days of randomization. The Day 85 scan can be taken up to 14 days post Day 85 visit. In case a subject discontinues treatment before schedule imaging will be performed only at the time of treatment discontinuation.

[q] The actibelt should be worn for 9 days before first treatment and again for 9 days during the period between visit 104 and 105, as close to Visit 105, as possible.

[r] For subjects who have discontinued treatment before Day 85, or roll over to the extension study directly, this information should be registered at their Day 85 visit, which will be their last Visit, for other subjects it will be registered at the EOS visit at Day 112.

Abbreviations: BID, twice daily; BL, baseline; CIQ, classroom impairment questions; COVID-19, coronavirus disease 2019; CT, computed tomography; CMV, cytomegalovirus; DNA, deoxyribonucleic acid; EBV, Epstein-barr virus; ECG, electrocardiogram; eCRF, electronic case report form; EOS, end of study; EOT, end of treatment; HIV, human immunodeficiency virus; hsCRP, high-sensitivity C-reactive protein; ICF, informed consent form; IMP, investigational medicinal product; LDH, lactate dehydrogenase; MRI, magnetic resonance imaging; miRNA, micro r bonucleic acid; PIK3CD, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit delta; PK, pharmacokinetic; PMBC, peripheral blood mononuclear cell; RNA, r bonucleic acid; SCR, screening; SF-36; short form health survey; WPAI, work productivity and activity impairment.

Imaging Assessments

Imaging modalities included contrast-enhanced MRI of neck, chest, abdomen, and pelvis or contrast-enhanced diagnostic-quality CT scan of the neck, chest, abdomen, and pelvis. Protocol defined imaging included MRI or CT of the neck, chest, abdomen, and pelvis, which was obtained at baseline and 12 weeks. Slice thickness of ≤5 mm was recommended for both modalities and the same modality was to be used at each time point. Images were sent to a central CRO for evaluation where a single radiologist, who was blinded to treatment, but not timepoint, was responsible for approving all imaging results. Up to 14 lesions (6 lymph nodes, 4 liver or spleen, and 4 other extranodal) were selected based on size, ease of accurate measurement, and ability to represent overall nodal disease burden. To be considered measurable, lesions were required to measure at least twice the slice thickness and be at least 1.5 cm long axis (for nodes) or be at least 1 cm in both long and short axis (for non-nodal lesions). A minimum of one and a maximum of six of the largest lymph nodes were selected to calculate the log10 SPD as part of the primary endpoint calculation. On follow up scans, lesions that disappeared were recorded as 0 cm and lesions that were less than 5 mm as 5 mm. The criteria used for lesion selection are from the 1999 International Working Group (Cheson et al. 1999).

Reviewer Comment: *Examining the change in the sum of the product of the diameters for assessing response in patients with lymphoma is a common practice. Additionally, the review team consulted the Division of Imaging and Radiation Medicine (DIRM) to assess the adequacy of the imaging methods used to measure primary and secondary endpoints. While CT may have better spatial resolution of lymph node measurements, both CT and MRI are capable of providing sufficient anatomic detail for lymph node diameter measurements. The method for selection of index lesions used by the Applicant incorporates the Cheson criteria along with additional criteria emphasizing ease of measurement and overall disease burden and is a reasonable approach. There is some degree of subjectivity in the selection of index lesions rather than all lesions; however, blinding the reader to treatment assignment (as done in this trial) should be sufficient to prevent biased lesion selection.*

Immunophenotyping Assessments

All immunophenotyping endpoints including the co-primary endpoint, the effect of leniolisib on the percentage of naïve B cells out of total B cells, were assessed by flow cytometry. Whole blood samples were collected from all subjects at the clinical sites as defined in the assessment schedule in Table 17. The samples were stained, lysed/fixed and frozen before shipment to the central analysis laboratory. Specific B cell immunophenotypes were defined as follows:

- Naïve B cells; CD19+CD27-CD10-
- Transitional B cells; CD19+CD27-CD10+

- Plasmablasts; CD19+CD27+CD38++
- Switched memory B cells; CD19+CD27+IgD-
- Nonswitched memory B cells; CD19+CD27+IgD+

Patient Reported Outcome Assessments

Subject-reported measures were completed at the center no less than 3 hours post dose.

Completed questionnaires were reviewed and examined by the blinded Investigator. To assess the physical and mental functioning of subjects, the SF-36, a 36-item questionnaire used to evaluate individual subject health status and monitor and compare disease burden, was administered per the schedule outlined in Table 17. Likewise, the amount of absence or presence in school and daily activity impairment attributable to APDS was measured via the WPAI-CIQ or WPAI, respectively. The patient's global assessment (PtGA) required subjects to rate their APDS related well-being using 100 mm visual analogue scale (VAS) ranging from "very poor" (0) to "very good" (100) and the physician's global assessment required investigators to rate the subject's disease activity on the same scale from "no disease activity" (0) to "maximal disease activity" (100). The physician and subject were blinded to the other's assessment. Copies of the questionnaires are provided in Appendix 16.4.1.1.

Reviewer Comment: *The Short Form 36 Survey (SF-36), Work Productivity and Activity Impairment plus Classroom Impairment Questionnaire (WPAI-CIQ), Patient Global Assessment Questionnaire (PtGA), and the Physician's Global Assessment Questionnaire (PGA) are not fit for purpose to support labeling claims for the context of use in this drug development program. However, data from these instruments were considered to provide contextual information for regulatory decision-making.*

Activity Parameter Assessments

Subject activity was tracked using a tri-axial accelerometer, which was placed inside a belt buckle. Subjects were instructed to wear the belt for 9 days at baseline before first treatment and again for 9 days during the treatment period as close to the Day 85 Visit as possible. Number of steps per minute, real world gait speed, and overall distance of walking were captured for the activity parameter analyses.

Markers of Viremia

Both latent and lytic burden of EBV and CMV in systemic circulation were evaluated by deoxyribose nucleic acid (DNA) copy number assessed by quantitative polymerase chain reaction (PCR) in a cell free matrix. Plasma samples were obtained at baseline and as described in Table 17 and sent to a central lab for processing. Viremia biomarker data was described as absolute values and as percent change from baseline.

Soluble Biomarkers

A panel of chemokines, cytokines, and immunoglobulins were quantified in serum at various timepoints throughout the study according to Table 17. The chemokine panels consisted of CXCL13, MIP-3a/CCL20, MDC/CCL22, MIP-1b/CCL4, and IP-10. The cytokine panel consisted of IFN-γ, and TNF-α. The immunoglobulin panel included IgG, IgM, IgA, IgE, and isotypes.

Trial Endpoints

Co-primary Efficacy Endpoints

- Change from baseline in the log10 transformed sum of product of diameters (SPD) in the index lesions selected as per the Cheson methodology from MRI/CT imaging at day 85.
- Change from baseline in percentage of naïve B cells out of total B cells at day 85.

Reviewer Comment: *Skewed distribution of the B cell subsets including a low proportions of naïve B cells and concomitant increased plasmablasts and transitional B cells due to the dysregulation of the PI3K pathway are thought to be responsible for infectious sequelae of APDS. Therefore, correction of immunophenotype with regard to B cell subsets, as measured by change from baseline in percentage of naïve B cells out of total B cells at day 85, is considered both a clinically meaningful measure of response to leniolisib as well as a direct measure of effect of leniolisib on the dysregulation of the PI3Kδ pathway inherent in APDS. Likewise, although not a traditional clinical endpoint that directly measures how a patient feels, functions, or survives, lymphoproliferation, as measured by degree of lymphadenopathy, is considered a reasonable substitute measure that reflects clinical course as it is a direct consequence of the underlying immune dysregulation, is a hallmark of the disease, and if left uncorrected, can lead to obstruction or lymphoma. Reduction in lymphadenopathy, which is not expected to occur spontaneously without intervention, would imply correction of the underlying immune dysregulation.*

Additionally, naïve B cells represented by CD38−IgM+IgD+CD27−CD10− surface markers differentiate into plasma or memory B cells after binding of antigen by specific Ig receptors. Normal percentages of naïve B cells vary with age and range from approximately 48% to 85% of total B cells (van Gent et al. 2009). Therefore, the Applicant's decision to evaluate subjects with abnormal naïve B cell percentages (i.e. those < 48%) was considered acceptable.

Secondary Efficacy Endpoints

- Change from baseline in 3D volume of index and measurable non-index lesions selected as per the Cheson methodology, and 3D volume and bi-dimensional size of the spleen at day 85.

- Single dose leniolisib PK parameters (including but not limited to C_{max} and AUC) and trough evaluations after multiple dose (see Section 6.3).
- Change from baseline in Short Form 36 (SF-36) Survey and Work Productivity Activity Impairment Classroom Impairment Questionnaire (WPAI-CIQ) at day 85.
- Change from baseline in the visual analogue scale (VAS) score for the physician global assessment (PGA) and patient global assessment (PtGA) at day 85.
- Change from baseline in C-reactive protein (CRP), lactate dehydrogenase (LDH), beta2 microglobulin, ferritin, fibrinogen, and erythrocyte sedimentation rate (ESR) at day 85.

Exploratory Efficacy Endpoints

- The difference in activity parameters (e.g., number of steps, real world gait speed, overall distance of walking) provided by a device placed inside a belt buckle (Actibelt) that uses a tri-axial accelerometer between baseline and day 85
- DNA assessments to examine whether individual genetic variation in genes relating to drug metabolism and transporting, the APDS the drug target pathway, or other relevant genetic pathways confer differential response to study drug
- Change from baseline in hepatomegaly, 3D liver volume and bi-dimensional liver size from MRI or CT scans at day 85
- Change from baseline in frequency distribution measured by fluorescence activated cell sorting (FACS) as well as T-cell function measured by ex vivo stimulation of PBMC at day 85
- Difference between leniolisib and placebo in the frequency of antibiotics use over the course of the treatment period as measured by difference in number of episodes of antibiotic treatment between leniolisib and placebo.
- Change from baseline in soluble biomarkers including immunoglobulins, cytokines, chemokines, 4βOH-cholesterol, and cellular biomarkers at day 85
- Model PK parameters and associated intra- and/or inter-individual variability. Parameters include, but are not limited to, absorption rate constant, apparent oral clearance, and volume of distribution

Safety Endpoints

Descriptive summary statistics were provided for the following:

- Incidence of adverse events
- Vital signs
- Laboratory assessments

- ECGs
- Physical examination

Statistical Analysis Plan

Population Analysis Sets

- The safety analysis set included all subjects who received any study drug.
- The PK analysis set included all subjects who had at least one PK concentration measurement, who received any study drug, and who did not experience any protocol deviations that would affect PK data.
- The PD analysis set was comprised of those subjects who received any study drug and experienced no protocol deviations that would impact PD data.

Sample Size Calculation

The sample size (20 subjects in active and 10 subjects in placebo for a total of 30 subjects) was mostly driven by the analysis of one of the two co-primary efficacy variables, the change from baseline in the log10-transformed SPD of index lesions. Data from 43 subjects in the uncontrolled everolimus study CRAD001N2201 in mantel cell lymphoma after 2 months of treatment were used to estimate the variability of the change from baseline in log10-transformed SPD of index lesions. Assuming comparable variability in this APDS patient population (standard deviation of 0.2 on the log10 scale), with 30 subjects this part of the study had 80% power to detect a treatment difference of -0.225 between leniolisib and placebo groups in the change from baseline in log10-transformed SPD using a 5% type I error in a two-sided t-test. On the original scale, this corresponds to a decrease of around 40%. The observed standard deviation was slightly lower (0.14) in the six subjects from the Part I of this study. Assuming a standard deviation of 0.14, with 30 subjects, Part 2 of the study had a 97% power to detect a statistically significant difference.

Assuming that 10% of subjects are excluded from the analysis of the other co-primary variable, the change from baseline in the percentage of naïve B cells out of total B cells, due to no reduced percentage of naïve B cells at baseline, it was anticipated that 27 subjects would have provided data for the analysis. Assuming an increase of 25% points and comparable variability as in Part I for the change from baseline to Week 12 in the percentage of naïve B cells (standard deviation of 14), with 27 subjects, Part 2 of the study had 98% power to detect a statistically significant difference at the 5% level. With the above assumptions, the power to achieve a statistical significance in both endpoints was at least 78%.

Estimands

The estimands for the co-primary endpoints are described in Table 18.

Table 18. Estimands for Primary Endpoints

	Lymphadenopathy	Immune Function	
Attribute	**Primary**	**Primary**	**Supportive**
Treatment	Leniolisib 70 mg Placebo	Leniolisib 70 mg Placebo	Leniolisib 70 mg Placebo
Population	Subjects in PD analysis set and without 0 lesions at baseline	Subjects in PD analysis set and with a percentage of less than 48% of naïve B cells at baseline	Subjects in PD analysis set
Variable	Change of log10 transformed SPD from baseline	Change from baseline in the naïve B cells	Change from baseline in the naïve B cells
Intercurrent event	N/A	N/A	N/A
Population-level summary	Difference in variable means between treatments	Difference in variable means between treatments	Difference in variable means between treatments

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 9-2.
Abbreviations: N/A, not applicable; PD, pharmacodynamic; SPD, sum of product of diameters.

The estimands for secondary endpoints are described in Table 19.

Table 19. Estimands for Secondary Endpoints

Attribute	Lymphadenopathy - 3D Volume Index and Non-index Lesions (in log10 Transformation) Bi Dimensional Size and 3D Volume of Spleen PtGA/ PGA/SF-36/WPAI-CIQ
Treatment	Leniolisib 70 mg
	Placebo
Population	All subjects in PD analysis set
Variable	Change from baseline in respective endpoint
Intercurrent event	N/A
Population-level summary	Difference in variable means between treatments

Source: Applicant's Documentation of Statistical Methods for Study 2201 Part 2, Table 6-2.
Abbreviations: N/A, not applicable; PD, pharmacodynamic; PGA, physician global assessment; PtGA, patient global assessment; SF-36; short form health survey; WPAI, work productivity and activity impairment, CIQ, classroom impairment questions

Primary Efficacy Analysis Model

Change from baseline in the log10 transformed sum of product of diameters (SPD) in the index lesions selected as per the Cheson methodology from MRI/CT imaging at day 85

An analysis of covariance was performed to compare the change of the log10 transformed SPD from baseline between the two treatment groups, with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The baseline intake of glucocorticoids as well as the information about being treated with intravenous immunoglobulin G (IgG) were both included as categorical (Yes/No) covariates. The comparison of the two treatment groups was conducted at a two-sided 5% significance level. Subjects with zero lesions at baseline were excluded from the analysis.

Change from baseline in percentage of naïve B cells out of total B cells at day 85

An analysis of covariance was performed to compare the change from baseline in the naïve B cells at the end of treatment (i.e., Day 85 assessment for subjects who complete the 12-week treatment period or the treatment discontinuation visit for subjects who discontinued treatment prematurely prior to Day 85 visit) between the two treatment groups, adjusted for baseline naïve B cells frequencies. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) fixed effects. Comparison of the two treatment groups was conducted at a two-sided 5% significance level. Only subjects with a reduced percentage of naïve B cells at baseline (defined as below 48%) were included in the analysis (van Gent et al. 2009). Baseline was defined as the arithmetic mean of the baseline and Day 1 values when both were available, and if either baseline or the Day 1 value was missing, the existing value was used.

As a sensitivity analysis, the change from baseline in naïve B cells was analyzed using a longitudinal repeated-measures mixed model, with treatment, time, treatment by time interaction, baseline, and baseline by time interaction as fixed effects. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) fixed effects. An unstructured covariance matrix was fitted to adjust for correlations among the measurements made on the same subject. The difference between the treatment groups in the change from baseline after 12 weeks of treatment was assessed at a two-sided 5% significance level. Only subjects with a reduced percentage of naïve B cells at baseline were included in the analysis. For subjects who complete the treatment period, this repeated-measures analysis included all measurements in the treatment period (Baseline, Day 29, Day 57, and Day 85).

A supportive analysis of covariance was performed to compare the change from baseline in the naïve B cells at the end of treatment (i.e., Day 85 assessment for subjects who completed the 12-week treatment period or the treatment discontinuation visit for subjects who discontinued treatment prematurely prior to the Day 85 visit) between the two treatment groups, adjusted for baseline naïve B cells frequencies. Comparison of the two treatment groups was conducted

at a two-sided 5% significance level. All subjects in the PD analysis set were included in this analysis.

Reviewer Comment: *It was stated in the protocol that intravenous or subcutaneous immunoglobulin replacement therapy (IRT) was allowed in the trial. However, the statistical model in the SAP only specified baseline intravenous immunoglobulin G (IgG) as a categorical (Yes/No) covariate. The Applicant performed a modified sensitivity analysis on the co-primary endpoints repeating the primary analysis with the use of glucocorticoids and any IRT at baseline as categorical covariates. The clinical team considered that from a clinical perspective, it was reasonable to use any IRT regardless of route of administration, as the impact is expected to be the same. Therefore, the efficacy analyses for the primary and secondary endpoints in this review included the use of glucocorticoids and any IRT at baseline as categorical covariates in the models. The analyses were based on the efficacy datasets submitted in eCTD sequence 0040. See Appendix 16.5 for the prespecified analyses per the SAP.*

Secondary Efficacy Analysis Model

An analysis of covariance was performed for the 3D volume of index and measurable non-index lesions and 3D volume and bi-dimensional spleen size to compare the change of the log10 transformed value from baseline between the two treatment groups, with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) fixed effects.

An analysis of covariance was performed for SF-36 and WPAI-CIQ scores, and visual analogue scale scores for PGA and PtGA, to compare the change from baseline at the end of treatment (i.e., the Day 85 assessment for subjects who completed the 12-week treatment period or the treatment discontinuation visit for subjects who discontinued treatment prematurely prior to Day 85 visit) between the two treatment groups, with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were included as categorical (Yes/No) fixed effects.

Reviewer Comment: *By the same reason as above, all the key secondary efficacy analyses in this review are based on the use of glucocorticoids and any IRT at baseline as categorical covariates. See Appendix 16.5* for the prespecified analyses per the SAP.

Protocol Amendments

The protocol was amended ten times as outlined below. The amendments implemented to address regulatory body feedback or to adjust the protocol based on updated information from Part 1. None of the amendments impact the interpretation of the results or the review.

Table 20. Trial 2201, Part 2 Amendment History

Version Number	Date	Reason for Update	Summary of Key Changes
Original	November 11, 2014		
1.0	February 6, 2015	Address feedback received by the IRB, NIH, FDA and the MHRA	• Inconsistencies removed • Clarifying information added • Monitoring of thyroid function added • Requirement for contraception adjusted
2.0	March 25, 2015	Update information related to safety findings	• Risk benefit information updated to discuss potential risk of infections
3.0	June 9, 2015	Enable the use of the new capsule dose	• 70 mg capsule for the 70 mg bid dose level, rather than 7 capsules allowed
4.0	September 2, 2015	Align contraception requirements with European HMA	
5.0	December 21, 2015	Revise restrictions on concomitant medication use	• Revised medication restrictions based on the nonclinical studies and updated the final results and data for the rabbit embryo-fetal section
6.0	April 20, 2016	Update safety information based on another PI3Kδ inhibitor	• Excluded co-medication with immunosuppressives • Required a washout period for such drugs prior to inclusion • Minimum age for inclusion in the study was revised from 16 to 12 years of age
7.0	July 24, 2017	Adjust the design, endpoints and biomarkers of Part 2 based on the results obtained Part 1	• Extension component turned into separate study • Contraception requirements adjusted • Endpoints, biomarkers updated
8.0	February 7, 2019	Change eligibility criteria to allow for inclusion of APDS subjects with more severe disease phenotype	• Vital signs, criteria for QT, and restricted medication section updated to all for treatment with higher dose of corticosteroids • Allowed subjects with varying doses of Ig replacement
9.0	June 23, 2020	Address changes to trial conduct in the case of an epidemic or pandemic	• Protocol was adapted to allow alternative methods of providing continuing care as well as remote collection of efficacy endpoints

Version Number	Date	Reason for Update	Summary of Key Changes
10.0	October 13, 2020	Address questions and comments raised by the Health Authority in Germany	• Risk benefit section updated for adolescents • COVID-19 test prior to enrollment required

Source: Applicant's CSR for Study 2201 Part 2 Protocol Amendments, p.50
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; COVID-19, coronavirus disease 2019; CSR, clinical study report; FDA, food and drug administration; IRB, institutional review board; HMA; heads of medicine; Ig; immunoglobulin; MHRA; medicines and healthcare products regulatory agency; NIH, national institutes of health; PI3Kδ; phosphoinositide 3-kinase-δ; QT, QT interval.

Table 21. Document History – Changes Compared to Previous Final Version of SAP

Date	Time Point	Reason for Update	Outcome for Update
08 DEC 2017	Prior to DtB lock	Creation of initial version	N/A – First Version
05 SEP 2021	Prior to DtB lock	Inclusion of Gender Split Summaries	Inclusion of additional summary Tables and Figures
		Update to study reference documentation	Reference to protocol version corrected to version V10
		Updates to subjects exclusion from analysis sets	Updated to the primary and secondary PK and PD related outputs • INCL03 and EXCL03 are included for both PK, PD analysis
05 JAN 2022	After the DtB lock	Inclusion of additional subgroup analysis	Post hoc analysis to compare the APDS1 vs APDS2 subjects
		Updates to subjects exclusion from analysis sets	Updated to the primary and secondary PK and PD related outputs • INCL03 and EXCL03 are included for both PK, PD analysis
		Inclusion of additional subgroup analysis	Post hoc analysis to compare the APDS1 vs APDS2 subjects

Source: Applicant's Statistical Analysis Plan for Study 2201 Part 2 Document History, p.3
Abbreviations: APDS1, activated phosphoinositide 3-kinase delta syndrome 1; APDS2, activated phosphoinositide 3-kinase delta syndrome 2; DtB, database; N/A, not applicable; PD, pharmacodynamic; PK, pharmacokinetic; SAP, statistical analysis plan.

8.1.4. Study Results

Compliance With Good Clinical Practices

Trial 2201, Part 2 was conducted in accordance with the International Council for Harmonisation (ICH) Harmonised Tripartite Guideline for Good Clinical Practice (GCP) E6. A statement of compliance with Good Clinical Practices is located in the clinical trial reports.

Financial Disclosure

Details of financial disclosure are presented in Section 16.2.

Data Quality and Integrity

The NDA submission was appropriately indexed and complete to allow for review. There were no issues with submission quality or data integrity. During the inspection, it was noted that for 11 of the 16 randomized subjects, hsCRP lab results were missing for Baseline (Day of Randomization) and Visit 101 (Day 1 Dosing Day). However, these missing values did not impact the review. The Applicant provided the Clinical Investigator's corrective and preventative action (CAPA) plan to institute a check list to ensure all tests results are completed per protocol.

Subject Disposition

Disposition information is presented in Table 22. A total of 32 subjects were screened for the trial, 31 of whom were randomized to study treatment (21 to leniolisib; 10 to placebo). All randomized subjects completed the study.

Table 22. Subject Screening and Disposition (Trial 2201, Part 2)

Status	Screen Failure N=1 n (%)	Leniolisib N=21 n (%)	Placebo N=10 n (%)	Total N=32 n (%)
Screen failure[a]	1 (100)	0	0	1 (3)
Randomization	0	21 (100)	10 (100)	31 (97)
Completed to week 12	0	21 (100)	10 (100)	31 (97)
Analysis Sets				
Safety analysis set		21 (100)	10 (100)	31 (97)
PK analysis set		19 (91)	0	19 (59)
PD analysis set		19 (91)	8 (80)	27 (84)

Source: Clinical Reviewer generated using OCS Analysis Studio with DS and DM datasets.
[a] Subject was hospitalized for fever prior to randomization.
Abbreviations: N, total number of subjects; n, number of subjects in category; PD, pharmacodynamic; PK, pharmacokinetic

Subject (b) (6) was excluded from the PD analysis set after discovering that she took glucocorticoid above 25 mg prednisone equivalent within the 14 days leading up to the first dose of study medication leading to the deviation from exclusion criteria 3 (described in Table

23). Subject (b) (6) randomized to the leniolisib arm was excluded from both the PK and PD analysis set due to a failure to collect informed consent before the MRI was obtained resulting in a protocol deviation for inclusion criteria 1 and 3. Subject (b) (6) randomized to the leniolisib arm was excluded from the PK and PD analysis set for deviation from inclusion criteria 3 as there were no measurable nodal lesions on imaging at baseline. Subject (b) (6) randomized to the placebo group was excluded from the PD analysis set because the MRI was performed before written informed consent was obtained (inclusion criteria 1). Because the MRI was obtained before consent was signed the collected MRI data could not be used to evaluate subject eligibility, which also led to violation of inclusion criteria 3.

Protocol Violations/Deviations

Twenty subjects reported at least one protocol deviation during the trial outlined in Table 23.

Table 23. Protocol Deviations (Trial 2201, Part 2)

Deviation Description	Leniolisib (N=21)	Placebo (N=10)
Procedure changed due to COVID-19	2 (10)	3 (30)
Exclusion criterion 3[a]	0	1 (10)
Exclusion criterion 5[b]	1 (5)	0
Inclusion criterion 1[c]	1 (5)	1 (10)
Inclusion criterion 3[d]	2 (10)	1 (10)
Inclusion criterion 4[e]	1 (5)	1 (10)
Drug supply method changed due to COVID-19	1 (5)	0
Failure to perform key procedures in accordance with protocol requirements	9 (43)	3 (30)
GCP related deviation[f]	3 (14)	0
Missed visit due to COVID-19	1 (5)	0
Study treatment deviation	1 (5)	0
Use of prohibited medication during the study	2 (10)	1 (10)
Visit done outside of study site due to COVID-19	1 (5)	1 (10)

Source: Clinical Reviewer generated using OCS Analysis Studio with DS and DM datasets.
[a] Previous or concurrent use of immunosuppressive medication.
[b] Current use of medication known to be strong inh bitors, or moderate or strong inducers of isoenzyme CYP3A.
[c] Written informed consent must be obtained before any assessment is performed.
[d] Must have at least one measurable nodal lesion on a CT or MRI scan.
[e] Vital signs assessed in the sitting position after the subject has rested for at least three minutes and are within the prespecified ranges.
[f] Immunophenotyping blood sample was delivered out of quality condition, blood sample was sent to local instead of central laboratory, and one subject did not wear the belt for nine days during the screening period.
Abbreviations: COVID-19, coronavirus disease 2019; CT, computed tomography; CYP3A, cytochrome P450, family 3, subfamily A; GCP, good clinical practice; MRI, magnetic resonance imaging; N, total number of subjects.

Subject (b) (6) in the treatment group was found to be taking a moderate to strong CYP3A inducer prior to starting study treatment. This particular case was not categorized as a screen failure as it was not detected prior to enrollment and was therefore deemed to be a protocol deviation.

Reviewer Comment: *The aforementioned violations were treated as protocol deviations rather than inclusion/exclusion exceptions. This is reasonable based on the reasoning and justification provided by the Applicant.*

Demographic and Baseline Disease Characteristics

Table 24 provides baseline characteristics for the 31 subjects enrolled in Trial 2201 Part 2. Of note, ten subjects (47.6%) in the leniolisib arm and 6 subjects (60%) in the placebo arm were treated at site 1001, located within the U.S. Additionally, the majority of subjects enrolled in the trial were of Caucasian descent. Other demographic characteristics including enrollment of 40% adolescents with an average age of approximately 24 years was evenly distributed between the treatment arms and is reflective of the U.S. patient population with APDS.

Table 24. Baseline Demographic and Clinical Characteristics, Safety Population, Trial 2201, Part 2

Subgroup	Leniolisib (N = 21) n (%)	Placebo (N = 10) n (%)	Total (N = 31) n (%)
Sex			
Female	10 (48)	6 (60)	16 (52)
Male	11 (52)	4 (40)	15 (48)
Age (years), n (%)			
Mean (SD)	22.2 (10)	26.7 (13)	23.7 (11)
Median	20	19.5	20
Range	11, 54	15, 48	11, 54
Age group, n (%)			
<18	8 (38)	4 (40)	12 (39)
≥18	13 (62)	6 (60)	19 (61)
Race			
Asian	1 (5)	1 (10)	2 (7)
Black or African American	1 (5)	1 (10)	2 (7)
Caucasian	18 (86)	7 (70)	25 (80)
Other	1 (5)	1 (10)	2 (7)
Ethnicity			
Hispanic or Latino	0	1 (10)	1 (3)
Not Hispanic or Latino	14 (67)	7 (70)	21 (68)
Mutation			
PIK3CD	16 (76)	9 (90)	25 (81)
PIK3R1	5 (24)	1 (10)	6 (19)

Subgroup	Leniolisib (N = 21) n (%)	Placebo (N = 10) n (%)	Total (N = 31) n (%)
Baseline Medications[a]			
Glucocorticoids	12 (57)	6 (60)	18 (58)
Immunoglobulin replacement	14 (67)	7 (70)	21 (68)
Previous Rapamycin Use	4 (19)	3 (30)	7 (23)
Antibiotic Use	7 (33)	3 (30)	10 (32)

Source: Clinical Reviewer generated in OCS Analysis Studio, Custom Table Tool. Columns – Dataset: Demographics; Filter: TRT01A = 'CDZ173 70 mg bid' or 'Placebo'. Dataset: Demographics; Filter: None.
[a] Subjects taking rapamycin underwent a washout period of 6 weeks before entry. Glucocorticoid doses equivalent to ≤25 mg per day of prednisone were allowed within 2 weeks before first dosing and throughout the study
Abbreviations: BID, twice daily; N, total number of subjects; n, number of subjects in category; PIK3CD, Phosphatidylinositol-4,5-Bisphosphate 3-Kinase Catalytic Subunit Delta; PIK3R1, Phosphatidylinositol 3-kinase regulatory subunit alpha, SD, standard deviation

Other Baseline Characteristics (e.g., Disease Characteristics, Important Concomitant Drugs)

There were seven subjects (33%) randomized to the leniolisib treatment arm and three subjects (30%) randomized to the placebo arm that were on antibiotics prior to the initiation of the trial and continued during and beyond the treatment period. While the treatment groups appeared relatively balanced overall, it should be noted there there was an imbalance in the randomization of subjects with a past medical history of sinusitis with 13 (62%) subjects randomized to the leniolisib treatment group and 5 (50%) subjects randomized to the placebo group.

Treatment Compliance, Concomitant Medications, and Rescue Medication Use

Subjects completed a patient diary during the course of the trial to evaluate the level of compliance with study medication. Treatment compliance in both groups was near 100% with no reported dose adjustments or interruptions. However, a protocol deviation occurred when subject (b) (6) inadvertently took treatment for an additional three days after the end of the study. Overall, 17 subjects, nine subjects (43%) in the leniolisib treatment group and eight subjects (80%) in the placebo group, received at least one antibiotic medication prior to and/or after the start of study treatment.

Efficacy Results – Co-primary Endpoints

Two co-primary endpoints were evaluated at day 85: the change from baseline in the log10 transformed sum of product of diameters (SPD) in the index lesions selected as per the Cheson methodology from MRI/CT imaging and change from baseline in percentage of naïve B cells out of total B cells. The primary analysis was conducted in the PD analysis set (N = 27) which included subjects who received any study drug and experienced no protocol deviations that would impact PD data.

Leniolisib met statistical significance for both co-primary endpoints at α=0.05 significance level, demonstrating an improvement over placebo in change from baseline in the log10 transformed

SPD in the index lesions and percentage of naïve B cells out of total B cells at Day 85. The LS mean treatment difference estimate was -0.25 [95% CI: -0.38, -0.12; p-value = 0.0006] in change from baseline in the log10 transformed SPD in the index lesions at Day 85 and 37.30 [95% CI: 24.06, 50.54; p-value = 0.0002] in change from baseline at Day 85 in naïve B cells (Table 25 and Table 26).

Table 25. Primary Analysis of Change from Baseline at Day 85 in Log10 Transformed SPD of Index Lesions (PD Analysis Set)

				Leniolisib vs Placebo	
Treatment Group	n	Baseline Mean (SD)	LS Mean Change From Baseline (SE)	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	18	3.03 (0.42)	-0.27 (0.04)	-0.25 (-0.38, -0.12)	0.0006
Placebo (N = 8)	8	3.05 (0.39)	-0.02 (0.05)		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 1 subject in the leniolis b group having complete resolution of the index lesion identified at baseline.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; SD: Standard deviation; SE: Standard error; Diff, difference; IgG, Immunoglobulin G; LS, Least Squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SPD, sum of the products of diameters.

Table 26. Primary Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set and With a Percentage of Less Than 48% of naïve B Cells at Baseline)

				Leniolisib vs Placebo	
Treatment Group	n	Baseline Mean (SD)	LS Mean Change From Baseline (SE)	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	8	27.16 (13.16)	37.39 (5.34)	37.30 (24.06, 50.54)	0.0002
Placebo (N = 8)	5	30.51 (7.97)	0.09 (6.66)		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group who had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolisib group.
Abbreviations: ANCOVA, analysis of covariance; SD: Standard deviation; SE: Standard error; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

A planned sensitivity analysis using a longitudinal repeated-measures mixed model was performed by the Applicant to evaluate the change from baseline at Day 85 in naïve B cells including all measurements taken during the treatment period (Baseline, Day 29, Day 57, and Day 85). The results were statistically significant favoring the leniolisib group (n=8) group versus placebo (n=5) with the adjusted mean difference (95% CI) for change from baseline at Day 85 in naïve B cells of 37.41 (24.70, 50.11); the 2-sided p-value observed as p<0.01 (Table 27).

Table 27. Sensitivity Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set and With a Percentage of Less Than 48% of Naive B cells at Baseline)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	8	36.74	37.41 (24.70, 50.11)	<0.01
Placebo (N = 8)	5	-0.66		

Source: Statistical Reviewer.
Note: Data were analyzed using a longitudinal repeated-measures mixed model, with treatment, time, treatment by time interaction, baseline, and baseline by time interaction as fixed effects. An unstructured covariance matrix will be fitted to adjust for correlations among the measurements made on the same subject. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 5 subjects in the leniolis b group and 3 subjects in the placebo group who had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolisib group.
Abbreviations: CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Given the number of subjects excluded from the primary analysis due to a baseline percentage of naïve B cells greater than 48%, an additional supportive analysis was performed by the Applicant on all subjects in the PD analysis set to compare the frequencies of naïve B cells between treatment groups. The results were statistically significant for the leniolisib group (n=13) group vs placebo (n=8) with the adjusted mean difference (95% CI) for change from baseline at Day 85 in naive B cells of 27.94 (15.02, 40.85); the 2-sided p-value observed as p<0.01 (Table 28).

Table 28. Supportive Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	13	34.70	27.94 (15.02, 40.85)	<0.01
Placebo (N = 8)	8	6.76		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolisib group.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Reviewer Comment: *Normal percentages of naïve B cells vary with age. However, because the normal range varies from approximately 48% to 85%, the change from baseline seen in the leniolisib treatment group appears to be substantial with the majority of subjects in the leniolisib treatment group who entered the treatment period with a lower-than-average* naïve B cell percentage developing a normal naïve B cell percentage at Day 85.

Efficacy Results – Secondary Endpoints

3D Volume of Index and Non-Index Lesions

The LS mean change from baseline at Day 85 in the sum of the log10 transformed 3D volume of index lesions was -2.08 in the leniolisib group and -1.10 in placebo. The difference in adjusted mean change (95% CI) of leniolisib vs placebo was -0.97 (-2.74, 0.79) (Table 29).

The LS mean change from baseline at Day 85 in the sum of the log10 transformed 3D volume of non-index lesions was greater in the leniolisib group: -1.63 (leniolisib) vs 0.08 (placebo). The difference in adjusted mean change (95% CI) of leniolisib vs placebo was -1.71 (-2.77, -0.64), favoring leniolisib (Table 30).

Table 29. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in the Sum of the Log10 Transformed 3D Volume of Index Lesions (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	18	-2.08	-0.97 (-2.74, 0.79)	0.27
Placebo (N = 8)	8	-1.10		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and the log10 transformed baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; LS, least squares; IgG, Immunoglobulin G; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Table 30. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in the Sum of the Log10 Transformed 3D Volume of Non-Index Lesions (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	16	-1.63	-1.71 (-2.77, -0.64)	<0.01
Placebo (N = 8)	8	0.08		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and the log10 transformed baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 3 subjects with no measurements at baseline and Day 85 in the leniolisib group. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; LS, least squares; IgG, Immunoglobulin G; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

***Reviewer* Comment:** Examining the change in the sum of the product of the diameters for assessing response in patients with lymphoma is a common practice. The trends seen in the secondary endpoints, especially the sum of the log10 transformed 3D volume of the non-index lesions in the leniolisib treatment arm decreased while that of the placebo treatment arm increased, are consistent with results for the index lesion co-primary endpoint.

Spleen Size

The LS mean change from baseline at Day 85 in spleen bi-dimensional size (mm^2) was -1,428 in the leniolisib group and -77 in placebo. The difference in adjusted mean change (95% CI) was -1,350 (-2,409, -291), favoring leniolisib (Table 31). The LS mean change from baseline at Day 85 in spleen organ volume (mm^3) was -182,799 in the leniolisib group and 3,562 in placebo. The difference in adjusted mean change (95% CI) was -186,361 (-296,547, -76,175), favoring leniolisib (Table 32).

Table 31. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in Spleen Bi-dimensional Size (mm^2) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	-1,428	-1,350 (-2,409, -291)	0.02
Placebo (N = 8)	8	-77		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; LS, least squares; IgG, Immunoglobulin G; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Table 32. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in Spleen Organ Volume (mm^3) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	-182,799	-186,361 (-296,547, -76,175)	<0.01
Placebo (N = 8)	8	3,562		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; LS, least squares; IgG, Immunoglobulin G; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Reviewer Comment: *Imaging to determine spleen size may be helpful in monitoring disease activity. Several imaging methods can be used to assess splenic size including ultrasound, CT, and MRI. Because spleen size may vary when measured by different methods or at different times, due to changes in hydration status, body position, or measurement approach, a three-dimensional imaging modality is preferable to a bi-dimensional measurement. Generally, a variation by 10 to 20 percent is not considered significant unless there is a trend over time. Estimates of average spleen size in adults range from 105,000 mm^3 to 458,000 mm^3 depending on height and sex (*Linguraru et al. 2013*;* Chow et al. 2016*;* Mohammed et al. 2022*).*The degree of change seen in the treatment group is greater than natural fluctuations and, therefore, appear to be convincing of a leniolisib treatment effect.

Patient Reported Outcomes

As previously stated, the Patient Global Assessment Questionnaire (PtGA), the Physician's Global Assessment Questionnaire (PGA), Short Form 36 Survey (SF-36), and Work Productivity and Activity Impairment plus Classroom Impairment Questionnaire (WPAI-CIQ) are not fit for purpose to support labeling claims for the context of use in this drug development program. For the completion of presentation, the results from these instruments are presented and discussed in Appendix 16.4.1.2.

Subgroup Analyses Conducted on the Individual Trial

Although numbers within each subgroup were small**, the treatment effect for the co-primary endpoints was preserved across subgroups by age, sex, region, and genotype as shown below. Because the proportion of non-white subjects in the study was small, it is difficult to draw any conclusions from the subgroup analysis by race; however, there were no apparent differences.**

Age Group Analyses

The co-primary endpoints were analyzed for the pre-defined subgroups of pediatric subjects < 18 years of age and adults ≥ 18 years of age. The treatment effect for change from baseline in the log10 transformed SPD of index lesions and percent of naïve B cells at Day 85 was preserved in both age groups as shown in Table 33 and Table 34.

Table 33. Subgroup Analysis of Change from Baseline at Day 85 in Log10 Transformed SPD of Index Lesions (PD Analysis Set)

				Leniolisib vs Placebo	
Age Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
< 18 years	Leniolisib (N = 7)	7	-0.37	-0.27 (-0.57, 0.02)	0.07
	Placebo (N = 3)	3	-0.10		
≥ 18 years	Leniolisib (N = 12)	11	-0.25	-0.31 (-0.48, -0.14)	<0.01
	Placebo (N = 5)	5	0.06		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 1 subject in the leniolis b group having complete resolution of the index lesion identified at baseline. P-values are nominal as the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SPD, sum of product of diameters

Table 34. Subgroup Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set with a Percentage of Less Than 48% of Naive B Cells at Baseline)

				Leniolisib vs Placebo	
Age Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
< 18 years	Leniolisib (N = 7)	4	44.53	61.06 (12.59, 109.53)	0.04
	Placebo (N = 3)	2	-16.53		
≥ 18 years	Leniolisib (N = 12)	4	28.40	29.47 (15.99, 42.95)	<0.01
	Placebo (N = 5)	3	-1.07		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolis b group. P-values are nominal as the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Sex

A post-hoc subgroup analysis for comparing the co-primary endpoints between the leniolisib and placebo groups in female and male subjects was conducted. The estimated treatment effect for change from baseline in the log10 transformed SPD of index lesions and percent of naïve B cells at Day 85 was consistent with the overall population for both female and male subjects as shown in Table 35 and Table 36.

Table 35. Subgroup Analysis of Change from Baseline at Day 85 in Log10 Transformed SPD of Index Lesions (PD Analysis Set)

				Leniolisib vs Placebo	
Sex Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Female	Leniolisib (N = 10)	9	-0.21	-0.13 (-0.28, 0.01)	0.07
	Placebo (N = 5)	5	-0.07		
Male	Leniolisib (N = 9)	9	-0.34	-0.50 (-0.64, -0.36)	<0.01
	Placebo (N = 3)	3	0.16		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 1 subject in the leniolis b group having complete resolution of the index lesion identified at baseline. P-values are nominal because the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, Diff, difference; confidence interval; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SPD, sum of product of diameters

Table 36. Subgroup Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set With a Percentage of Less Than 48% of Naive B Cells at Baseline)

				Leniolisib vs Placebo	
Sex Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Female	Leniolisib (N = 10)	3	25.51	28.45 (NC, NC)	NC
	Placebo (N = 5)	2	-2.94		
Male	Leniolisib (N = 9)	5	35.01	37.77 (23.07, 52.47)	<0.01
	Placebo (N = 3)	3	-2.75		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolis b group. P-values are nominal because the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; NC, not calculated; PD, pharmacodynamic.

Country

A post-hoc subgroup analysis for comparing the co-primary endpoints between the leniolisib and placebo groups in USA and non-USA (Belarus, Czech Republic, Germany, Italy, Russia, United Kingdom) was conducted. The estimated treatment effect for change from baseline in the log10 transformed SPD of index lesions and percent of naïve B cells at Day 85 was consistent with the overall population for USA and non-USA subgroups as shown in Table 37 and Table 38.

Table 37. Subgroup Analysis of Change from Baseline at Day 85 in Log10 Transformed SPD of Index Lesions (PD Analysis Set)

				Leniolisib vs Placebo	
Country Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
USA	Leniolisib (N = 10)	9	-0.33	-0.28 (-0.46, -0.09)	0.01
	Placebo (N = 5)	5	-0.05		
Non-USA	Leniolisib (N = 9)	9	-0.21	-0.26 (-0.53, 0.01)	0.06
	Placebo (N = 3)	3	0.05		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 1 subject in the leniolis b group having complete resolution of the index lesion identified at baseline. P-values are nominal because the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, Diff, difference; confidence interval; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SPD, sum of product of diameters

Table 38. Subgroup Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set With a Percentage of Less Than 48% of Naive B Cells at Baseline)

				Leniolisib vs Placebo	
Country Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
USA	Leniolisib (N =10)	3	29.94	29.10 (-40.69, 98.90)	0.12
	Placebo (N = 5)	3	0.84		
Non-USA	Leniolisib (N = 9)	5	43.37	50.46 (5.21, 95.71)	0.04
	Placebo (N = 3)	2	-7.09		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolisib group. P-values are nominal because the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic.

Genetic Diagnosis (APDS1 and APDS2)

A post-hoc subgroup analysis for comparing the co-primary endpoints between the leniolisib and placebo groups in APDS1 and APDS2 diagnosis subjects was conducted. The estimated treatment effect for change from baseline in the log10 transformed SPD of index lesions and percent of naïve B cells at Day 85 was consistent with the overall population for ADPS1 diagnosis subgroup as shown in Table 39 and Table 40. No comparison was made for APDS2 subjects as there was no subject in the placebo group.

Table 39. Subgroup Analysis of Change from Baseline at Day 85 in Log10 Transformed SPD of Index Lesions (PD Analysis Set)

				Leniolisib vs Placebo	
Genetic Diagnosis Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
APDS1	Leniolisib (N = 15)	14	-0.26	-0.24 (-0.37, -0.10)	<0.01
	Placebo (N = 8)	8	-0.02		
APDS2	Leniolisib (N = 4)	4	-0.33	NC (NC, NC)	NC
	Placebo (N = 0)	0	NC		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 1 subject in the leniolisib group having complete resolution of the index lesion identified at baseline. P-values are nominal because the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; APDS, activated phosphoinositide 3-kinase delta syndrome; CI, Diff, difference; confidence interval; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; NC, not calculated; PD, pharmacodynamic; SPD, sum of product of diameters

Table 40. Subgroup Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set With a Percentage of Less Than 48% of Naive B Cells at Baseline)

				Leniolisib vs Placebo	
Genetic Diagnosis Subgroups	Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
APDS1	Leniolisib (N =15)	7	39.79	40.99 (25.13, 56.85)	<0.01
	Placebo (N = 8)	5	-1.19		
APDS2	Leniolisib (N = 4)	1	23.00	NC (NC, NC)	NC
	Placebo (N = 0)	0	NC		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolis b group. P-values are nominal because the analyses were not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; APDS, activated phosphoinositide 3-kinase delta syndrome; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; NC, not calculated; PD, pharmacodynamic.

Efficacy Results – Exploratory Endpoints

Exploratory endpoints were descriptive in nature. Outcomes were either similar to placebo (EBV and CMV PCR, liver volume, cytokine and chemokine levels, non-IgM immunoglobulins, IRT usage, most activity parameters for the tri-axial accelerometer data), had too few events to interpret (malignancy) or trended in favor of leniolisib (liver area, antibiotic usage, running steps per day derived from the tri-axial accelerometer data, and IgM levels).

Dose/Dose Response

The dose and dose response were evaluated in Trial 2201, Part 1. The Applicant evaluated a single nominal dose and dosing regimen in Trial 2201, Part 2. Refer to Section 6.3 for details.

Durability of Response

As discussed previously, the treatment effect, measured by the log10 SPD of index lesions, of leniolisib was durable through the Day 85 visit and appeared to continue through the OLE (See Figure 38). The durability of response of the increase in naïve B cells experienced by individual subjects in Trial 2201, Part 2 appeared to continue through day 85. However, there were too few data to assess durability of response through the OLE.

Persistence of Effect

There are no data to evaluate persistence and maintenance of leniolisib effect after cessation of treatment. All subjects enrolled in Trial 2201, Part 1 and 2 were entered into the OLE where they continued to received treatment with leniolisib.

Efficacy Results – Additional Reviewer Analyses

Additional reviewer analyses were performed to evaluate other immune parameters that may be affected by PI3K pathway dysregulation in APDS and contribute to the clinical features of disease. In addition, reviewer generated analyses explored other measures of lymphoproliferative disease, autoimmunity, and antibiotic usage.

T Cell Compartment

Given that persistent viral infections due to pathogens such as EBV and CMV occur in APDS patients, it is likely that T cell defects contribute to the immunodeficiency. Within Appendix 16.4.2, Figure 35 shows changes in each individual subject in the CD4+ and CD8+ T cell subsets, respectively. In the leniolisib treatment arm, the percent of CD4+ cells out of CD3+ T cells on average increased from 38.24% at baseline (N=17) to 46.5% at Day 85 (N=15). In the placebo group, the percent of CD4+ cells out of CD3+ T cells decreased from 43.75% at baseline (N=8) to 40.21% at Day 85 (N=7). The reversal in ratio of CD4+ to CD8+ T cells suggests a correction of the underlying immune dysregulation.

Additionally, leniolisib appeared to have an impact on senescent T cell subsets, including CD57+ CD4+, CD57+ CD8+, PD-1+ CD4+, and PD-1+ CD8+ T cells, where more subjects in the leniolisib treatment group experienced a decrease in senescent T cell markers than the placebo group as shown in Appendix 16.4.2, Figure 36, again suggesting an improvement in the underlying immune dysregulation.

B Cell Compartment

Within the B cell compartment, the characteristic immune phenotype consists of increased proportions of transitional B cells and plasmablasts (PB), progressive B cell loss, and elevated levels of serum IgM (Wang et al. 2022). Appendix 16.4.2, Figure 37 shows the change from baseline in naïve B cells, transitional B cells, plasmablasts, and immunoglobulin M levels respectively, for each individual subject over the course of Trial 2201, Part 2. There were no apparent differences in change from baseline between the treatment groups in IgG and IgA levels (data not shown). These results appear to suggest that leniolisib corrects some component of the underlying immune dysregulation inherent to APDS within the B cell compartment.

Lymphoproliferation

Nonneoplastic lymphoproliferation typically manifests as chronic or reactive lymphadenopathy, splenomegaly, and/or hepatomegaly are common findings in APDS, often apparent in early childhood, and is thought to be caused by constitutive activation of PI3Kδ signaling that stimulates abnormal proliferation of white blood cells (Coulter et al. 2017; Pham and

Cunningham-Rundles 2018). Not only can progressive lymphadenopathy cause issues due to mass effect, but uncontrolled lymphoproliferation predisposes individuals to lymphomas with the risk of malignancies increasing with age (Kracker et al. 2014). In addition to the imaging data collected to evaluate the co-primary endpoint of change from baseline in the log10 transformed SPD in the index lesions, additional measurements were collected from index lymph nodes, non-index lymph nodes, liver, and spleen.

The average longest perpendicular lesion diameter was calculated for each subject by summing the measurements for each identified lesion and dividing by the number of lesions. Similarly, the average longest lesion diameter was calculated for each subject by summing the measurements for each identified lesion and dividing by the number of lesions. Time trends by individual in the leniolisib and placebo treatment arms for longest perpendicular lesion diameter are shown in Appendix 16.4.2 (Figure 39 and Figure 40, respectively). In general, lesion measurements appeared to decrease in most subjects in the leniolisib treatment group and increase or remain the same in the placebo treatment group. Similar trends were visually apparent in additional measurements including longest lesion diameter, organ volume, area, 3D index volume, and 3D non-index volume (data not shown).

Cytopenias

Some form of autoimmunity is present in approximately one third of APDS cases. The most common autoimmune manifestations are cytopenias accounting for 76% of all autoimmune complications and include immune thrombocytopenia purpura (ITP) and autoimmune hemolytic anemia (AIHA) (Jamee et al. 2020). There were three subjects with a past medical history of thrombocytopenia enrolled in the trial. Although there were too few subjects by which to draw substantive conclusions, the platelet levels in the two subjects who received placebo appeared to remain at baseline while the platelet levels of the one subject receiving leniolisib appeared to increase over the duration of the trial. There were six subjects with a past medical history of anemia not found to be caused by iron deficiency. Hemoglobin values for the four subjects receiving leniolisib appeared to increase over the course of the trial while those for the two subjects receiving placebo remained unchanged (data not shown).

Antibiotic Usage

Finally, frequency of usage of antibiotics was used as an exploratory endpoint to evaluate infection burden in Trial 2201, Part 2. Two subjects (10%) in the leniolisib treatment group required antibiotic medication post treatment start compared to six subjects (60%) in the placebo group. While the number of episodes of antibiotic usage are small, the trend in Trial

2201, Part 2 suggests fewer subjects in the leniolisib treatment group required antibiotic use for an infection after the initiation of treatment.

Pharmacodynamic Markers of Inflammation

There were several measurements obtained at various timepoints throughout the study to assess the effect of leniolisib on reducing systemic inflammation in APDS. The Applicant evaluated hsCRP, beta-2 microglobulin, ESR, fibrinogen, ferritin, and LDH. There were too few subjects to adequately interpret the changes in hsCRP. While there appeared to be a reduction in beta-2 microglobulin and ESR, there was no meaningful changes in fibrinogen, ferritin, or LDH.

The results of change from baseline in various cytokine and chemokine levels thought to be relevant to APDS, including CXCL13, IP-10 (CXCL10), TNF-alpha, MDC, MIP-1B, MIP3A, immunoglobulins (discussed above), and TNF-alpha, were evaluated as part of the exploratory analysis. Treatment with leniolisib caused a decrease in the concentration of CXCL13, IP-10, MIP-1B, and TNF-alpha concentrations.

8.1.5. CCDZ173X2201E1

Title and Administrative Information

Trial CCDZ173X2201E1, hereafter referred to as 2201E1, is the open label extension (OLE) to Trial 2201 entitled, “An open-label, non-randomized extension study to evaluate the long-term safety, tolerability, efficacy, and pharmacokinetics of leniolisib in subjects with APDS (Activated phosphoinositide 3-kinase delta syndrome/p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency).” The trial was conducted at eight centers in seven countries including the US. The trial has been ongoing since September 8, 2016. The data cutoff date for this interim analysis was December 13, 2021. The Sponsor additionally submitted the 120-day safety update with data through September 30, 2022. Findings from the 120-day safety update are described in applicable sections where relevant.

Trial Objective

The primary objective was to evaluate the long-term safety and tolerability of leniolisib in subjects with APDS.

Trial Design

Trial 2201E1 is the ongoing, open-label, single arm study to extend active oral treatment with leniolisib 70 mg taken orally twice daily to those subjects with APDS who either participated in Trial 2201 Part 1 or Part 2 or who were previously treated with PI3Kδ inhibitors other than leniolisib, in order to collect long-term safety, tolerability, efficacy, and PK data. Subjects could

be enrolled in this extension study either directly at the end of treatment (EOT) or end of study (EOS) visit of study 2201 or later in time. Subjects previously treated with PI3Kδ inhibitors other than leniolisib could be enrolled if they met the eligibility criteria at the screening visit.

Details of each study visit are summarized below:

- Screening Visit: If a subject rolled over directly from Study 2201, the EOT/EOS visit from 2201 was considered the screening visit for the OLE study. Otherwise, eligibility was assessed and reviewed, physical exam was performed, vital signs, laboratory evaluations, and ECGs were obtained.
- During the Treatment Period, subjects initiated daily dosing of leniolisib on Day 1 and were assessed for safety at 2 weeks, 6 weeks, 12 weeks, 24 weeks, 36 weeks, 52 weeks, 78 weeks, and every 26 weeks thereafter. Treatment with leniolisib could last up to six years for an individual subject.
- Subjects who discontinued treatment were assessed again for safety at the EOT visit followed by another safety assessment at the EOS visit, 12 weeks later.
- Safety assessments were performed after 14 days of treatment, every 6 weeks for the first 3 months, then every 3 months until Week 36, after 1 year of treatment and then every 26 weeks until 6 years of treatment for each individual.

Study Population

Subjects between the ages of 12 and 75 who had a documented APDS-associated genetic PI3K delta mutation who participated in Trial 2201, Part 1 or 2, or who were treated previously with PI3Kδ inhibitors other than leniolisib and were deemed by the investigator to benefit from PI3Kδ inhibitor therapy were enrolled in the trial. Eligibility criteria were similar to those of Trials 2201, Part 1 and 2. Minor additions or adjustments included the requirement to have participated in Trial 2201 or been treated previously with PI3Kδ inhibitors other than leniolisib. Subjects were excluded if they had withdrawn consent, were non-compliant or demonstrated a serious protocol deviation in Trial 2201.

Concomitant Therapy

Prohibited therapies and drugs to be used with caution during the course of the study were the same as those of Trial 2201, Part 1 and Part 2.

Study Assessments

Study assessments were performed according to the schedule shown in Table 41. Imaging, immunophenotyping, and patient reported outcome assessments were performed as described in Section 8.1.4 .

Table 41. Schedule of Assessments for Trial 2201E1

Epoch	Extension-Screening	Extension-Treatment																	
Visit Name	Screening	Treatment																EOT	EOS
Visit Numbers[1]	501[2]	502[3]	502.1	503	504	505	506	507	508	509	510	511	512	513	514	515	516	517	599
Study Day(s)	-7 -3 +7	1	14 -3 +3	42 -7 +7	84 -10 +10	168 -10 +10	252 -10 +10	364 -10 +10	546 ±2 weeks	728 ±2 weeks	910 ±2 weeks	1092 ±2 weeks	1274 ±2 weeks	1456 ±2 weeks	1638 ±2 weeks	1820 ±2 weeks	2002 ±2 weeks	2184 ±2 weeks	2268 ±2 weeks
Written informed consent[4]	X																		
Inclusion/exclusion criteria	X																		
Medical history/current medical conditions	X																		
Demography	X																		
Physical examination*	X[5]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Tanner staging[6]	X[5]							X		X		X		X		X		X	
Pregnancy test[7]	X[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital signs and body measurements																			
Body height[8]*	X[5]			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Body weight*	X[5]	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Body temperature*	X[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
BP/pulse rate[9]*	X[5]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
ECG evaluation*	X[5]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Hematology*	X[5]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry[10]*	X[5]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Urinalysis*	X[5]		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
HIV/hepatitis/ QuantiFERON test	X[11]																		
Trough PK blood collection[12]		X			X	X	X												

Source: Applicant provided Trial 2201E1 CSR
Abbreviations: BP, blood pressure; CSR, clinical study report; ECG, electrocardiogram; EOS, end of study, EOT, end of treatment; HIV, human immunodeficiency virus; PK, pharmacokinetic.

Trial Endpoints

Primary Endpoint

The primary endpoint was change from baseline in all safety parameters including AEs, physical exam, vital signs, ECG, and safety laboratories at Day 14, followed by every 6 weeks until Week 36, at 1 year of treatment and then every 26 weeks until 6 years of treatment for each individual subject.

Secondary Endpoints

The secondary endpoints were any change in the following assessments from Extension Day 1 at regular timepoints according to the schedule provided in Table 41:

- SF-36 Survey and WPAI-CIQ, Visual analogue scales for PGA and PtGA, and subject narratives by Investigator.
- CRP, LDH, frequencies of infections and other disease complications.
- Steady-state trough concentrations of leniolisib.
- PK parameters.

Exploratory Endpoints

- Change in soluble biomarkers from Extension Day 1 to Extension Day 252.
- Changes in T and B cell immunophenotyping at Extension Days 1, 84, and 252.
- Changes in EBV and CMV viremia, EBV and CMV lytic and latent in blood, and EBV DNA in saliva at Extension Days 1, 84, and 252.
- Changes in serum IgM, IgG, and IgA levels at Extension at Days 1, 84, 168, and 252.
- Changes in degree of lymphoproliferation as measured by MRI/CT imaging or ultrasound (e.g., 3D volume of index and measurable non-index lesions selected as per the Cheson methodology and 3D volume and bidimensional sizes of spleen and liver between Extension Day 1 and Extension Days 168 and 252.

***Reviewer Comment*: All data collected during Trial2201E1 was considered exploratory and supportive given lack of a control arm.

8.1.6. Study Results

Compliance With Good Clinical Practices

Trial 2201E1 was conducted in accordance with the International Council for Harmonisation (ICH) Harmonised Tripartite Guideline for Good Clinical Practice (GCP) E6. A statement of compliance with Good Clinical Practices is located in the clinical trial reports.

Financial Disclosure

Details of financial disclosure are presented in Section 16.2.

Results – Primary Endpoints

The primary objective of Trial 2201E1 was to gather further long-term safety data in subjects with APDS. Due to a lack of control group, the efficacy analyses were descriptive in nature. Therefore, results pertaining to the primary endpoint including changes from baseline in all safety parameters including AEs, physical exam, vital signs, ECGs, and safety laboratory values are discussed in Section 8.2.3.

Efficacy Results – Secondary and Exploratory Endpoints

Imaging for lymphoproliferation, B and T cell immunophenotyping, clinical chemistry, PRO measurements, soluble biomarkers, EBV and CMV PCR, and serum immunoglobulins were evaluated at regular intervals throughout the OLE. Additionally, annualized rate of IRT and antibiotic usage were followed for the duration of the trial. Due to lack of a control group,

findings were descriptive in nature and are summarized in Appendix 16.4.3. However, several important trends, particularly pertaining to imaging for the degree of lymphoproliferation and change in B cell immunophenotype, appeared to continue during the OLE.

8.1.7. Assessment of Efficacy Across Trials

Not applicable to this review as there was only one randomized, placebo-controlled trial included in this submission.

Additional Efficacy Considerations

No additional efficacy data are considered other than the data described in Section 8.1.4.

8.1.8. Integrated Assessment of Effectiveness

The Applicant conducted a single adequate and well-controlled trial (Trial 2201, Part 2) of 12 weeks duration in 31 adult and adolescent subjects with APDS. Per the statistical testing strategy, Trial 2201, Part 2 was successful based on the change from baseline at day 85 in the co-primary endpoints of percentage of naïve B cells out of total B cells and log10 transformed SPD in the index lesions on MRI/CT imaging, both of which demonstrated highly significant improvements for leniolisib subjects over placebo. The LS mean treatment difference estimate was -0.25 [95% CI: -0.38, -0.12; p-value =0.0006] in change from baseline in the log10 transformed SPD in the index lesions at Day 85 and 37.30 [95% CI: 24.06, 50.54; p-value=0.0002] in change from baseline at Day 85 in percentage of naïve B cells out of total B cells. The treatment effect on the co-primary endpoints was preserved across supplementary and subgroup analyses, including by age group. Other pharmacodynamic endpoints such as pAKT inhibition, spleen size/volume, volumetric measures of lymphadenopathy, and additional T cell and B cell subset immune parameters showed improvements with leniolisib treatment compared to placebo.

Regarding the co-primary endpoints, there was agreement between the Applicant and Agency that the selected endpoints may be acceptable, but the Agency noted that neither were direct measures of a clinical outcome reflecting how a patient feels, functions, or survives. Therefore, the Agency anticipated reviewing the 'totality of evidence' and that mechanistic data and correction of the underlying immune defect would be important supportive data. While there is no established threshold for a clinically meaningful change index lesion size in response to an intervention, the normal percentage of naïve B cells ranges from 48-84% of total B cells. Further, neither measure is expected to spontaneously improve, and the demonstration of a highly statistically significant increase in percentage of naïve B cells and decrease in lymphadenopathy provides strong support that leniolisib treatment results in a PD effect.

In conclusion, the review team has determined that Trial 2201, Part 2 is an adequate and well controlled trial that demonstrates the efficacy of leniolisib for the treatment of APDS. In

addition to meeting statistical significance for the co-primary efficacy endpoints, the trial demonstrated improvements in spleen size and volume, non-index lesion measures of lymphadenopathy, and additional immune parameters. We concluded that these are clinically meaningful because a correction of the abnormal immunophenotype associated with APDS is expected to lead to normalization of immune function and improvement in clinical sequelae such as fewer infections, autoimmune manifestations, and lymphoproliferative disease.

Substantial evidence of effectiveness, the regulatory requirement for approval, generally consists of evidence from at least two adequate and well-controlled trials. However, in certain circumstances, a single adequate and well-controlled trial demonstrating efficacy together with appropriate confirmatory evidence may be sufficient to generate substantial evidence of effectiveness. Such circumstances are described in the FDA draft guidance *Demonstrating Substantial Evidence of Effectiveness for Human Drug and Biological Products* (December 2019). For this NDA, the Applicant conducted a single adequate and well-controlled trial in adult and adolescent subjects (Trial 2201, Part 2). Confirmatory evidence is derived from the well-established etiology of the disease, the mechanism of action of the therapy, and PD biomarker data from leniolisib clinical trials in adult and adolescent subjects with APDS.

8.2. Review of Safety

8.2.1. Safety Review Approach

All clinical studies conducted as part of the leniolisib development program for APDS were evaluated for safety; however, Trial 2201, Part 2 provided the only blinded, controlled safety data and therefore is the focus of this safety review. Trial 2201, Part 1 and the OLE, 2201E1 were both single-arm, open-label, and uncontrolled, but provide supportive information regarding dose response (Part 1) and long-term safety (2201E1). Relevant information from Part 1 and the OLE will be provided in each section following results from Part 2. For a detailed summary of the protocols, refer to Section 8.1.

Overall Exposure

Across the clinical development program for leniolisib (phase 1, phase 2, and phase 3 studies), 196 healthy volunteers, 20 subjects with Sjogren's syndrome, and 38 subjects with APDS received at least one dose of leniolisib. Table 42 lists the entire population of subjects exposed to leniolisib in the development program for APDS specifically from Trials CCDZ173X2201, Part 1 and 2, and CCDZ173X2201E1. Analyses were conducted in the safety population which includes all subjects who received at least one dose of study treatment.

Table 42. Summary of Exposure to Leniolisib in Trials 2201, Parts 1 and 2, and 2201E1

	Trial 2201 Part 1			Trial 2201 Part 2		Trial 2201E1	
	Leniolisib 10 mg BID N=6	Leniolisib 30 mg BID N=6	Leniolisib 70 mg BID N=6	Leniolisib N=21	Placebo N=10	Previous Leniolisib N=26	No Previous Leniolisib N=11
Subgroup	**n (%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**	**n (%)**
Duration of exposure, weeks							
Mean (SD)	3.93 (0.078)	4.02 (0.058)	4.00 (0.090)	12.04 (0.121)	12.01 (0.196)	120.52 (84.9)	76.78 (44.4)
Median	3.93	4.00	4.00	12.00	12.00	103.9	79.14
Min, max	3.9, 4.0	4.0, 4.1	3.9, 4.1	11.7, 12.3	11.7, 12.3	12, 262	12.1, 144.9
Person-years	0.45	0.46	0.46	4.84		60.3	16.2
Subjects treated, by duration, n (%)							
≥ 2 weeks	6 (100)	6 (100)	6 (100)	21 (100)	10 (100)	26 (100)	11 (100)
≥ 4 weeks	3 (50)	6 (100)	5 (83)	21 (100)	10 (100)	26 (100)	11 (100)
≥ 11 weeks	0	0	0	21 (100)	10 (100)	26 (100)	11 (100)
≥ 12 weeks	0	0	0	19 (91)	6 (60)	26 (100)	11 (100)
≥ 24 weeks	0	0	0	0	0	22 (85)	9 (82)
≥ 52 weeks	0	0	0	0	0	21 (81)	7 (64)
≥ 104 weeks	0	0	0	0	0	13 (50)	3 (27)
≥ 156 weeks	0	0	0	0	0	7 (27)	0
≥ 208 weeks	0	0	0	0	0	5 (19)	0

Source: FDA Clinical Reviewer calculated in JMP 16.1.0 using ADSL dataset selecting subjects by SAFFL(Y) and calculating treatment duration by subtracting 'TR01SDT' from 'TR01EDT'.
Abbreviations: BID, twice daily; N, total number of subjects; n, number of subjects in category; SD, standard deviation.

Within Trial 2201 Part 2, all subjects were exposed to treatment for the full 12 weeks in both the leniolisib and the placebo arms and mean and median exposure times were similar between the two treatment arms. Duration of exposure was analyzed in the safety population over the entire length of the trial.

The Applicant submitted the 120-Day Safety Update summarizing new safety information obtained after the data cut-off for the OLE of September 20, 2021, presented in the original NDA submission. The mean duration of exposure for the 37 subjects who remained in the OLE was 130 weeks and four subjects were exposed to leniolisib for more than five years.

Adequacy of the Safety Database

Given the rarity of the disease, the pooled safety database, while small, is of sufficient size and duration to assess the safety of the proposed dose and dosing regimen of leniolisib for the indication of ADPS in adult and adolescent patients.

8.2.2. Adequacy of Applicant's Clinical Safety Assessments

Issues Regarding Data Integrity and Submission Quality

There were no issues regarding data integrity or submission quality.

Categorization of Adverse Events

The submission is appropriately indexed and complete to permit review. The Applicant used definitions of adverse events (AEs) and serious adverse events (SAEs) that were consistent with requirements outlined in 21 Code of Federal Regulations 312.32. Reports of all AEs and SAEs, regardless of Investigator attribution, were collected from the time of signing of the informed consent through to the last study visit. Treatment-emergent adverse events (TEAEs) were defined as any AE that increased in severity or that was newly developed at or after the first dose of study drug through the final follow-up visit. Adverse events were classified using the Medical Dictionary for Regulatory Activities (MedDRA) Version 24.0. The Common Terminology Criteria for AE (CTCAE) grading system was used for AE reporting. If the CTCAE grading did not exist for an AE, the grading of AE severity was determined as mild (Grade 1), moderate (Grade 2), severe (Grade 3), or life-threatening (Grade 4).

Routine Clinical Tests

Clinical tests were assessed as per Table 17. Changes in vital signs, physical examination, and laboratory test results were only reported as AEs if judged to be clinically relevant by the investigator.

8.2.3. Safety Results

Treatment-Emergent Adverse Events

Table 43 below summarizes the overall number of treatment-emergent adverse events (TEAEs) and serious adverse events (SAEs) experienced by subjects during Trial 2201 Part 2. The overall frequencies of TEAEs and SAEs were balanced between groups with a slightly higher percentage of SAEs in the placebo group.

Table 43. Summary of Treatment-Emergent Adverse Events (Trial 2201 Part 2, Safety Population)

TEAE	Leniolisib (N=21) n (%)	Placebo (N=10) n (%)
Any TEAE	18 (86)	9 (90)
TEAEs by severity grade		
Grade 1 (mild)	15 (71)	8 (80)
Grade 2 (moderate)	9 (43)	5 (50)
Grade 3 (severe)	2 (10)	3 (30)
Grade 4 (life-threatening)	2 (10)	1 (10)
Grade 5 (fatal)	0	1 (10)

TEAE	Leniolisib (N=21) n (%)	Placebo (N=10) n (%)
TEAEs leading to discontinuation	0	0
Non-fatal SAEs	3 (14)	2 (20)
Fatal SAEs	0	1 (10)

Source: FDA Clinical Reviewer generated table using OCS Analysis Studio, Custom Table Tool.
Note: Columns - Dataset: Demographics; Filter: SAFFL = 'Y', Any TEAE - Dataset: Adverse Events; Filter: TRTEMFL = 'Y', Any SAE - Dataset: Adverse Events; Filter: AESER = 'Y', TRTEMFL = 'Y', Fatal SAE - Dataset: Adverse Events; Filter: AEOUT = 'FATAL', TRTEMFL = 'Y'.
Abbreviations: N, total number of subjects; n, number of subjects in category; SEAs, serious adverse events; TEAE, treatment-emergent adverse events.

Serious Adverse Events including Deaths

Across the APDS clinical development program, a total of two deaths have been reported. One death occurred in a placebo-treated subject during the follow-up period of 2201 Part 2 and one death occurred in a leniolisib-treated subject during the OLE as shown in Table 44 and as detailed in the brief narratives that follow.

Table 44. SAEs by Subject, Safety Population, Trial 2201 and 2201E1

Subject ID#	Age (yrs)	Sex	Treatment Arm	Preferred Term	Study Day	Reason Considered Serious
(b) (6)	12	F	Leniolisib	• Mastoiditis	2	Hospitalized
	22	M	Leniolisib	• Lipase increased	29	Hospitalized; Fatal
				• Failure to thrive	29	
				• Cardiac Arrest	879	
	23	F	Leniolisib	• Coma	4	Hospitalized
				• Alcohol poisoning	4	
	44	F	Placebo	• Urinary tract infection	23	Medically important
	44	F	Placebo	• Lymphadenopathy	84	Hospitalized; Fatal
				• Dyspnea	110	
				• Infective exacerbation of bronchiectasis	148	
				• Dependence on oxygen therapy	187	
				• Pulmonary Hypertension	224	

Source: Clinical Reviewer generated using OCS Analysis Studio, Safety Explorer. ADAE and ADSL datasets selecting by TRT01A = "CDZ173 70 mg bid" and SAFFL = "Y" (CDZ173 70 mg bid); TRT01A = "Placebo" and SAFFL = "Y" (Placebo); TRTEMFL = "Y" and AESER = "Y" (Adverse Events).
Abbreviations: BID, twice daily; F, female; M, male.

The frequency of nonfatal SAEs was similar between treatment groups in Trial 2201 Part 2 with three (14%) of subjects experiencing an SAE in the leniolisib group versus two (20%) in the placebo group. Subject (b) (6), a 12-year-old female with a past medical history of recurrent

bronchitis, otitis media, pneumonia, and sinusitis experienced an episode of Grade 2 mastoiditis during Trial 2201 Part 2 and was hospitalized for treatment. Elevated liver enzymes and failure to thrive resulting in hospitalization were considered SAEs in Subject (b) (6) (described in more detail below). Subject (b) (6), a 23-year-old female with a past medical history of respiratory tract infections, lymphadenopathy, hypogammaglobulinemia, and autoimmune thyroiditis suffered from Grade 4 alcohol intoxication resulting in coma on Day 4 of Trial 2201 Part 2. She subsequently recovered and completed the study after which she entered the OLE.

Deaths

Subject (b) (6)

44-year-old female with a history of bronchiectasis, pseudomonas infections, and lymphadenopathy, who was randomized to the placebo arm of Trial 2201 Part 2 and did not enroll in the OLE study. After receiving the last dose of study drug on Day 85, she developed dyspnea on Day 110. She was diagnosed with an exacerbation of bronchiectasis that was unresponsive to antibiotic therapy or diuretics. On Day 187, the subject developed pulmonary hypertension to which she succumbed on Day 224.

Subject (b) (6)

22-year-old male with a complicated past medical history that included recurrent/chronic pneumonia and disseminated mycoplasma orale who received leniolisib 70 mg BID in Trial 2201 Part 2. The subject was subsequently enrolled in the OLE, 2201E1. On post-randomization Day 877, treatment was discontinued when he developed a significant elevation in liver function tests relative to his baseline. He died of cardiac arrest on Day 879 thought to be direct sequelae of his worsening chronic disseminated mycoplasma orale infection and underlying cardiomyopathy.

There were no deaths or SAEs reported during Trial 2201, Part 1. Six subjects experienced 23 SAEs during the OLE. Overall, no new safety concerns in SAEs were raised by Trial 2201, Part 1 or the OLE compared to Trial 2201, Part 2.

Dropouts and/or Discontinuations Due to Adverse Effects

There were no dropouts or premature treatment discontinuations due to adverse effects in Trial 2201, Part 1 or 2. Subject (b) (6), initially randomized to placebo in Trial 2201, Part 2 was subsequently enrolled in Trial 2201E1 and discontinued treatment on Day 877 when he developed significant elevation in liver function tests and died of cardiac arrest on Day 879 as described previously. Two additional subjects experienced AEs leading to discontinuations in the 120-safety update to Trial 2201E1. Approximately two years after her first dose of leniolisib, subject (b) (6), a 19-year-old female developed anemia, febrile neutropenia, and suspected

viral infection and was hospitalized on the same date for the events. Subject (b) (6), a 15-year-old female experienced an asthma exacerbation secondary to COVID-19 infection 2 years and 9 months after her first dose of leniolisib. Leniolisib was temporarily withheld while receiving dexamethasone and remdesivir for COVID-19 infection, and restarted six days later.

Other Significant Adverse Events – Grade 3 and 4 TEAEs

Six subjects out of 31 patients experienced 11 Grade 3 and Grade 4 AEs; six of which occurred in the leniolisib treatment group and five of which occurred in the placebo treatment group. All but one of the Grade 3 or 4 TEAEs in the leniolisib group were also considered SAEs and described above; there was a case of syncope in the leniolisib treatment group from which the subject fully recovered. In the placebo treatment group, one subject developed a Grade 3 urinary tract infection and one experienced Grade 3 weight gain. Overall, there were no apparent differences in the frequency and pattern of severe or life-threatening AEs in Trial 2201, Part 1 or the OLE nor any new safety signals in the significant adverse events compared to Trial 2201, Part 2.

Common Treatment Emergent Adverse Events

The common TEAEs that occurred more frequently in the leniolisib treatment are summarized in Table 45.

Regarding infections, sinusitis occurred more frequently in the leniolisib group; however, all cases were of Grade 1 or 2 and the difference between the treatment groups may be explained by the imbalance in the randomization of subjects with a past medical history of sinusitis as described in Section 8.1.4. Furthermore, within the infection SOC, all AEs in the leniolisib treatment arm were of Grade 1 or 2. In the placebo treatment arm, most of the infections were of Grade 1 or 2, with one Grade 3 AE of urinary tract infection. Notably, there were more cases of exacerbations of bronchiectasis or lower respiratory tract infection in the placebo group than the leniolisib treatment group, and apart from sinusitis, the frequency of infections in the leniolisib treatment arm, including SAEs in the infection SOC, was lower. Evaluating antibiotic usage as a proxy for infection, there were seventeen subjects overall, 9 (43%) of those receiving leniolisib and 8 (80%) of those treated with placebo, received antibiotic medication during the study. After day 85, two (10%) subjects in the leniolisib treatment arm required antibiotic medication compared to 6 (60%) subjects in the placebo group. While there continued to be frequent infections in both treatment arms, there were fewer serious infections in the leniolisib group, particularly in the types of serious infections expected in the APDS population such as lower respiratory infections, and less antibiotics prescribed overall in the leniolisib treatment group.

During Trial 2201, Part 2, seven subjects (33.3%) in the leniolisib treatment group and four subjects (40 %) in the placebo treatment group reported AEs within the gastrointestinal disorder SOC. Only diarrhea (10%) was reported in more than one subject and in greater

frequency than placebo. There were four (11%) additional cases of diarrhea reported during Trial 2201E1. Overall, no safety concerns were identified in the analysis of common TEAEs.

Table 45. TEAEs Reported in ≥2 Subjects and Greater Than Placebo, Safety Population, Trial 2201, Part 2

Preferred Term	Leniolisib (N=21) n (%)	Placebo (N=10) n (%)
Headache	5 (24)	2 (20)
Sinusitis	4 (19)	0
Atopic dermatitis[a]	3 (14)	0
Alopecia	2 (10)	0
Back pain	2 (10)	0
Diarrhea	2 (10)	0
Fatigue	2 (10)	1 (10)
Neck pain	2 (10)	0
Pyrexia	2 (10)	0
Tachycardia[b]	2 (10)	0

Source: Clinical Reviewer Generated with OCS Analysis Studio, Safety Explorer. Filters: TRT01A = "CDZ173 70 mg bid" and SAFFL = "Y" (CDZ173 70 mg bid); TRT01A = "Placebo" and SAFFL = "Y" (Placebo); TRTEMFL = "Y" (Adverse Events).
[a] Includes “eczema” and “dermatitis, atopic”
[b] Includes “tachycardia” and “sinus tachycardia”
Abbreviations: BID, twice daily; N, total number of subjects; n, number of subjects in category, TEAEs, treatment-emergent adverse events

Although there were no control groups in Trials 2201, Part 1 and 2201E1, including the 120-day safety update, the incidence of TEAEs did not raise any new safety concerns. The most common AEs were similar to those described in the controlled portion of the trial, with the exception of COVID-19 infections, which were understandably more frequently reported during the OLE and consistent with worldwide epidemiological reporting.

Laboratory Findings

Clinical Chemistry

Laboratory tests were collected at screening, baseline, at visit day 1, 15, 29, and 57, as well as at the end of treatment and end of study visits as shown in Table 17. Mean values for clinical chemistry patterns were similar across treatment arms throughout the study and between baseline and postbaseline values. No subjects had reports of on- or post-treatment AEs related to changes in chemistry laboratory tests.

Hematology

Neutrophil counts were lower in the leniolisib group compared to placebo with the lowest values noted at or around the Day 15 visit. Treatment-emergent neutropenia (ANC <1.5 x 10^9/L) occurred in seven subjects in the leniolisib group versus none in the placebo group. There were

no reports of ANC < 0.5 x 10^9/L nor reports of infections associated with these laboratory findings.

Four subjects in the leniolisib treatment group had neutrophil levels < 1.0 x 10^9/L at a single visit that recovered to > 1.0 x 10^9/L at subsequent visits.

- Subject (b) (6) had an absolute neutrophil count (ANC) of 0.80 x 10^9/L on Day 15, which increased to 1.1 x 10^9/L on Day 29.
- Subject (b) (6) had an ANC of 0.70 x 10^9/L on Day 29, which increased to 2.2 x 10^9/L on Day 57.
- Subject (b) (6) had an ANC of 0.90 x 10^9/L on Day 85. No further values were obtained for this subject who subsequently entered the OLE and received treatment with leniolisib for an additional 18 months with no reports of infections other than SARS-CoV-2 on days 557 through 559 of the OLE.
- Subject (b) (6) had an ANC of 0.79 x 10^9/L on Day 29, which increased to 1.97 x 10^9/L on Day 57.

Three subjects in the leniolisib treatment group had ANC levels between 1.0 x 10^9/L and 1.5 x 10^9/L:

- Subject (b) (6) had an ANC of 1.0 x 10^9/L on Day 15, which increased to 1.1 x 10^9/L on Day 29 and 2.4 x 10^9/L on Day 57, but decreased to 1.7 x 10^9/L at Day 85.
- Subject (b) (6) had an ANC of 1.2 x 10^9/L on Day 15, which was 2.3 x 10^9/L on Day 29, 1.7 x 10^9/L on Day 57, and 1.2 x 10^9/L at Day 85.
- Subject (b) (6) had an ANC of 1.3 x 10^9/L on Day 85, which increased to 1.48 x 10^9/L on the following visit, the first value obtained during the OLE.

Two subjects experienced a decreased neutrophil count during the OLE. Subject (b) (6), described above, had a decreased neutrophil count on Day 2 of the OLE that resolved without intervention on Day 46. An additional case of febrile neutropenia was included in the 120-day update in a 19-year-old female (Subject (b) (6)) who developed febrile neutropenia on day 606 of the OLE. The subject continued to take leniolisib and the AE was attributed to a suspected viral infection.

Vital Signs

No clinically significant changes in vital signs were identified in Trial 2201, Part 1 or 2. Time trend analysis, box plots, and waterfall plots (JMP Clinical 8.0) were used to assess systolic and diastolic blood pressure, heart rate, temperature, and body mass index. Two subjects treated with leniolisib, one with a past medical history of tachycardia, experienced AEs of asymptomatic tachycardia. Overall, no new safety concerns were identified in the analysis of vital signs.

Electrocardiograms (ECGs)

There were no apparent trends in ECG intervals observed in Trial 2201, Part 1 or 2.

QT

Evaluation of effect of leniolisib on QTcF interval as part of Trial 2201 described in Section 6.3.1. Data were analyzed using exposure-response analysis as the primary analysis, which did not suggest that leniolisib is associated with significant QTc prolonging effect.

Immunogenicity

Not applicable.

8.2.4. Analysis of Submission-Specific Safety Issues

To explore labeled adverse reactions with other PI3Kδ inhibitors, the Applicant provided additional safety analyses for infections, malignancies, neutropenias and other cytopenias, skin rashes, gastrointestinal tolerability, cardiovascular disorders or QT prolongation, elevated liver enzymes, and fatigue. No additional safety signals were identified in these analyses, and the safety profile of approved PI3Kδ inhibitors is likely influenced by the concomitant therapies and severity of illness of the target population of oncology patients.

8.2.5. Safety Analyses by Demographic Subgroups

Additional subgroup analyses of TEAEs, including SAEs and AEs leading to treatment discontinuation were conducted by the Clinical Reviewer using JMP Clinical 8.1. The overall incidence of TEAEs was comparable between the two age groups with 83.3% of subjects in age group <18 years versus 89.5% of subjects in age group ≥18 years reporting at least one AE. The majority of subjects were white and the number of subjects of other races was limited; however, no meaningful differences in the occurrences of adverse events by race were identified. By sex, 73% of males in the leniolisib treatment group versus 100% of males in the placebo group experienced at least one AE and 100% of females in the leniolisib treatment group versus 83.3% of females in the placebo group experienced at least one AE; however, there were no major differences in pattern or type of TEAEs between these subgroups. The overall incidence of AEs was comparable between APDS1 (88.0%) and APDS2 subjects (83.3%). Although the number of subjects within each subgroup limits the interpretation of these analyses, there were no apparent differences in the safety profile of leniolisib by age group, sex, race, region, or genotype.

8.2.6. Additional Safety Explorations

Human Carcinogenicity or Tumor Development

No human carcinogenicity studies have been conducted on leniolisib. See Section 5.5.3 for further details on carcinogenicity.

Human Reproduction and Pregnancy

There were no pregnancies reported during Trial 2201, Part 1 or 2, or in Trial 2201E1.

Pediatrics and Assessment of Effects on Growth

The Applicant did not include bone mineral density measurements, formal hypothalamic–pituitary–adrenal (HPA) axis and growth studies in the adult or pediatric clinical development program for leniolisib. However, based on MOA and nonclinical data, leniolisib's effect on growth is not a concern.

Overdose, Drug Abuse Potential, Withdrawal, and Rebound

No overdose cases were reported in Trial 2201, Part 1 or 2, or in Trial 2201E1. There is no known abuse or addiction potential for leniolisib. Withdrawal or rebound effects have not been formally evaluated but are not expected based on the mechanism of action of leniolisib.

8.2.7. Safety in the Postmarket Setting

Safety Concerns Identified Through Postmarket Experience

There is no postmarketing experience with leniolisib

Expectations on Safety in the Postmarket Setting

It is possible that adverse reactions from leniolisib not observed in the clinical development program may occur in the postmarket setting due to longer duration of use in a larger population compared to what was seen in the clinical trials. However, the population included in Trial 2201 is representative of the APDS population as a whole, making results generally applicable to the intended population. Additionally, the duration of the OLE has provided longer term data that has not revealed significant safety concerns for a drug intended to be taken chronically.

8.2.8. Integrated Assessment of Safety

The safety data submitted with this NDA were sufficient to assess the safety of leniolisib at the proposed dose and in the proposed APDS patient population. The safety information for a

leniolisib was primarily derived from the pivotal, placebo-controlled Trial 2201, Part 2 along with long-term safety from the OLE study Trial 2201E1.

The occurrence of SAEs, TEAEs leading to discontinuation, and severe or life-threatening TEAEs in the clinical trial were infrequent and not indicative of a particular safety risk with leniolisib treatment. Extended exposure in the OLE did not reveal concerning safety signals after prolonged use. Common TEAEs and lab abnormalities were generally mild or moderate in severity and can be adequately mitigated through product labeling. Although sinusitis was reported more frequently in the leniolisib group, this may potentially be due to baseline differences in prior sinus disease; other treatment emergent infections and new antibiotic usage during the trial were more common in the placebo group. Based on the clinical data submitted for the APDS program, the overall safety profile of leniolisib for the treatment of APDS is favorable.

8.3. Statistical Issues

Robustness of Efficacy Data

Missing Data Sensitivity Analysis

In Trial 2201, Part 2, there were fourteen subjects who were excluded from the PD analysis set for the analysis of the co-primary endpoint of change from baseline in percentage of naïve B cells: 8 subjects were excluded due to a greater than 48% of naïve B cells at baseline and 6 subjects from the leniolisib treatment group were excluded due to missing data. To examine the robustness of the primary analysis results to missing data, the reviewer performed a sensitivity analysis for the co-primary endpoint to assess the robustness to variations of the missing data assumption underlying the primary analysis. The analysis explored the impact of missing data by imputing missing data with the placebo group completers' mean value at baseline (30.51) and change from baseline at Day 85 (-3.23). The sensitivity analysis demonstrated a similar treatment effect compared to the primary analysis and supported the conclusion of the co-primary efficacy analysis of change from baseline in percentage of naïve B cells in Trial 2201 (Table 46).

The sensitivity analysis using placebo mean imputation supported treatment effect on the co-primary endpoint. While this analysis explored only one alternative missing assumption, this

assumption was fairly conservative, and the result was statistically persuasive. Therefore, the result of effectiveness is considered reasonably robust against the missing data assumption.

Table 46. Missing Data Sensitivity Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set Including Subjects With Naive B Cells <48% of Total B Cells at Baseline)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	14	16.28	27.38 (7.14, 47.62)	0.01
Placebo (N = 8)	5	-11.10		

Source: Statistical Reviewer.

Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolis b group and 3 subjects in the placebo group who had more than 48% naïve B cells at baseline. There were 6 subjects with missing data in the leniolis b group (5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement) and the missing data were imputed with placebo group completer's mean value at baseline (30.51) and change from baseline at Day 85 (-3.23).

Abbreviations: CI, confidence interval; Diff, difference; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic. IgG, Immunoglobulin G;

Post-hoc Modified Analysis on the Co-primary Endpoints

The applicant initially performed a primary analysis on the co-primary endpoints with the use of glucocorticoids and IRT as categorical covariates. However, it was retrospectively recognized that only IV IRT rather than both SC and IV IRT were counted as covariates in the analysis. The Applicant subsequently performed a post-hoc modified analysis on the co-primary endpoints repeating the primary analysis with the use of glucocorticoids and any concomitant IRT as a categorical covariate. From a statistical perspective, this was not the prespecified analysis agreed to in the statistical analysis plan. However, the clinical and statistical reviewers agreed that it was preferable to include the actual number of subjects receiving IRT because the effect should be the same regardless of the route of administration. See Appendix 16.5 for the pre-specified analyses per the SAP.

8.4. Conclusions and Recommendations

The Applicant conducted a single adequate and well-controlled trial (Trial 2201, Part 2) of 12 weeks duration in 31 adult and adolescent subjects with APDS. The trial demonstrated statistically significant improvements on the change from baseline at day 85 in the co-primary endpoints of percentage of naïve B cells out of total B cells and log10 transformed SPD in the index lesions on MRI/CT imaging for leniolisib subjects over placebo. The LS mean treatment difference estimate was -0.25 [95% CI: -0.38, -0.12; p-value =0.0006] in change from baseline in the log10 transformed SPD in the index lesions at Day 85 and 37.30 [95% CI: 24.06, 50.54; p-value=0.0002] in change from baseline at Day 85 in percentage of naïve B cells out of total B cells. The treatment effect on the co-primary endpoints was preserved across supplementary and subgroup analyses, including by age group. Other pharmacodynamic endpoints such as

pAKT inhibition, spleen size/volume, volumetric measures of lymphadenopathy, and additional T cell and B cell subset immune parameters showed improvements with leniolisib treatment compared to placebo. The results are clinically meaningful because a correction of the abnormal immunophenotype associated with APDS is expected to lead to normalization of immune function and improvement in clinical sequelae such as fewer infections, autoimmune manifestations, and lymphoproliferative disease.

Substantial evidence of effectiveness was demonstrated by the results from the single adequate and well-controlled trial and confirmatory evidence consisting of the well-established etiology of the disease, the mechanism of action of the therapy, and PD biomarker data from leniolisib clinical trials in adult and adolescent subjects with APDS.

The safety data submitted with this NDA were sufficient to assess the safety of leniolisib at the proposed dose and in the proposed APDS patient population. The occurrence of SAEs, TEAEs leading to discontinuation, and severe or life-threatening TEAEs in the clinical trial were infrequent and not indicative of a particular safety risk with leniolisib treatment. Extended exposure in the OLE did not reveal concerning safety signals after prolonged use. Common TEAEs and lab abnormalities were generally mild or moderate in severity and can be adequately mitigated through product labeling. Although sinusitis was reported more frequently in the leniolisib group, this may potentially be due to baseline differences in prior sinus disease; other treatment emergent infections and new antibiotic usage during the trial were more common in the placebo group.

Based on the clinical data submitted for the APDS program, the overall benefit/risk assessment of leniolisib for the treatment of APDS is favorable.

9. Advisory Committee Meeting and Other External Consultations

A Pulmonary and Allergy Drug Advisory Committee (PADAC) Meeting was not convened for this application. In determining the need for a PADAC meeting, the Division considered several factors. Although leniolisib will be the first approved drug product for APDS and thus may well become the standard of care, there are few controversial issues requiring external input and discussion given the favorable benefit-risk assessment which considers the lack of available therapies and the serious nature of the disease. Additional considerations centered around the use of novel co-primary endpoints. While the co-primary endpoints of lymph node size and percentage of naïve B cells out of total B cells are not direct reflections of clinically meaningful outcomes and have not been used by the Division to support prior approvals, they are based on a clear understanding of the pathophysiology of the disease and mechanism of action of leniolisib. Therefore, there is clear mechanistic rationale for the use of the chosen endpoints in APDS and the study met statistical significance for both of these co-primary endpoints. Furthermore, there are supportive trends in secondary endpoints.

There are no apparent subgroup differences in efficacy that would limit the indication, the benefits and risks are similar across subgroups, the proposed indication is specific and narrow, and no major approvability issues were identified. Finally, there is public support for the approval of leniolisib in the APDS community.

10. Pediatrics

Leniolisib was granted Orphan Drug Designation on January 30, 2018. PREA requirements do not apply to this orphan drug product.

11. Labeling Recommendations

11.1. Prescription Drug Labeling

A summary of major revisions to the proposed PI are described in the table below.

Table 47. Prescription Drug Labeling

Full Prescribing Information Sections[1]	Rationale for Major Changes Incorporated into the Finalized Prescribing Information (PI)[2]
All Sections	The labeling was revised to provide recommendations and format of the Prescribing Information (PI) to help ensure that the PI was compliant with Physician Labeling Rule (PLR) and current labeling practice.
1 INDICATIONS AND USAGE	JOENJA is indicated for the treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adult and pediatric patients 12 years of age and older.
2 DOSAGE AND ADMINISTRATION	• Added Testing Prior to Treatment of JOENJA subsection because of Embryo-Fetal Toxicity risk. • Deleted (b) (4) • Deleted (b) (4)
5 WARNINGS AND PRECAUTIONS	Vaccination information relocated from Section 7 because vaccinations are considered a precaution and not a drug interaction.
6 ADVERSE REACTIONS	• Added a common adverse reaction table from the placebo-controlled portion of Study 2201 • Added subsection for lab abnormalities to describe decreased neutrophil count
8 USE IN SPECIFIC POPULATIONS (e.g., Pregnancy, Lactation, Females and Males of Reproductive Potential, Pediatric Use, Geriatric Use, Renal Impairment, Hepatic Impairment)	**8.1 Pregnancy and 8.2 Lactation** No human data is available but included animal data. **8.4 Pediatric Use** Revised the pediatric use statement and removed (b) (4) Added language that conveys support for use in the pediatric population and added that there is no recommended dosage for pediatric patients less than 12 years of age or who weigh less than 45 kg. Also

	included Juvenile Animal Toxicity Data.
14 CLINICAL STUDIES	• Removed [redacted] (b) (4) • Added demographics/baseline characteristics table • Added forest plot to display co-primary endpoints by age group (adults vs adolescents) • [redacted] (b) (4) were removed

12. Risk Evaluation and Mitigation Strategies (REMS)

A REMS was not deemed necessary for this application.

13. Postmarketing Requirements and Commitment

The following postmarketing requirements (PMR) and commitments (PMC) have been agreed upon with the Applicant.

13.1. Postmarketing Requirements

13.1.1. PMR-1, 6-Month Carcinogenicity Study in Transgenic (Rash2) Mice

Requirement

Conduct a 6-month carcinogenicity study in transgenic (rasH2) mice.

Table 48. PMR-1 Milestones Schedule, Carcinogenicity Study in Transgenic (rasH2) Mice

Milestone	Status
Final protocol submission	Submitted
Study completion date	01/2023
Final report submission date	04/2023

Abbreviations: PMR, postmarketing requirement

13.1.2. PMR-2, 2-Year Carcinogenicity Study in Rats

Requirement

Conduct a 2-year carcinogenicity study in rats.

Table 49. PMR-2 Milestones Schedule, 2-Year Carcinogenicity Study in Rats

Milestone	Status
Final protocol submission	Submitted
Study completion date	07/2025
Final report submission date	03/2026

Abbreviations: PMR, postmarketing requirement.

13.1.3. PMR-3, Pre- and Postnatal Development Study in Rats

Requirement

Conduct a pre- and post- natal development study in rats.

Table 50. PMR-3 Milestones Schedule, Pre- and Postnatal Development Study in Rats

Milestone	Status
Study completion date	12/2022
Final report submission date	04/2023

Abbreviations: PMR, postmarketing requirement.

13.1.4. PMR-4, Cocktail DDI Clinical Study

Requirement

Conduct a cocktail drug-drug interaction (DDI) clinical study to assess the effect of leniolisib on the pharmacokinetics of CYP1A2 substrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and MATE/ OCT2 substrate (metformin).

Table 51. PMR-4 Milestones Schedule, Conduct a Cocktail DDI Clinical Study

Milestone	Status
Draft protocol submission	05/2023
Final protocol submission	09/2023
Study completion date	03/2024
Final report submission date	09/2024

Abbreviations: DDI, drug-drug interaction; PMR, postmarketing requirement.

13.1.5. PMR-5, Hepatic Impairment Clinical Study

Requirement

Conduct a hepatic impairment clinical study to assess the impact of moderate and severe hepatic impairment on the pharmacokinetics of leniolisib.

Table 52. PMR-5 Milestones Schedule, Hepatic Impairment Clinical Study

Milestone	Status
Draft protocol submission	06/2023
Final protocol submission	10/2023
Study completion date	04/2024
Final report submission date	10/2024

Abbreviations: PMR, postmarketing requirement

13.2. Postmarketing Commitment

13.2.1. PMC-1, Submission of Final Clinical Study Report From OLE Study CCDZ732201E1

Commitment

Submit the final clinical study report (CSR) from the ongoing open-label extension study CCDZ732201E1 with leniolisib in subjects 12 years and older with activated phosphoinositide 3-kinase delta syndrome/p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency (APDS) to provide long-term safety and clinical outcomes, including all clinical laboratory and imaging test results, frequency of infections and antibiotic use, occurrence of adverse events, hospitalizations, and deaths.

Table 53. PMC-1 Milestones Schedule, Submission of Final Clinical Study Report From OLE Study CCDZ732201E1

Milestone	Status
Final protocol submission	Submitted
Study completion date	11/2027
Final report submission date	05/2028

Abbreviations: OLE, open-label extension; PMC, postmarketing commitment.

14. Division Director (DPACC) Comments

Activated phosphoinositide-3 kinase δ syndrome (APDS) is a rare autosomal dominant immunodeficiency disease that was first described in 2013. Clinical manifestations of APDS include recurrent sinopulmonary bacterial infections, bronchiectasis, chronic herpesvirus viremia, lymphoproliferation, autoimmunity, enteropathy, and lymphoma. The clinical presentation is heterogeneous, but the most common features are recurrent sinopulmonary bacterial infections and lymphoproliferation. Laboratory features of APDS include:

- Hypogammaglobulinemia;
- B cell defects, including B cell lymphopenia with reduced naïve B cells and class-switched memory B cells, and an increase in transitional B cells; and
- T cell defects, including reduced naïve T cells and expansion of terminally differentiated effector memory T cells expressing RA (T_{EMRA}) and senescent CD8+ T cells.

PI3Kδ is a heterodimeric protein that consists of a p110δ catalytic subunit and a p85 regulatory subunit. PI3Kδ is expressed primarily in hematopoietic cells and its activation by antigen receptors, co-receptors, growth factor receptors, and cytokine receptors leads to phosphorylation of phosphatidylinositol 3,4-bisphosphate (PIP2) to generate phosphatidylinositol 3,4,5-triphosphate (PIP3), with subsequent Akt phosphorylation and activation of the mTOR pathway; activation through this pathway leads to cell activation, growth, differentiation, metabolism, and inhibition of apoptosis. APDS results from autosomal dominant gain-of-function mutations in the *PIK3CD* gene encoding the p110δ catalytic subunit (APDS1) or loss-of-function mutations in the *PIK3R1* gene encoding the regulatory p85α subunit (APDS2). These mutations result in activation of PI3Kδ and the resulting downstream events, including increased Akt phosphorylation, which are critical to the immune defects and lymphoproliferation that are central to disease pathophysiology.

Pharming Technologies B.V. submitted this NDA to support the approval of leniolisib for the "treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adult and pediatric patients 12 years of age and older." Leniolisib (CDZ173) is a small molecule inhibitor of the p110δ subunit of PI3Kδ that reduces signaling through this pathway. It is a new molecular entity (NME) that is not approved for any indication in the U.S. or any other country.

Product Quality

The Office of Pharmaceutical Quality (OPQ) reviewed the CMC package submitted with this application and recommended approval. Assessment of the drug substance, drug product, labeling, manufacturing, and biopharmaceutics were all considered adequate.

Leniolisib tablets (70 mg) are for oral administration and immediate release. The drug is a free base that has low solubility and high permeability (BCS Class II). The drug substance has one chiral center (b) (4). All regulatory starting materials satisfy the ICH Q11 guidelines, and the Applicant has performed adequate evaluations of the potential impurities in the drug substance based on the manufacturing process. The drug substance container closure system components are typical of what the Agency accepts, (b) (4)

The drug product is formulated with compendial grade excipients and is manufactured (b) (4) It contains the active ingredient in its phosphate salt form, leniolisib phosphate, which is a new molecular entity. The Applicant provided up to 12 months of long-term stability studies and 6 months of accelerated stability studies for three registration batches with supporting stability results of development and scale-up batches. There are no trends observed for any of the stability parameters under both accelerated and long-term conditions for studied periods and the Applicant committed to continue the stability studies for primary stability batches. The proposed shelf-life of 24 months for the leniolisib film-coated tablets with the container closure system of HDPE bottles with an aluminum induction seal and (b) (4) screw cap closures with a storage condition of do not store above 25°C, do not refrigerate is acceptable.

Nonclinical Pharmacology-Toxicology

The Nonclinical Pharmacology-Toxicology review team recommended approval of this application from a nonclinical perspective. The Applicant characterized leniolisib in safety pharmacology, genetic toxicology, general toxicology, and developmental/reproductive toxicology studies. Adequate exposure margins for leniolisib were obtained in all species that exceed the predicted human exposure at the MRHD of 70 mg BID. Leniolisib administration to pregnant rats and rabbits was associated with embryofetal toxicity, including malformations at exposures approximately 2-6 times the MRHD on an AUC basis. Labeling includes the need to verify pregnancy status in females of reproductive potential prior to initiating treatment and the recommendation to use highly effective contraception during leniolisib treatment.

Three Nonclinical Pharmacology-Toxicology Postmarketing Requirements (PMR) have been agreed upon with the Applicant and are included with this approval action:

- Conduct a 6-month carcinogenicity study in transgenic (rasH2) mice.
- Conduct a 2-year carcinogenicity study in rats.
- Conduct a pre- and post- natal development study in rats.

Clinical Pharmacology

The Office of Clinical Pharmacology, Division of Inflammation and Immune Pharmacology (DIIP) reviewed the clinical pharmacology data relevant to this application and recommend approval from a Clinical Pharmacology perspective.

The clinical pharmacology program for leniolisib includes 8 clinical studies (Studies 2101, 2102, 2104, 2203, 2201, 2201E1, LE1101, and LE2101) (Table 5). Of the clinical studies in the program, 5 were conducted in healthy participants (Studies LE1101, LE2101, 2101, 2102, and 2104), 1 was conducted in subjects with Sjögren's syndrome (Study 2203), and 2 were conducted in subjects with APDS (Studies 2201 and 2201E1). Phase 1 clinical studies provided plasma and urine leniolisib pharmacokinetic (PK) information following the single- and multiple-dose in healthy participants; assessed absorption, distribution, metabolism, excretion (ADME) of leniolisib; and evaluated the drug-drug interaction (DDI) potential with cytochrome P450 (CYP)3A4/CYP2D6/P-glycoprotein (P-gp) inhibitors (itraconazole and quinidine) and CYP3A/UDP-glucuronosyl transferase (UGT)1A1/sulfotransferase (SULT)1E1 substrates (oral contraceptives ethinylestradiol and levonorgestrel). Phase 2 and 3 clinical studies provided single- and multiple-dose PK data on plasma leniolisib as well as pharmacodynamic (PD) data on % phosphorylated protein kinase B (pAkt)-positive B cells in subjects with Sjögren's syndrome and subjects with APDS.

Dose Selection

Dose selection was based on the PD endpoint (*ex vivo* %pAkt-positive B cells) following leniolisib treatment, as Akt is a direct downstream target of activated PI3Kδ. Inhibition of pAkt in *ex vivo* non-stimulated and stimulated B cells was observed shortly after dosing with leniolisib in a dose- and concentration-dependent manner in both the first-in-human study at leniolisib doses from 10 to 400 mg (Study 2101 Parts 1 and 3) and the dose-finding part of the pivotal study in subjects with APDS evaluating leniolisib at 10, 30, and 70 mg twice daily (Study 2201 Part 1).

In healthy participants, inhibition of *ex vivo* stimulated pAkt-positive B cells was observed shortly after dosing with leniolisib, while negligible reduction was observed with placebo. Percent pAkt inhibition was highest in the 70 and 140 mg twice daily dose groups and peaked at approximately 1-hour post-dose. Percent pAkt-positive B cells gradually returned close to baseline levels in approximately 24 hours from the last dose. The maximal pAkt inhibition appeared to be reached at doses of 70 mg and 140 mg BID (approximately 90%). Therefore, doses up to 70 mg BID were evaluated in Study 2201 (Part 1) in APDS subjects.

In subjects with APDS from Part 1 of Study 2201, a clear and apparent maximum reduction in both stimulated and non-stimulated pAkt-positive B cells was observed as early as 1-hour post-dose. The early onset of pAkt inhibition occurred at approximately the same time as plasma leniolisib T_{max}. The pAkt inhibition started to rebound after T_{max} (1 hr) and returned to baseline

level after 12 hours thereby providing support for the proposed BID dosing regimen. Although a single dose of 10 mg generally reduced %pAkt-positive B cells to less than 20%, the inhibition generally waned over the ensuing 12 hours. While the 30- and 70-mg dose levels did not differ in their maximum effect, inhibition was more sustained over time at 70 mg, which may therefore be more optimal in terms of time-averaged inhibition and inhibition at trough (12 hours post-dose).

T**wo Clinical Pharmacology Post Marketing Requirements (PMR) have been agreed upon with the Applicant and are included with this approval action:**

- Conduct a hepatic impairment clinical study to assess the impact of moderate and severe hepatic impairment on the pharmacokinetics of leniolisib.
- Conduct a cocktail drug-drug interaction (DDI) clinical study to assess the effect of leniolisib on the pharmacokinetics of CYP1A2 sustrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and MATE/ OCT2 substrate (metformin).

Substantial Evidence of Effectiveness

Substantial evidence of effectiveness for leniolisib in subjects with APDS was established in the leniolisib development program with one adequate and well-controlled trial, with confirmatory evidence provided by a well-understood pathophysiology of disease, relative selective inhibition of PI3Kδ by leniolisib, and demonstration of a decrease in phosphorylation of the downstream target of PI3Kδ, Akt, in subjects treated with leniolisib.

The single adequate and well-controlled trial was a 12-week, randomized, double-blind, placebo-controlled, multi-center phase 3 trial (Study 2201, Part 2) to assess the efficacy and safety of leniolisib in adults and children ≥12 years of age with APDS. For assessment of efficacy, the trial included 12 adolescents (12-17 years of age) and 19 adults (≥18 years of age) who were randomized 2:1 to leniolisib 70 mg orally twice daily (n=21) or placebo twice daily (n=10) for a 12-week treatment period.

Since APDS is a rare disease and the clinical presentation is heterogeneous, the accepted co-primary efficacy endpoints were based on the lymphoproliferation that is a common clinical feature of APDS (and a required inclusion criteria) and the characteristic B cell defect of diminished naïve B cells due to a block of B cell development at the transitional B cell stage. Based on these considerations, the agreed co-primary endpoints were:

- Change from baseline in the log10 transformed sum of product of diameters (SPD) in the index lesions selected as per the Cheson methodology from MRI/CT imaging at day 85.
- Change from baseline in percentage of naïve B cells (CD19+CD27-CD10-) out of total B cells at day 85.

The primary efficacy analysis included 27 subjects who received leniolisib or placebo and did not have significant protocol deviations that impacted assessment of the co-primary endpoints.

The trial met statistical significance for both co-primary endpoints at α=0.05 significance level. The LS mean treatment difference estimate was:

- -0.25 [95% CI: -0.38, -0.12; p-value = 0.0006] in change from baseline in the log10 transformed SPD in the index lesions at Day 85.
- 37.30 [95% CI: 24.06, 50.54; p-value = 0.0002] in change from baseline at Day 85 in naïve B cells.

The treatment effects were preserved across sensitivity and subgroup analyses. Secondary endpoints, including reductions in index and non-index lesion (lymphoid) volumes and spleen volume favored leniolisib. All other clinical endpoints were exploratory and not considered in assessment of efficacy due to small numbers.

Confirmatory evidence of effectiveness is provided by a well-understood pathophysiology based on over-activation of PI3Kd, inhibition of p110d by leniolisib, and ex vivo demonstration of an 80% reduction in pAkt positive B cells in APDS patients treated with leniolisib 70 mg twice a day.

Safety Assessment

Across the clinical development program for leniolisib (phase 1, phase 2, and phase 3 studies), 196 healthy volunteers, 20 subjects with Sjogren's syndrome, and 38 subjects with APDS received at least one dose of leniolisib. The safety data from Study 2201, Part 2 submitted with this NDA were sufficient to assess the safety of leniolisib at the proposed dose and in the proposed APDS patient population.

The occurrence of SAEs, TEAEs leading to discontinuation, and severe or life-threatening TEAEs in the clinical trial were infrequent and not indicative of a particular safety risk with leniolisib treatment. Extended exposure in the OLE did not reveal concerning safety signals after prolonged use. Common TEAEs and lab abnormalities were generally mild or moderate in severity and can be adequately mitigated through product labeling. Although sinusitis was reported more frequently in the leniolisib group, this may potentially be due to baseline differences in prior sinus disease; other treatment emergent infections and new antibiotic usage during the trial were more common in the placebo group.

One Clinical Safety Post**marketing Commitment (PMC) has been agreed upon with the Applicant and is included with this approval action:**

- Submit the final clinical study report (CSR) from the ongoing open-label extension study CCDZ732201E1 with leniolisib in subjects 12 years and older with activated phosphoinositide 3-kinase delta syndrome/p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency (APDS) to provide long-term safety and clinical outcomes, including all clinical laboratory and imaging test results, frequency of infections and antibiotic use, occurrence of adverse events, hospitalizations, and deaths.

Conclusions

Substantial evidence of effectiveness for leniolisib in the treatment of APDS has been demonstrated based on the results from the single adequate and well-controlled trial, as well as confirmatory evidence consisting of the well-established pathophysiology of the disease, the mechanism of action of the therapy, and PD biomarker data from the leniolisib clinical trial in adult and adolescent subjects with APDS. Overall, there is a favorable benefit-risk assessment for leniolisib in the treatment of adults and adolescents ≥12 years of age at a dose of 70 mg orally twice a day to support **approval** of this application. Approval of leniolisib provides the first treatment specifically for APDS and meets an unmet need for treatment of this rare and severe disease.

15. Office Director (OII) Comments

I concur with the recommendation from the Division of Pulmonology, Allergy and Critical Care to approve NDA 217759, leniolisib, for the treatment of APDS, activated phosphoinositide-3 kinase δ (PI3Kδ) syndrome, in adult and pediatric patients 12 years of age and older. APDS is an ultra-rare autosomal dominant immunodeficiency disease associated with increased morbidity and potentially fatal sequelae, including bronchiectasis and B cell lymphoma. In this condition, PI3Kδ is constitutively active, resulting in dysregulated B and T cell development, and clinical features of immunodeficiency and overactive or inappropriate immune responses. There are no approved therapies for APDS. Leniolisib is administered orally twice daily at the 70 mg dose, the dose that resulted in sustained inhibition of pAKT-positive B cells compared to lower doses.

The demonstration of substantial evidence of effectiveness of leniolisib, a kinase inhibitor and new molecular entity, is supported by one 12-week, placebo-controlled trial conducted in 19 adults and 12 adolescents, and confirmatory evidence from mechanistic and pharmacodynamic data. Clinically meaningful and statistically significant improvements in the co-primary endpoints of change from baseline in 1) percent of naïve B cells (out of total B cells), and 2) index lymph node size, were observed in subjects treated with leniolisib compared to placebo. Leniolisib systemic exposures and effects on the co-primary endpoints were comparable between adult and adolescent subjects. Improvement in index lesions appeared to continue through the open label extension of the trial.

Favorable effects were also observed for several pharmacodynamic endpoints (pAKT-positive B cell inhibition, improvements in spleen size/volume, lymph node volume, and T and B cell subsets) with leniolisib treatment compared to placebo.

Taken together, these findings support that leniolisib's normalization of the underlying immune abnormality, i.e., change from baseline in naïve B cells, is expected to result in normalization of immune function and improvement in clinical sequelae (e.g., fewer infections, decreased risk of lymphoproliferative disease).

Leniosilib treatment was well tolerated; prolonged exposure in the open label extension trial did not reveal new concerning safety signals. The risks of leniolisib can be adequately addressed in product labeling.

Given that hepatomegaly is a commonly reported feature of APDS, the Applicant will be required to conduct a hepatic impairment study to assess the impact of moderate and severe hepatic impairment on the pharmacokinetics of leniolisib. In addition, a postapproval cocktail DDI clinical study will be required to assess the effects of leniolisib on the pharmacokinetics of the following: CYP1A2 substrate (caffeine), CYP3A4 substrate (midazolam), BCRP/OATB1B1/1B3 substrate (rosuvastatin), and MATE/ OCT2 substrate (metformin).

With the approval of this NDA, the Applicant will be granted a rare pediatric disease priority review voucher.

16. Appendices

16.1. References

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Draft Guidance for Industry *Pharmacokinetics in Patients with Impaired Renal Function — Study Design, Data Analysis, and Impact on Dosing and Labeling* (September 2020)

16.2. Financial Disclosure

From Trials CCDZ2201X2201, Parts 1 and 2, and CCDZ2201X2201E1, the Applicant certified that the clinical investigators did not participate in any financial arrangement whereby the value of compensation to the investigator could be affected by the outcome of the study, had no proprietary interest in the product, nor was a recipient of significant payments.

Covered Clinical Study: CCDZ2201X2201, Parts 1 and 2, and CCDZ2201X2201E1

Was a list of clinical investigators provided:	Yes ☒	No ☐ (Request list from Applicant)
Total number of investigators identified: 34		
Number of investigators who are Sponsor employees (including both full-time and part-time employees): 0		
Number of investigators with disclosable financial interests/arrangements (Form FDA 3455): 0		

If there are investigators with disclosable financial interests/arrangements, identify the number of investigators with interests/arrangements in each category (as defined in 21 CFR 54.2(a), (b), (c) and (f)): Compensation to the investigator for conducting the study where the value could be influenced by the outcome of the study: _____ Significant payments of other sorts: _____ Proprietary interest in the product tested held by investigator: _____ Significant equity interest held by investigator in S Sponsor of covered study: _____		
Is an attachment provided with details of the disclosable financial interests/arrangements:	Yes ☐	No ☐ (Request details from Applicant)
Is a description of the steps taken to minimize potential bias provided:	Yes ☐	No ☐ (Request information from Applicant)
Number of investigators with certification of due diligence (Form FDA 3454, box 3) _____		
Is an attachment provided with the reason:	Yes ☐	No ☐ (Request explanation from Applicant)

16.3. OCP Appendices

16.3.1. Overview of Bioanalytical Methods

Bioanalytical methods supporting the biopharmaceutics and clinical pharmacology assessment of leniolisib are outlined in Table 54.

Table 54. Bioanalytical Methods Supporting the Clinical Pharmacology Assessment of Leniolisib in Clinical Studies

Analyte	Biomatrix	Bioanalytical Method; Laboratory	Clinical Studies Supported
Leniolisib	Human plasma	1100679b; (b) (4)	2101 and 2201 Part 1
		15BASM071V1; (b) (4)	2102, 2104, and 2203
		PKH/MOA/1031 and 1464; (b) (4)	LE1101, LE2101, 2201 Part 2, and 2201E1
		PKH/MOA/1411; (b) (4)	2201E1
	Human urine	(b) (4) 1100679g	2101
Itraconazole	Human plasma	15BAS0302; (b) (4)	2102
Ethinylestradiol and levonorgestrel	Human plasma	Draft10BASM032V2; (b) (4)	2104
pAkt-positive B cells	Human whole blood	ICDFC 6; (b) (4)	2101
		ICDFC 6;	2203 and 2201

pAkt=phosphorylated Akt.

Source: Summary of Biopharmaceutical Studies, Table 7.

16.3.1.1. Determination of Leniolisib in Human Plasma

Throughout clinical development, assay for plasma leniolisib in clinical samples was performed by the following laboratories: (b) (4) Initially, bioanalytical methods for determination of plasma leniolisib were developed and validated by (b) (4) (Method 1100679b) and (b) (4) (Method 15BASM071V1). The (b) (4) method was later transferred to (b) (4) (b) (4) (Method PKH/MOA/1031) and validated. Following the method transfer, a subsequent partial validation to extend the quantitation range of Method PKH/MOA/1031 was performed at (b) (4) (Method PKH/MOA/1464).

A chiral bioanalytical method for the determination of (S)- and (R)-enantiomers of leniolisib in plasma was also developed and validated by (b) (4) (Method PKH/MOA/1411).

16.3.1.1.1. (b) (4) (1100679b) Method

A liquid chromatography-tandem mass spectrometry (LC-MS/MS) method for the assay of leniolisib in K_3EDTA-treated human plasma was developed and validated at (b) (4) Validation results met acceptance criteria and are summarized in Table 55. The validated quantitation range was from 3.00 to 1000 ng/mL; quality control (QC) samples from 3.00 to 750 ng/mL were used for the accuracy and precision assessments. Dilution integrity was assessed at 100-fold dilution of 2000 ng/mL.

Table 55. Summary of Method Performance for Determination of Leniolisib in Human Plasma (1100679b)

Bioanalytical method validation report name, amendments, and hyperlinks	Quantitative determination of CDZ173 in human plasma by LC-MS/MS Method Validation Report DMPK R1100679b		
Method description	Protein precipitation from human plasma (K_3EDTA) samples followed by pellet filtration using solid phase extraction and analysis of filtrate by LC-MS/MS in SRM mode using ESI in positive mode as the ionization technique		
Materials used for standard calibration curve and concentration	Leniolisib phosphate, (b) (4) Batches 1110001 and 1251002 REF Calibration standards: 3.00, 30.0, 150.0, 375.0, 600, 800, and 1000 ng/mL Internal standard: $^{13}CD_3$-leniolisib, (b) (4) Batches SSE 208-1 and SSE 261-1		
Validated assay range	3.00 to 1000 ng/mL		
Material used for QCs and concentration	Leniolisib phosphate, (b) (4) Batches 1110001 and 1251002 REF QC levels: 3.00 (LLOQ), 9.00 (LQC), 500 (MQC), 750 (HQC), and 2000 (Dilution QC) ng/mL Internal standard: $^{13}CD_3$-leniolisib, (b) (4) Batches SSE 208-1 and SSE 261-1		
Minimum required dilutions	NA		
Source and lot of reagents	Human plasma, K_3EDTA-treated (b) (4) Methanol, Lichrosolv® gradient grade (b) (4) Acetonitrile, Chromasolv® grade (b) (4) Tetrahydrofuran, Chromasolv® grade (b) (4) Formic acid (b) (4) Purified deionized water using (b) (4)		
Regression model and weighting	Quadratic ($y=ax^2+bx+c$), weighted ($1/x^2$)		
Validation parameters	**Method validation summary**		**Source location**
Standard calibration curve performance during accuracy and precision runs	Number of standard calibrators from LLOQ to ULOQ	7	DMPK R1100679b, Table 6-12
	Cumulative accuracy (%bias) from LLOQ to ULOQ	-1.30% to 2.10%	
	Cumulative precision (%CV) from LLOQ to ULOQ	≤7.80%	

Performance of QCs during accuracy and precision runs	**Cumulative accuracy (%bias) in 4 QC levels**	1.90% to 6.20%	DMPK R1100679b, Table 6-14
	Interbatch %CV	≤7.90 %	
	Total error	NR	
Selectivity and matrix effect	Selectivity for leniolisib was considered acceptable in 6 batches of plasma tested. Interference from blank matrix was 0% of response at LLOQ for leniolisib and 0% of response at working concentration for IS. Matrix effect from LQC to HQC: Mean matrix factor: 0.98 to 1.01, precision (%CV): ≤5.94% Mean IS-normalized matrix factor: 0.98 to 1.02, precision (%CV): ≤6.90%		DMPK R1100679b, Table 6-1, Table 6-2, Table 6-5, Table 6-6, and Table 6-7
Interference and specificity	Contribution of IS to the signal of leniolisib (4.00% interference) and of leniolisib to the signal of IS (0.30% interference) was not significant.		DMPK R1100679b, Table 6-3 and Table 6-4
Hemolysis effect	NA		NA
Lipemic effect	NA		NA
Dilution linearity and hook effect	100-fold dilution of 2000 ng/mL, accuracy (%bias): 1.50%, precision (%CV): 3.89% Hook effect: NA		DMPK R1100679b, Table 6-17
Bench-top/process stability	71 h in plasma at room temperature. 25.2 h for processed extract in autosampler at room temperature.		DMPK R1100679b, Table 6-18 and Table 6-20
Freeze-thaw stability	3 cycles at <-15°C.		DMPK R1100679b, Table 6-19
Long-term storage	182 days in spiked plasma at <-15°C. 461 days in incurred plasma samples at <-15°C.		DMPK R1100679b, Table 6-21 and Table 6-22
Parallelism	NR		NA
Carryover	Carryover was evaluated by injecting blank matrix samples after the ULOQ standard. There was no significant carryover evident for leniolisib (≤16.9% carryover) or IS (0% carryover).		DMPK R1100679b, Table 6-10
	Method Performance in CCDZ173X2101 **Bioanalytical Report: DMPK RCCDZ173X2101**		
Assay passing rate	54 of 67 runs (81%) in plasma met acceptance criteria		DMPK RCCDZ173X2101, Table 5-1
Standard curve performance	Cumulative accuracy (%bias): -0.5% to 0.3% Cumulative precision (%CV): ≤6.3%		DMPK RCCDZ173X2101, Table 5-7
QC performance	Cumulative accuracy (%bias): -0.6% to 9.2% Cumulative precision (%CV): ≤98.7%* Total error: NR * The high CV is due to 2 datapoints of 135 datapoints (121 and -0.513 ng/mL) at LQC (9.00 ng/mL). Excluding these 2 datapoints, the cumulative precision across the QC range is ≤8.7% CV.		DMPK RCCDZ173X2101, Table 5-9

Method reproducibility	Incurred sample reanalysis was performed on 441 samples, and 368 (83%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2101, Table 9-4
Study sample analysis stability	Samples were received frozen and stored at ≤-15°C. Samples were analyzed within the established stability limit of 182 days in spiked plasma at <-15°C and 461 days in incurred plasma samples at <-15°C.	DMPK RCCDZ173X2101, Section 1 and Section 6
Standard calibration curve performance during accuracy and precision	53 calibrator sets with 2 replicates at each of 7 concentration levels from 3.00 to 1000 ng/mL (total 742 datapoints) – 35 excluded from curve fitting due to no value, technical issue, or value deviating by ≥15%.	
	Method Performance in CCDZ173X2201 Part 1 Bioanalytical Report: DMPK RCCDZ173X2201	
Assay passing rate	LIMS Study CCDZ173X2201_A1: 2 of 2 runs (100%) met acceptance criteria. LIMS Study CCDZ173X2201_A2: 12 of 13 runs (92%) met acceptance criteria.	DMPK RCCDZ173X2201, Section 5.2
Standard curve performance	Cumulative accuracy (%bias) LIMS Study CCDZ173X2201_A1: -3.8% to 4.3% LIMS Study CCDZ173X2201_A2: -1.3% to 2.7% Cumulative precision (%CV) LIMS Study CCDZ173X2201_A1: NR LIMS Study CCDZ173X2201_A2: ≤7.2%	DMPK RCCDZ173X2201, Table 5-3 and Table 5-4
QC performance	Cumulative accuracy (%bias) LIMS Study CCDZ173X2201_A1: -2.2% to 2.8% LIMS Study CCDZ173X2201_A2: -3.0% to 5.0% Cumulative precision (%CV) LIMS Study CCDZ173X2201_A1: ≤11.7% LIMS Study CCDZ173X2201_A2: ≤7.9% Total error: NR	DMPK RCCDZ173X2201, Table 5-5 and Table 5-6
Method reproducibility	Not performed	DMPK RCCDZ173X2201, Section 7
Study sample analysis stability	Samples were received frozen and stored at ≤-15°C. Samples were analyzed within the established stability limit of 182 days in spiked plasma at <-15°C and 461 days in incurred plasma samples at <-15°C.	DMPK RCCDZ173X2201, Section 1 and Section 6
Standard calibration curve performance during accuracy and precision	LIMS Study CCDZ173X2201_A1: 2 calibrator sets with 2 replicates at each of 7 concentration levels from 3.00 to 1000 ng/mL (total 28 datapoints) – none excluded from curve fitting. LIMS Study CCDZ173X2201_A2: 12 calibrator sets with 2 replicates at each of 7 concentration levels from 3.00 to 1000 ng/mL (total 168 datapoints) – 4 datapoints rejected due to deviation of 15% or 20% from nominal.	

CV=coefficient of variation; ESI=electrospray ionization; HQC=high QC; IS=internal standard; LC-MS/MS=liquid chromatography coupled with tandem mass spectroscopy; LLOQ=lower limit of quantification; LQC=low QC; MQC=middle QC; NA=not applicable; NR=not reported; QC=quality control; SRM=selective reaction monitoring; ULOQ=upper limit of quantification.

Source: DMPK Report R1100679b

16.3.1.1.2. [redacted] (15BASM071V1) Method

An LC-MS/MS method for the assay of leniolisib in K_3EDTA-treated human plasma was developed and validated at [redacted] (DMPK R1501017). Validation results met acceptance criteria and are summarized in Table 56. The validated quantitation range was from 3.00 to 1000 ng/mL; QC samples from 3.00 to 750 ng/mL were used for the accuracy and precision assessments. Dilution integrity was assessed at 10-fold dilution of 5000 ng/mL. Interference from itraconazole, quinidine, ethinylestradiol, and levonorgestrel have been evaluated; no significant chromatographic interference at the retention time of leniolisib was observed for these compounds. Assessment of interference from quinidine, ethinylestradiol, and levonorgestrel were within-study validation experiments.

Cross-validation was performed between the [redacted] and [redacted] (15BASM071V1) methods for assay of leniolisib in plasma. Validation results met acceptance criteria with 39 of 50 (78%) samples assayed by [redacted] found to be within ±20.0% difference from the reference laboratory [redacted] determined value.

Table 56. Summary of Method Performance for Determination of Leniolisib in Human Plasma (15BASM071V1), Report DMPK R1501017

Bioanalytical method validation report name, amendments, and hyperlinks	Quantitative determination of CDZ173 in human plasma by LC-MS/MS Method Validation Report DMPK R1501017 Amendment 1 DMPK R1501017-01 Assessment of interference from itraconazole, quinidine, ethinylestradiol, and levonorgestrel were performed as within-study validation and are reported in DMPK RCCDZ173X2102 and DMPK RCCDZ173X2104.	
Method description	Protein precipitation of human plasma (K_3EDTA) samples and analysis of diluted samples by LC-MS/MS in MRM mode using ESI as the ionization technique	
Materials used for standard calibration curve and concentration	Leniolisib phosphate, (b) (4) Batches 1251002 REF, 1251002, and 1010006837 Calibration standards: 3.00, 6.00, 15.0, 75.0, 300, 800, and 1000 ng/mL Internal standard $^{13}CD_3$-leniolisib, (b) (4) Batch SSE 261-1	
Validated assay range	3.00 to 1000 ng/mL	
Material used for QCs and concentration	Leniolisib phosphate, (b) (4) Batches 1251002 REF, 1251002, and 1010006837 QC levels: 3.00 (LLOQ), 7.50 (LQC), 60.0 (GMQC), 500 (MQC), 750 (HQC), and 5000 (Dilution QC) ng/mL Internal standard $^{13}CD_3$-leniolisib, (b) (4) Batch SSE 261-1	
Minimum required dilutions	NA	
Source and lot of reagents	Human plasma, K_3EDTA-treated (b) (4) Lots 15BAS0051B.0001, 15BAS0051B.0002, 15BAS0075B.0006, 15BAS0051B.0003, 14BAS0178B.0001, 15BAS0129B.0002, and 16BAS0251B.0001 Itraconazole (b) (4) lot IRNNI Methanol, HPLC grade (b) (4) Acetonitrile, HPLC grade (b) (4) Isopropanol, HPLC grade (b) (4) Formic acid, ACS grade (b) (4) Purified deionized water using (b) (4)	
Regression model and weighting	Linear (y=ax+b), weighted ($1/x^2$)	
Validation parameters	**Method validation summary**	**Source location**
Standard calibration curve performance	Number of standard calibrators from LLOQ to ULOQ: 7	DMPK R1501017-01, Table 4-16

during accuracy and precision runs	Cumulative accuracy (%bias) from LLOQ to ULOQ	-0.7% to 1.2%	
	Cumulative precision (%CV) from LLOQ to ULOQ	≤3.4%	
Performance of QCs during accuracy and precision runs	**Cumulative accuracy (%bias) in 5 QC levels**	1.7% to 12.0%	DMPK R1501017, Table 4-17
	Interbatch %CV	≤4.9%	
	Total error	NR	
Selectivity and matrix effect	Selectivity for leniolisib was considered acceptable in 6 individual lots of plasma tested. Interference from blank matrix was ≤1.0% of response at LLOQ for leniolisib and 0% of response at working concentration for IS. Matrix effect from LQC to HQC: Mean matrix factor: 1.07 to 1.09, precision (%CV): ≤4.8% Mean IS-normalized matrix factor: 0.95 to 1.03, precision (%CV): ≤1.4%		DMPK R1501017, Table 4-1, Table 4-2, Table 4-6, Table 4-7, and Table 4-8
Interference and specificity	No significant chromatographic interference from IS, itraconazole (2500 ng/mL), quinidine (5000 ng/mL), ethinylestradiol (5.00 ng/mL), or levonorgestrel (0.500 ng/mL) was detected at the retention time of leniolisib.		DMPK R1501017, Table 4-3, Table 4-4, and Table 4-5 DMPK RCCDZ173X2102, Table 5-14 DMPK RCCDZ173X2104, Table 5-5
Hemolysis effect	No significant effect of hemolysis (2%[v/v] whole blood) at LQC (7.5 ng/mL) and HQC (750 ng/mL). Accuracy (%bias): 1.7% to 2.9% Precision (%CV): ≤1.7%		DMPK R1501017, Table 4-9
Lipemic effect	No significant effect of lipemia (≥20 mg/mL lipid concentration) at LQC (7.5 ng/mL) and HQC (750 ng/mL). Accuracy (%bias): 1.5% to 5.5% Precision (%CV): ≤3.2%		DMPK R1501017, Table 4-10
Dilution linearity and hook effect	10-fold dilution of 5000 ng/mL, accuracy (%bias): 4.8%, precision (%CV): 5.2% Hook effect: NA		DMPK R1501017, Table 4-19
Bench-top/process stability	21 h in plasma at room temperature. 115 h for processed extract in autosampler at 4°C. 2 h in whole blood at 37°C and in ice bath.		DMPK R1501017, Table 4-23, Table 4-26, and Table 4-30
Freeze-thaw stability	6 cycles at -20°C and -80°C.		DMPK R1501017, Table 4-24 and Table 4-25

Long-term storage	104 days in plasma at -20°C. 783 days in plasma at -80°C.	DMPK R1501017, Table 4-28 DMPK R1501017-01, Table 4-29
Parallelism	NR	NA
Carryover	Carryover was evaluated by injecting blank matrix samples after the ULOQ standard. There was no significant carryover evident for leniolisib (≤16.8% carryover) or IS (≤0.3% carryover) except for Run 18 where leniolisib carryover was 30.9%.	DMPK R1501017-01, Table 4-13 and Table 4-14
Cross-validation	Comparison of 50 samples analyzed: 39 of 50 (78%) samples were within ±20.0% difference from the reference laboratory (b) (4) (b) (4) determined value.	DMPK R1501017, Section 5
	Method Performance in CCDZ173X2102 **Bioanalytical Report: DMPK RCCDZ173X2102**	
Assay passing rate	16 of 17 runs (94%) met acceptance criteria; stock checks and 1 run with syringe wash malfunction excluded from calculation.	DMPK RCCDZ173X2102, Table 5-1 and Table 5-2
Standard curve performance	Cumulative accuracy (%bias): -1.1% to 0.7% Cumulative precision (%CV): ≤4.7%	DMPK RCCDZ173X2102, Table 5-6
QC performance	Cumulative accuracy (%bias): -0.2% to 2.5% Cumulative precision (%CV): ≤4.9% Total error: NR	DMPK RCCDZ173X2102, Table 5-10
Method reproducibility	Incurred sample reanalysis was performed on 50 samples, and 49 (98.0%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2102, Table 9-7
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at -80°C for up to 43 days (06 Aug 2015 to 18 Sep 2015).	DMPK RCCDZ173X2102, Section 1 and Section 6
Standard calibration curve performance during accuracy and precision	16 calibrator sets with 2 replicates at each of 7 concentration levels from 3.00 to 1000 ng/mL (total 224 datapoints) – none excluded from curve fitting	
	Method Performance in CCDZ173X2104 **Bioanalytical Report: DMPK RCCDZ173X2104**	
Assay passing rate	16 of 18 runs (89%) met acceptance criteria; stock checks and 1 run using incorrect column excluded from calculation.	DMPK RCCDZ173X2104, Table 5-1 and Table 5-2
Standard curve performance	Cumulative accuracy (%bias): -3.8% to 3.3% Cumulative precision (%CV): ≤4.0%	DMPK RCCDZ173X2104, Table 5-3
QC performance	Cumulative accuracy (%bias): -2.6% to 0.8% Cumulative precision (%CV): ≤3.8% Total error: NR	DMPK RCCDZ173X2104, Table 5-4

Method reproducibility	Incurred sample reanalysis was performed on 33 samples, and 32 (96.97%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2104, Table 9-3
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at -80°C for up to 82 days (08 Apr 2016 to 29 Jun 2016).	DMPK RCCDZ173X2104, Section 1 and Section 6
Standard calibration curve performance during accuracy and precision	15 calibrator sets with 2 replicates at each of 7 concentration levels from 3.00 to 1000 ng/mL (total 210 datapoints) – none excluded from curve fitting	
	Method Performance in CCDZ173X2203 **Bioanalytical Report: DMPK RCCDZ173X2203**	
Assay passing rate	10 of 10 runs (100%) met acceptance criteria; stock checks excluded from calculation.	DMPK RCCDZ173X2203, Table 5-1 and Table 5-2
Standard curve performance	Cumulative accuracy (%bias): -1.8% to 1.3% Cumulative precision (%CV): ≤5.7%	DMPK RCCDZ173X2203, Table 5-3
QC performance	Cumulative accuracy (%bias): -0.3% to 4.3% Cumulative precision (%CV): ≤5.7% Total error: NR	DMPK RCCDZ173X2203, Table 5-4
Method reproducibility	Incurred sample reanalysis was performed on 30 samples, and 29 (96.7%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2203, Table 9-3
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at ≤-70°C for up to 329 days (05 Jul 2016 to 30 May 2017).	DMPK RCCDZ173X2203, Section 1 and Section 6
Standard calibration curve performance during accuracy and precision	10 calibrator sets with 2 replicates at each of 7 concentration levels from 3.00 to 1000 ng/mL (total 140 datapoints) – none excluded from curve fitting.	

CV=coefficient of variation; ESI=electrospray ionization; GMQC=geometric QC; HPLC=high-performance liquid chromatography; HQC=high QC; IS=internal standard; LC-MS/MS=liquid chromatography coupled with tandem mass spectroscopy; LLOQ=lower limit of quantification; LQC=low QC; MQC=middle QC; MRM=multiple reaction monitoring; NA=not applicable; NR=not reported; QC=quality control; ULOQ=upper limit of quantification.

Source: DMPK Report R1501017.

16.3.1.1.3. [redacted] (b) (4) (PKH/MOA/1031 and 1464)

Method

The [redacted] (b) (4) (15BASM071V1) method assay of leniolisib in plasma was transferred to [redacted] (b) (4) and validated (DMPK R1701469). The original method validation report was amended once to include additional long-term stability data and to include results from cross-validation against [redacted] (b) (4) (DMPK R1701469-01). Validation results met acceptance criteria and are summarized in Table 57. The validated quantitation range at

[redacted] was from 3.00 to 1000 ng/mL; QC samples from 3.00 to 800 ng/mL were used for the accuracy and precision assessments. Dilution integrity was assessed at 10- and 100-fold dilution.

Cross-validation of the method was performed between the [redacted] and [redacted] laboratories. Validation results met acceptance criteria, and 20 of 24 (83%) samples assayed by [redacted] were within ±15% difference (±20.0% difference at LLOQ) from the reference laboratory [redacted] determined value. A subsequent partial validation to extend the quantitation range of the method from 3.00 to 1000 ng/mL to 5.00 to 5000 ng/mL was conducted. QC samples from 5.00 to 4000 ng/mL were used for the accuracy and precision assessments. Dilution integrity was assessed at 5-fold dilution. Validation results met acceptance criteria.

Table 57. Summary of Method Performance for Determination of Leniolisib in Human Plasma (15BASM071V1), Report DMPK R1701469

Bioanalytical method validation report name, amendments, and hyperlinks	Validation of a LC-MS/MS analytical method for CDZ173 assay in human plasma samples Method Validation Report DMPK R1701469 Amendment 1 DMPK R1701469-01 Partial validation to extend the quantitation range from 3.00-1000 ng/mL to 5.00-5000 ng/mL is summarized in Appendix 3d.		
Method description	Protein precipitation of human plasma (K_3EDTA) samples and analysis of diluted samples by LC-MS/MS in MRM mode using ESI as the ionization technique		
Materials used for standard calibration curve and concentration	Leniolisib phosphate, (b) (4) Batches 1010013786 and 1010013796 Calibration standards: 3.00, 6.00, 25.0, 125, 250, 500, 900, and 1000 ng/mL Internal standard: $^{13}CD_3$-leniolisib, (b) (4) Batch SSE261-1/SVI-ALS-18-056		
Validated assay range	3.00 to 1000 ng/mL		
Material used for QCs and concentration	Leniolisib phosphate, (b) (4) Batch 1010013786 QC levels: 3.00 (LLOQ), 9.00 (LQC), 500 (MQC), 800 (HQC), and 5000 (Dilution QC) ng/mL Internal standard: $^{13}CD_3$-leniolisib, (b) (4) Batch SSE261-1/SVI-ALS-18-056		
Minimum required dilutions	NA		
Source and lot of reagents	Methanol, HPLC grade (b) (4) Acetonitrile, HPLC grade (b) (4) Isopropanol, HPLC grade Formic acid, HPLC grade Purified deionized water using (b) (4) Human plasma, K_3EDTA-treated (b) (4) batches 8D0779-01 to 06 R270418 and BRH1490291 to 296 R30518, BRH1593242 and 244 R291118, 0383975 R050320, BRH 1593242 R291118, 0383842 R05032020, and BRH1515308 R260618, respectively		
Regression model and weighting	Linear (y=ax+b), weighted ($1/x^2$)		
Validation parameters	**Method validation summary**		**Source location**
Standard calibration curve performance during accuracy and precision runs	Number of standard calibrators from LLOQ to ULOQ	8	DMPK R1701469-01, Table 5-14
	Cumulative accuracy (%bias) from LLOQ to ULOQ	-3.10% to 2.70%	

	Cumulative precision (%CV) from LLOQ to ULOQ	≤4.61%	
Performance of QCs during accuracy and precision runs	**Cumulative accuracy (%bias) in 4 QC levels**	-2.42% to -1.11%	DMPK R1701469-01, Table 5-15
	Interbatch %CV	≤7.07 %	
	Total error	NR	
Selectivity and matrix effect	Selectivity for leniolisib was considered acceptable in 6 batches of plasma tested. No peak for leniolisib or IS was detected in blank samples. Matrix effect from LQC (9.00 ng/mL) to HQC (800 ng/mL): Mean matrix factor: 1.0 to 1.1, precision (%CV): ≤2.29% Mean IS-normalized matrix factor: 1.0, precision (%CV): ≤3.12%		DMPK R1701469-01, Table 5-1, Table 5-2, Table 5-5, Table 5-6, and Table 5-7
Interference and specificity	No significant chromatographic interferences from IS on the detection of leniolisib (no interfering peaks) and from leniolisib on detection of IS (interference ≤0.1%).		DMPK R1701469-01, Table 5-3 and Table 5-4
Hemolysis effect	No significant effect of hemolysis (5% hemolyzed plasma) from LQC (9.00 ng/mL) to HQC (800 ng/mL). Accuracy (%bias): -1.69% to -0.99% Precision (%CV): ≤3.97%		DMPK R1701469-01, Table 5-20
Lipemic effect	No significant effect of lipemia (hyperlipidemic plasma) from LQC (9.00 ng/mL) to HQC (800 ng/mL). Accuracy (%bias): -0.83% to 0.11% Precision (%CV): ≤3.86%		DMPK R1701469-01, Table 5-21
Dilution linearity and hook effect	10- and 100-fold dilution of 5000 ng/mL, accuracy (%bias): -6.46% to -2.44%, precision (%CV): ≤6.67% Hook effect: NA		DMPK R1701469-01, Table 5-17
Bench-top/process stability	4 h 7 min in plasma at room temperature. 57 h 59 min for processed extract at 4°C. 1 h in whole blood at room temperature.		DMPK R1701469-01, Table 5-23, Table 5-24, and Table 5-28
Freeze-thaw stability	3 cycles at -20°C and 4 cycles at -80°C.		DMPK R1701469-01, Table 5-25 and Table 5-26
Long-term storage	733 and 1183 days in plasma at -20°C and -80°C, respectively.		DMPK R1701469-01, Table 5-29 and Table 5-30
Parallelism	NR		NA
Carryover	Carryover was evaluated by injecting blank matrix samples after the ULOQ standard. There was no carryover evident for leniolisib and IS (no peaks observed).		DMPK R1701469-01, Table 5-11 and Table 5-12

Cross-validation	Comparison of 24 samples analyzed: 20 of 24 (83%) samples were within ±15.0% difference (±20.0% difference at LLOQ) from the reference laboratory (b) (4) determined value.	DMPK R1701469-01, Appendix C
	Method Performance in LE1101 **Bioanalytical Report: LE 1101 BAR LE**	
Assay passing rate	10 of 10 runs (100%) met acceptance criteria (excluding a run for preparation of standards and QCs from calculation)	LE 1101 BAR LE, Table 3
Standard curve performance	Cumulative accuracy (%bias): -1.04% to 0.94% Cumulative precision (%CV): ≤5.39	LE 1101 BAR LE, Table 4
QC performance	Cumulative accuracy (%bias): -2.40% to -0.22% Cumulative precision (%CV): ≤5.02% Total error: NR	LE 1101 BAR LE, Table 7
Method reproducibility	Incurred sample reanalysis was performed on 79 samples, and 76 (96.20%) of which had <20% difference from the original results.	LE 1101 BAR LE, Table 8
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at ≤-20°C for up to 84 days (14 Sep 2021 to 13 Jan 2022).	LE 1101 BAR LE, Table 9
Standard calibration curve performance during accuracy and precision	11 calibrator sets with 1 replicate at each of 8 concentration levels from 5.00 to 5000 ng/mL (total 88 datapoints) – none excluded from curve fitting.	
	Method Performance in LE2101 **Bioanalytical Report: LE 2101 BAR LE**	
Assay passing rate	4 of 4 runs (100%) met acceptance criteria (excluding a run for preparation of frozen QCs from calculation)	LE 2101 BAR LE, Table 2
Standard curve performance	Cumulative accuracy (%bias): -2.56% to 3.99% Cumulative precision (%CV): ≤3.90%	LE 2101 BAR LE, Table 3
QC performance	Cumulative accuracy (%bias): -2.59% to 0.87% Cumulative precision (%CV): ≤4.55% Total error: NR	LE 2101 BAR LE, Table 6
Method reproducibility	Incurred sample reanalysis was performed on 32 samples, and 23 (71.88%) of which were within ±20% difference from original results.	LE 2101 BAR LE, Table 9
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at -20°C for up to 32 days (12 Nov 2021 to 14 Dec 2021).	LE 2101 BAR LE, Table 10
Standard calibration curve performance during accuracy and precision	4 calibrator sets with 1 replicate at each of 8 concentration levels from 5.00 to 5000 ng/mL (total 32 datapoints) – none excluded from curve fitting.	

Method Performance in CCDZ173X2201 Part 2 Bioanalytical Report: DMPK RCCDZ173X2201a		
Assay passing rate	8 of 8 runs (100%) met acceptance criteria	DMPK RCCDZ173X2201a, Table 9-3
Standard curve performance	Cumulative accuracy (%bias): -2.14% to 1.39% Cumulative precision (%CV): ≤8.67%	DMPK RCCDZ173X2201a, Table 5-4
QC performance	Cumulative accuracy (%bias): -3.26% to -1.86% Cumulative precision (%CV): ≤6.90% Total error: NR	DMPK RCCDZ173X2201a, Table 5-6
Method reproducibility	Incurred sample reanalysis was performed on 30 samples, and 25 (83.3%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2201a, Table 9-4
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at -80°C for up to 727 days (11 Sep 2018 to 07 Sep 2020).	DMPK RCCDZ173X2201a, Section 6 and Table 9-5
Standard calibration curve performance during accuracy and precision	8 calibrator sets with 1 replicate at each of 8 concentration levels from 3.00 to 1000 ng/mL (total 64 datapoints) – none excluded from curve fitting.	
Method Performance in CCDZ173X2201E1 Bioanalytical Report: DMPK RCCDZ173X2201E1		
Assay passing rate	11 of 11 runs (100%) met acceptance criteria	DMPK RCCDZ173X2201E1, Table 9-3
Standard curve performance	Cumulative accuracy (%bias): -1.15% to 1.25% Cumulative precision (%CV): ≤5.53%	DMPK RCCDZ173X2201E1, Table 5-4
QC performance	Cumulative accuracy (%bias): -1.82% to -1.41% Cumulative precision (%CV): ≤5.22% Total error: NR	DMPK RCCDZ173X2201E1, Table 5-6
Method reproducibility	Incurred sample reanalysis was performed on 30 samples, and 25 (83.3%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2201E1, Table 9-4
Study sample analysis stability	Stability data in matrix cover the sample collection, storage, and analysis period in this study. Samples were stored at -80°C for up to 1088 days (07 Sep 2018 to 30 Aug 2021).	DMPK RCCDZ173X2201E1, Section 6 and Table 9-5
Standard calibration curve performance during accuracy and precision	11 calibrator sets with 1 replicate at each of 8 concentration levels from 3.00 to 1000 ng/mL (total 88 datapoints) – none excluded from curve fitting.	

CV=coefficient of variation; ESI=electrospray ionization; HPLC=high-performance liquid chromatography; HQC=high QC; IS=internal standard; LC-MS/MS=liquid chromatography coupled with tandem mass spectroscopy; LLOQ=lower limit of quantification; LQC=low QC; MQC=middle QC; MRM=multiple reaction monitoring; NA=not applicable; NR=not reported; QC=quality control; ULOQ=upper limit of quantification.

Source: DMPK Report R1701469

16.3.1.2. Determination of Leniolisib in Human Urine

16.3.1.2.1. [redacted](b) (4)(1100679g) Method

An LC-MS/MS method for the assay of leniolisib in human urine was developed and validated at [redacted](b) (4) (DMPK R1100679g). Validation results met acceptance criteria and are summarized in Table 58. The validated quantitation range was from 10.0 to 10000 ng/mL; QC samples from 10.0 to 7500 ng/mL were used for the accuracy and precision assessments. Dilution integrity was assessed at 100-fold dilution of 100,000 ng/mL. Of note, adsorption of leniolisib on urine sample tubes was discovered while clinical samples from Study 2101 were being analyzed. The issue was remedied by addition of 1% Tween 20. Urine samples in Cohort 20 (140 mg single-dose leniolisib) of Study 2101 analyzed with and without 1% Tween 20 showed systemically higher leniolisib concentrations when treated with Tween 20. Hence, all urine leniolisib concentration data for Cohorts 1 to 19 were underestimated by approximately 30% and should be interpreted with caution.

Table 58. Summary of Method Performance for Determination of Leniolisib in Human Urine (1100679g)

Bioanalytical method validation report name, amendments, and hyperlinks	Quantitative determination of CDZ173 in human urine by LC-MS/MS Method Validation Report DMPK R1100679g		
Method description	Human urine samples were diluted with ethanol and water. Then, the supernatant was analyzed by LC-MS/MS in SRM mode using ESI in positive mode as the ionization technique.		
Materials used for standard calibration curve and concentration	Leniolisib phosphate, (b) (4) Batches 1251002 REF and 1010006837 Calibration standards: 10.0, 50.0, 100, 500, 1000, 5000, and 10000 ng/mL Internal standard: $^{13}CD_3$-leniolisib, (b) (4) Batch SSE 261-1		
Validated assay range	10.0 to 10000 ng/mL		
Material used for QCs and concentration	Leniolisib phosphate, (b) (4) Batches 1251002 REF and 1010006837, QC levels: 10.0 (LLOQ), 30.0 (LQC), 5000 (MQC), 7500 (HQC), and 100000 (Dilution QC) ng/mL Internal standard: $^{13}CD_3$-leniolisib, (b) (4) Batch SSE 261-1		
Minimum required dilutions	NA		
Source and lot of reagents	Human urine Ethanol, Lichrosolv® gradient grade (b) (4) Formic acid (b) (4) Tetrahydrofuran, Chromasolv® grade (b) (4) Purified deionized water using (b) (4)		
Regression model and weighting	Quadratic ($y=ax^2+bx+c$), weighted ($1/x^2$)		
Validation parameters	**Method validation summary**		**Source location**
Standard calibration curve performance during accuracy and precision runs	Number of standard calibrators from LLOQ to ULOQ	7	DMPK R1100679g, Table 6-13
	Cumulative accuracy (%bias) from LLOQ to ULOQ	-2.4% to 2.0%	
	Cumulative precision (%CV) from LLOQ to ULOQ	≤4.8%	

Performance of QCs during accuracy and precision runs	**Cumulative accuracy (%bias) in 4 QC levels**	-5.3% to 9.3%	DMPK R1100679g, Table 6-14
	Interbatch %CV	≤7.9%	
	Total error	NR	NA
Selectivity and matrix effect	Selectivity for leniolisib and IS was considered acceptable in 6 batches of urine tested. Interference from blank matrix was ≤12.3% of response at LLOQ for leniolisib and ≤0.3% of response at working concentration for IS. Matrix effect from LQC to HQC: Mean matrix factor: 1.61 to 1.95, precision (%CV): ≤17.9% Mean IS-normalized matrix factor: 1.25 to 1.37, precision (%CV): ≤10.4%		DMPK R1100679g, Table 6-1, Table 6-2, Table 6-5, Table 6-6, and Table 6-7
Interference and specificity	Contribution of IS to the signal of leniolisib (8.0% interference) and of leniolisib to the signal of IS (0.3% interference) was not significant.		DMPK R1100679g, Table 6-3 and Table 6-4
Hemolysis effect	NA		NA
Lipemic effect	NA		NA
Dilution linearity and hook effect	100-fold dilution of 100000 ng/mL, accuracy (%bias): -14.3%, precision (%CV): 3.9% Hook effect: NA		DMPK R1100679g, Table 6-16
Additional experiments in urine with 1% Tween: Standard calibration curve performance	Number of standard calibrators from LLOQ to ULOQ	7	DMPK R1100679g, Table 7-5
	Cumulative accuracy (%bias) from LLOQ to ULOQ	-4.8% to 3.2%	
Additional experiments in urine with 1% Tween: Performance of QCs	**Cumulative accuracy (%bias) in 4 QC levels**	-10.2% to 8.4%	DMPK R1100679g, Table 7-6
	Intrabatch %CV	≤7.0%	
Additional experiments in urine with 1% Tween: Selectivity and matrix effect	Selectivity for leniolisib and IS was considered acceptable in 6 batches of urine tested. Interference from blank matrix was ≤2.3% of response at LLOQ for leniolisib and 0.0% of response at working concentration for IS.		DMPK R1100679g, Table 7-1 and Table 7-2
Additional experiments in urine with 1% Tween: Interference and specificity	Contribution of IS to the signal of leniolisib (0.0% interference) and of leniolisib to the signal of IS (0.0% interference) was not significant.		DMPK R1100679g, Table 7-3 and Table 7-4
Bench-top/process stability	5 h in urine at room temperature 160 h for processed extract in autosampler at 5°C		DMPK R1100679g, Table 6-17 and Table 6-20
Freeze-thaw stability	13 cycles at <-15°C in urine 13 cycles at <-65°C in urine 2 cycles at ≤-15°C in urine with 1% Tween 20		DMPK R1100679g, Table 6-18, Table 6-19, and Table 7-7

Long-term storage	67 days in urine with 1% Tween 20 at ≤-15°C	DMPK R1100679g, Table 7-8
Parallelism	NR	NA
Carryover	Carryover was evaluated by injecting blank matrix samples after the ULOQ standard. There was no significant carryover evident for leniolisib (≤6.5% carryover) or IS (≤0.2% carryover).	DMPK R1100679g, Table 6-11
	Method Performance in CCDZ173X2101 **Bioanalytical Report: DMPK RCCDZ173X2101**	
Assay passing rate	16 of 18 runs (89%) in urine met acceptance criteria (excluding runs analyzed with Method 1100679f rather than Method 1100679g).	DMPK RCCDZ173X2101, Table 5-1
Standard curve performance	Cumulative accuracy (%bias): -1.1% to 1.2% Cumulative precision (%CV): ≤6.6%	DMPK RCCDZ173X2101, Table 5-8
QC performance	Cumulative accuracy (%bias): -1.7% to 6.2% Cumulative precision (%CV): ≤21.7% Total error: NR	DMPK RCCDZ173X2101, Table 5-10
Method reproducibility	Incurred sample reanalysis was performed on 28 samples, and 23 (82%) of which were within ±20% difference from original results.	DMPK RCCDZ173X2101, Table 9-5
Study sample analysis stability	Samples were received frozen and stored at ≤-15°C. Long-term stability could not be demonstrated in samples not treated with Tween 20. Samples treated with Tween 20 were analyzed within the established stability limit of 67 days at <-15°C.	DMPK RCCDZ173X2101, Section 1 and Section 6
Standard calibration curve performance during accuracy and precision	16 calibrator sets with 2 replicates at each of 7 concentration levels from 10.0 to 1000 ng/mL (total 224 datapoints) – 4 excluded from curve fitting due to deviation ≥15% of nominal.	

CV=coefficient of variation; ESI=electrospray ionization; HQC=high QC; IS=internal standard; LC-MS/MS=liquid chromatography coupled with tandem mass spectroscopy; LLOQ=lower limit of quantification; LQC=low QC; MQC=middle QC; NA=not applicable; NR=not reported; QC=quality control; SRM=selective reaction monitoring; ULOQ=upper limit of quantification.

Source: DMPK Report R1100679g

16.3.1.3. Characterization of Leniolisib Absorption, Metabolism, and Excretion

^{19}F-NMR Analysis of Human Plasma, Urine, and Feces

^{19}F-nuclear magnetic resonance (NMR) analysis is a viable option for metabolite investigations for drug molecules containing fluorine atoms because NMR is fully quantitative, giving equimolar response for drug and metabolites under appropriate measurement conditions. In addition, fluorine is not present in endogenous molecules allowing for quantification of drug-related material without endogenous background interference. Because leniolisib contains 3

fluorine atoms, the ^{19}F-NMR approach was considered for the analysis of leniolisib absorption, metabolism, and excretion. Following a single dose of 400 mg leniolisib in healthy participants, the total drug-related material in individual plasma, urine, and feces samples were quantified by ^{19}F-NMR based on a calibration curve of leniolisib in each matrix extracted and analyzed in the same way.

Leniolisib and metabolites in plasma, urine, and fecal extract pools of pooled participants were quantified by 19F-NMR after liquid chromatography separation based on a calibration curve of leniolisib and internal standard in NMR solvent. Quantification of metabolites was performed using the leniolisib calibration curves under the assumption of equimolar NMR response for leniolisib and metabolites compared with an internal standard. Total drug-related material was determined based on the sum of all NMR peak integrations in each sample.

The structural characterization of metabolites in plasma and excreta was carried out by MS/MS analysis after liquid chromatography separation of analytes. Single stage and product ion spectra with exact mass measurements were obtained in positive ion mode. The structures of the metabolites were derived from their product ion mass spectra and the elemental composition determined by exact mass measurement. Sites of metabolism were localized based on MS/MS fragments containing the m/z of the moiety of interest or an unambiguous fragment thereof.

^{14}C-Reactivity in Human Plasma, Urine, and Feces

Validated liquid-scintillation counting methods were used to determine the ^{14}C-radioactivity in plasma, urine, and feces samples from the mass balance study of a single dose of 70 mg ^{14}C-leniolisib (40 µCi of ^{14}C-radioactivity) in healthy male participants (Study LE2101). The validated quantitation ranges were 30.0 to 80000 dpm/mL for the plasma method, 40000 to 1,000,000 dpm/g for the urine method in normal count mode, and 100,000 to 1,000,000 dpm/g for the feces method in normal count mode. Plasma and urine samples were prepared for analysis by mixing with scintillation cocktail; fecal samples were prepared by homogenization and combustion.

16.3.1.4. Determination of Pakt-Positive B Cells in Human Whole Blood

Throughout clinical development, assay for pAkt-positive B cells in clinical samples was performed by [redacted]$^{(b)(4)}$ Initially, bioanalytical methods for determination of pAkt-positive B cells in nonstimulated and ex vivo stimulated whole blood were developed and validated by [redacted]$^{(b)(4)}$ (Method 0750979). The [redacted]$^{(b)(4)}$ method was transferred to [redacted]$^{(b)(4)}$ (Method ICDFC 6) and validated. Afterward, the method was transferred from [redacted]$^{(b)(4)}$ to [redacted]$^{(b)(4)}$ and partially validated.

16.3.1.4.1. [redacted] (0750979) Method

An intracellular flow cytometry assay for the enumeration of pAkt-positive B cells in ex vivo stimulated heparinized human whole blood was developed and validated at [redacted] (0750979). In brief, the whole blood was lysed and fixed with pre-warmed (37 °C) BD PhosFlow-Lyse/Fix buffer for 20 minutes in a 37 °C water bath protected from light. The lysed and fixed blood was stored at -80 °C ± 10 °C and was processed within one week to mimic sample storage and shipment from clinical site to lab. The frozen samples were thawed and stained with antibody cocktail as per the method for flow cytometry analysis.

The assay consisted of both direct staining (total AKT and CD20) and indirect staining for phospho-AKT. Samples were processed in duplicate for FACS analysis. Percent phospho-AKT positive B cells were measured as % of all CD20 positive B cells. For stimulation of blood B cells, whole blood sample containing sodium heparin was stimulated with 7.50 µg/mL anti-IgM and 7.50 ng/mL IL-4 for 30 minutes at 37 °C and 5% CO2 before the lysis. Stimulated cells show at least a three-fold increase in pAKT staining in CD19 positive B cells as compared to non-stimulated cells.

16.3.1.4.2. [redacted] (ICDFC 6) Method

The [redacted] (0750979) method was transferred to [redacted] and validated (ICDFC 6). Afterward, the ICDFC 6 method was transferred from [redacted] to [redacted] and partially validated. The validated intra-assay precision coefficient of variation (CV) based on 3 donors, 6 replicates ranged from 3.1% to 10.2% for stimulated samples, and the validated inter-assay precision CV based on 2 donors, 6 runs ranged from 13.4% to 27.7% for stimulated samples at [redacted]. Validation results met acceptance criteria.

16.3.2. Human Biomaterial Studies

Plasma Protein Binding and Partitioning in Blood

In vitro plasma protein binding and blood partitioning were assessed in human blood and plasma incubated with leniolisib. Mean blood-to-plasma ratio of 0.643 and plasma protein binding of 94.5% were observed at 10, 100, 1000, and 10,000 ng/mL leniolisib. No concentration dependencies were observed.

Intestinal Uptake Mechanisms and Transport

In vitro permeability of leniolisib was assessed at 6 to 600 µM using Caco-2 cell monolayers. Concentration-dependent increase in apical-to-basolateral permeability and decrease in basolateral-to-apical permeability were observed, suggesting saturation of an apically directed efflux transporter process, with an apparent Michaelis-Menten constant ($K_{m,app}$) of 65.3 µM. At concentrations ≥40 µM, leniolisib exhibited high permeability even in the presence of efflux transporter activity. Human intestinal absorption >90% is expected based on the passive

leniolisib permeability for concentrations ≥100 µM. Assessment of intestinal uptake mechanisms at 0.5 to 300 µM indicated that luminal membrane uptake of leniolisib in the intestine occurs by a passive permeation process modulated by one or several efflux pumps, most likely P-gp.

In Vitro Hepatic Metabolism

Comparison of in vitro hepatic metabolism using animal (mouse, rat, dog, and monkey) and human hepatocytes identified similar metabolic pathways across species. Observed metabolites were from phase 1 metabolism; no phase 2 metabolism was observed. One metabolite, M18 (O-demethylation with hydration on pyrrolidine), was observed only in human hepatocytes. In vivo, M18 was also found in rat excreta and accounted for <2% of the drug-related material in human plasma. In vitro human metabolism was evaluated using pooled human liver microsomes and recombinant CYP and flavin-containing monooxygenase (FMO) enzymes.

In human liver microsomes, biotransformation of leniolisib was characterized by a $K_{m,app}$ of 20.3 µM, maximum reaction velocity (V_{max}) of 1000 pmol/min/mg, and hepatic intrinsic clearance (CL_{int}) of 49.3 µL/min/mg. Leniolisib was mainly metabolized to M6 and M7 (by hydroxylation), and in vitro metabolism was completely inhibited by ketoconazole and azamulin (CYP3A4/5 inhibitors) at 10 µM leniolisib. In incubations of recombinant human CYP (CYP1A1, CYP1A2, CYP1B1, CYP2A6, CYP2B6, CYP2C8, CYP2C9, CYP2C18, CYP2C19, CYP2D6, CYP2E1, CYP2J2, CYP3A4, CYP3A5, CYP4A11, CYP4F2, CYP4F3B, and CYP4F12) and FMO (FMO1, FMO3, and FMO5) isoenzymes with leniolisib at 10 and 50 µM, CYP1A1 and CYP3A4 showed the highest turnover, low metabolic activity was observed with CYP3A5 and CYP2D6, and only trace or no metabolism was detected for the other CYP and FMO isoenzymes.

Taken together, results suggest that hepatic metabolism of leniolisib is driven by CYP3A4 (95.4%) with minimal contribution from other enzymes (3.5%, 0.7%, and 0.4% from CYP3A5, CYP1A2, and CYP2D6, respectively), and CYP1A1 is possibly involved in its extrahepatic biotransformation.

In Vitro Inhibition/Induction of Drug Metabolizing Enzymes

The potential of leniolisib to inhibit CYP enzymes was assessed in pooled human liver microsomes. CYP-selective probe substrates were incubated with increasing concentrations of leniolisib up to 100 µM. In vitro, leniolisib showed inhibitory potency for CYP2C8 (IC_{50} corrected by unbound fraction [$IC_{50,u}$]=83 µM; K_i corrected by unbound fraction [$K_{i,u}$]=23 µM), CYP2C9 ($K_{i,u}$=29 µM), CYP2C19 ($K_{i,u}$=29 µM), and CYP2D6 ($K_{i,u}$=36 µM). Very little or no reversible inhibition of CYP1A2, CYP2A6, CYP2B6, CYP2E1, and CYP3A4/5 was observed. Time-dependent (irreversible) inhibition was observed for CYP1A2 ($K_{I,u}$=303 µM, maximal rate of enzyme inactivation [k_{inact}]=0.017 min-1) but not CYP2C9, CYP2D6, and CYP3A4/5. Clinically relevant inhibition of CYP2C8, CYP2C9, CYP2C19, and CYP2D6 by leniolisib is unlikely because the $I_{max,ss,u}$ at the therapeutic dose (0.449 µM at 70 mg twice daily is ≥50-fold lower than the $K_{i,u}$ values,

leading to R_1 values <1.02. However, clinically relevant inhibition of CYP1A2 could not be excluded, and the R_2 value calculated was ≥1.25.

The effect of leniolisib on UGT- and SULT-mediated metabolism of ethinylestradiol was tested in vitro using pooled human liver microsome and liver cytosol. Leniolisib showed concentration-dependent inhibition of UGT1A1-mediated glucuronidation ($IC_{50,u}$=7.24 µM), while no inhibitory effects on SULT1E1-mediated sulfation were observed at leniolisib concentrations up to 200 µM. The potential of leniolisib to induce CYP enzymes (activity and mRNA) was assessed in human hepatocytes. Leniolisib was incubated with primary human hepatocyte cultures at concentrations up to 75 µM and induction of mRNA was evaluated relative to vehicle and positive control while enzyme activity was evaluated from metabolism of CYP-selective probe substrates.

Concentration-dependent induction of CYP2B6, CYP2C9, and CYP3A4 mRNA and activity was observed, but there was no significant induction (<20% of maximal positive control induction) for CYP1A2 mRNA or activity. However, leniolisib is unlikely to be an inducer of these enzymes at therapeutic doses based on the R_3 values (R_3 >0.8). In PBPK model-based simulations, the fifth percentile of model-predicted ratio of midazolam (a prototypical CYP3A4 substrate) exposure with/without concomitant leniolisib ranged from 0.32 to 0.77, indicating leniolisib is a weak-to-moderate inducer of CYP3A4.

In vitro activation of pregnane X-receptor (PXR) and aryl hydrocarbon receptor (AhR) by leniolisib was evaluated using human hepatoma cell lines DPX-2 and CYP1A2-DRE, respectively. Significant activation of PXR was observed in the presence of ≥25 µM leniolisib, indicating possible PXR-mediated induction of CYP3A4 in vivo at sufficiently high concentrations. No significant activation of AhR at leniolisib concentrations ≤100 µM was observed. Hence, AhR-mediated induction of CYP1A2 is not expected.

In Vitro Interaction With Transporters

In vitro, leniolisib was not a substrate of OATP, OCT, or MRP2 but was a substrate of P-gp, BCRP, and OAT. Active biliary secretion via BCRP or P-gp was observed to contribute to the total clearance of leniolisib. In a clinical DDI study with quinidine (a strong CYP2D6 and P-gp inhibitor), leniolisib exposure was unchanged with quinidine coadministration (Study 2102). Thus, leniolisib was not found to be a substate of P-gp in vivo. In vitro, leniolisib-mediated inhibition was observed for BCRP (IC_{50}=18.9 µM), OATP1B1 (IC_{50}=3.0 µM), OATP1B3 (IC_{50}=13.4 µM), OCT1 (IC_{50}=39.9 µM; K_i=39.8 µM), OCT2 (IC_{50}=3.4 µM; K_i=3.4 µM), OAT1 (IC_{50}=73.8 µM; K_i=69 µM), OAT3 (IC_{50}=15.2 µM; K_i=10 µM), MATE1 (IC_{50}=6.70 µM; K_i=6.70 µM), and MATE2K (IC_{50}=0.85 µM; K_i=0.85 µM). However, based on the anticipated $I_{max,ss,u}$ at the therapeutic dose (70 mg BID), clinically relevant interactions are not expected for these transporters ($I_{max,u}/IC_{50}$ <0.1) except for BCRP, OATP1B1, OATP1B3, OCT2, and MATE2K, which had a calculated $I_{max,u}/IC_{50}$, I_{gut}/IC_{50}, or R value above the cutoff.

Although the calculated $I_{max,u}/IC_{50}$ for the renal transporters OCT2 and MATE2K was >0.1, indicating potential in vivo inhibition, the Applicant states that clinically relevant interactions between leniolisib and substrates of these transporters are unlikely because leniolisib is only approximately 6% renally cleared. However, this justification is not valid as leniolisib could still inhibit these renal transporters even with low renal clearance. In vitro, leniolisib was found to inhibit BSEP (IC_{50}=38.03 µM, K_i=35.92 µM). However, clinically relevant inhibition is unlikely because the K_i value is ≥80-fold higher than $I_{max,ss,u}$ at therapeutic doses of leniolisib.

16.3.3. Individual Clinical Pharmacology Study Review

16.3.3.1. Study 2101

- Single- and multiple-dose PK, PD, food effect, dose proportionality, explorative mass balance, metabolite profiling, and concentration-QTC analysis in healthy participants.

Study 2101 was a phase 1, multicenter, randomized, double-blind, placebo-controlled, single and multiple ascending dose study assessing leniolisib PK following single and multiple oral doses of leniolisib and under fed and fasted conditions, and investigating the PD effect of leniolisib on pAkt in ex vivo stimulated B cells in healthy participants. This study was conducted in 3 parts: a randomized, double-blind, placebo-controlled, single ascending dose study (Part 1); a randomized, open-label, 2-way crossover, food effect study (Part 2); and a randomized, double-blind, placebo-controlled, multiple ascending dose study (Part 3).

In Part 1, participants were randomized to receive a single oral dose of leniolisib in the fasted state at ascending dose levels of 10, 20, 40, 80, 110, 140, 200, 300, and 400 mg or placebo. In Part 2, participants were randomized to treatment sequence and received a single oral dose of leniolisib 70 mg (1 × 70 mg capsule) in the fasted and fed state. In Part 3, participants were randomized to receive oral leniolisib twice daily in the fasted state at ascending dose levels of 20, 40, 70, and 140 mg or placebo for 15 days. A total of 64, 12, and 42 participants received leniolisib and contributed PK data in Parts 1, 2, and 3, respectively.

There were 44 and 26 participants randomized to the placebo group in Parts 1 and 3, respectively. Treatments were separated by a ≥14-day washout period in Part 2. Study drugs were manufactured by Novartis Pharma AG. Parts 1 and 3 of the study used 10, 70 (Part 1, Cohort 20 only), and 100 mg capsules to make the required dose. The 10 and 100 mg capsules in the study were (b) (4) formulation, while the 70 mg capsules were (b) (4) formulation of Novartis hard gelatin capsule (Novartis HGC).

Single- and Multiple-Dose Pharmacokinetics

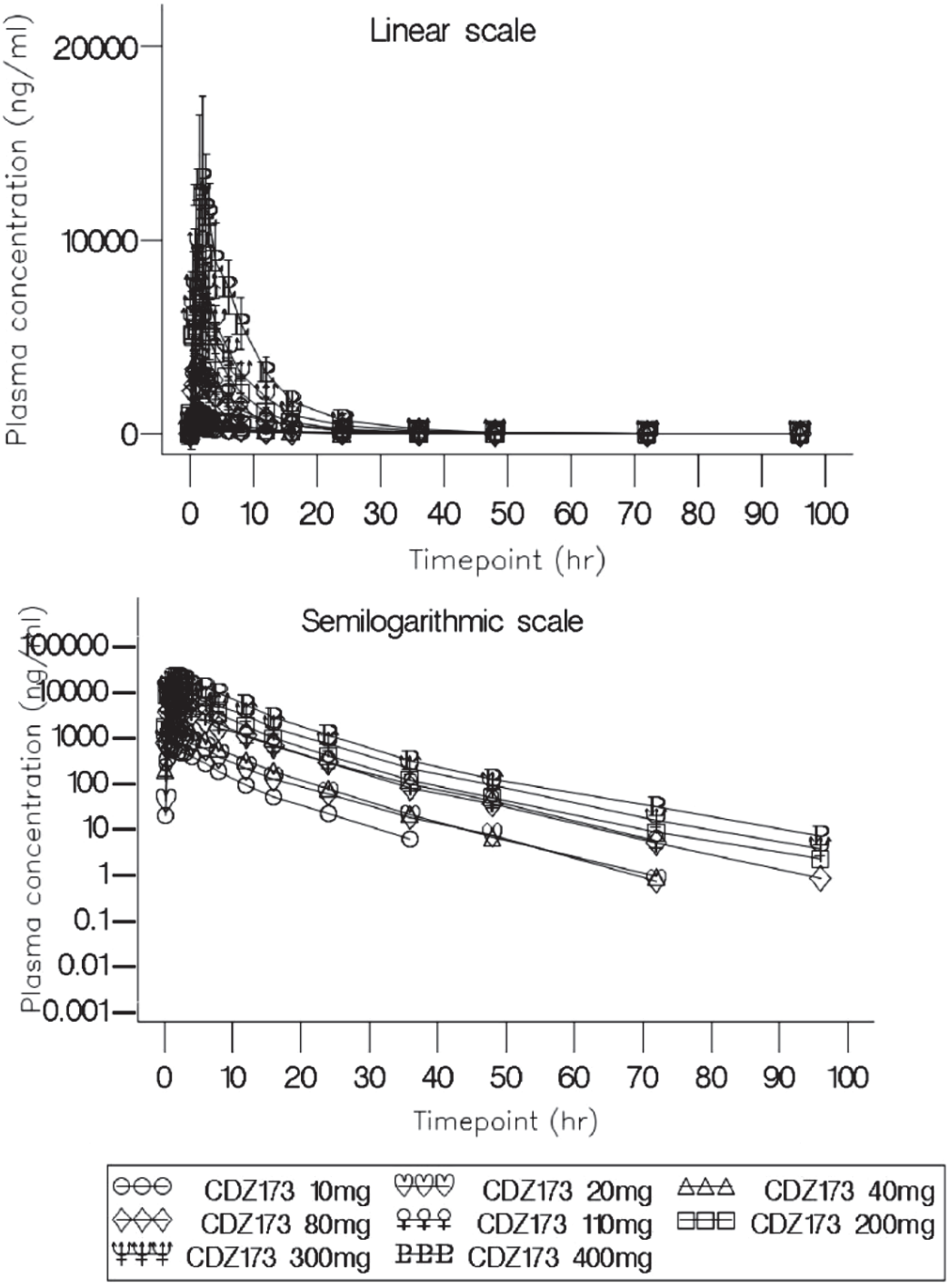
Following single-dose administration in the fasted state, oral leniolisib was rapidly absorbed, reaching peak plasma concentration approximately 1-hour post-dose, and showed biexponential disposition, with most of the drug eliminated in the distribution phase (Figure 12). Terminal elimination $t_{1/2}$ was variable (geometric mean 5.32 to 10.9 hours) (Table 59).

Modest drug accumulation was observed with twice daily dosing, consistent with an effective half-life of approximately 7 hours (Figure 1, Table 60). Steady-state of plasma leniolisib concentrations was achieved in most participants within 2 to 3 days of dosing, as evident from stable C_{trough} values (Figure 13). Leniolisib PK was dose proportional over the entire dose range investigated for both single- and multiple-dose (twice daily) administration (20 to 140 mg twice a day dosing and single doses of 10 to 400 mg).

Oral plasma geometric mean CL_{ss}/F ranged from 3.33 to 3.69 L/h. Based on the in vitro human blood-to-plasma ratio of 0.643, the population median systemic hepatic blood clearance was calculated as approximately 6 L/h, well below 30% of human liver blood flow (approximately 90 L/h), indicating leniolisib is a drug with low hepatic clearance. Accordingly, first-pass metabolic extraction is not expected to restrict the absolute oral bioavailability of leniolisib to a meaningful extent (<10% of oral dose).

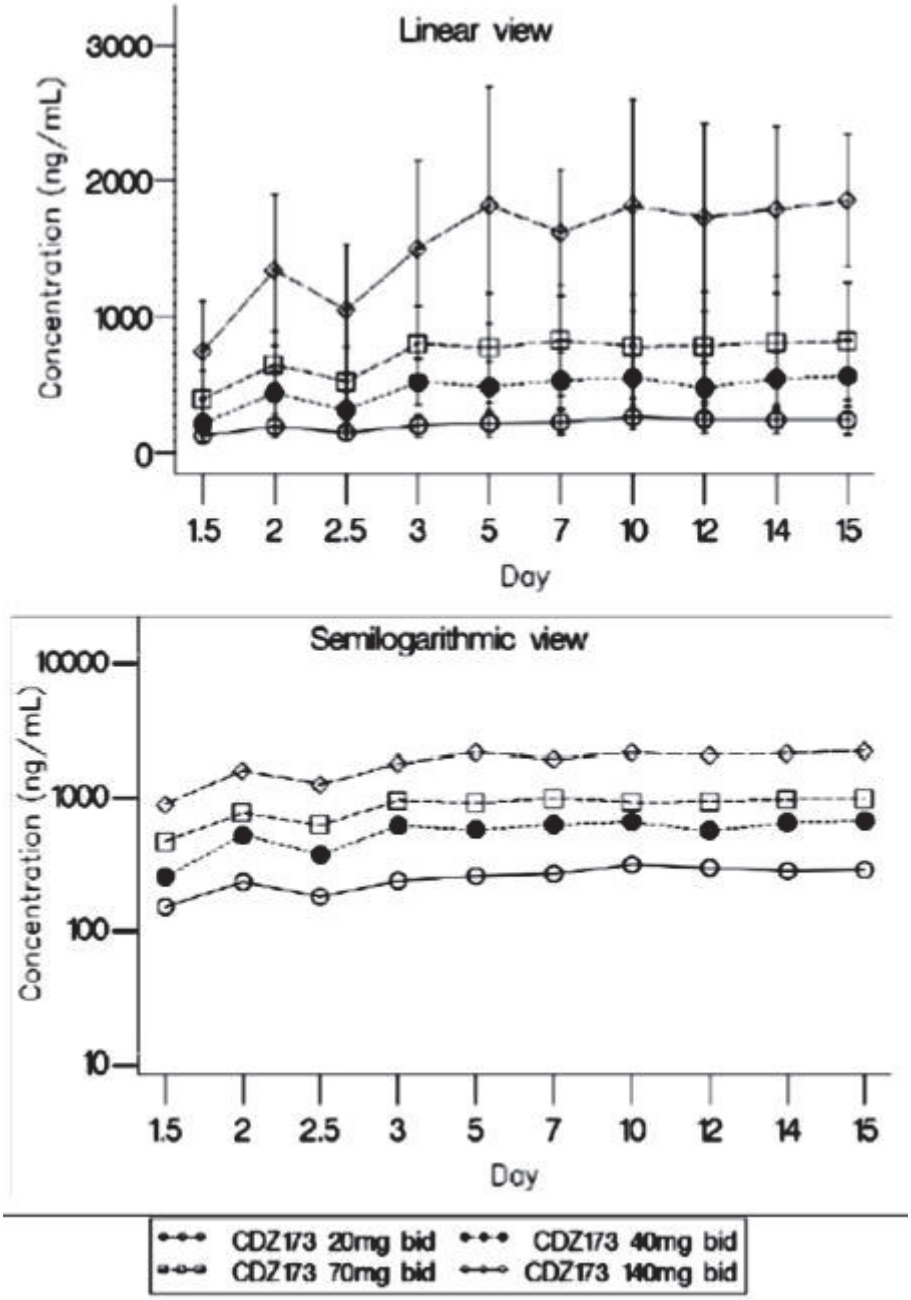
Geometric mean CL_R ranged from 0.0861 to 0.251 L/h at steady state, approximating 6%of the oral dose. Hence, direct secretion of parent drug into urine is not expected to be a major elimination route for leniolisib. Of note, the Applicant states that adsorption of leniolisib to sample tubes was discovered while clinical urine samples were being analyzed. The Applicant estimated that the urine leniolisib measurements for Cohorts 1 through 19 in this study are underestimated by approximately 30% and should be interpreted with caution (DMPK RCCDZ173X2101).

Figure 12. Mean (SD) Leniolisib Plasma Concentration-Time Profiles Following Single-Dose Administration of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 1)



Source: Study 2101 CSR, Figure 14.2-2.1a.
Abbreviations: CSR, clinical study report; SD, standard deviation.

Figure 13. Mean (SD) Leniolisib Trough Plasma Concentration-Time Profiles Following Twice Daily Dosing of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 3)



Source: Study 2101 CSR, Figure 14.2-4.
Abbreviations: BID, twice daily; CSR, clinical study report; BID, twice daily; SD, standard deviation

Table 59. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following Single-Dose Administration of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 1)

		Geometric Mean (Geometric CV%)				
Leniolisib Dose (mg)	n	C_{max} (ng/mL)	T_{max} (h)	AUC_{inf} (ng*h/mL)	$t_{1/2}$ (h)	CL_R (L/h)
10	8	402 (35.5)	1.00 (0.5, 2.5)	2380 (43.5)	5.32 (37.1)	0.159 (51.7)
20	8	790 (20.1)	1.00 (1.00, 2.00)	5200 (30.9)	6.49 (29.6)	0.144 (51.6)
40	6	1470 (33.0)	0.875 (0.5, 1.50)	8590 (39.2)	6.53 (24.4)	0.173 (13.4)
80	6	3400 (42.4)	1.25 (0.5, 2.50)	23800 (44.5)	9.15 (50.5)	0.146 (17.5)
110	6	4010 (33.9)	1.25 (0.5, 6.00)	28300 (29.6)	8.83 (38.5)	0.147 (36.7)
140	6	5980 (27.4)	1.00 (0.517, 3.00)	28400 (20.2)	5.94 (71.7)	0.178 (36.7)
200	12	7110 (22.6)	1.31 (0.5, 2.17)	45700 (33.2)	8.40 (37.7)	0.206 (19.6)
300	6	9780 (17.6)	1.52 (1.50, 2.05)	70400 (20.9)	8.61 (26.0)	0.153 (38.6)
400	6	13800 (29.3)	2.00 (1.50, 3.00)	111000 (21.9)	10.4 (18.8)	0.187 (55.0)

Source: Study 2101 CSR, Figure 14.2-2.1a and Table 14.2-2.2a.
Abbreviations: AUC_{inf}, area under the curve to infinity; CL_R, renal clearance; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; T_{max}, time to maximum observed plasma concentration; $t_{1/2}$, half-life.

Table 60. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following Twice Daily Dosing of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 3)

		Geometric Mean (Geometric CV%)						
Dose Regimen	n	C_{max} (ng/mL)	T_{max} (h)	AUC_{0-12} (ng*h/mL)	CL/F (L/h)	$t_{1/2}$ (h)	R_{acc}	CL_R (L/h)
20 mg bid								
Day 1	8	811 (21.9)	1.05 (0.75, 2.00)	4470 (17.1)	-	-	-	-
Day 15	6	1030 (24.4)	1.00 (0.583, 2.50)	5890 (28.7)	3.40 (28.7)	9.85 (46.7)	1.32 (16.1)	0.0861 (24.9)
40 mg bid								
Day 1	6	1910 (21.7)	0.875 (0.5, 1.50)	9030 (14.9)	-	-	-	-
Day 15	6	2680 (25.3)	1.00 (0.6, 1.00)	12500 (30.8)	3.20 (30.8)	9.66 (47.5)	1.38 (21.4)	0.193 (53.5)
70 mg bid								
Day 1	22	3020 (22.5)	1.00 (0.5, 4.00)	13900 (28.0)	-	-	-	-
Day 15	16	3650 (29.6)	1.00 (0.5, 2.00)	19000 (43.5)	3.69 (43.5)	9.38 (43.6)	1.45 (18.8)	0.179 (55.5)

		Geometric Mean (Geometric CV%)						
140 mg bid								
Day 1	8	5620 (10.4)	0.875 (0.5, 2.50)	27400 (28.6)	-	-	-	-
Day 15	7	7640 (21.6)	0.75 (0.5, 2.55)	42000 (27.8)	3.33 (27.8)	10.9 (29.8)	1.57 (12.4)	0.251 (31.1)

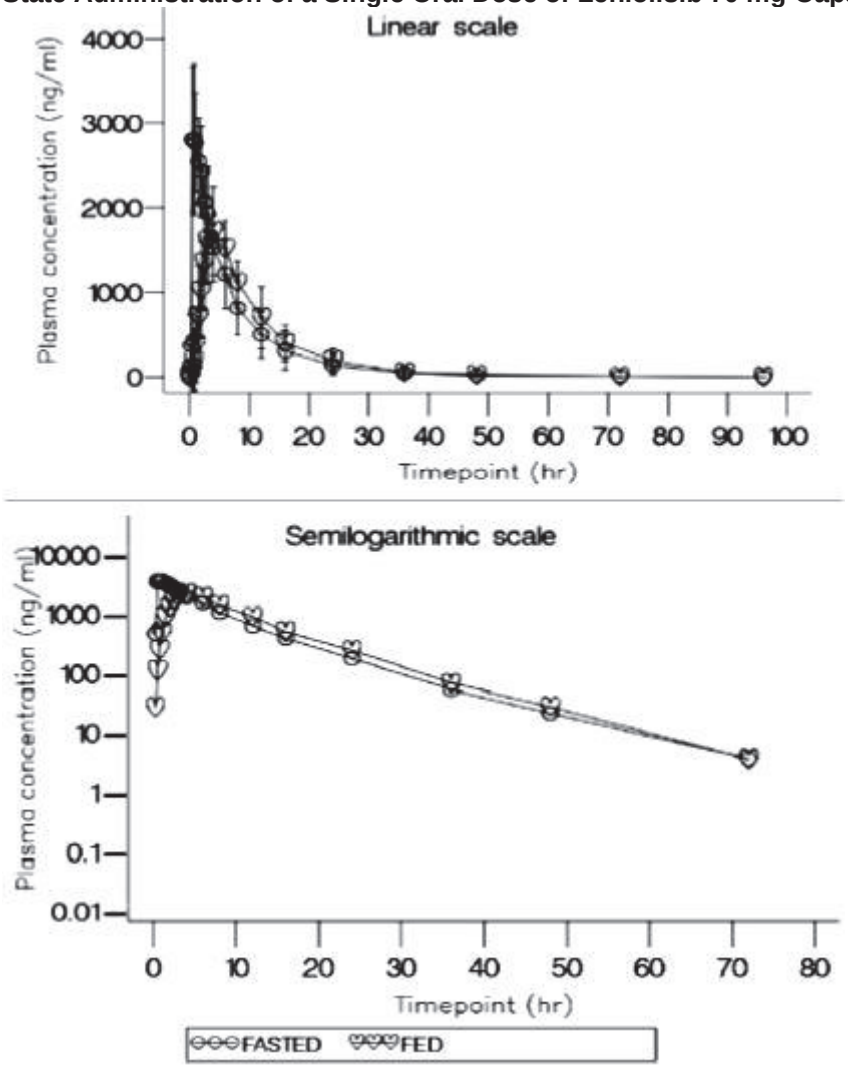
Source: Study 2101 CSR, Figure 14.2-2.1c and Table 14.2-2.2c.
Abbreviations: AUC_{0-12}, area under the curve from 0 to 12 hours; BID, twice daily; CL/F, apparent clearance; CL_R, renal clearance; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; R_{acc} accumulation ratio; T_{max}, time to maximum observed plasma concentration; $t_{1/2}$, half-life.

Food Effect

Food effect on the capsule ([(b) (4)] Novartis HGC) formulation was evaluated in Part 2 of Study 2101. Healthy participants were randomized to treatment sequence and received a single oral dose of leniolisib 70 mg (1 × 70 mg capsule) in the fasted and fed state. Mean plasma concentration-time profiles following a single dose of leniolisib 70 mg in the fed and fasted states are presented in Figure 14. Co-administration with a high-fat meal delayed the median T_{max} (3.51 versus 0.64 hours) and produced a lower geometric mean C_{max} (1890 versus 3190 ng/mL) relative to fasted-state administration but had no substantial effects on AUC_{inf}, apparent clearance (CL/F), and elimination half-life ($t_{1/2}$) (Table 61).

Statistical analysis of the individual ratios of leniolisib exposure (AUC from time 0 to the last measurable concentration [AUC_{0-t}], AUC_{inf}, and C_{max}) in the presence and absence of a high-fat meal showed that the 90% CIs for fed/fasted GMR were contained within 0.80 to 1.25 for all exposure metrics except C_{max} (Table 62). The fed/fasted GMR [90% CI] for C_{max} and $AUC_{0-\infty}$ were 0.59 [0.53, 0.66] and 1.00 [0.93, 1.07], respectively, indicating a decreased rate but unaffected extent of absorption with fed-state administration, consistent with the similar CL/F and $t_{1/2}$ observed regardless of food status

Figure 14. Mean (SD) Plasma Leniolisib Concentration-Time Profiles Following Fasted and Fed-State Administration of a Single Oral Dose of Leniolisib 70 mg Capsule (Study 2101, Part 2)



Source: Study 2101 CSR, Figure 14.2-2.1b.
Abbreviations: CSR, clinical study report; SD, standard deviation.

Table 61. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following Fasted- and Fed-State Administration of a Single Oral Dose of Leniolisib 70 mg Capsule (Study 2101, Part 2)

Treatment Group	n	Geometric Mean (Geometric CV%)[a]						
		C_{max} (ng/mL)	T_{max} (h)	$AUC_{0-\infty}$ (ng*h/mL)	CL/F (L/h)	V_z/F (L)	$t_{1/2}$ (h)	t_{lag} (h)
Fed	12	1890 (21.2)	3.51 [2.50, 6.00]	19200 (33.2)	3.64 (33.2)	35.4 (15.1)	6.75 (32.2)	0.500 [0.00, 1.02]
Fasted	12	3190 (18.0)	0.642 [0.500, 2.02]	19300 (42.0)	3.63 (42.0)	34.6 (27.2)	6.61 (47.4)	0.00 [0.00, 0.250]

Source: Study 2101 CSR, Table 14.2-2.1b.
Abbreviations: $AUC_{0-\infty}$, area under the curve to infinity; CL/F, apparent clearance; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; T_{max}, time to maximum observed plasma concentration; t_{lag}, lag time; $t_{1/2}$, half-life; V_z/F, apparent volume of distribution during terminal phase.

Table 62. Summary of Statistical Assessment for Food Effect on Plasma Leniolisib Exposure Following a Single Oral Dose of Leniolisib 70 mg Capsule (Study 2101, Part 2)

Parameter	Treatment	n	Geometric LS Mean[a]	Fed/Fasted Comparison[b]	
				Geometric LS Mean Ratio (%)	90% Confidence Interval (%)
AUC_{0-t} (ng*h/mL)	Fed	12	19105.35	99.43	92.40, 107.00
	Fasted	12	19214.41		
$AUC_{0-\infty}$ (ng*h/mL)	Fed	12	19223.28	99.66	92.52, 107.35
	Fasted	12	19288.30		
C_{max} (ng/mL)	Fed	12	1886.32	59.20	52.92, 66.23
	Fasted	12	3186.31		

Source: Study 2101 CSR, Table 14.2-4.1.
Abbreviations: AUC_{0-t}, area under the curve to the last quantifiable time point; $AUC_{0-\infty}$, area under the curve to infinity; C_{max}, maximum plasma concentration; CSR, clinical study report; LS, least squares; n, number of subjects in category.

Effect of Ethnicity on Pharmacokinetics

An exploratory analysis showed that the range of individual dose-normalized AUC_{inf} (AUC_{inf}/Dose) observed in the Japanese-only cohort (N=6, 169 to 281 ng*h/mL/mg) overlapped with the geometric mean range in the Caucasian-only cohorts (277 to 297 ng*h/mL/mg) in Part 1 of Study 2101. However, the geometric mean AUC_{inf}/Dose in the Japanese-only cohort (203 ng*h/mL/mg) was approximately 30% lower relative to Caucasian-only cohorts (Table 63). In regard to dose-normalized C_{max} (C_{max}/Dose), the Japanese-only cohort showed a tendency for higher peak exposure (geometric mean 42.7 ng/mL) compared to the Caucasian-only cohorts (geometric mean 34.5 to 42.4 ng/mL) in Part 1. This difference may be explained by the lower body weight in the Japanese participants and thus lower Vz/F. Because the leniolisib capsule formulation used by the Japanese-only cohort in Part 1 of the study ((b) (4) was different from the other cohorts in Parts 1 and 3 ((b) (4) formulation cannot be excluded as a confounding factor in the analysis. However, comparison of fasted-state administration data in Parts 1 and 2 of the study showed similar dose-normalized exposures between (b) (4) and (b) (4) formulations.

Based on the relatively minor differences seen, clinically relevant differences warranting dose adjustments relative to Caucasians are not expected for Japanese individuals. However, it should be noted that there was a limited number (n=6) of Japanese participants in Study 2101.

Table 63. Summary of Dose-Normalized Plasma Leniolisib Exposure Following Single-Dose Administration of Leniolisib at Escalating Doses in Healthy Participants (Study 2101, Part 1)

Leniolisib Dose	n	Geometric Mean (Geometric CV%)			
		C_{max}/Dose (ng/mL/mg)	$AUC_{0-\infty}$/Dose (ng*h/mL/mg)	AUC_{0-t}/Dose (ng*h/mL/mg)	AUC_{0-12}/Dose (ng*h/mL/mg)
10 mg	8	40.2 (35.5)	238 (43.5)	234 (43.6)	198 (34.2)
20 mg	8	39.5 (20.1)	260 (30.9)	258 (31.0)	204 (18.7)
40 mg	6	36.8 (33.0)	215 (39.2)	214 (39.3)	177 (32.7)
80 mg[a]	6	42.4 (42.4)	297 (44.5)	295 (44.6)	222 (40.8)
110 mg	6	36.5 (33.9)	257 (29.6)	256 (29.4)	199 (28.2)
140 mg[b]	6	42.7 (27.4)	203 (20.2)	203 (20.2)	183 (16.9)
200 mg	12	35.5 (22.6)	228 (33.2)	228 (33.2)	185 (28.1)
300 mg	6	32.6 (17.6)	235 (20.9)	234 (21.0)	180 (16.8)
400 mg[a]	6	34.5 (29.3)	277 (21.9)	277 (21.9)	210 (21.0)

Source: Study 2101 CSR, Table 14.2-2.1a.
[a] Caucasian-only cohort.
[b] Japanese-only cohort.
Abbreviations: AUC_{0-t}, area under the curve to the last quantifiable time point; AUC_{0-12}, area under the curve from 0 to 12 hours; $AUC_{0-\infty}$, area under the curve to infinity; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category.

Effect of Formulation

Leniolisib initially entered clinical development as a capsule formulation (Novartis HGC) before a tablet formulation was developed by Novartis (Novartis FCT). Two formulations of Novartis HGC were used in clinical studies, (b) (4) and (b) (4) All Novartis HGC at the 70 mg leniolisib dosage strength were of the (b) (4) formulation. Following acquisition by Pharming, the manufacturing process for the tablet formulation was transferred from Novartis Pharma AG to Pharming's intended manufacturer of the commercial product, Skyepharma (Pharming FCT). Healthy participants who received (b) (4) and (b) (4) formulations of Novartis HGC in Study 2101 showed similar dose-normalized leniolisib exposures, suggesting no clinically relevant differences between the 2 capsule formulations. The geometric mean dose-normalized AUC_{inf} and C_{max} following a single dose of (b) (4) in the fasted state were 276 (19300/70) ng*h/mL/mg and 45.6 (3190/70) ng/mL/mg, respectively, comparable to those observed with (b) (4) in the fasted state (215 to 297 ng*h/mL/mg and 32.6 to 42.4 ng/mL/mg, respectively) (Table 63).

Explorative Mass Balance and Metabolite Profiling

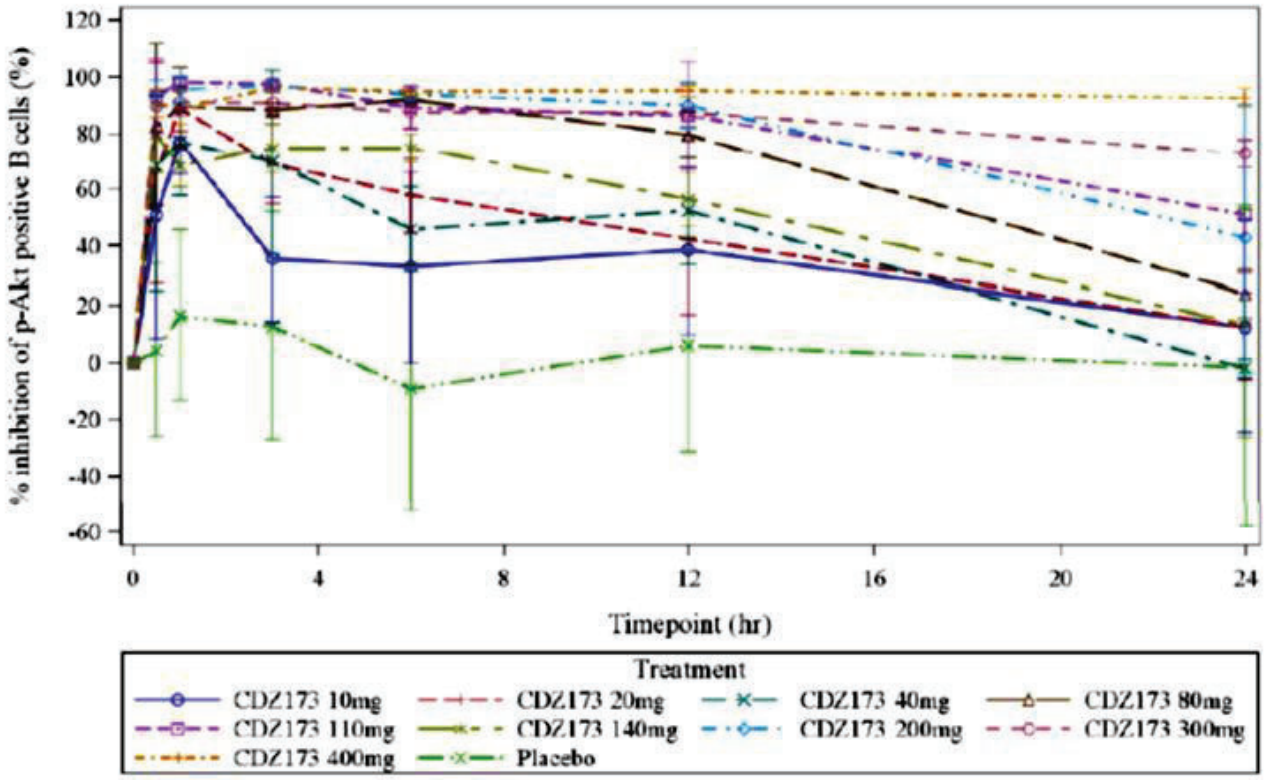
The ADME of leniolisib was investigated in the 400 mg single dose cohort through ^{19}F-NMR and liquid chromatography-tandem mass spectroscopy analysis of serial samples collected up to 96

(blood) and 120 (urine and feces) hours post-dose in Study 2101. Mass balance was not complete in the study, and only 65.5% of the administered dose was recovered (23.0% and 42.4% in urine and feces, respectively). The incomplete mass balance was mainly attributed to inadequate length of fecal sampling because cumulative recovery in the urine remained approximately constant from 24 to 120 hours post-dose, indicating complete urinary excretion of leniolisib and its metabolites after 24 hours. Thus, a subsequent ADME study using ^{14}C-leniolisib with longer urinary and feces collection period was conducted to elucidate the mass balance of leniolisib (Study LE2101).

Pharmacodynamic Results (%pAkt Inhibition in Ex Vivo Stimulated B cells)

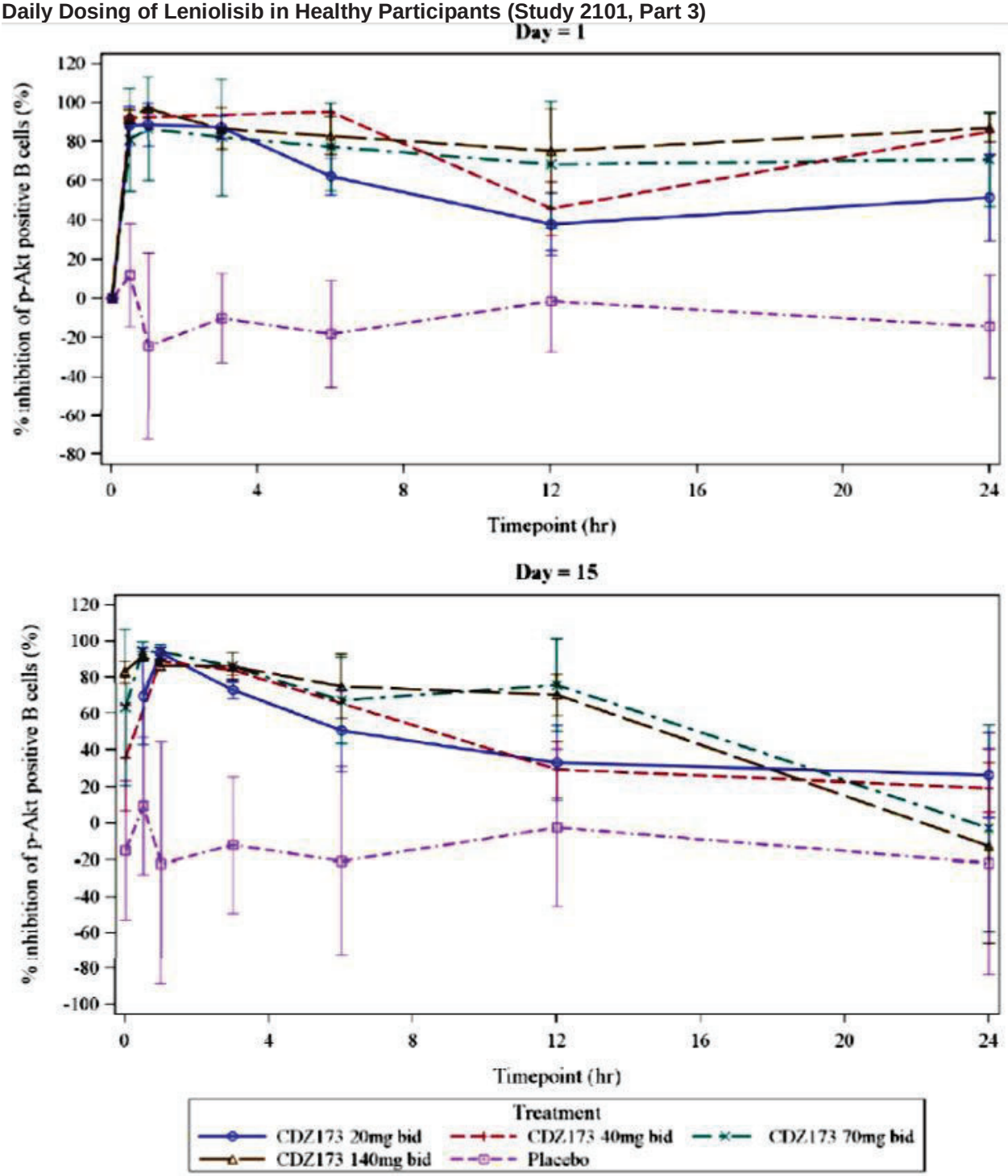
The PD effect of leniolisib was measured using ex vivo stimulated %pAkt-positive B cells as readout as Akt is a direct downstream target of activated PI3Kδ. Inhibition of pAkt in ex vivo stimulated B cells was observed shortly after dosing with leniolisib, while negligible reduction was observed in the placebo group Figure 15 and Figure 16. Percent pAkt inhibition was highest in the 70 and 140 mg twice daily dose groups and peaked at approximately 1-hour post-dose, suggesting maximal pAkt inhibition was achieved after doses 70 mg BID in healthy subjects. On treatment discontinuation following 15 days of a twice daily regimen, mean %pAkt-positive B cells recovered to their baseline level in approximately 24 hours. As evident from the scatterplots of %pAkt-positive B cells versus plasma leniolisib concentration in a real-time fashion (Figure 17), %pAkt-positive B cells rapidly declined to <10% as plasma concentration increased to approximately 200 to 300 ng/mL.

Figure 15. Mean (SD) %pAkt Inhibition in Ex Vivo Stimulated B Cells Over Time Following Single-Dose Administration of Leniolisib in Healthy Participants (Study 2101, Part 1)



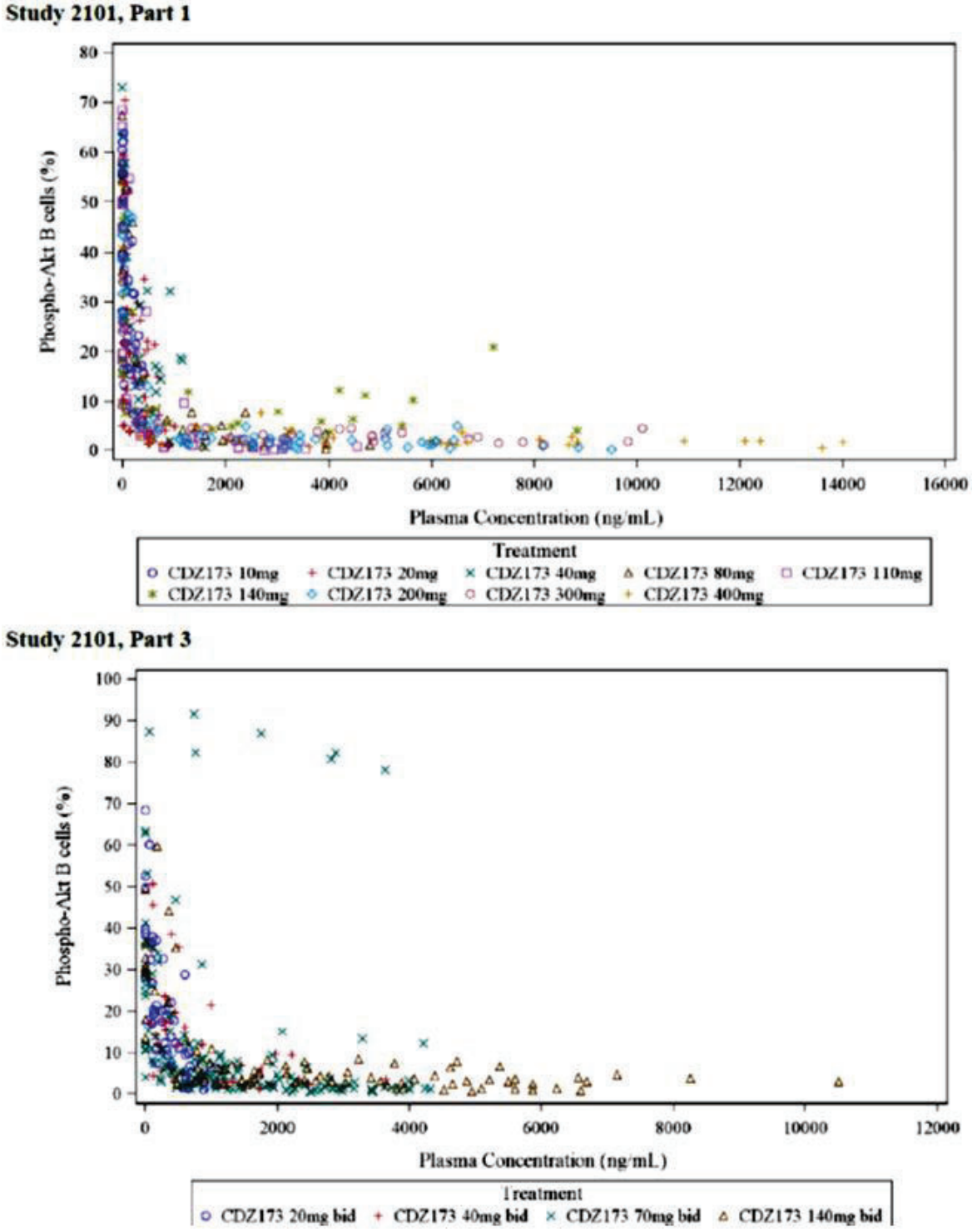
Source: Study 2101 CSR, Figure 14.2-10a.
Abbreviations: CSR, clinical study report; pAkt, phosphorylated protein kinase B; SD, standard deviation

Figure 16. Mean (SD) %pAkt Inhibition in Ex Vivo Stimulated B Cells Over Time Following Twice Daily Dosing of Leniolisib in Healthy Participants (Study 2101, Part 3)



Source: Study 2101 CSR, Figure 14.2-10c.
Abbreviations: BID, twice daily; CSR, clinical study report; pAkt, phosphorylated protein kinase B; SD, standard deviation

Figure 17. Scatterplots of Percent pAkt-Positive Ex Vivo Stimulated B Cells Versus Plasma Leniolisib Concentration in Healthy Participants (Study 2101, Parts 1 and 3)



Source: Study 2101 CSR, Figure 14.2-11a and Figure 14.2-12c.
Abbreviations: BID, twice daily; CSR, clinical study report; Phospho-Akt, phosphorylated protein kinase B.

Reviewer Comment: *Following a single dose from 10 to 400 mg in healthy participants, leniolisib was rapidly absorbed with the median T_{max} ranging from 1 to 2 hours postdose. With twice daily dosing from 20 to 140 mg, median T_{max} was mostly unchanged at approximately 1-hour post-dose. Leniolisib exposure increased proportional to dose across the dose ranges evaluated. With twice daily dosing, there was low accumulation (accumulation ratio 1.32 to 1.57) in healthy participants, and steady state was achieved within 2 to 3 days from onset of therapy. PK of leniolisib were similar between healthy participants and APDS subjects. Co-administration with a high-fat meal decreased the rate but not the extent of leniolisib absorption following a single dose of 70 mg leniolisib capsule, as indicated by a delayed time to maximum concentrations (T_{max}) from approximately 1 to 4 hours postdose and decreased C_{max} but unaffected AUC_{inf} upon fed-state administration (fed/fasted geometric mean ratio [90% CI] of 0.59 [0.53, 0.66] and 1.00 [0.93, 1.07], respectively), suggesting that the effect of food on leniolisib PK is low.*

The effect of food on leniolisib exposure is not expected to compromise on-target pathway inhibition because fed-state Cmax (1890 ng/mL) at the therapeutic dose was well over the EC50 for pAkt inhibition in B cells (300 ng/mL). However, it should be noted that food effect study was conducted with the capsule formulation and the food effect on the to-be-marketed tablet formulation has not been evaluated. The Applicant provided in vitro dissolution data to show similarity in dissolution profiles between the capsule and to-be-marketed tablet formulations. The dosage form change between the tablet and capsule formulations was not considered significant (b) (4)

In addition, bioequivalence was achieved between the capsule and tablet formulations in Study LE1101. Furthermore, leniolisib was administered without regard to food in the pivotal Study 2201. In the extension study, both capsule and tablet formulations were given irrespective of food and based on the results from a recently conducted food intake survey by the Applicant, the majority of the subjects in Study 2201E1 took the drug with food. Therefore, based on these observations, the lack of food effect data with the to-be-marketed formulation may be acceptable.

The maximal pAkt inhibition in healthy subjects appeared to reach at doses of 70 mg and 140 mg BID (around 80%) from the ex vivo stimulation data in Study 2101, and, therefore, doses up to 70 mg BID were evaluated in Study 2201 (Part 1) in APDS *subjects* which appears reasonable from Clinical Pharmacology perspective.

16.3.3.2. Study LE1101

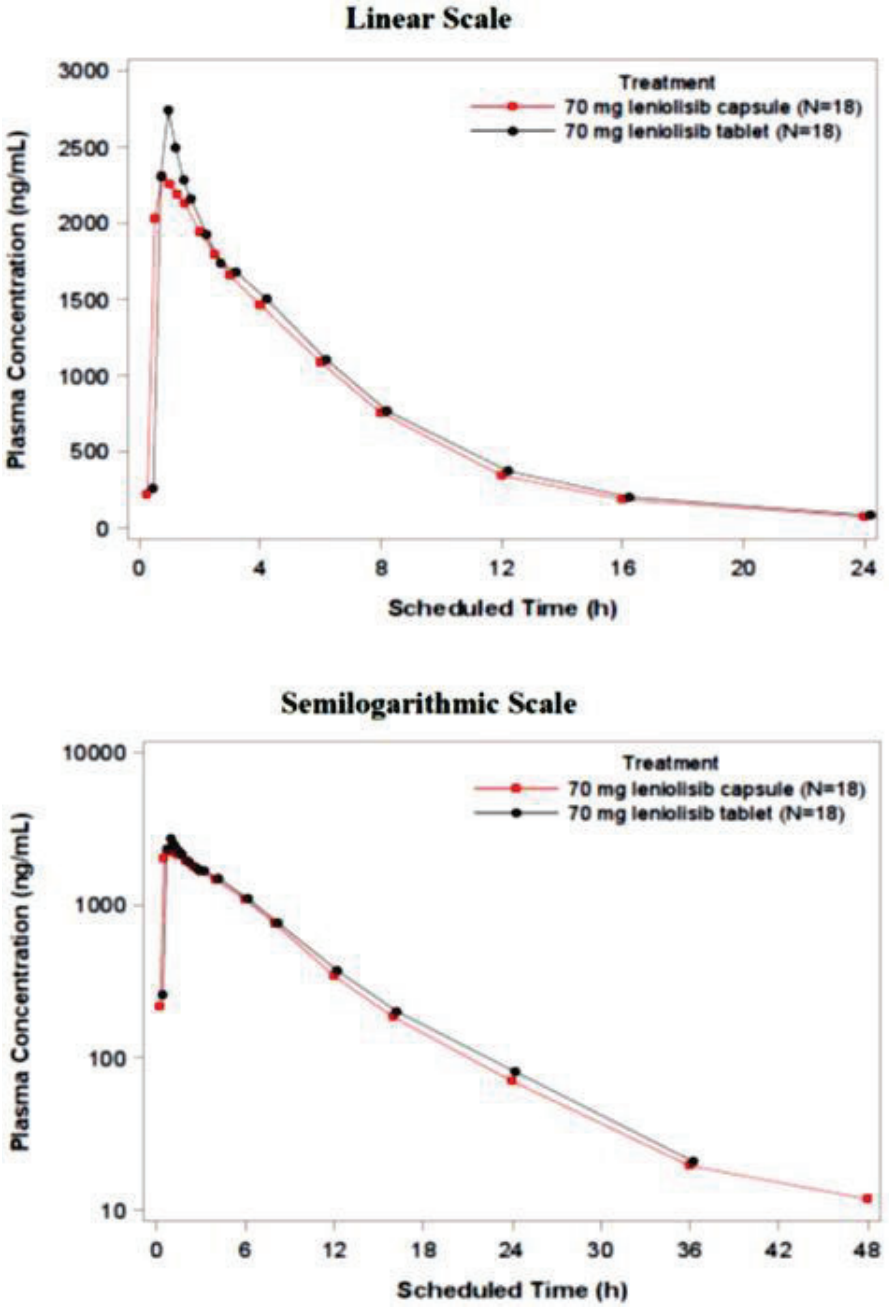
- Single-dose PK and bioequivalence between capsule and tablet formulations in healthy participants.

Study LE1101 was a phase 1, single-center, randomized, open-label, 2-way crossover study to demonstrate that Pharming 70 mg leniolisib film-coated tablets (Pharming FCT) were

bioequivalent to Novartis 70 mg leniolisib HGC (Novartis HGC (b) (4) in healthy participants. A total of 20 participants were randomized to treatment sequence, each receiving a single oral dose of leniolisib 70 mg as capsule (1 × 70 mg capsule) and as tablet (1 × 70 mg tablet) formulation. There were 18 participants who completed both treatment periods and received both formulations. Capsules were manufactured by Novartis Pharma AG (Novartis HGC), and tablets were manufactured by Skyepharma (Pharming FCT). Treatments were given in the fasted state and were separated by a ≥7-day washout period.

Geometric mean plasma concentration-time profiles following a single dose of leniolisib 70 mg as Pharming FCT and Novartis HGC formulations are presented in Figure 18. The rate and extent of leniolisib exposure appeared comparable between formulations (Table 64). T_{max} was 0.63 and 0.75 hours postdose with Pharming FCT and Novartis HGC, respectively. Geometric mean C_{max} and AUC_{inf} were 3102 ng/mL and 17327 ng*h/mL, respectively, with Pharming FCT and 2873 ng/mL and 16701 ng*h/mL, respectively, with Novartis HGC. Statistical analysis for bioequivalence showed that the 90% CI of GMRs for leniolisib exposure (C_{max} and AUC) with test/reference formulation were well contained within 0.80 to 1.25 for all exposure metrics (Table 65). These results show that Pharming FCT (test) and Novartis HGC (reference) formulations of leniolisib 70 mg were bioequivalent.

Figure 18. Geometric Mean Plasma Leniolisib Concentration-Time Profiles Following a Single Oral Dose of Leniolisib 70 mg as Pharming FCT and Novartis HGC Formulations (Study LE1101)



Source: Study LE1101 CSR, Figure 15.2.4.
Abbreviations: CSR, clinical study report; N, total number of subjects.

Table 64. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following a Single Oral Dose of Leniolisib 70 mg as Pharming FCT and Novartis HGC Formulations (Study LE1101)

Treatment Group	n	Geometric Mean (Geometric CV%)[a]					
		C_{max} (ng/mL)	T_{max} (h)	$AUC_{0-\infty}$ (ng*h/mL)	CL/F (L/h)	V_z/F (L)	$t_{1/2}$ (h)
Pharming FCT	18	3102 (21.8)	0.63 [0.50, 1.25]	17327 (33.8)	4.04 (33.8)	38.9 (24.4)	6.67 (24.1)
Novartis HGC	18	2873 (21.5)	0.75 [0.50, 2.00]	16701 (36.0)	4.19 (36.0)	39.9 (26.1)	6.60 (26.6)

Source: Study LE1101 CSR, Table 15.2.2.
Abbreviations: $AUC_{0-\infty}$, area under the curve to infinity; CL/F, apparent clearance; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category, T_{max}, time to maximum observed plasma concentration; $t_{1/2}$, half-life; V_z/F, apparent volume of distr bution during terminal phase.

Table 65. Summary of Statistical Assessment for Bioequivalence Between Pharming FCT and Novartis HGC Formulations of Leniolisib 70 mg (Study LE1101)

Parameter	Treatment	n	Geometric LS Mean	Test/Reference Comparison	
				Geometric LS Mean Ratio (%)	90% Confidence Interval
C_{max} (ng/mL)	Pharming FCT (test)	18	3102	1.0799	0.9737, 1.1977
	Novartis HGC (reference)	18	2873		
AUC_{0-t} (ng*h/mL)	Pharming FCT (test)	18	17222	1.0375	0.9756, 1.1032
	Novartis HGC (reference)	18	16600		
$AUC_{0-\infty}$ (ng*h/mL)	Pharming FCT (test)	18	17327	1.0374	0.9756, 1.1032
	Novartis HGC (reference)	18	16701		

Source: Study LE1101 CSR, Table 15.2.3.
Abbreviations: AUC_{0-t}, area under the curve to the last quantifiable time point; $AUC_{0-\infty}$, area under the curve to infinity; C_{max}, maximum plasma concentration; CSR, clinical study report; LS, least squares; n, number of subjects in category.

Reviewer Comment: *In Study LE1101, leniolisib exposure was comparable between a single dose of 70 mg leniolisib as Pharming FCT (to-be-marketed formulation) and as Novartis HGC (used in pivotal phase 2/3 study). The 90% CI of the geometric mean ratios for exposure metrics (C_{max} and AUC) with two formulations were well contained within the limits of 0.80 to 1.25, showing that the two formulations are bioequivalent.*

16.3.3.3. Study LE2101

- Single-dose PK and mass balance in healthy male participants

Study LE2101 was a Phase 1, single-center, open-label, ADME study to determine the total recovery and relative excretion of radioactivity in urine and feces after a single oral dose of 70 mg ^{14}C-leniolisib in healthy male participants. A total of 6 participants were enrolled and received a single dose of 70 mg ^{14}C-leniolisib (40 µCi) oral solution in the fasted state.

Following administration of 70 mg ^{14}C-leniolisib, absorption was rapid and the median T_{max} was 0.5 hours for both plasma leniolisib and total ^{14}C-radioactivity. Geometric mean $t_{1/2}$ was substantially longer for plasma total ^{14}C-radioactivity than plasma leniolisib (32.8 versus 6.39 hours), suggesting presence of metabolites with longer $t_{1/2}$ than parent (Table 66). The GMR for plasma leniolisib exposure (AUC and C_{max})/plasma total ^{14}C-radioactivity ranged from 0.68 to 0.87, indicating that the majority (approximately 70%) of total ^{14}C-radioactivity in plasma was associated with unchanged leniolisib.

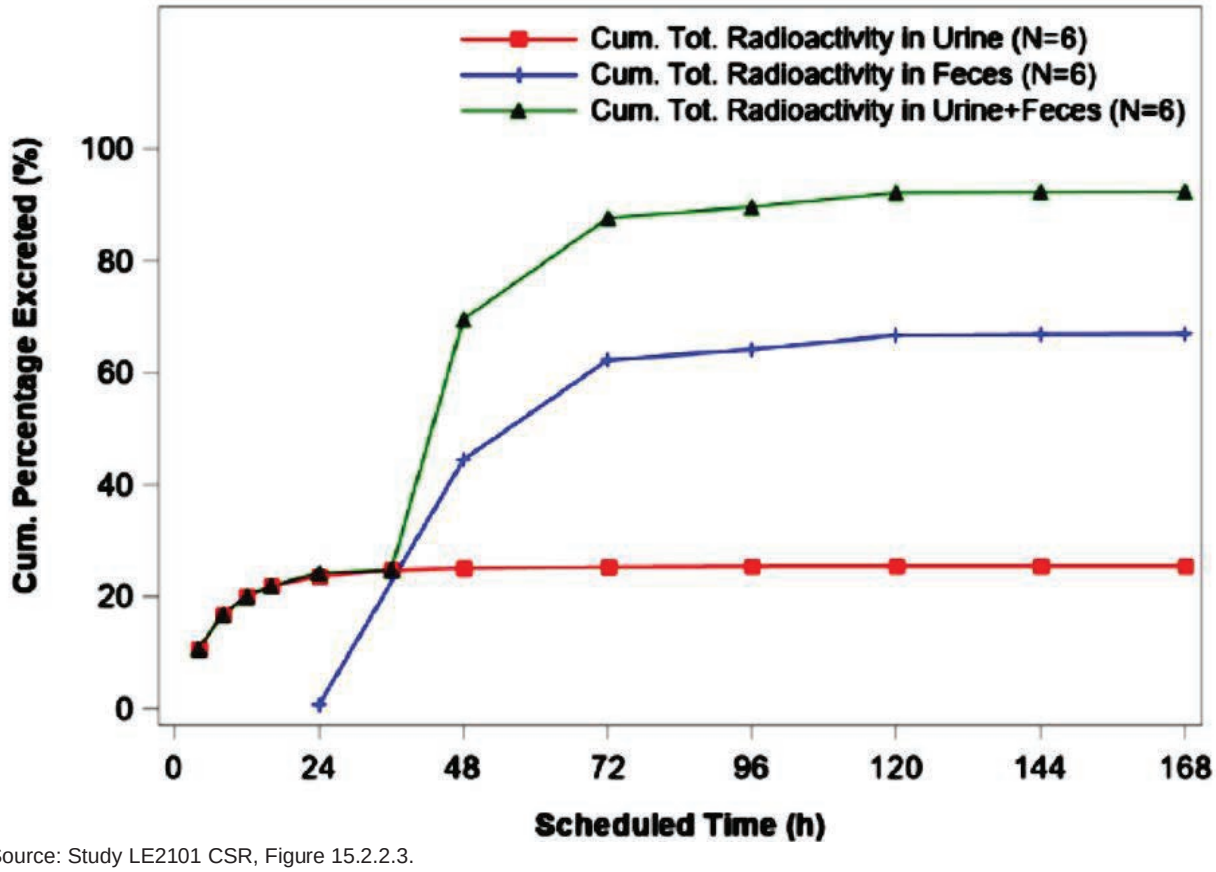
Table 66. Summary of Plasma Leniolisib and Total ^{14}C-Radioactivity Pharmacokinetic Parameters Following Single-Dose 70 mg ^{14}C-Leniolisib in Healthy Male Participants (Study LE2101)

		Geometric Mean (Geometric CV%)[a]						
Analyte	n	C_{max}[b] (ng/mL)	T_{max} (h)	AUC_{0-tz}[c] (ng*h/mL)	AUC_{0-t}[c] (ng*h/mL)	CL/F (L/h)	V_z/F (L)	$t_{1/2}$ (h)
Plasma leniolisib	6	3153 (36.6)	0.50 [0.25, 0.75]	16008 (36.5)	15862 (36.9)	4.38 (36.2)	40.3 (36.9)	6.39 (30.3)
Plasma total ^{14}C-radioactivity	6	3606 (23.5)	0.50 [0.25, 0.75]	23371 (26.5)	21500 (23.8)	–	–	32.8 (176.9)

Source: Study LE2101 CSR, Table 15.2.1.7 and Table 15.2.1.8.
Abbreviations: AUC_{0-t}, area under the curve to the last quantifiable time point; $AUC_{0-\infty}$, area under the curve to infinity; CL/F, apparent clearance; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; T_{max}, time to maximum observed plasma concentration; $t_{1/2}$, half-life; V_z/F, apparent volume of distribution during terminal phase.

The mean cumulative mass balance of total ^{14}C-radioactivity was 92.5% at 168 hours postdose. Excretion of leniolisib and its metabolites was predominately via feces (67.0%), followed by urine (25.5%; Figure 19).

Figure 19. Mean Cumulative Total ^{14}C-Radioactivity in Urine and Feces Versus Time Following Single-Dose 70 mg ^{14}C-Leniolisib in Healthy Male Participants (Study LE2101)



Source: Study LE2101 CSR, Figure 15.2.2.3.
Abbreviations: CSR, clinical study report; Cum. Tot, cumulative total; N, total number of subjects.

Reviewer Comment: *The mean recovery of total ^{14}C-radoactivity following a single oral dose of 70 mg ^{14}C-leniolisib was 92.5% (67.0% and 25.5% recovered via feces and urine, respectively) 168 hours postdose. The results suggested that the majority (approximately 70%) of total ^{14}C-radioactivity in plasma was associated with unchanged leniolisib.*

16.3.3.4. Study 2102

- DDI potential with a strong dual CYP3A4 and P-gp inhibitor (Itraconazole) and a strong P-gp and CYP2D6 inhibitor (quinidine) in healthy male participants.

Study 2102 was a phase 1, single-center, open-label, single-sequence, 3-period crossover DDI study assessing the effect of itraconazole and quinidine co-administration on the single-dose PK of oral leniolisib in healthy male participants. A total of 20 participants were enrolled to enter the following treatment periods:

1. Single-dose leniolisib 10 mg on Day 1.

2. Itraconazole 200 mg once daily on Days 1 through 9 + single-dose leniolisib 10 mg on Day 5 approximately 3 hours after the itraconazole dose.
3. Two doses of quinidine 300 mg 4 hours apart on Day 1 + single-dose leniolisib 10 mg on Day 1 approximately 1 hour after the first quinidine dose.

All morning doses were administered after an overnight fast, a standard light breakfast, and another fasting period. Leniolisib was administered at least 3.5 hours after the start of the breakfast. Study drugs were supplied as 10 mg leniolisib capsules, 100 mg itraconazole capsules, and 300 mg quinidine tablets. A washout period of ≥1 week separated Period 1, Day 1 and Period 2, Day 1, while a washout period of ≥4 weeks separated Period 2, Day 1 and Period 3, Day 1.

Co-administration with itraconazole increased leniolisib AUC_{inf} by approximately 2-fold and C_{max} by 25% (Table 67). Therefore, leniolisib is not a sensitive substrate of CYP3A, and concomitant use with weak/moderate CYP3A inhibitors may be acceptable without dose adjustments, while concomitant use with strong CYP3A inhibitors should be avoided. Mean plasma itraconazole C_{trough} was above 100 ng/mL by Period 2, Day 5 and confirmed adequate itraconazole exposure to exert strong CYP3A/P-gp inhibition by the time leniolisib was given in the study.

Co-administration with quinidine did not affect leniolisib exposure (AUC and C_{max}). The GMR 90% CIs for leniolisib exposure metrics with/without quinidine coadministration fell within 0.80 to 1.25 (Table 67). Based on the observed DDI between leniolisib and itraconazole (a strong dual CYP3A/P-gp inhibitor) but the absence of interaction between leniolisib and quinidine (a strong dual CYP2D6/P-gp inhibitor), it can be concluded that CYP3A plays a relevant role in the in vivo disposition of leniolisib but CYP2D6 and P-gp do not. Hence, concomitant use of leniolisib with CYP2D6 or P-gp inhibitors is acceptable without any dose adjustments.

Table 67. Summary of Statistical Assessment for Effect of Itraconazole and Quinidine Coadministration on Plasma Leniolisib Exposure in Healthy Male Participants (Study 2102)

Leniolisib Exposure Metric	Treatment Period	n	Adjusted Geometric Mean	Treatment Comparison	
				Test/Reference Period	Geometric Mean Ratio [90% CI]
C_{max} (ng/mL)	1 (leniolisib alone)	20	441	–	–
	2 (leniolisib + itraconazole)	20	552	2/1	1.25 [1.15, 1.36]
	3 (leniolisib + quinidine)	18	406	3/1	0.92 [0.84, 1.00]
$AUC_{0-\infty}$ (ng*h/mL)	1 (leniolisib alone)	20	2700	–	–
	2 (leniolisib + itraconazole)	20	5730	2/1	2.12 [1.97, 2.28]
	3 (leniolisib + quinidine)	18	2760	3/1	1.02 [0.95, 1.10]
AUC_{0-t} (ng*h/mL)	1 (leniolisib alone)	20	2660	–	–
	2 (leniolisib + itraconazole)	20	5620	2/1	2.12 [1.97, 2.28]
	3 (leniolisib + quinidine)	18	2710	3/1	1.02 [0.94, 1.10]
AUC_{0-24} (ng*h/mL)	1 (leniolisib alone)	20	2500	–	–
	2 (leniolisib + itraconazole)	20	4440	2/1	1.78 [1.67, 1.90]
	3 (leniolisib + quinidine)	18	2560	3/1	1.02 [0.96, 1.10]

Source: Study 2102 CSR, Table 14.2-1.3.1.
Abbreviations: AUC_{0-t}, area under the curve to the last quantifiable time point; $AUC_{0-\infty}$, area under the curve to infinity; AUC_{0-24}, area under the curve from 0 to 24 hours; CI, confidence interval; C_{max}, maximum plasma concentration; CSR, clinical study report; n, number of subjects in category.

Reviewer Comment: *Co-administration with itraconazole (a strong dual inhibitor of CYP3A4 and P-gp) increased leniolisib AUC_{inf} by approximately 2-fold and C_{max} by 25%, while co-administration with quinidine (a strong dual inhibitor of CYP2D6 and P-gp) did not affect leniolisib exposure. Hence, leniolisib may be co-administered with CYP2D6 and P-gp inhibitors without dose adjustment, but concomitant use with strong CYP3A4 inhibitors should be avoided.*

16.3.3.5. Study 2104

- DDI Potential With CYP3A4, UGT1A1, and SULT1E1 substrate oral contraceptives (ethinylestradiol and levonorgestrel) in healthy female participants

Study 2104 was a phase 1, single-center, open-label, fixed-sequence, 2-period DDI study assessing the effect of multiple-dose leniolisib administration on the single-dose PK of a monophasic combination oral contraceptive (ethinylestradiol and levonorgestrel) in healthy female participants. A total of 30 participants were enrolled to enter the following treatment periods:

1. Single dose of oral contraceptive (30 µg ethinylestradiol/150 µg levonorgestrel) on Day 1.
2. Leniolisib 70 mg twice daily on Days 1 through 17 + single dose of oral contraceptive (30 µg ethinylestradiol/150 µg levonorgestrel) on Day 15.

All morning doses were administered after an overnight fast. Evening doses of leniolisib were given at least 1 hour after dinner was consumed. Study drugs were supplied as 70 mg leniolisib capsules and 30 µg ethinylestradiol/150 µg levonorgestrel tablets. A washout period of ≥1 week separated Period 1, Day 1 and Period 2, Day 1.

Co-administration with leniolisib at steady state increased ethinylestradiol exposure (AUC and C_{max}) by approximately 30% relative to when oral contraceptive (ethinylestradiol/levonorgestrel) was given alone but had no effect on levonorgestrel exposure (Table 68). The 30% increase in ethinylestradiol exposure is modest and unlikely to constitute a relevant increase in the risk of adverse events associated with low-dose oral contraceptives containing 20 to 35 µg ethinylestradiol.

In both treatment periods, AUC_{inf} was not calculated for several PK profiles because it would have required >20% extrapolation from the last quantifiable concentration to infinity. Hence, AUC from time 0 to 48 hours (AUC_{0-48}) was the favored exposure metric for statistical analysis of total drug exposure because it was available for most participants in both treatment periods and for both analytes (n≥27).

Table 68. Summary of Statistical Assessment for Effect of Leniolisib Co-administration on Plasma Ethinylestradiol and Levonorgestrel Exposure in Healthy Female Participants (Study 2104)

Analyte Exposure Metric	Treatment Period	n	Adjusted Geometric Mean	Treatment Comparison	
				Test/Reference Period	Geometric Mean Ratio [90% CI]
Ethinylestradiol					
C_{max} (pg/mL)	1 (leniolisib alone)	30	72.0	–	–
	2 (leniolisib + oral contraceptive)	27	89.7	2/1	1.25 [1.19, 1.30]
$AUC_{0-\infty}$ (pg*h/mL)	1 (oral contraceptive alone)	23	665	–	–
	2 (leniolisib + oral contraceptive)	22	879	2/1	1.32 [1.26, 1.39]
AUC_{0-t} (pg*h/mL)	1 (leniolisib alone)	30	513	–	–
	2 (leniolisib + oral contraceptive)	27	690	2/1	1.35 [1.29, 1.41]
AUC_{0-48} (pg*h/mL)	1 (leniolisib alone)	30	547	–	–
	2 (leniolisib + oral contraceptive)	27	723	2/1	1.32 [1.27, 1.38]
Levonorgestrel					
C_{max} (pg/mL)	1 (leniolisib alone)	29	3300	–	–
	2 (leniolisib + oral contraceptive)	27	2950	2/1	0.896 [0.823, 0.974]
$AUC_{0-\infty}$ (pg*h/mL)	1 (oral contraceptive alone)	15	39400	–	–
	2 (leniolisib + oral contraceptive)	9	39900	2/1	1.01 [0.860, 1.20]
AUC_{0-t} (pg*h/mL)	1 (leniolisib alone)	29	32600	–	–
	2 (leniolisib + oral contraceptive)	27	30800	2/1	0.945 [0.875, 1.02]
AUC_{0-48} (pg*h/mL)	1 (leniolisib alone)	29	27700	–	–
	2 (leniolisib + oral contraceptive)	27	25900	2/1	0.935 [0.867, 1.01]

Source: Study 2104 CSR, Table 14.2-3.1.
Abbreviations: AUC_{0-t}, area under the curve to the last quantifiable time point; $AUC_{0-\infty}$, area under the curve to infinity; AUC_{0-48}, area under the curve from 0 to 48 hours; CI, confidence interval; C_{max}, maximum plasma concentration; CSR, clinical study report; n, number of subjects in category.

Reviewer Comment: *Co-administration of leniolisib at steady state with CYP3A4, SULT1E1, and UGT1A1 substrate oral contraceptives (ethinylestradiol + levonorgestrel) increased ethinylestradiol exposure by approximately 30% and had no effect on levonorgestrel exposure. The increase in ethinylestradiol exposure is unlikely to compromise the efficacy of a combined oral contraceptive composed of ethinylestradiol and levonorgestrel. Hence, leniolisib may be co-administered with oral contraceptives without dose adjustment.*

16.3.3.6. Study 2201

- Single- and multiple-dose PK, PD, and dose proportionality in subjects with APDS

Study 2201 was a Phase 2/3, multicenter, multinational study assessing the PK, dose-PD, and PK/PD relationships of leniolisib in subjects with APDS. The study was conducted in 2 parts: an open-label, nonrandomized, dose-finding study (Part 1) and a participant-, investigator-, and sponsor-blinded, randomized, placebo-controlled, fixed-dose study (Part 2). In Part 1, a total of 6 subjects received oral leniolisib twice daily at sequentially increasing dose levels every 4 weeks from 10 mg to 30 mg to 70 mg. Evening doses of 10 and 30 mg on Part 1, Days 28 and 56, respectively, were not taken to allow for pAkt washout. In Part 2, a total of 31 subjects were randomized in a ratio of 2:1 to receive oral leniolisib 70 mg (N=21) or placebo (N=10) twice daily for 12 weeks. Study drug was supplied as leniolisib 10 and 70 mg capsules and taken without regard to food in both study parts.

Leniolisib Pharmacokinetics in Part 1 (Dose Finding)

Leniolisib was rapidly absorbed following oral administration in subjects with APDS (Figure 20). Individual T_{max} was variable likely because doses were taken without regard to food (Table 69). Food is known to decrease the rate but not the extent of leniolisib absorption based on the previous clinical experience (Study 2102 Part 2). A time-invariant C_{trough} was reached by Day 8 of treatment at each dose level, indicating achievement of steady state ().

In Part 1 of the study, individual steady-state PK parameters were derived from a 1-compartment model, where disposition was characterized by a single oral drug clearance (TVCL) and volume (TVV) parameter (Table 70). The estimated TVCL geometric mean was 3.72 L/h with interindividual variability (geometric CV) of 27.7%.

Dose proportionality was observed for leniolisib exposure over the entire dose range investigated (10, 30, and 70 mg twice daily). Leniolisib exposure metrics (AUC, C_{max}, and C_{trough}) increased in an approximately dose-proportional manner as the leniolisib dose was sequentially increased. Furthermore, a linear 1-compartmental model with a single clearance term provided adequate fit to longitudinal data across the 3 dose levels.

Table 69. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following Sequentially Escalating Doses of Leniolisib Capsule in Subjects With APDS (Study 2201, Part 1)

		Geometric Mean (Geometric CV%)			
Leniolisib Dose Regimen	**n**	**C_{max} (ng/mL)**	**T_{max} (h)**	**AUC_{0-8} (ng*h/mL)**	**C_{trough} (ng/mL)**
10 mg bid					
Day 1	6	372 (38.6)	2.01 (0.92, 4.78)	1730 (28.9)	-
Day 8	5	-	-	-	109 (61.1)
Day 15	6	-	-	-	116 (39.1)

Leniolisib Dose Regimen	n	Geometric Mean (Geometric CV%) C_{max} (ng/mL)	T_{max} (h)	AUC_{0-8} (ng*h/mL)	C_{trough} (ng/mL)
30 mg bid					
Day 29	6	1040 (21.7)	3.05 (0.25, 4.87)	4770 (18.4)	-
Day 36	5	-	-	-	340 (53.6)
Day 43	6	-	-	-	333 (60.1)
70 mg bid					
Day 57	6	2440 (31.7)	1.93 (0.97, 4.95)	11800 (21.6)	-
Day 64	6	-	-	-	872 (42.7)
Day 71	6	-	-	-	916 (50.1)
Day 84	5	-	-	-	870 (51.3)

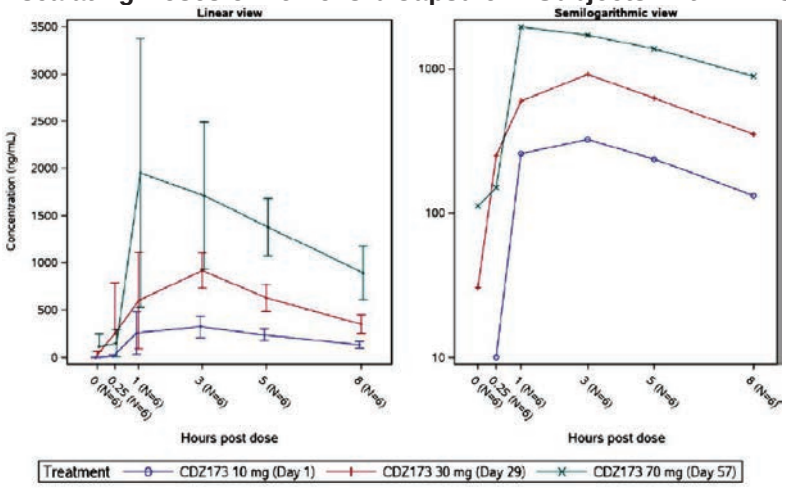
Source: Study 2201 Part 1 CSR, Table 14.2-1.2.1.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{0-8}, area under the curve from 0 to 8 hours; BID, twice daily; C_{max}, maximum plasma concentration; C_{trough}, trough concentration at steady-state; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; T_{max}, time to maximum observed plasma concentration.

Table 70. Summary Statistics of Leniolisib Pharmacokinetic Parameters Estimated by a 1-Compartment Model in Subjects With APDS (Study 2201, Part 1)

Statistic	TVCL (L/h)	TVV (L)	TVKA (h^{-1})
n	6	6	6
Mean (SD)	3.83 (0.897)	29.0 (5.76)	4.25 (3.66)
Geometric mean (Geometric CV%)	3.72 (27.7)	28.5 (19.6)	3.33 (84.2)
Median [Min, Max]	4.18 [2.26, 4.75]	28.3 [22.9, 38.2]	3.47 [1.42, 11.4]

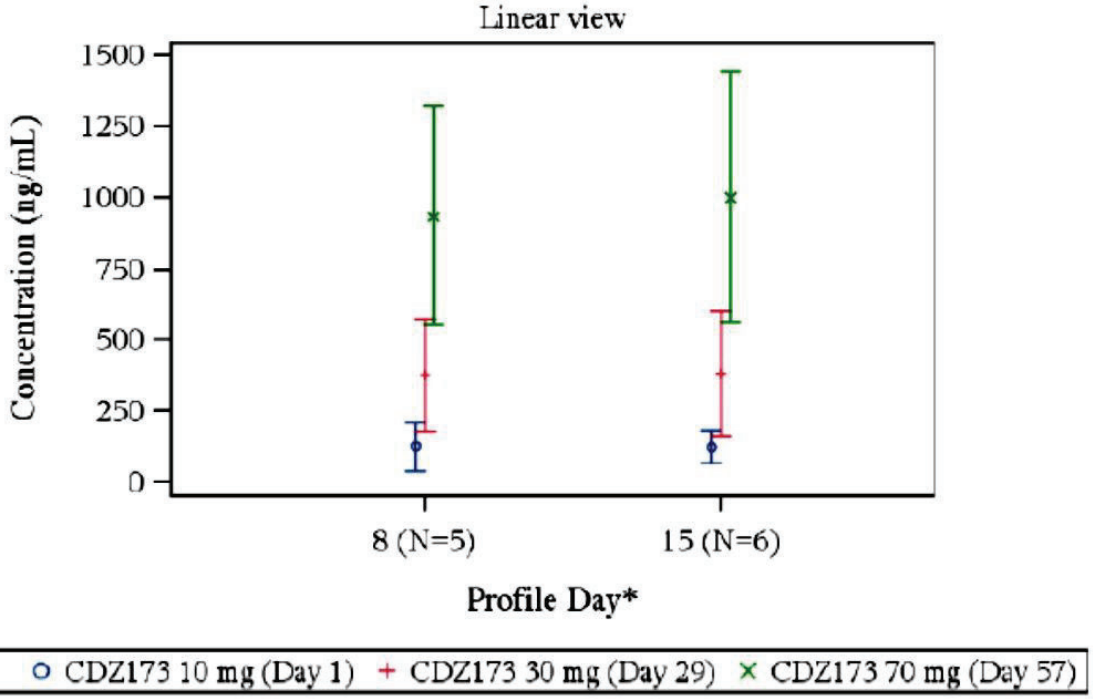
Source: Study 2201 Part 1 CSR, Table 14.2-1.2.2.
Abbreviations: CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; SD, standard deviation; TVCL, typical value of clearance; TVKA, typical value of drug absorption rate; TVV; typical value of volume of distribution.

Figure 20. Mean (SD) Leniolisib Plasma Concentration-Time Profile Following Sequentially Escalating Doses of Leniolisib Capsule in Subjects With APDS (Study 2201, Part 1)



Source: Study 2201 Part 1 Amendment 1 CSR, Figure 4-2.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Figure 21. Mean (SD) Leniolisib Plasma Trough Concentration-Time Profile Following Sequentially Escalating Doses of Leniolisib Capsule in Subjects With APDS (Study 2201, Part 1)

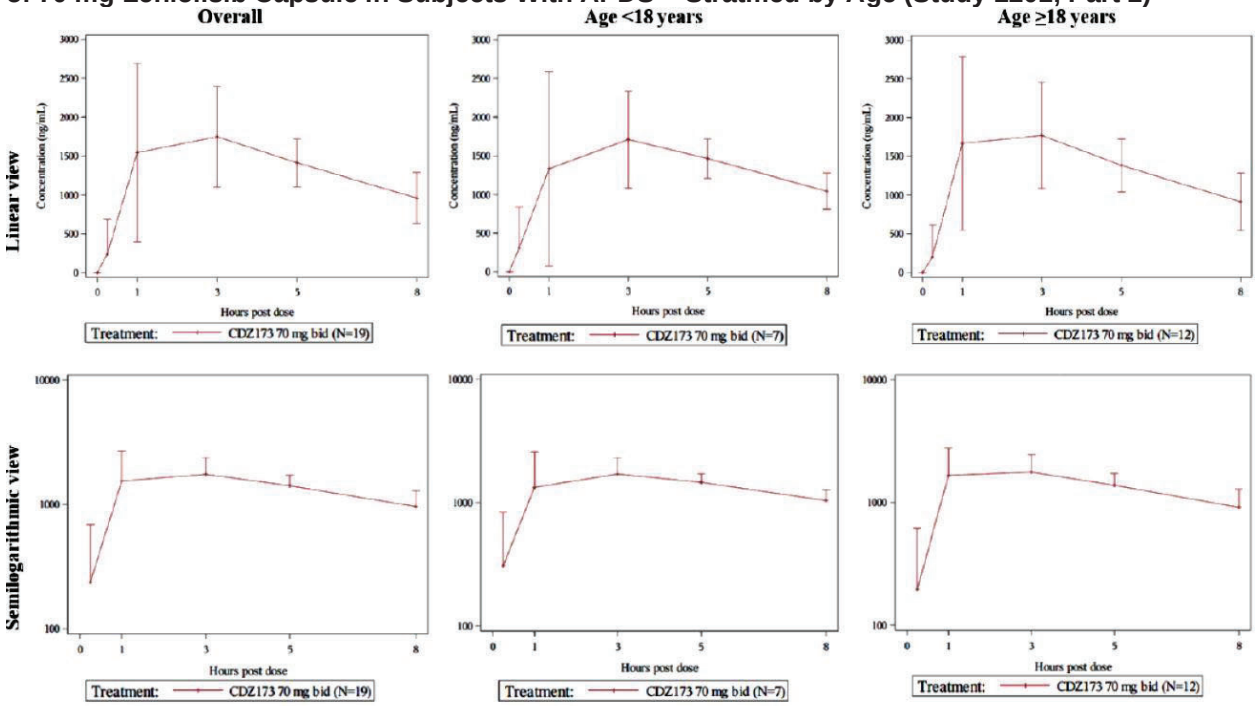


Source: Study 2201 Part 1 CSR, Figure 14.2-1.3.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Leniolisib Pharmacokinetics in Part 2 (Fixed Dose)

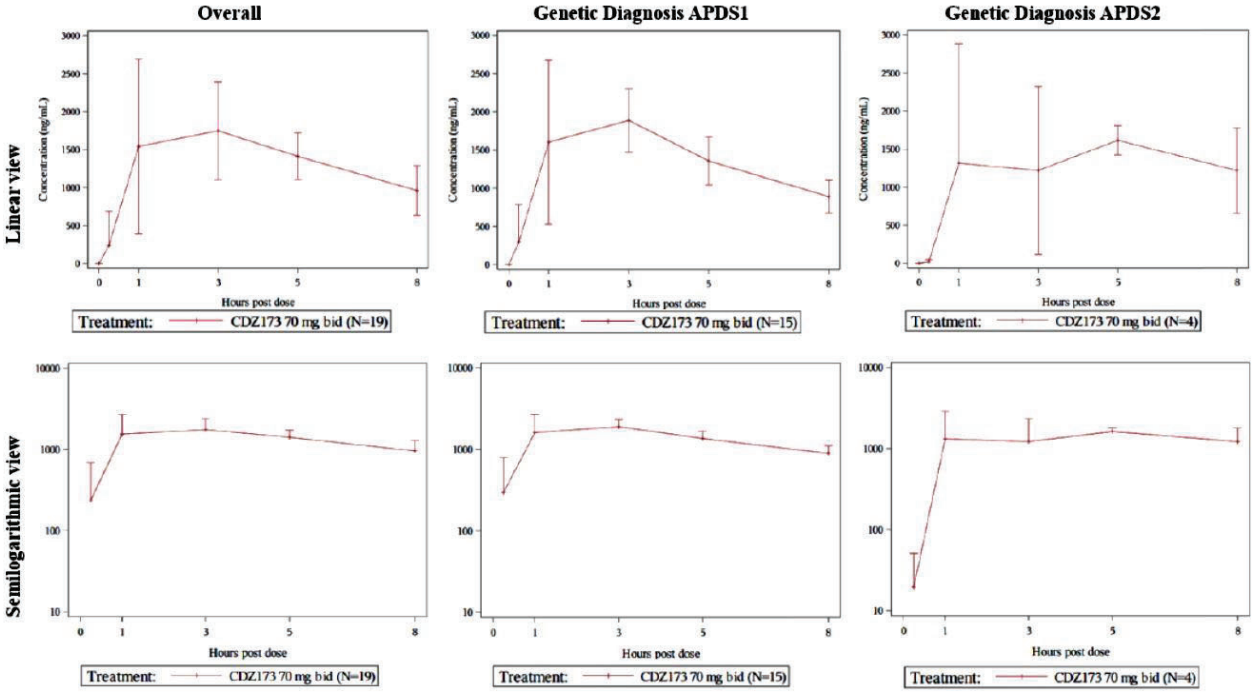
In Part 2 of the study, leniolisib plasma PK profiles appeared comparable between age groups (<18 years versus ≥18 years; Figure 22) and genetic diagnoses (APDS1 versus APDS2; Figure 23) following the first dose of twice daily leniolisib 70 mg in subjects with APDS. On Day 1 of dosing, geometric mean leniolisib plasma AUC_{0-8} and C_{max} were 10400 ng*h/mL, 2080 ng/mL for the overall group; 10200 ng*h/mL, 2070 ng/mL for <18-year-old subjects; and 10600 ng*h/mL, 2090 ng/mL for ≥18-year-old subjects (Table 71). Stratified by genetic diagnosis, Day 1 geometric mean AUC_{0-8} and C_{max} were 10500 ng*h/mL, 2060 ng/mL in subjects with APDS1 and 10300 ng*h/mL, 2180 ng/mL in subjects with APDS2. Sparse PK sampling was performed on Days 29, 57, and 85 of treatment in Part 2 of the study (Figure 24). Geometric mean (geometric CV%) leniolisib plasma C_{trough} at these time points were 926 (73.2%), 844 (75.7%), and 804 (67.5%) ng/mL, respectively, in the overall group.

Figure 22. Mean (SD) Leniolisib Plasma Concentration-Time Profile on Day 1 of Twice Daily Dosing of 70 mg Leniolisib Capsule in Subjects With APDS – Stratified by Age (Study 2201, Part 2)



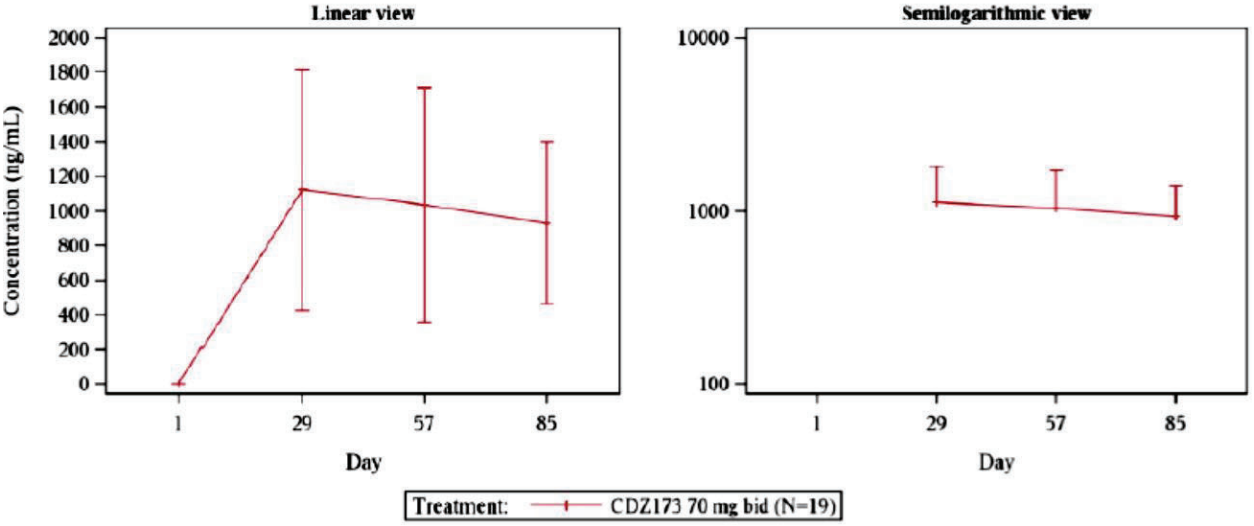
Source: Study 2201 Part 2 CSR, Figure 14.2-1.1.1b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Figure 23. Mean (SD) Leniolisib Plasma Concentration-Time Profile on Day 1 of Twice Daily Dosing With 70 mg Leniolisib Capsule in Subjects With APDS – Stratified by Genetic Diagnosis (Study 2201, Part 2)



Source: Study 2201 Part 2 CSR, Figure 14.2-1.1.2b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; APDS 1, activated phosphoinositide 3-kinase delta syndrome 1; APDS 2, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Figure 24. Mean (SD) Leniolisib Plasma Trough Concentration-Time Profile Following Twice Daily Dosing of 70 mg Leniolisib Capsule in Subjects With APDS (Study 2201, Part 2)



Source: Study 2201 Part 2 CSR, Figure 14.2-1.3.1b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CSR, clinical study report; N, total number of subjects; SD, standard deviation.

Table 71. Summary of Plasma Leniolisib Pharmacokinetic Parameters Following the First Dose of 70 mg Leniolisib Capsule in Subjects With APDS – Stratified by Age Group (Study 2201, Part 2)

Age Group Visit	n	Geometric Mean (Geometric CV%)[a]			
		C_{max} (ng/mL)	T_{max} (h)	AUC_{0-t} (ng*h/mL)	AUC_{0-8} (ng*h/mL)
Overall					
Day 1	19[b]	2080 (25.4)	2.87 [0.92, 7.78]	10100 (27.6)	10400 (26.3)
<18-year-old					
Day 1	7	2070 (25.4)	3.00 [1.00, 5.05]	9890 (31.1)	10200 (28.4)
≥18-year-old					
Day 1	12[c]	2090 (26.6)	1.16 [0.92, 7.78]	10100 (26.9)	10600 (26.3)

Source: Study 2201 Part 2 CSR, Table 14.2-1.2.1.1b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{0-t}, area under the curve to the last quantifiable time point; AUC_{0-8}, area under the curve from 0 to 8 hours; C_{max}, maximum plasma concentration; CSR, clinical study report; CV, coefficient of variation; n, number of subjects in category; T_{max}, time to maximum observed plasma concentration.

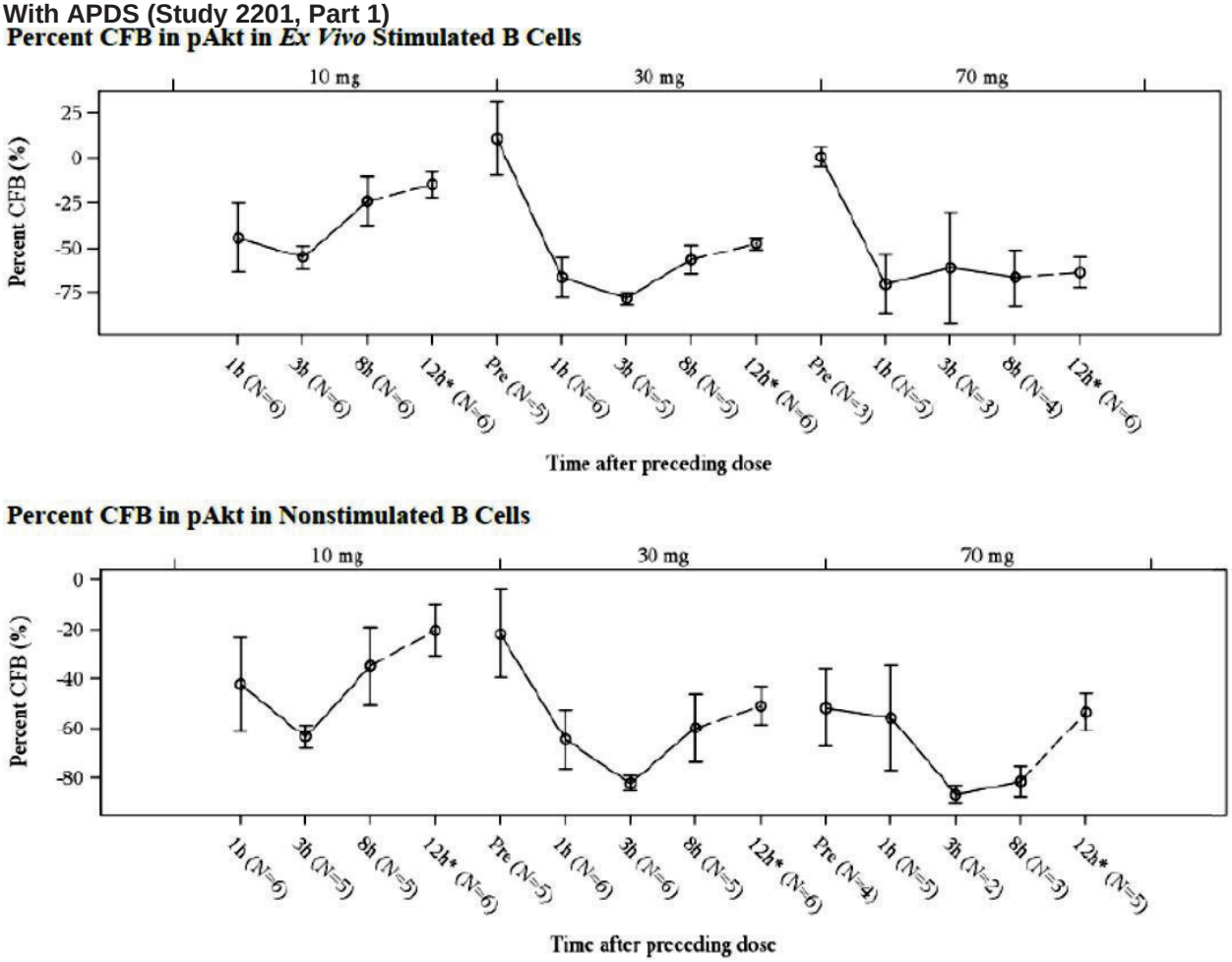
Pharmacodynamic Results

Percent Change From Baseline in pAkt-Positive B Cells

In Part 1 of the study, the PD effect of leniolisib was measured using %pAkt-positive B cells as read out; Akt is a direct downstream target of activated PI3Kδ. A clear and apparent maximum reduction in pAkt-positive B cells was observed as early as 1-hour post-dose for most of the subjects with APDS (Figure 25). The early onset of pAkt inhibition occurred at approximately the same time as plasma leniolisib T_{max} (Table 69) suggesting a direct PD response. Furthermore, %pAkt inhibition in B cells and leniolisib plasma concentration were well fitted to an E_{max} model (Figure 26).

Although a single dose of 10 mg generally reduced %pAkt-positive B cells to less than 20%, the inhibition generally waned over the ensuing 12 hours. In contrast, a slightly more pronounced peak drug effect and sustained inhibition was observed at the higher doses (Figure 25). While the 30- and 70-mg dose levels did not differ in their maximum effect, inhibition was clearly more sustained over time at 70 mg, which may therefore be most optimal in terms of time-averaged inhibition and inhibition at trough (12 hours post-dose).

Figure 25. Mean (SE) % Change From Baseline in %pAkt-Positive B Cells Over Time in Subjects With APDS (Study 2201, Part 1)

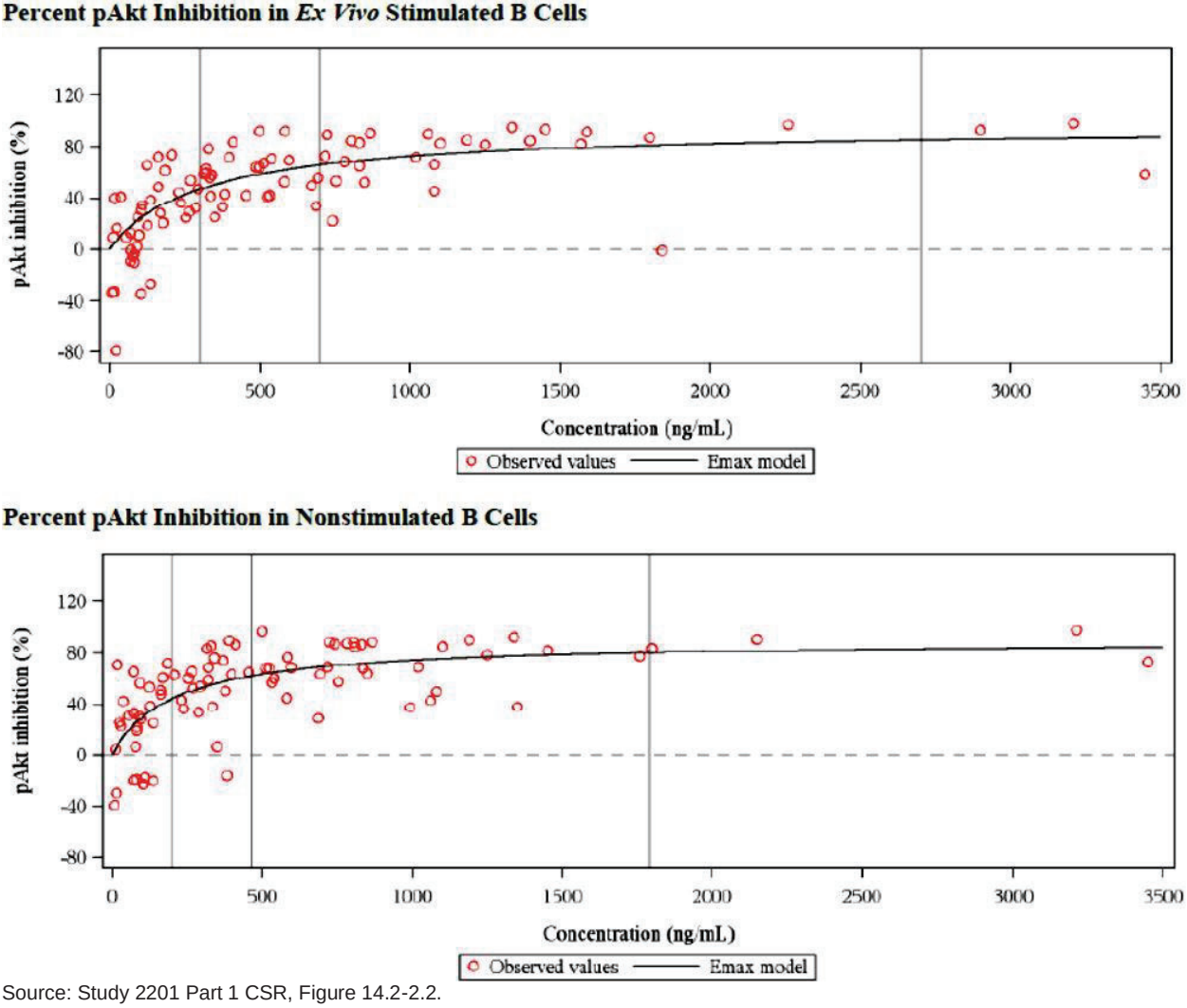


Source: Study 2201 Part 1 CSR, Figure 14.2-2.1.2.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; CFB, change from baseline; CSR, clinical study report; pAkt, phosphorylated protein kinase B; N, total number of subjects; SE, standard error.

Concentration-Response Analysis of Percent pAkt Inhibition in B Cells:

Percent pAkt inhibition in B cells versus leniolisib plasma concentration was well fitted to an E_{max} model (Figure 26). Similar model parameters for EC_{50} and E_{max} were obtained for %pAkt inhibition in both ex vivo stimulated and nonstimulated B cells (Table 72), suggesting that apart from increasing the overall signal, ex vivo stimulation may be irrelevant for the determination of model parameters related to the potency and maximum effect of leniolisib. Based on the E_{max} model, treatment with leniolisib 10, 30, and 70 mg twice daily is expected to result in time-averaged pAkt inhibition in B cells of approximately 38%, 63%, and 78%, respectively, and time-averaged pAkt inhibition at 50% (ED_{50}), 70% (ED_{70}), and 90% (ED_{90}) of E_{max} are expected with leniolisib doses of 15, 35, and 136 mg twice daily, respectively, in ex vivo stimulated B cells (Table 72).

Figure 26. Scatterplot of %pAkt Inhibition in B Cells Versus Leniolisib Plasma Concentration in Subjects With APDS (Study 2201, Part 1)



Source: Study 2201 Part 1 CSR, Figure 14.2-2.2.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; CSR, clinical study report; E_{max}, maximal effect; pAkt, phosphorylated protein kinase B.

Table 72. E_{max} Model of Relationship Between Leniolisib Plasma Concentration and %pAkt Inhibition in B Cells in Subjects With APDS (Study 2201, Part 1)

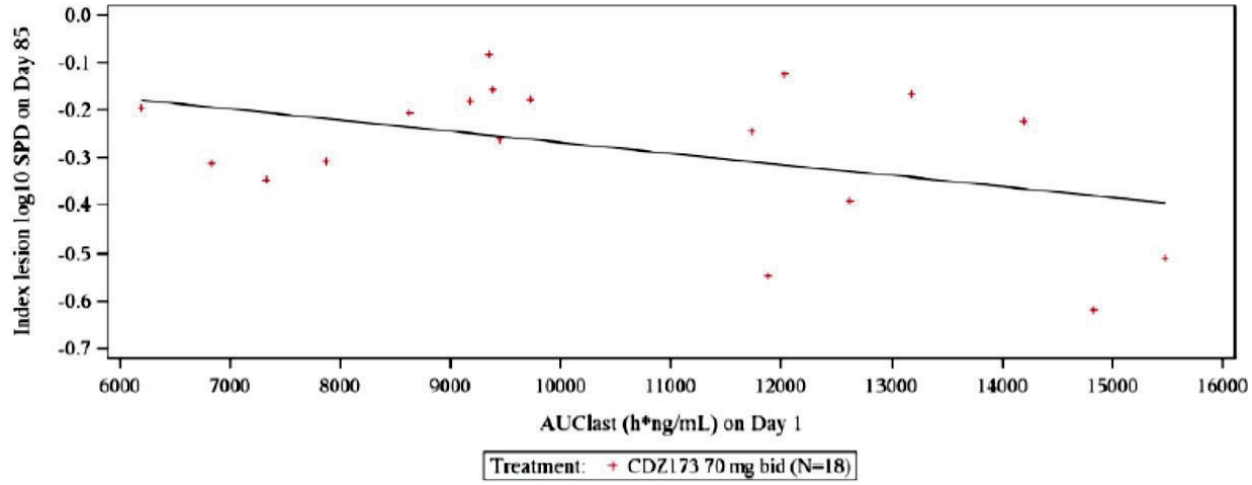
Parameter	%pAkt Inhibition in *Ex Vivo* Stimulated B Cells		%pAkt Inhibition in Nonstimulated B Cells	
	Estimate (SE)	2-sided 95% CI	Estimate (SE)	2-sided 95% CI
E_{max} (%)	94.60 (5.80)	78.49, 110.72	88.93 (7.57)	69.47, 108.39
EC_{50} (ng/mL)	300.32 (55.68)	145.72, 454.92	199.19 (68.36)	23.45, 374.92
EC_{70} (ng/mL)	700.75 (129.93)	340.02, 1061.49	464.77 (159.51)	54.72, 874.81
EC_{90} (ng/mL)	2702.91 (501.14)	1311.51, 4094.30	1792.67 (615.27)	211.08, 3374.26
ED_{50} (mg)	15.06 (2.79)	7.31, 22.82	9.99 (3.43)	1.18, 18.81
ED_{70} (mg)	35.15 (6.52)	17.06, 53.24	23.31 (8.00)	2.75, 43.88
ED_{90} (mg)	135.58 (25.14)	65.79, 205.37	89.92 (30.86)	10.59, 169.25
Time-averaged %pAkt inhibition at 10 mg bid (%)	37.74 (2.23)	31.57, 43.92	44.48 (5.50)	30.35, 58.62
Time-averaged %pAkt inhibition at 30 mg bid (%)	62.98 (1.46)	58.92, 67.04	66.71 (4.17)	55.99, 77.43
Time-averaged %pAkt inhibition at 70 mg bid (%)	77.85 (2.58)	70.69, 85.01	77.82 (4.76)	65.59, 90.05

Source: Study 2201 Part 1 CSR, Table 14.2-2.2.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CI, confidence interval; CSR, clinical study report; EC_{50}, effect concentration 50; EC_{70}, effect concentration 70; EC_{90}, effect concentration 90; ED_{50}, effective dose 50; ED_{70}, effective dose 70; ED_{90}; effective dose 90; E_{max}, maximal effect; pAkt, phosphorylated protein kinase B; SE, standard error.

PK/PD Analysis of Effect on Lymphoproliferation and Immunophenotype Normalization

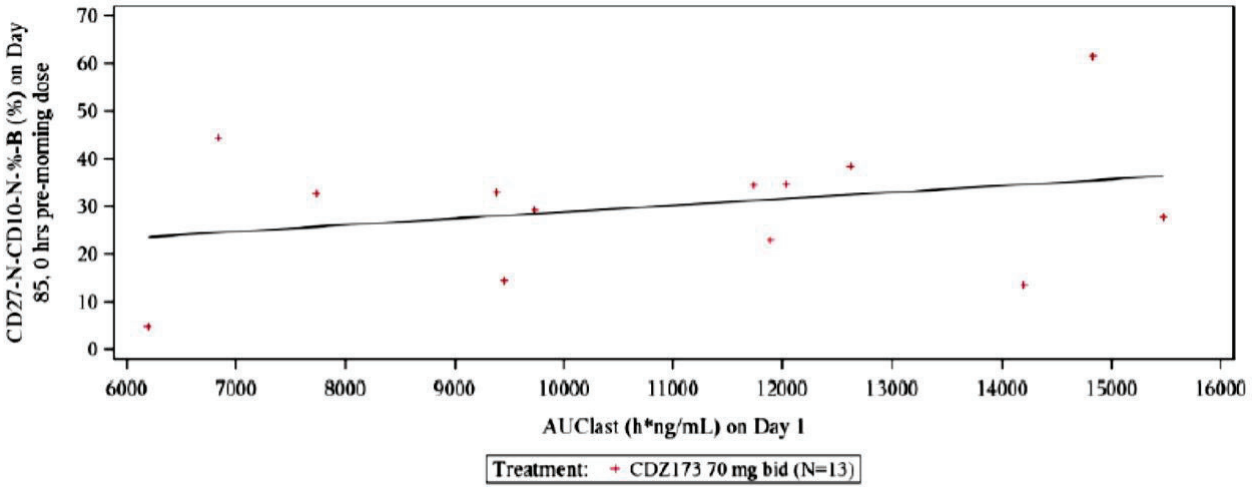
In Part 2 of the study, the effect of 70 mg leniolisib twice daily on lymphoproliferation and immunophenotype normalization in subjects with APDS was assessed by the primary efficacy endpoints, $\log_{10}$ SPD of index lesions and % naive B cells. Scatterplots of the change from baseline in primary efficacy endpoints at Day 85 of treatment against leniolisib plasma AUC_{0-t} on Day 1 show slight trends for greater reduction in $\log_{10}$ SPD of index lesions (Figure 27) and increase in % naive B cells (Figure 28) at higher AUC_{0-t} values.

Figure 27. Scatterplot of %pAkt Inhibition in B Cells Versus Leniolisib Plasma Concentration in Subjects With APDS (Study 2201, Part 1)



Source: Study 2201 Part 2 CSR, Figure 14.2-13.1.1b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{last}, area under the curve to the last quantifiable time point; BID, twice daily; CSR, clinical study report; pAkt, phosphorylated protein kinase B; SPD, sum of product of diameters.\

Figure 28. Scatterplot of %pAkt Inhibition in B Cells Versus Leniolisib Plasma Concentration in Subjects With APDS (Study 2201, Part 1)



Source: Study 2201 Part 2 CSR, Figure 14.2-13.1.1b.
Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{last}, area under the curve to the last quantifiable time point; BID, twice daily; CSR, clinical study report; pAkt, phosphorylated protein kinase B.

Reviewer Comment: *In subjects with APDS, steady state of leniolisib was achieved at 1 week from onset of therapy. Steady-state in subjects with APDS was also likely achieved within 2 to 3 days of twice daily dosing but not observed in trough concentration-time profiles because there were no PK sampling timepoints from Days 2 through 7 of treatment in Study 2201. In subjects with APDS, the median T_{max} following a single dose of 10 to 70 mg leniolisib ranged from approximately 2 to 3 hours post-dose. PK of leniolisib were similar between healthy participants and APDS subjects. Dose proportionality was observed for leniolisib exposure over the entire*

dose range investigated (10, 30, and 70 mg twice daily). In Part 2 of the study, leniolisib plasma PK profiles appeared comparable between age groups (<18 years versus ≥18 years). PK of leniolisib was similar between subjects with genetic diagnosis APDS1 and APDS2.

A clear and apparent maximum reduction in pAkt-positive B cells was observed as early as 1-hour post-dose. The early onset of pAkt inhibition occurred at approximately the same time as plasma leniolisib T_{max}. The pAkt inhibition started to rebound after T_{max} (1 hr) and returned to baseline level after 12 hours thereby providing support for the proposed BID dosing regimen. Based on the E_{max} model, treatment with leniolisib 10, 30, and 70 mg twice daily is expected to result in time-averaged pAkt inhibition in B cells of approximately 38%, 63%, and 78%, respectively, with an EC_{50} of 300 ng/mL, and time-averaged pAkt inhibition at 50% (ED_{50}), 70% (ED_{70}), and 90% (ED_{90}) of E_{max} are expected with leniolisib doses of 15, 35, and 136 mg twice daily, respectively, in ex vivo stimulated B cells. Although a single dose of 10 mg generally reduced %pAkt-positive B cells to less than 20%, the inhibition generally waned over the ensuing 12 hours. While the 30- and 70-mg dose levels did not differ in their maximum effect, inhibition was more sustained over time at 70 mg, which may therefore be most optimal in terms of time-averaged inhibition and inhibition at trough (12 hours post-dose).

Overall, based on the PK/PD relationship and on the E_{max} concentration-response model, while both 30 and 70 mg may achieve near complete pathway inhibition shortly post-dose, 70 mg BID is expected to confer a more sustained and higher time-averaged inhibition relative to lower doses which provides support for the recommended dose of 70 mg BID of leniolisib from a Clinical Pharmacology perspective.

16.3.3.7. Study 2201E1

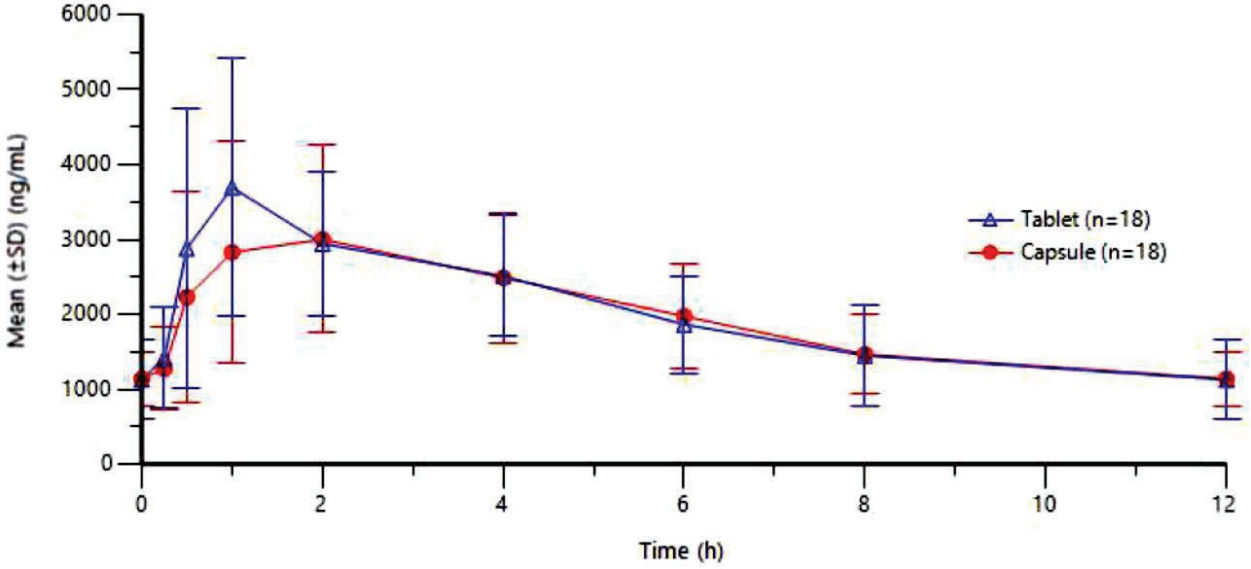
- Multiple-dose PK and relative bioavailability between capsule and tablet formulations in subjects with APDS

Study 2201E1 is an ongoing Phase 2/3, multicenter, multinational, open-label, nonrandomized, active-treatment extension study characterizing the PK of leniolisib and evaluating the relative bioavailability of Novartis leniolisib film-coated tablets (Novartis FCT) compared to Novartis HGC (b) (4) in subjects with APDS who participated in Study 2201 or who were previously treated with PI3Kδ inhibitors other than leniolisib. A total of 35 subjects received oral leniolisib 70 mg twice daily initially as capsule (1 × 70 mg capsule) then as tablet (1 × 70 mg tablet) formulation through the end of treatment period lasting up to a maximum of 6 years. Study drugs (Novartis HGC and Novartis FCT) used were manufactured by Novartis Pharma AG. Treatment was given in the fasted state or 0.5 hours after a light breakfast for morning doses when serial PK sampling was performed and irrespective of food for all other doses. Two subjects from Russia remained on the Novartis HGC formulation per feedback from the Russian Health Authority and were thus excluded from the PK assessment in this study. All other subjects transitioned from the capsule to tablet formulation as described above.

Study 2201E1 was initially planned to include assessment of relative bioavailability between Novartis capsule (Novartis HGC (b) (4)) and tablet (Novartis FCT) formulations to support a change in formulation in lieu of a classical bioequivalence study. However, the study design and PK sampling scheme from this study would not sufficiently support the bridging between the two formulations after the reclassification of leniolisib to a Biopharmaceutics Classification System (BCS) class 2 (low solubility, high permeability) compound (b) (4) necessitated comparison of exposure through a formal bioequivalence study. Hence, a dedicated clinical study, Study LE1101, assessing the bioequivalence between 70 mg leniolisib as the formulation used in the pivotal Phase 2/3 study (Novartis HGC (b) (4)) and the intended to-be-marketed formulation (Pharming FCT) in healthy participants was performed instead.

As a response to the Information Request, the Applicant submitted the PK data from Study 2201E1 as an addendum to the CSR on November 8, 2022. Figure 29 shows the mean plasma concentration-time profiles and Table 73 shows the PK parameters for both formulations at steady-state. Only subjects with paired observations (a PK profile for both capsule and tablet) were included in this analysis. The mean concentration-time profiles for both formulations were similar except for a higher C_{max} for the tablet formulation, with T_{max} also reached earlier.

Figure 29. Leniolisib Mean (SD) Plasma Concentration-Time Profiles After Administration of Leniolisib 70 mg BID Formulated as Film-Coated Tablet or Hard Gelatin Capsule



Source: Addendum to the Study CCDZ173X2201E1 CSR, Figure 1.
Abbreviations: BID, twice daily; CSR, clinical study report; n, number of subjects in category; SD, standard deviation.

Table 73. Leniolisib PK Parameters After Administration of Leniolisib 70 mg BID Formulated as Film-Coated Tablet or Hard Gelatin Capsule

	Tablet (N=18)	Capsule (N=18)
	Mean ± SD	Mean ± SD
C_{max} (ng/mL)	4156 ± 1538	3482 ± 1164
t_{max} (h)	1.00 (0.47 - 3.96)	1.93 (0.00 - 4.02)
AUC_{0-12h} (ng.h/mL)	23873 ± 7754	23220 ± 7756

Note: Median, min-max are presented for t_{max}.
Source: Addendum to the Study CCDZ173X2201E1 CSR, Table 1.
Abbreviations: AUC_{0-12h}, area under the curve from 0 to 12 hours; BID, twice daily; C_{max}, maximum plasma concentration; CSR, clinical study report; N, total number of subjects; PK, pharmacokinetic; SD, standard deviation; T_{max}, time to maximum observed plasma concentration.

When comparing leniolisib tablet (test) with leniolisib capsule (reference), the 90% confidence intervals (Cis) of the least squares (LS) means ratio for AUC_{0-12} (estimate [90% CI]: 103.39% [85.83% – 124.53%]) were contained within the standard bioequivalence range (i.e., 80.00% - 125.00%), while for C_{max} (estimate [90% CI]: 119.42% [97.63% – 146.08%]) this criterion was not met (Table 74).

Table 74. Assessment of the Relative Bioavailability of Leniolisib After Administration of Leniolisib 70 mg BID Formulated as Film-Coated Tablet Versus Hard Gelatin Capsule

	LSMeans		LSM Ratio	
	Tablet (Test)	Capsule (Reference)	Estimate (%)	90% CI (%)
N	18	18		
C_{max} (ng/mL)	3928	3288	119.42	97.63 - 146.08
AUC_{0-12h} (ng.h/mL)	22758	22013	103.39	85.83 - 124.53

Note: Evaluation 1: all exclusions listed.
Source: Addendum to the Study CCDZ173X2201E1 CSR, Table 2.
Abbreviations: AUC_{0-12h}, area under the curve from 0 to 12 hours; BID, twice daily; CI, confidence interval; C_{max}, maximum plasma concentration; CSR, clinical study report; LS, least squares; LSM, least squares means; N, total number of subjects.

Overall, the results demonstrated that the total drug exposure at steady-state (as determined by AUC_{0-12}) was comparable between the capsule and tablet formulation. The point estimate of the LS mean ratio was close to 100%, and the 90% CI fell within the bio-equivalence limits of 80-125%. In addition, leniolisib peak concentrations at steady-state (as determined by C_{max}) was only slightly higher for the tablet formulation as the least squares mean ratios for tablet versus capsule ranged between 110% and 120%.

To further understand the effect of food on the PK of leniolisib using the to-be-marketed formulation, and Information Request was sent to the Applicant to provide information on whether the subjects in Study 2201E1 routinely took the drug with or without food. The Applicant was asked to conduct a brief subject survey in the PK population to obtain information about administration of drug with regards to food in this study if such information

was not available. The Applicant provided the results of this food intake survey from Study 2201E1 and data from 28 subjects were included in the survey results.

- 12 subjects selected, "I never took food with study medication."
  - 6 of the 12 subjects who stated they did not take the medication with food also indicated a time before/after eating. These subjects may be most appropriately categorized as taking the medication with food but not eating and taking the medication at the same time.
  - 6 subjects' replies are consistent (they did not indicate a time for the before/after eating).
- 22 subjects (including 6 mentioned above) responded that they did take food with the medicine.
  - 1 subject reported that they took the medicine while eating.
  - There was an approximately equal distribution of subjects indicating taking the medicine before eating and after eating.

Overall, the results of the survey demonstrate that the majority of the subjects took leniolisib with food in Study 2201E1 where both capsule and tablet formulations were administered to the subjects.

An explorative retrospective analysis of the data evaluating the effects of concomitant PPI use on leniolisib PK was performed. Mean plasma leniolisib concentration-time profiles appeared nearly comparable between APDS subjects with reported concomitant PPI use and those without for both Novartis HGC and FCT formulations (Figure 9, Table 75). Thus, concomitant use of PPIs did not have a marked effect on leniolisib exposure requiring dose adjustment.

Table 75. Summary of Statistical Assessment for Effect of PPI Use on Leniolisib Exposure Following Twice Daily Dosing of 70 mg Leniolisib in Subjects With APDS, Stratified by Formulation (Study 2201E1)

Formulation Parameter	Treatment	n	LS Mean	Treatment Comparison	
				LS Mean Ratio (%)	90% Confidence Interval
Novartis hard gelatin capsule formulation					
C_{max} (ng/mL)	PPI use reported (test)	11	3260	101.93	80.54, 129.00
	No PPI use reported (reference)	19	3198		
AUC_{0-8} (ng*h/mL)	PPI use reported (test)	10	16478	98.25	77.22, 125.00
	No PPI use reported (reference)	19	16945		
Novartis film-coated tablet formulation					
C_{max} (ng/mL)	PPI use reported (test)	9	3628	109.07	88.61, 134.25
	No PPI use reported (reference)	18	3327		
AUC_{0-8} (ng*h/mL)	PPI use reported (test)	9	17033	104.03	84.59, 127.93
	No PPI use reported (reference)	17	16373		

Source: Summary of Clinical Pharmacology Studies, Table 16.
Abbreviations: Abbreviations: APDS, activated phosphoinositide 3-kinase delta syndrome; AUC_{0-8}, area under the curve from 0 to 8 hours; CI, confidence interval; C_{max}, maximum plasma concentration; LS, least squares; n, number of subjects in category; PPI, protein pump inh bitor.

Reviewer Comment: *Overall, the results demonstrated that the total drug exposure at steady-state was comparable between the capsule and tablet formulation in the extension study. Mean plasma leniolisib concentration-time profiles appeared nearly comparable between APDS subjects with reported concomitant PPI use and those without for both Novartis HGC and FCT formulations. Thus, concomitant use of PPIs did not have a marked effect on leniolisib exposure requiring dose adjustment. The results of the food intake survey conducted by the Applicant showed that the majority of the subjects took leniolisib with food in Study 2201E1 where both capsule and tablet formulations were administered to the subjects, providing evidence support for the administration of the to-be-marketed tablet formulation of leniolisib regardless of food.*

16.3.4. Physiologically Based Pharmacokinetic Modeling Review

Division of Pharmacometrics, Office of Clinical Pharmacology

NDA Number	217759
Generic Name	Leniolisib
Trade Name	JOENJA®
Submission Type	505(b)(1)
Applicant	Pharming Inc.
Dosage Form and Strengths	Oral tablets, 70 mg
Proposed Indication	Indicated for the treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adults and adolescents aged 12 or older.

Proposed Dose Regimen	70 mg orally twice daily approximately 12 hours apart, with or without food
Primary PBPK Reviewer	Jianghong Fan, Ph.D.
Secondary PBPK Reviewer	Manuela Grimstein, Ph.D.

16.3.5. Executive Summary

The objective of this review is to evaluate the adequacy of the Applicant's following PBPK reports to support the intended uses.

- 1600092: Predictions of CDZ173 (leniolisib) pharmacokinetics and drug-drug interactions with typical CYP perpetrators using Simcyp V15
- 1500108: Simcyp prediction of the drug interaction effect of CDZ173 (leniolisib) on the pharmacokinetics of midazolam

The Division of Pharmacometrics has reviewed the PBPK reports, supporting modeling files, and the Applicant's response to FDA's information request (IR) submitted on Nov. 29, 2022, and concluded the following:

- The leniolisib PBPK model is adequate to predict the PK of leniolisib following a single oral dose administration (10, 40, 80, 110, 200 and 400 mg), or multiple oral dose administration (20, 40, 70 and 140 mg BID) in healthy subjects.
- The leniolisib PBPK model is inadequate to accurately predict the magnitude of effect of erythromycin (a moderate CYP3A4 inhibitor) on leniolisib PK following multiple dose administration of leniolisib (70 mg BID) in healthy subjects. However, the modeling analysis, along with mass balance study results, indicated that coadministration of multiple doses of erythromycin (300 mg BID) is expected to increase steady-state exposure (70 mg BID) of leniolisib by less than 75%.
- The leniolisib PBPK model is inadequate to accurately predict the magnitude of effect of rifampin (a strong CYP3A4 inducer) or efavirenz (a moderate CYP3A4 inducer) on leniolisib PK following multiple dose administration of leniolisib (70 mg BID) in healthy subjects. However, the modeling analysis along with mass balance study results, indicated that coadministration of multiple doses of rifampin (600 mg QD) or efavirenz (600 mg QD) is expected to decrease steady-state exposure (70 mg BID) of leniolisib by less than 78% or 58%, respectively.
- The leniolisib PBPK model is inadequate to accurately predict the magnitude of effect of leniolisib on midazolam PK following multiple dose administration of leniolisib (70 mg BID) in healthy subjects. (b) (4) Due to the uncertainty associated with the in vitro to in vivo extrapolation of enzyme induction parameter values, the219 current model predicted effect of leniolisib on midazolam PK should be interpreted cautiously.

16.3.5.1. Applicant's PBPK Modeling Effort

PBPK Software

Simcyp V15 (Simcyp Ltd, UK) was used to develop the PBPK models and predict the effects of itraconazole, erythromycin, rifampin and efavirenz on the PK of leniolisib and the effects of leniolisib on the PK of midazolam in healthy subjects. The reviewer used Simcyp V21 for analyses.

16.3.5.1.1. Model Development

(b) (4)

In vitro phenotyping study 1200021 suggested that hepatic metabolism of leniolisib was primarily mediated by CYP3A4 with minimal contribution from other enzymes (3.5%, 0.7%, and 0.4% from CYP3A5, CYP1A2, and CYP2D6, respectively). CYP1A1 was involved in the extrahepatic biotransformation. The contribution of CYP3A4 to the overall metabolism of leniolisib (fmCYP3A4=0.60) was refined based on the clinical DDI study with itraconazole (CDZ173X2102). The contribution of CYP1A1 to the overall metabolism of leniolisib (fmCYP1A1) was estimated to be about 0.1-0.29 based on the information of the metabolite (M1) formed by CYP1A1 in mass balance study (Study 1500425).

A non-pathway specific additional hepatic clearance in HLM (fmCLadditional=0.30) was assigned in the Applicant's PBPK model to account for CYP1A1 mediated metabolism of leniolisib and biliary clearance. Biliary clearance may occur to a limited extend based on the amount of parent drug in bile (7% of i.v. dose) in the bile duct cannulated rats (DMPK R1100673) and the parent drug in the bile may also undergo reabsorption due to its high permeability.

Following oral administration (400 mg), the unchanged parent drug accounted for 6.32% and 2.94% of the dose administered in urine (23% of total administered radioactivity) and feces

(42% of total administered radioactivity), respectively (Study 1500425). Renal clearance of 0.2 L/h was obtained from the clinical PK study CDZ173X2101.

Leniolisib exhibited linear PK and has dose-proportional exposures across the dose range of 10 to 400 mg following a single dose administration and 20 to 140 mg following multiple doses administration (BID) (study CDZ173X2101).

In vitro study indicated that leniolisib was an inducer of CYP3A4 (DMPK R1300388) and no significant inhibition on CYP3A was observed (DMPK R1100170). The CYP3A4 induction parameter value assigned in the model were based on CYP3A4 activity induction data and calibrated against the induction parameter value for positive control rifampicin.

Victim Drug Models

The default PBPK models of itraconazole, erythromycin, rifampin, efavirenz and midazolam in Simcyp (V15) were used for DDI predictions. The reviewer used Simcyp V21 for analyses.

FDA's Assessment

1. Simcyp V15 was used in the Applicant's model prediction, while Simcyp V21 was used in the FDA reviewer's analysis. The reviewer's analysis showed that the predicted AUC and C_{max} values using Simcyp V21 were within 1.20-fold of the clinically observed data following a single dose administration of leniolisib (Table 76). The predictive performance of leniolisib model appears to be improved using Simcyp V21 as compared to those predicted values using Simcyp V15 (Table 76). In addition, the leniolisib PK profiles were better captured using Simcyp V21 verse Simcyp 15. Therefore, the model predicted data using Simcyp V21 were presented in this PBPK review with respective to the model validation and DDI evaluation.

Table 76. Comparison of Observed and Simulated Leniolisib C_{max} and AUC_{inf} Values Using the Model in Simcyp V15 and V21 Following a Single Dose Administration of Leniolisib in Healthy Subjects

	C_{max} (ng/mL)			AUC_{inf} (ng*h/mL)		
Dose	**Predicted**	**Observed**	**Pred./Obs.**	**Predicted**	**Observed**	**Pred./Obs.**
Predicted result using Simcyp V21*						
10 mg	392	420	0.93	2653	2550	1.04
40 mg	1570	1540	1.02	10606	9100	1.17
80 mg	3141	3610	0.87	21199	25400	0.83
110 mg	4319	4200	1.03	29135	29400	0.99
200 mg	7851	7270	1.08	52896	47900	1.10
400 mg	15700	14300	1.10	105466	113000	0.93

Dose	C_{max} (ng/mL) Predicted	Observed	Pred./Obs.	AUC_{inf} (ng*h/mL) Predicted	Observed	Pred./Obs.
Predicted result using Simcyp V15						
10 mg	442	420	1.05	3113	2550	1.22
40 mg	1769	1540	1.15	12553	9100	1.38
80 mg	3538	3610	0.98	25086	25400	0.99
110 mg	4864	4200	1.16	34473	29400	1.17
200 mg	8843	7270	1.22	62566	47900	1.31
400 mg	17681	14300	1.24	124648	113000	1.10

Sources: PBPK report 1600092, Table 6-3, *FDA reviewer's analysis.

Abbreviations: AUC_{inf}, area under the curve to infinity; C_{max}, maximum plasma concentration, Pred., predicted; Obs., observed.

2. The in vitro study report 1200021 indicated that CYP1A1 is one of the enzymes involved in the metabolism of leniolisib and the fmCYP1A1 value derived from the Applicant's PBPK report was about (b) (4). Literature report showed that itraconazole can induce CYP1A1 activity through the aryl hydrocarbon receptor (AhR)-dependent mechanism, and itraconazole was also a strong competitive inhibitor for CYP1A1 in vitro (Korashy et al. 2007). However, the impact of itraconazole on CYP1A1 activity was not evaluated or discussed in the Applicant's PBPK report. Therefore, there was a concern on the fmCYP3A4 value assigned in the model that was optimized based on a single DDI study with itraconazole. An information request was issued requesting the Applicant to provide adequate justification (e.g., in house data or literature information such as the DDI of other CYP1A1 substrates) to support the fmCYP3A4 estimation.

Applicant's Response to FDA's IR and FDA's Assessment

(b) (4)

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16.3.5.1.2. Model Application

The developed PBPK model was used to simulate the DDI for leniolisib in the following scenarios:

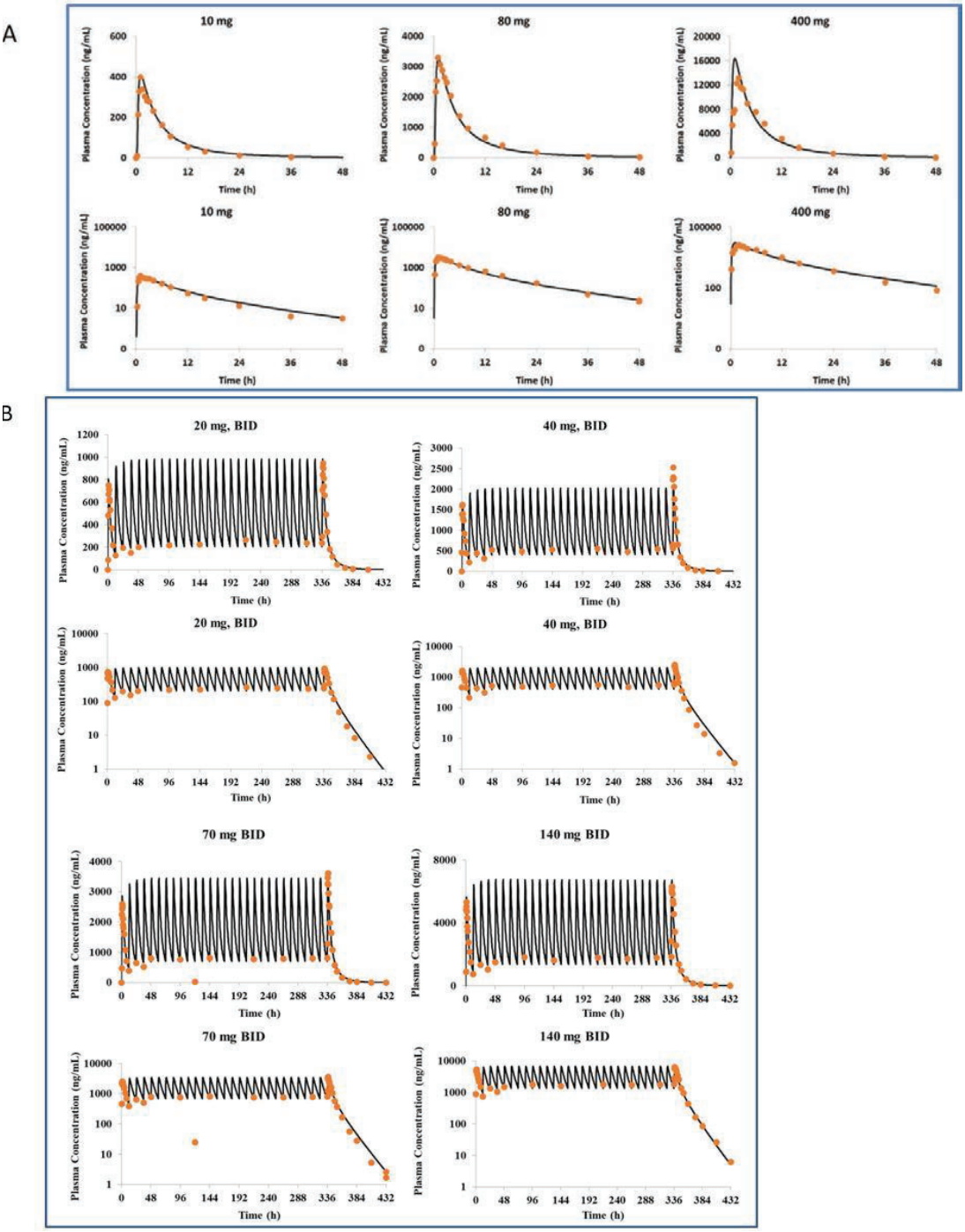
- To predict the effect of erythromycin (a moderate CYP3A inhibitor) on the PK of leniolisib following multiple dose administration of leniolisib (70 mg BID) in healthy subjects.
- To predict the effect of rifampin (a strong CYP3A inducer) and efavirenz (a moderate CYP3A4 inducer) on the PK of leniolisib following multiple dose administration of leniolisib (70 mg BID) in healthy subjects.
- To predict the effect of leniolisib on the PK of midazolam (a sensitive CYP3A4 substrate) following multiple dose administration of leniolisib (70 mg BID) in healthy subjects.

16.3.5.1.3. Results

Can Leniolisib PBPK Model Describe Leniolisib PK in Healthy Subjects?

Yes. The leniolisib model was able to capture the observed leniolisib PK profiles following a single oral dose administration (10, 40, 80, 110, 200 and 400 mg), or multiple oral dose administration (20, 40, 70 and 140 mg BID) in healthy subjects (Figure 30 and Table 78).

Figure 30. Observed (Dots) and Simulated (Lines) Leniolisib Plasma Concentration-Time Profiles Following a Single Dose (A) or Multiple Dose (B) Administration of Leniolisib in Healthy Subjects



Source: Observed data were from study CDZ173X2101; predicted data were from FDA reviewer's analysis using Simcyp V21.
Abbreviations: BID, twice daily.

Table 78. Simulated and Observed Leniolisib C_{max} and AUC Values Following a Single Dose or Multiple Dose Administration of Leniolisib in Healthy Subjects

	C_{max} (ng/mL)			AUC (ng*h/mL)		
Dose	**Predicted**	**Observed**	**Pred./Obs.**	**Predicted**	**Observed**	**Pred./Obs.**
10 mg, SD	392	420	0.93	2653	2550	1.04
40 mg, SD	1570	1540	1.02	10606	9100	1.17
80 mg, SD	3141	3610	0.87	21199	25400	0.83
110 mg, SD	4319	4200	1.03	29135	29400	0.99
200 mg, SD	7851	7270	1.08	52896	47900	1.10
400 mg, SD	15700	14300	1.10	105466	113000	0.93
20 mg, BID	958	1060	0.90	7332	6080	1.21
40 mg, BID	1971	2750	0.72	14781	13000	1.14
70 mg, BID	3357	3790	0.89	25334	20400	1.24
140 mg, BID	6563	7790	0.84	49326	43300	1.14

Source: Observed data were from study CDZ173X2101; predicted data were from FDA reviewer's analysis using Simcyp V21.
Abbreviations: AUC, area under the curve; BID, twice daily; C_{max}, maximum plasma concentration; Obs., observed; Pred., predicted; SD, standard deviation.

Can Leniolisib PBPK Model Predict the Effect of Erythromycin (a Moderate CYP3A Inhibitor) on the PK of Leniolisib Following Multiple Dose Administration of Leniolisib (70 mg BID) in Healthy Subjects?

No. The leniolisib PBPK model is inadequate to accurately predict the magnitude of effect of erythromycin on leniolisib PK following multiple dose administration of leniolisib (70 mg BID) in healthy subjects. (b) (4)

Can Leniolisib PBPK Model Predict the Effect of Rifampin (a Strong CYP3A Inducer) and Efavirenz (a Moderate CYp3A4 Inducer) on the PK of Leniolisib Following Multiple Dose Administration of Leniolisib (70 mg BID) in Healthy Subjects?

No. The leniolisib PBPK model is inadequate to accurately predict the magnitude of effect of rifampin or efavirenz on leniolisib PK following multiple dose administration of leniolisib (70 mg BID) in healthy subjects. (b) (4)

Can Leniolisib PBPK Model Predict the Effect of Leniolisib on PK of Midazolam (a Sensitive CYP3A4 Substrate) Following Multiple Dose Administration of Leniolisib (70 mg BID) in Healthy Subjects?

No. The leniolisib PBPK model is inadequate to accurately predict the magnitude of effect of leniolisib on midazolam PK following multiple dose administration of leniolisib (70 mg BID) in healthy subjects. (b) (4)

(b) (4)

16.4. Clinical Appendices

16.4.1. Patient Reported Outcomes (PROs)

16.4.1.1. Patient and Physician Reported Outcome Assessment Questionnaires

(b) (4)

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16.4.1.2. PRO Results

Patient Global Assessment (PtGA)

The PtGA questionnaire asks patients about their APDS related well-being using 100 mm visual analogue scale (VAS) ranging from “very poor” (0) to “very good” (100). The LS mean change from baseline at day 85 in PtGA scores was 10.59 in the leniolisib group and 1.34 in placebo. The difference in adjusted mean change (95% CI) of leniolisib vs placebo was 9.25 (-5.65, 24.14) (Table 80).

Table 80. Secondary Efficacy Endpoint – Analysis of Change from Baseline at Day 85 in Patient's Global Assessment (PtGA) of APDS (VAS scale) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	10.59	9.25 (-5.65, 24.14)	0.21
Placebo (N = 8)	8	1.34		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; APDS, activated phosphoinositide 3-kinase delta syndrome; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; PtGA, patient global assessment; VAS, visual analogue scale.

Physician Global Assessment (PGA)

In the PGA questionnaire the Investigator rates the disease activity of their patient using 100 mm VAS ranging from "no disease activity" (0) to "maximal disease activity" (100). The LS mean change from baseline in PGA questionnaire in the form of 100 mm VAS demonstrated less disease activity by Day 85 of the treatment in both the leniolisib treatment group (-22.09) and placebo (-20.76). The difference in adjusted mean change (95% CI) of leniolisib vs placebo was -1.33 (-19.48, 16.81) (Table 81).

Table 81. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in Physician's Global Assessment (PGA) of APDS (VAS scale) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	-22.09	-1.33 (-19.48, 16.81)	0.88
Placebo (N = 8)	8	-20.76		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; APDS, activated phosphoinositide 3-kinase delta syndrome; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; PGA, physician global assessment; VAS, visual analogue scale.

SF-36

The LS mean change from baseline at Day 85 in SF-36 scores (Mental Component Summary) was 2.69 in the leniolisib group and 1.56 in placebo. The difference in adjusted mean change (95% CI) was 1.13 (-5.50, 7.76) (Table 82). The LS mean change from baseline at Day 85 in SF-36 scores (Physical Component Summary) was 2.89 in the leniolisib group and 3.24 in placebo. The difference in adjusted mean change (95% CI) was -0.35 (-4.31, 3.62) (Table 83). In addition, the domain-level analyses of the SF-36 median scores showed greater numeric improvements from baseline for the physical functioning scale, general health scale, and physical component summary scores, for subjects on leniolisib compared to placebo at Week 12, while the SF-36

role physical and bodily pain scale median scores showed no change over the 12-week treatment period in both treatment groups.

Table 82. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in SF-36 (Mental Component Summary) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	2.69	1.13 (-5.50, 7.76)	0.73
Placebo (N = 8)	8	1.56		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SF-36; short form health survey.

Table 83. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in SF-36 (Physical Component Summary) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	2.89	-0.35 (-4.31, 3.62)	0.86
Placebo (N = 8)	8	3.24		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SF-36; short form health survey.

WPAI-CIQ

The LS mean change from baseline at Day 85 in WPAI-CIQ on percent activity impairment due to health was -9.18 for the leniolisib treatment group and -8.61 in placebo. The difference in adjusted mean change (95% CI) of leniolisib vs placebo was -0.57 (-22.72, 21.58) (Table 84). In addition, the domain-level analyses of the WPAI-CIQ median scores showed greater numeric improvements for work impairment (i.e., “Work Time Missed Due To Health”, “ Impairment While Working Due To Health”, and “Overall Work Impairment Due To Health”), but greater numeric worsening for classroom impairment (i.e., “Class Time Missed Due To Health”, “Impairment While In The Classroom Due To Health”, and “Overall Classroom Impairment Due To Health”), for subjects on leniolisib compared to placebo from baseline to Week 12. However, a high rate of missing data for the WPAI-CIQ was observed due to not taking the questionnaire as some questions or domains did not apply to subjects, thus, limiting the interpretation of the results.

Table 84. Secondary Efficacy Endpoint - Analysis of Change from Baseline at Day 85 in WPAI-CIQ (% Activity Impairment Due to Health) (PD Analysis Set)

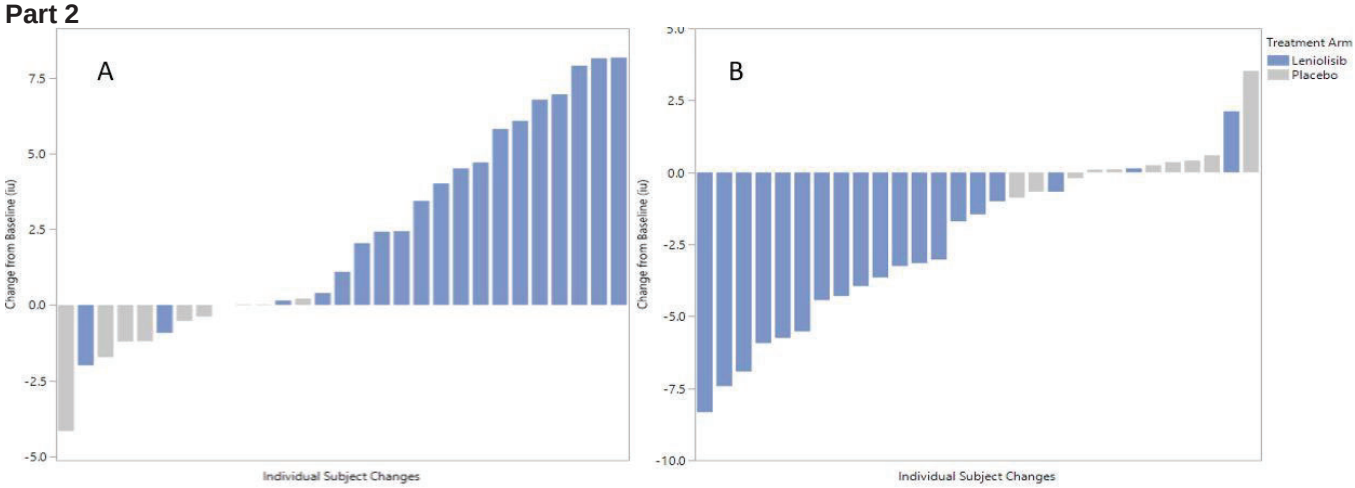
			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change from Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	-9.18	-0.57 (-22.72, 21.58)	0.96
Placebo (N = 8)	8	-8.61		

Source: Statistical Reviewer
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal because the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; CIQ, classroom impairment questions; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; WPAI, work productivity and activity impairment.

***Reviewer Comment**: Although there was substantial missing data in each domain of the WPAI-CIQl, the "% activity impairment due to health" domain (Table 84) did not have any missing data.*

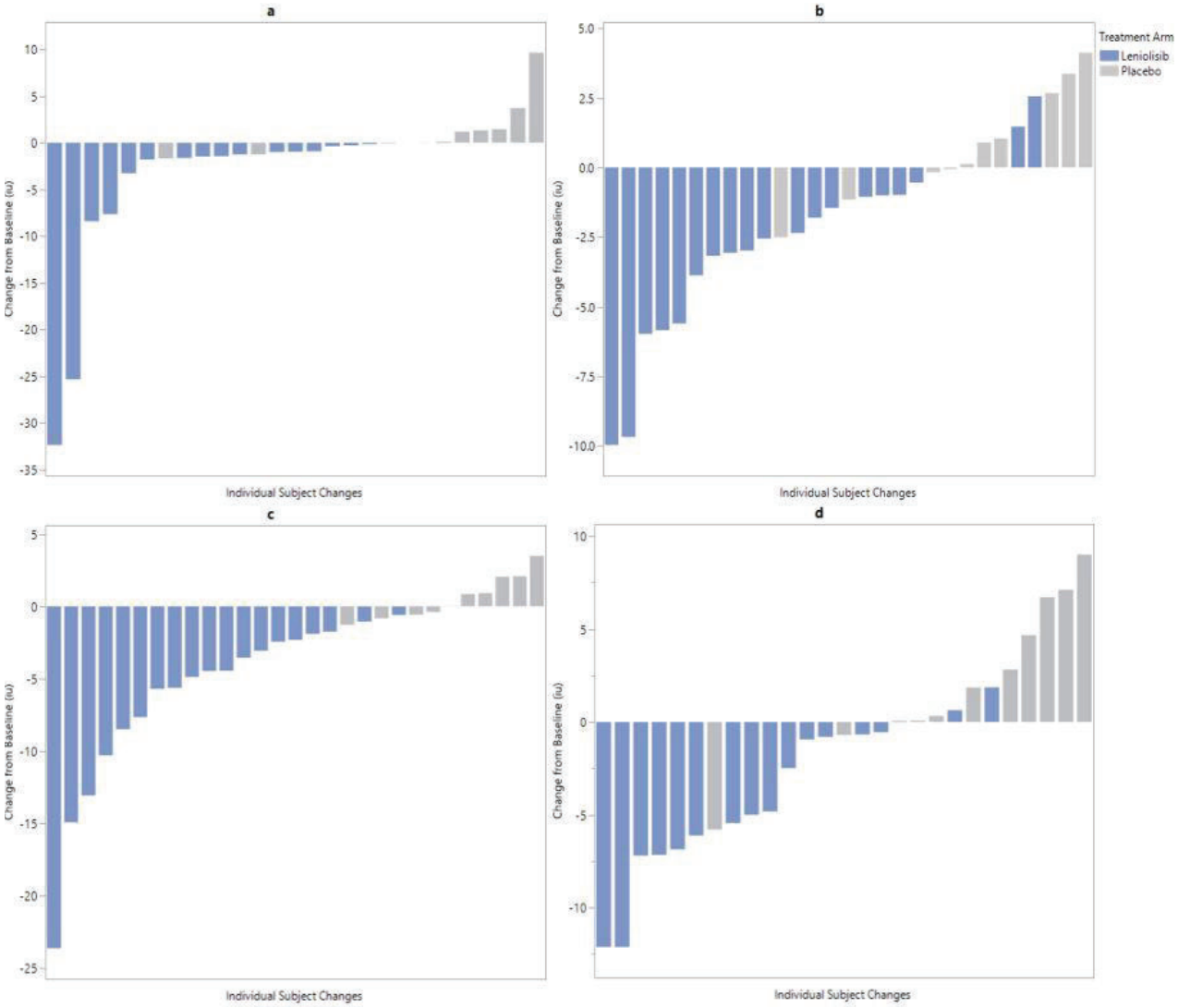
16.4.2. Trial 2201, Part 2 Exploratory Analyses

Figure 35. Individual Subject Changes From Baseline in CD4+ and CD8+ T Cell Counts, Trial 2201, Part 2



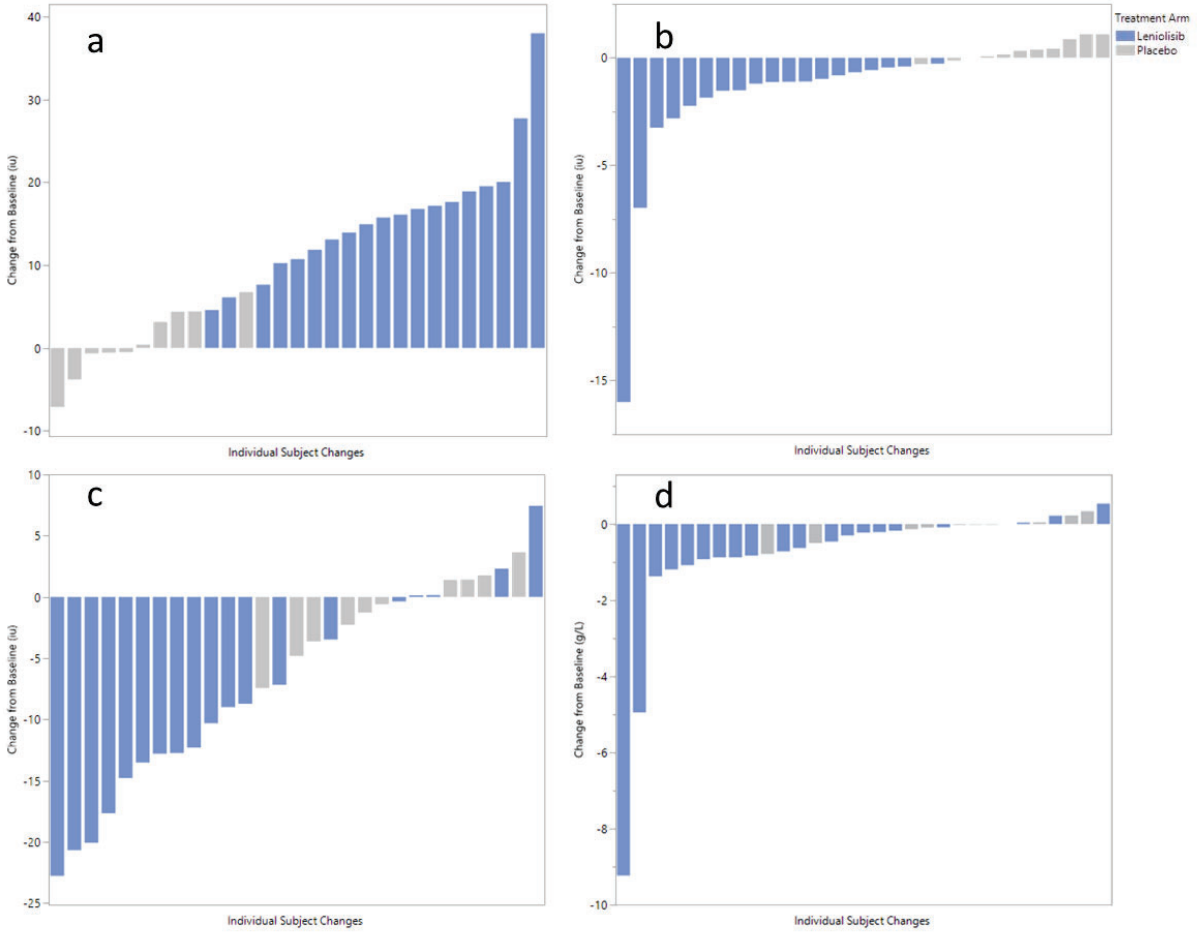
Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADLB dataset. Individual Subject changes over the course of the study in a) CD4+ T cells and b) CD8+ T cells.

Figure 36. Individual Subject Changes from Baseline in Senescent T Cell Markers, Trial 2201, Part 2



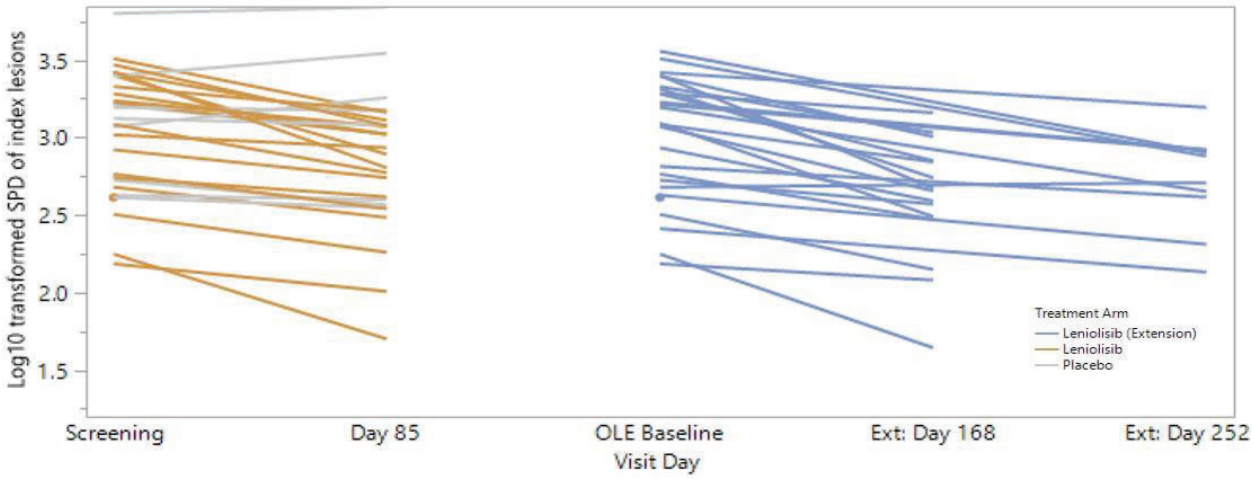
Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADLB dataset.
Note: Individual Subject changes over the course of the study in a) CD57+CD4+, b) CD57+CD8+, c) PD1+CD4+, and d) PD1+CD8+ Tcells.

Figure 37. Individual Sub ect Chan es From Baseline in the B Cell Com artment, Trial 2201, Part 2



Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADLB dataset.
Note: Individual Subject changes over the course of the study in a) naïve B cells b) Plasmablasts, c) Transitional B cells and d) IgM levels.

Figure 38. Log10 Transformed SPD of Index Lesions by Individual Subject, Trial 2201, Part 2 and Trial 2201E1

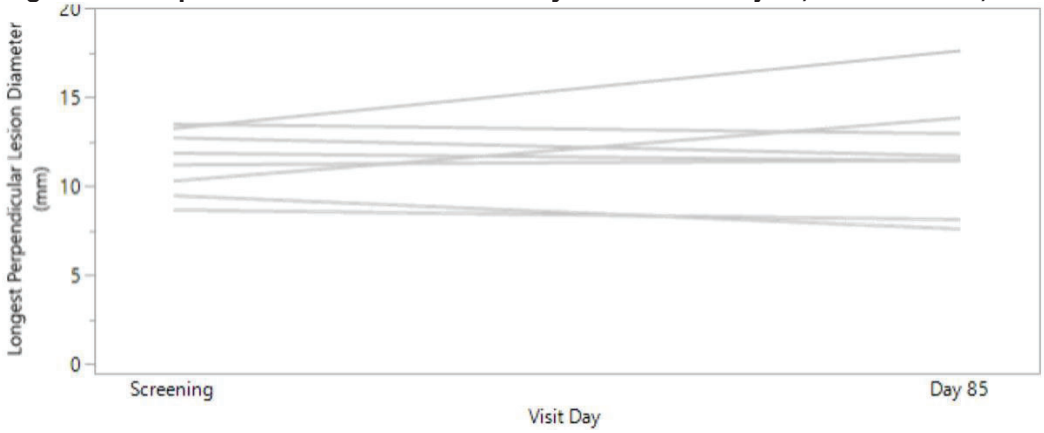


Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset.
Abbreviations: SPD, sum of product of diameters.

Figure 39. Perpendicular Lesion Diameter by Individual Subject, Leniolisib Arm, Trial 2201, Part 2

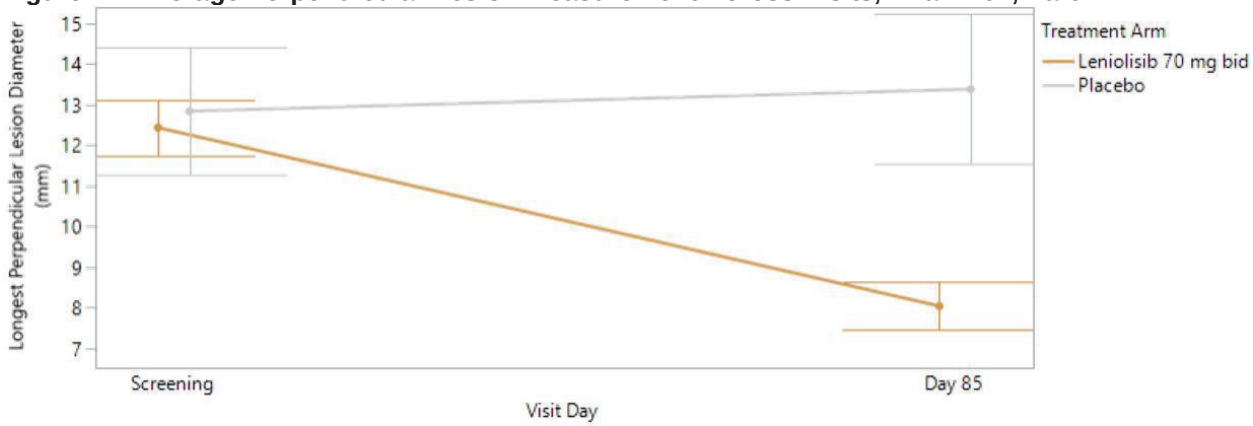
Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset

Figure 40. Perpendicular Lesion Diameter by Individual Subject, Placebo Arm, Trial 2201, Part 2



Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset

Figure 41. Average Perpendicular Lesion Measurement Across Visits, Trial 2201, Part 2



Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset.
Abbreviations: BID, twice daily.

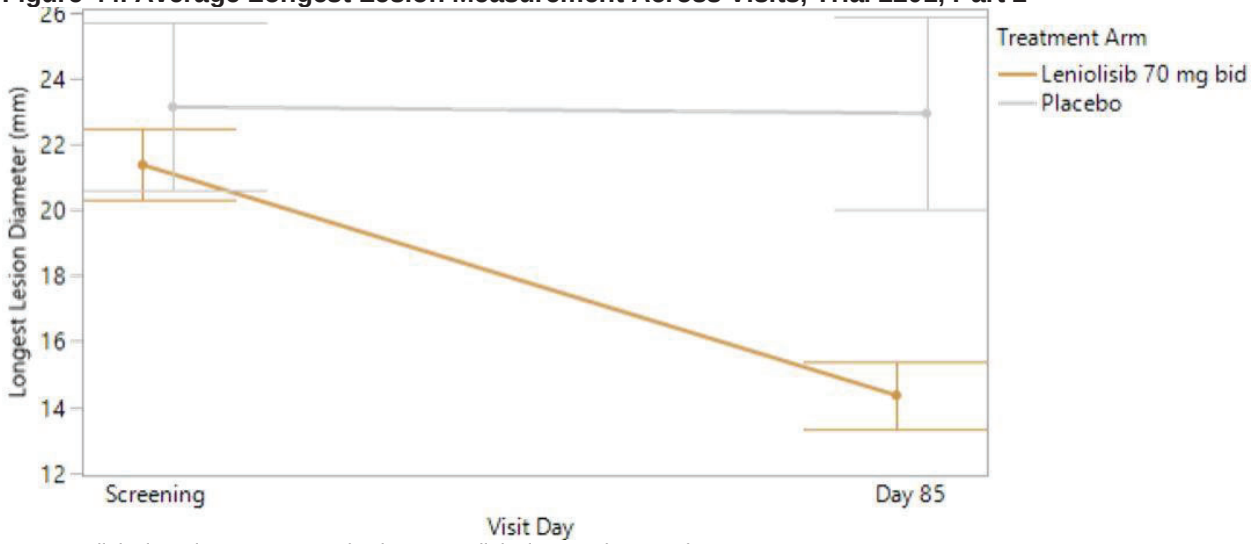
Figure 42. Longest Lesion Diameter by Individual Subject, Leniolisib Arm, Trial 2201, Part 2

Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset

Figure 43. Longest Lesion Diameter by Individual Subject, Placebo Arm, Trial 2201, Part 2

Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset

Figure 44. Average Longest Lesion Measurement Across Visits, Trial 2201, Part 2



Source: Clinical Reviewer Generated using JMP Clinical 8.1 and ADZR dataset.
Abbreviations: BID, twice daily.

Pharmacodynamic Markers of Inflammation

There were several measurements obtained at various timepoints throughout the study to assess the effect of leniolisib on reducing systemic inflammation in APDS. The Applicant evaluated hsCRP, beta-2 microglobulin, ESR, fibrinogen, ferritin, and LDH. There were too few subjects to adequately interpret the changes in hsCRP. While there appeared to be a reduction in beta-2 microglobulin and ESR, there was no meaningful changes in fibrinogen, ferritin, or LDH.

The results of change from baseline in various cytokine and chemokine levels thought to be relevant to APDS, including CXCL13, IP-10 (CXCL10), TNF-alpha, MDC, MIP-1B, MIP3A, immunoglobulins (discussed above), and TNF-alpha, were evaluated as part of the exploratory

analysis. Treatment with leniolisib caused a decrease in the concentration of CXCL13, IP-10, MIP-1B, and TNF-alpha concentrations.

16.4.3. OLE Study 2201E1 Secondary and Exploratory Endpoints

Imaging for Lymphoproliferation

Most subjects enrolled in Trial 2201E1 experienced a reduction from baseline in the imaging parameters including volume of index lesions, SPD of index lesions, volume and area of the spleen and liver through Extension Day 252. All 15 subjects who underwent imaging for evaluation of SPD of index lesions at Day 168 experienced a reduction from baseline. Eleven of 12 subjects who underwent imaging for evaluation of SPD of index lesions at Day 252 experienced a reduction from baseline. Similarly, of the 16 subjects who received imaging to evaluate spleen organ volume at Day 168, 15 experienced a reduction in volume. All subjects (11/11) who received imaging at Day 252 showed a reduction in spleen organ volume.

B Cell Immunophenotype

At baseline, the mean (SD) naïve B cell percentage was 58.16% (20.922; n=5). The mean (SD) change from baseline for all extension study subjects showed the percentage of naïve B cells increased at each timepoint throughout the extension study starting at 23.58% (16.177) at Extension Day 84 (n=5) to a maximum of 32.42% (25.293) at Extension Day 252 (n=5).

T Cell Immunophenotype

An increase in naïve CD4- (2.51%), CD4+ (12.05%), naïve CD4+ (1.5%), and overall T cells (9.58%) with a concomitant decrease in the percent CD4- of 11.70% from baseline to Extension Day 252 was recorded.

Clinical Chemistry

Similar to Trial 2201, Part 1 hsCRP and LDH were measured to assess the effect of leniolisib on systemic inflammation as a component of APDS over time. For subjects with prior exposure to leniolisib, a reduction from baseline was observed in the mean hsCRP values through Extension Day 252, followed by an increase from baseline in mean hsCRP values at all other timepoints with the exception of Extension Day 546 (n=6) and Extension Day 1820 (n=1). For subjects with no prior exposure to leniolisib, an increase from baseline was observed in the mean hsCRP values at all timepoints with the exception of Extension Day 910 (n=1).

SF-36 and WPAI-CIQ

Overall, the mean change from baseline of the SF-36 and the WPAI-CIQ remained unchanged over the course of the extension study.

PtGA and PGA

Both the mean change from baseline in PtGA and PGA scores for all extension study subjects combined showed decreases at all timepoints throughout the extension study.

Soluble Biomarkers

Soluble protein markers evaluated as part of the extension study included chemokine ligand 13 (CXCL13), interferon gamma-induced protein 10 (TP-10), macrophage-derived chemokine (MDC), macrophage inflammatory protein (MIP)-1β, MIP-3a, interferon gamma (IFN-γ), and tumor necrosis factor alpha (TNF-a) and were evaluated through Day 252 of the extension study. CXCL13, IP-10, MIP-3a, and TNF-α decreased throughout the extension study. MDC and MIP-1β initially increased followed by a decrease and IFN-γ remained essentially unchanged.

EBV and CMV Assessment

Shifts in EBV DNA from negative at baseline to positive post-baseline were reported for 4/26 subjects (15.4%) at Extension Day 1, 3/25 subjects (12.0%) at Extension Day 84, 2/21 subjects (9.5%) at Extension Day 168, and 4/21 subjects (19.0%) at Extension Day 252.

Shifts in CMV DNA from negative at baseline to positive post-baseline were reported for 3/23 subjects (13.0%) at Extension Day 1, 0 subjects at Extension Day 4, 1/19 subject (5.3%) at Extension Days 168, and 252.

Serum Immunoglobulins

Serum IgA, IgE, IgG, and IgM were monitored as part of the extension study. The change from baseline in all classes of immunoglobulins appeared to increase at Extension Day 1 after which they all approached baseline by Extension Day 84 where they remained through Extension Days 168 and 252 with the exception of IgM which decreased below baseline.

Immunoglobulin Replacement Therapy

In Trial 2201E1, annualized rate of IRT trended down throughout the course of the study. However, there was no control group, and few subjects were available for analysis.

Antibiotic Usage

In Trial 2201E1, subjects continued to receive antibiotics at a similar rate as those randomized to the leniolisib treatment arm in Trial 2201, Part 2. This trend was particularly evident in the first four years of treatment in the extension study.

16.5. OB Appendices

The pre-specified key efficacy analyses agreed to in the SAP are presented in this appendix.

Table 85. Primary Analysis of Change From Baseline at Day 85 in Log10 Transformed SPD of Index Lesions (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	18	-0.30	-0.24 (-0.37, -0.11)	<0.01
Placebo (N = 8)	8	-0.06		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and log10 transformed baseline SPD as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 1 subject in the leniolis b group having complete resolution of the index lesion identified at baseline.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SPD, sum of product of diameters.

Table 86. Primary Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set and With a Percentage of Less Than 48% of Naive B Cells at Baseline)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	8	34.76	40.13 (28.51, 51.75)	<0.01
Placebo (N = 8)	5	-5.37		

Source: Statistical Reviewer.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. Baseline is defined as the arithmetic mean of the baseline and Day 1 values when both are available, and if either baseline or the Day 1 value is missing, the existing value is used. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group who had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolis b group.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 87. Sensitivity Analysis of Change from Baseline at Day 85 in Naïve B Cells (PD Analysis Set and With a Percentage of Less Than 48% of Naive B Cells at Baseline)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	8	32.45	37.25 (25.91, 48.59)	<0.01
Placebo (N = 8)	5	-4.80		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-3.3.2b.
Note: Data were analyzed using a longitudinal mixed model, with treatment, time, treatment by time interaction, baseline, and baseline by time interaction as fixed effects. An unstructured covariance matrix will be fitted to adjust for correlations among the measurements made on the same subject. The use of glucocorticoids and intravenous IgG at baseline. The analysis excluded 5 subjects in the leniolisib group and 3 subjects in the placebo group who had more than 48% naïve B cells at baseline. The analysis also excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolisib group.
Abbreviations: CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 88. Supportive Analysis of Change From Baseline at Day 85 in Naïve B Cells (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	13	27.87	26.95 (13.93, 39.96)	<0.01
Placebo (N = 8)	8	0.92		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-3.2.2b.
Note: LS means, difference in LS means, 95% CI for difference in LS means, and p-values are estimated using ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. The analysis excluded 5 subjects with no measurement at Day 85 and 1 subject with no baseline measurement in the leniolis b group.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 89. Secondary Efficacy Endpoint – Analysis of Change From Baseline at Day 85 in the Sum of the Log10 Transformed 3D Volume of Index Lesions (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	18	-2.20	-0.90 (-2.67, 0.88)	0.31
Placebo (N = 8)	8	-1.31		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-2.6.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and the log10 transformed baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 90. Secondary Efficacy Endpoint – Analysis of Change From Baseline at Day 85 in the Sum of the Log10 Transformed 3D Volume of Non-Index Lesions (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	16	-1.42	-1.84 (-2.95, -0.84)	<0.01
Placebo (N = 8)	8	0.43		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-2.6.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and the log10 transformed baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.

Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 91. Secondary Efficacy Endpoint – Analysis of Change From Baseline at Day 85 in Spleen Bi-Dimensional Size (mm^2) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	-1062	-1473 (-2517, -428)	<0.01
Placebo (N = 8)	8	411		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-2.8.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 92. Secondary Efficacy Endpoint – Analysis of Change From Baseline at Day 85 in Spleen Organ Volume (mm^3) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	-148834	-194666 (-300125, -89207)	<0.01
Placebo (N = 8)	8	45833		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-2.8.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic

Table 93. Secondary Efficacy Endpoint – Analysis of Change From Baseline at Day 85 in Patient Global Assessment of APDS (VAS scale) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib 70 mg BID (N = 19)	19	11.10	9.64 (-5.56, 24.84)	0.20
Placebo (N = 8)	8	1.47		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-6.2.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; VAS, visual analogue scale.

Table 94. Secondary Efficacy Endpoint - Analysis of Change From Baseline at Day 85 in Physician Global Assessment of APDS (VAS scale) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib 70 mg BID (N = 19)	19	-12.62	-4.74 (-19.94, 10.46)	0.53
Placebo (N = 8)	8	-7.88		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-7.2.1b.
Note:
Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; APDS, activated phosphoinositide 3-kinase delta syndrome; BID, twice daily; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; VAS, visual analogue scale.

Table 95. Secondary Efficacy Endpoint - Analysis of Change From Baseline at Day 85 in SF-36 (Mental Component Summary) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	2.01	1.54 (-5.07, 8.15)	0.63
Placebo (N = 8)	8	0.47		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-4.2.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SF-36; short form health survey

Table 96. Secondary Efficacy Endpoint - Analysis of Change From Baseline at Day 85 in SF-36 (Physical Component Summary) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib (N = 19)	19	3.72	-0.50 (-4.66, 3.65)	0.80
Placebo (N = 8)	8	4.23		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-4.2.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; SF-36; short form health survey

Table 97. Secondary Efficacy Endpoint - Analysis of Change From Baseline at Day 85 in WPAI-CIQ (% Activity Impairment Due to Health) (PD Analysis Set)

			Leniolisib vs Placebo	
Treatment Group	n	LS Mean Change From Baseline	Diff (95% CI) in LS Means	P-value
Leniolisib 70 (N = 19)	19	-4.68	-2.66 (-23.89, 18.57)	0.80
Placebo (N = 8)	8	-2.02		

Source: Applicant's Clinical Trial Report for Study 2201 Part 2, Table 14.2-5.2.1b.
Note: Data were analyzed using an ANCOVA model with treatment as a fixed effect and baseline as a covariate. The use of glucocorticoids and intravenous IgG at baseline were both included as categorical (Yes/No) covariates. P-value is nominal as the analysis was not adjusted for multiplicity.
Abbreviations: ANCOVA, analysis of covariance; CI, confidence interval; CIQ, classroom impairment questions; Diff, difference; IgG, Immunoglobulin G; LS, least squares; N, total number of subjects; n, number of subjects in the analysis; PD, pharmacodynamic; WPAI, work productivity and activity impairment

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/s/

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03/24/2023 09:20:58 AM

JULIE G BEITZ
03/24/2023 10:20:29 AM

DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH

PHARMACOLOGY/TOXICOLOGY NDA/BLA REVIEW AND EVALUATION

Application number:	NDA 217759
Supporting document/s:	1, 39
Applicant's letter date:	07/29/2022
CDER stamp date:	07/29/2022, 01/27/2023
Product:	JOENJA® (leniolisib) oral tablets
Indication:	Activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS)
Applicant:	Pharming Technologies B.V.
Review Division:	Division of Pulmonology, Allergy and Critical Care (DPACC)
Reviewer:	Wei Sun, PhD, DABT
Team Leader:	Jessica A. Bonzo, PhD
Division Director:	Sally Seymour, MD
Project Manager:	Elaine Sit, PharmD

Template Version: September 1, 2010

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Except as specifically identified, all data and information discussed below and necessary for approval of NDA217759 owned by Pharming Technologies B.V. or are data for which Pharming Technologies B.V. has obtained a written right of reference. Any information or data necessary for approval of NDA217759 that Pharming Technologies B.V. does not own or have a written right to reference constitutes one of the following: (1) published literature, or (2) a prior FDA finding of safety or effectiveness for a listed drug, as reflected in the drug's approved labeling. Any data or information described or referenced below from reviews or publicly available summaries of a previously approved application is for descriptive purposes only and is not relied upon for approval of NDA217759.

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1 Executive Summary

1.1 Introduction

Pharming Technologies B.V. submitted an Original 505(b)(1) New Drug Application (NDA) on July 29, 2022, for JOENJA® (leniolisib), which is proposed for the treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adults and adolescents aged 12 or older. This review evaluates the labeling submitted to NDA 217759 to support leniolisib for approval. Refer to Unireview under NDA 217759 and Nonclinical reviews submitted under IND 124045 and IND (b) (4) for all related Pharmacology and Toxicology studies.

1.2 Brief Discussion of Nonclinical Findings

Pharmacological studies demonstrated that leniolisib binds to the active site of human PI3Kδ. In human cell-free isolated enzyme assays, leniolisib was selective for PI3Kδ over PI3Kα (28-fold), PI3Kβ (43-fold), and PI3Kγ (257-fold), as well as the broader kinome. In human cell-based assays, leniolisib reduced pAKT pathway activity and inhibited proliferation and activation of B and T cell subsets.

The Applicant has conducted subchronic and chronic toxicology studies with leniolisib in both rats and cynomolgus monkeys. Leniolisib-related toxicities included immune suppression (rats & monkeys), skin lesions (rats & monkeys), germ cell depletion (rats), GI tract inflammation (monkeys), and QTc prolongation (monkeys). These toxicities were generally pharmacodynamic-related and/or clinically monitorable and not considered dose-limiting. These findings were observed at plasma exposures approximately 3 times or equivalent to the human exposure at the maximum recommended human dose (MRHD) in rats and monkeys, respectively. In animal reproduction studies, oral administration of leniolisib to pregnant rats and rabbits during the period of organogenesis at exposures approximately 2-6 times the MRHD on an AUC basis, produced embryofetal toxicity including malformations. Leniolisib was not mutagenic or clastogenic in *in vitro* and *in vivo* assays. The pre- and postnatal developmental study in rats as well as the 6-month Tg-RasH2 mouse and 2-year rat carcinogenicity studies will be completed as post-marketing requirements.

1.3 Recommendations

1.3.1 Approvability

This review describes the recommended changes for labeling. NDA 217759 is recommended for approval from the nonclinical perspective.

1.3.2 Additional Nonclinical Recommendations

None.

1.3.3 Labeling

3 Pages of Draft Labeling have been Withheld in Full as b4 (CCI/TS) immediately following this page

(b) (4)

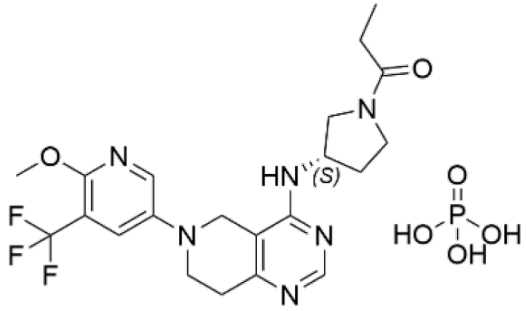
2 Drug Information

2.1 Drug

CAS Registry Number: 1354691-97-6

Generic Name: Leniolisib

Structure or Biochemical Description:



Pharmacologic Class: Kinase inhibitor

2.2 Relevant INDs, NDAs, BLAs and DMFs

The following is a list of relevant INDs and NDAs:

IND 124045: Leniolisib (Pharming Technologies B.V.)
IND (b) (4) Leniolisib (Pharming Technologies B.V.)

2.3 Drug Formulation

Leniolisib tablet components for the 70 mg strength are presented in Table 1. The drug product is supplied as film-coated tablets and is composed of leniolisib phosphate and the inactive ingredients lactose monohydrate, microcrystalline cellulose, hydroxypropyl methylcellulose, sodium starch glycolate (b) (4) magnesium stearate, colloidal silicon

dioxide, and [redacted]. Each tablet contains leniolisib equivalent to 70 mg of the free base compound (Table 1).

Table 1 Quantitative Composition of Leniolisib Tablets, 70 mg

Table 2 Qualitative and quantitative composition of leniolisib 70 mg film-coated tablets

Component	Quantity per tablet (mg)	Function	Reference to quality standards
Leniolisib phosphate[a]	85 [redacted]	Active ingredient	In-house
Lactose monohydrate	[redacted]	[redacted]	Ph. Eur./USP-NF
Microcrystalline cellulose			Ph. Eur./USP-NF
Hydroxypropyl methylcellulose			Ph. Eur./USP-NF
Sodium starch glycolate [redacted]			Ph. Eur./USP-NF
Magnesium stearate			Ph. Eur./USP-NF
Colloidal silicon dioxide			Ph. Eur./USP-NF
[redacted]			In-house
[redacted]			Ph. Eur./USP-NF

a. [redacted]

The qualitative composition [redacted] used in film-coating of leniolisib tablets is provided in Table 3.

Table 3 Composition of [redacted]

Component	Reference to standards
Hypromellose/Hydroxypropyl methylcellulose (HPMC [redacted]	Ph. Eur., USP-NF
Titanium dioxide [redacted]	USP-NF, Regulation (EU) 231/2012 [b]
Iron oxide yellow [redacted]	USP-NF, Regulation (EU) 231/2012 [b]
Iron oxide red [redacted]	USP-NF, Regulation (EU) 231/2012 [b]
Talc	Ph. Eur., USP-NF
[redacted] Polyethylene glycol	Ph. Eur., USP-NF
[redacted]	

[b] Commision Regulation (EU) 231/2012: of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council.

(Excerpted from the Applicant's submission, SD1-M3-32p1-description-and-composition-lnlbtab, pg1)

There is no novel excipient in the drug product formulation. The compendial excipients of the drug product comply with the respective current compendial monographs of USP-NF and Ph. Eur.

Leniolisib film-coated tablets are packaged per 60 tablets in a high density polyethylene (HDPE) bottle with an aluminum induction seal and [redacted] screw cap closure.

2.6 Proposed Clinical Population and Dosing Regimen

JOENJA® (leniolisib) is proposed for the treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adults and pediatric patients aged 12 or older.

The recommended dosage of JOENJA® (leniolisib) tablet is 70 mg orally twice daily approximately 12 hours apart, with or without food.

2.7 Regulatory Background

The original IND (b) (4) and IND 124,045 for APDS/PASLI (activated PI3Kδ syndrome/p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency) were submitted by Novartis Pharmaceuticals Corporation in 2012 and 2014, respectively. Pharming Technologies B.V. acquired the compound in 2019. Provided below is a summary of the regulatory background related to the nonclinical development program for leniolisib (under IND (b) (4) IND 124045, and NDA 217759).

- June 8, 2017: Type C meeting. “1. The microscopic findings of decreased germinal cell epithelium in the testes of male rats at CDZ173 doses ≥ 40 mg/kg/day in the 26-week rat toxicology study (Study 1470444) are considered to be adverse. CDZ173 exposure in rats at the 40 mg/kg/day dose is lower than the projected clinical AUC_{0-24} at the 70 mg BID dose. These findings are not clinically monitorable and did not appear to reverse after a 4-week recovery period. The findings should be described in the Informed Consent document for your proposed clinical study as well as in your updated Investigator’s Brochure. 2. QTc prolongation was observed in male and female monkeys at all CDZ173 doses tested (20 mg/kg/day and above) in your 39-week toxicology study (Study 1470445). These observations are consistent with findings in previously completed toxicology studies of shorter duration as well as the *in vitro* hERG assay and cardiovascular safety pharmacology studies. The findings should be described in the Informed Consent document for your proposed clinical study as well as in your updated Investigator’s Brochure.”
- November 17, 2020: Type C meeting WRO issued. Based on the indication for APDS/PASLI, the Division tentatively agreed to post-approval submission of the 2-year carcinogenicity study with rats and a pre- and postnatal development studies with rats.
- March 9, 2022: Type C meeting WRO issued. (b) (4)
- April 29,2022: PreNDA meeting issued. Nonclinical package is adequate for NDA filing and based on the indication for APDS/PASLI, agreed that the 6-month transgenic (rasH2) mouse carcinogenicity study reports will be provided post approval.
- July 29, 2022: The NDA for JOENJA® (leniolisib) was submitted on July 29, 2022, and the application was filed on September 27, 2022, for priority review.

3.3 Previous Reviews Referenced

See the unireview of NDA 217759 for a summary of nonclinical studies.

11 Integrated Summary and Safety Evaluation

The nonclinical review of the proposed product label recommended changes for labeling in Section 5.1 (Embryo-Fetal Toxicity), Section 8.1 (Pregnancy), Section 8.4 (Pediatric Use), Section 11 (DESCRIPTION), Section 12.1 (Mechanism of Action), and 13.1 (Carcinogenesis, Mutagenesis, Impairment of Fertility), that are described below. Additions are shown as underlined text and deletions are shown as strikeout ~~text~~.

Table 2. Labeling review for NDA 217759 (Leniolisib)

(b) (4)

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Appendix:

Table 3. Leniolisib Toxicokinetics (TK) and Exposure Margins in Developmental and Reproductive Studies

Study	Species	Dose (mg/kg)	$AUC_{0\text{-}24h}$, Male/Female or Average (ng·h/mL)	Exposure Margin
Fertility and early embryonic development (FEED) study[1]	Rat	10	5270/8470	0.1/0.2
		30	21600/40200	0.5/1.0
		90	90900/14300	2.2/3.5
Embryofetal development (EFD) studies	Rat	10	12800	0.3
		30	65000	1.6
		120	248000	6.1
	Rabbit	10	5210	0.1
		30	13200	0.3
		100	87300	2.1
Juvenile toxicology study	Rat	10	5270/8470	0.1/0.2
		30	21600/40200	0.5/1.0
		90	90900/14300	2.2/3.5

NOAELs are labeled in red.

Clinical exposure of leniolisib in healthy subjects at an oral dose of 70 mg BID at the steady state on Day 15 (Study 2101 CSR, part 3: AUCtau,ss = 20400 ng·h/mL, $AUC_{0\text{-}24hr}$ = 2 x 20400 ng·h/mL = 40800 ng·h/mL).

[1]AUCs of leniolisib in the rat FEED study are assessed on PND 77 in the juvenile toxicology study.

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/s/

WEI SUN
03/15/2023 10:41:52 AM

JESSICA A BONZO
03/15/2023 10:45:51 AM
I concur

DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH

PHARMACOLOGY/TOXICOLOGY NDA/BLA REVIEW AND EVALUATION

Application number:	NDA # 217759
Supporting document/s:	EDR, #1
Applicant's letter date:	07/29/2022
CDER stamp date:	07/29/2022
Product:	JOENJA® (leniolisib) oral tablets
Indication:	Activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS)
Applicant:	Pharming Technologies B.V.
Review Division:	Division of Pulmonology, Allergy, and Critical Care
Reviewer:	Wei Sun, PhD, DABT
Team Leader:	Jessica Bonzo, PhD,
Division Director:	Sally Seymour, MD
Project Manager:	Elaine Sit, PharmD

Template Version: September 1, 2010

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Any information or data necessary for approval of NDA 217759 that Pharming Technologies B.V. does not own or have a written right to reference constitutes one of the following: (1) published literature, or (2) a prior FDA finding of safety or effectiveness for a listed drug, as reflected in the drug's approved labeling. Any data or information described or referenced below from reviews or publicly available summaries of a previously approved application is for descriptive purposes only and is not relied upon for approval of NDA 217759.

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1.1 Introduction

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1.2 Brief Discussion of Nonclinical Findings

Seven mutagenic or potentially mutagenic impurities (b) (4) were negative in Ames assays. The other four mutagenic or potentially mutagenic impurities were appropriately controlled according to ICH M7. (b) (4) was appropriately controlled per FDA (b) (4) and EMA (b) (4) guidance. Other impurities that were identified are appropriately controlled per ICH Q3A, ICH Q3B, or United States Pharmacopeia (USP) Guidelines.

In the GLP compliant *in vitro* Balb/c 3T3 neutral red uptake phototoxicity assay, leniolisib was not phototoxic at concentrations up to 100 µg/mL. The risk of phototoxicity is considered minimal according to ICH S10.

1.3 Recommendations

1.3.1 Approvability

Approvability will be discussed in the unireview.

1.3.2 Additional Nonclinical Recommendations

None.

1.3.3 Labeling

Labeling recommendations will be provided in a separate review.

2 Drug Information

2.1 Drug

CAS Registry Number: 1354691-97-6

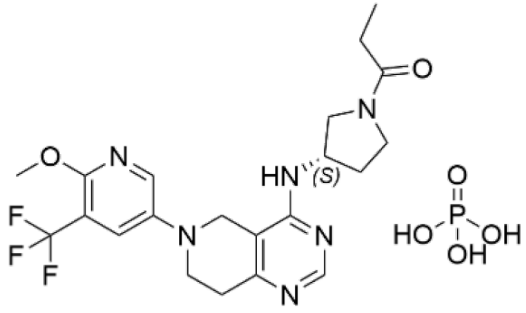
Generic Name: Leniolisib

Chemical Name: 1-[(3S)-3-[[5,6,7,8-Tetrahydro-6-[6-methoxy-5-(trifluoromethyl)-3-pyridinyl]pyrido[4,3-d]pyrimidin-4-yl]amino]-1-pyrrolidinyl]-1-propanone phosphate (1:1)

Other Name: CDZ173 phosphate, leniolisib phosphate

Molecular Formula/Molecular Weight: $C_{21}H_{25}F_3N_6O_2.H_3PO_4$/ 450.46 (free base) 548.45 (salt) g/mol

Structure or Biochemical Description:



Pharmacologic Class: PI3Kδ inhibitor

2.2 Relevant INDs, NDAs, BLAs and DMFs

The following is a list of relevant INDs and NDAs:

IND 124045: Leniolisib (Pharming Technologies B.V.)
IND (b) (4) Leniolisib (Pharming Technologies B.V.)

2.3 Drug Formulation

Leniolisib tablet components for the 70 mg strength are presented in Table 1. The drug product is supplied as film-coated tablets and is composed of leniolisib phosphate and the inactive ingredients lactose monohydrate, microcrystalline cellulose, hydroxypropyl methylcellulose, sodium starch glycolate (b) (4) magnesium stearate, colloidal silicon dioxide, and (b) (4). Each tablet contains leniolisib equivalent to 70 mg of the free base compound (Table 1).

Table 1 Quantitative Composition of Leniolisib Tablets, 70 mg

Table 2 Qualitative and quantitative composition of leniolisib 70 mg film-coated tablets

Component	Quantity per tablet (mg)	Function	Reference to quality standards
Leniolisib phosphate[a]	85 (b) (4)	Active ingredient	In-house
Lactose monohydrate	(b) (4)		Ph. Eur./USP-NF
Microcrystalline cellulose			Ph. Eur./USP-NF
Hydroxypropyl methylcellulose			Ph. Eur./USP-NF
Sodium starch glycolate (b) (4)			Ph. Eur./USP-NF
Magnesium stearate			Ph. Eur./USP-NF
Colloidal silicon dioxide			Ph. Eur./USP-NF
(b) (4)			In-house
			Ph. Eur./USP-NF

a. (b) (4)

(b) (4)

The qualitative composition (b) (4) used in film-coating of leniolisib tablets is provided in Table 3.

Table 3 Composition of (b) (4)

Component	Reference to standards
Hypromellose/Hydroxypropyl methylcellulose (HPMC (b) (4)	Ph. Eur., USP-NF
Titanium dioxide (b) (4)	USP-NF, Regulation (EU) 231/2012 [b]
Iron oxide yellow (b) (4)	USP-NF, Regulation (EU) 231/2012 [b]
Iron oxide red (b) (4)	USP-NF, Regulation (EU) 231/2012 [b]
Talc	Ph. Eur., USP-NF
(b) (4) Polyethylene glycol	Ph. Eur., USP-NF

(b) (4)

[b] Commision Regulation (EU) 231/2012: of 9 March 2012 laying down specifications for food additives listed in Annexes II and III to Regulation (EC) No 1333/2008 of the European Parliament and of the Council.

(Excerpted from the Applicant's submission, SD1-M3-32p1-description-and-composition-lnlbtab, pg1)

There is no novel excipient in the drug product formulation. The compendial excipients of the drug product comply with the respective current compendial monographs of USP-NF and Ph. Eur.

Leniolisib film-coated tablets are packaged per 60 tablets in a high density polyethylene (HDPE) bottle with an aluminum induction seal and (b) (4) screw cap closure.

2.4 Comments on Novel Excipients

There are no novel excipients. All the excipients are at levels less than or similar to levels in FDA approved oral drug products.

2.5 Comments on Impurities/Degradants of Concern

Potentially mutagenic impurities

The Applicant followed the recommendations of the ICH M7 Guideline and evaluated the mutagenic potential of impurities based on literature, internal databases, and in silico assessment including both expert rule-based (i.e., Derek Nexus (v.4.0.5, V4.1.0 or v.5.0.2)) and statistical-based systems (i.e., Case Ultra (v.1.5.0.1, v.1.5.2.0 or v.1.6.0.3) and/or Sarah Nexus (v.1.1.2, v1.2.0 or v.2.0.1)). The Applicant conducted additional non-GLP Ames assays for two positive predictions. Overall, the proposed impurity specifications are acceptable and there are no outstanding nonclinical safety concerns.

Table 2 List of potentially mutagenic impurities

(b) (4)

(b) (4)

A positive Ames test was shown for (b) (4) in Study 1419436. (b) (4) (b) (4) (b) (4) The Applicant proposed to control (b) (4) at (b) (4) ppm, which corresponds to (b) (4) µg/day at the highest anticipated clinical dose of 70 mg BID. The proposed acceptance criterion is below (b) (4) µg/day, according to the ICH M7 (R1) Guidance, (b) (4)

Table 3 Mutagenic impurity (b) (4) levels

(b) (4)

After an email consultation with the CMC review team dated October 12, 2022, it is noted that (b) (4) Not specifying these three impurities in the drug substance specification is acceptable.

(b) (4) An IR dated October 13, 2022, was sent to the Sponsor for additional details of the two non-GLP Ames tests. In the IR response dated October 21, 2022, the Sponsor noted the two Ames tests were conducted in accordance with the current GLP requirements of the UK MHRA and OECD guidelines of Ames assay, but without the GLP-Quality Assurance unit of the testing laboratories. In addition, (b) (4) was tested to be negative in a GLP-compliant bacterial mutagenicity assay according to OECD guidelines, (b) (4)

(b) (4)
were reported in literature to be negative in Ames assays. Overall, the weight of evidence supports assignment of these four impurities (b) (4) and control as ordinary impurities.

(b) (4)
The Applicant evaluated the risks of four (b) (4)
were not detected in any of the drug substance batches tested.
(b) (4)

Table 4 (b) (4) levels

(b) (4)

Other identified impurities are appropriately controlled per ICH Q3A, ICH Q3B, or United States Pharmacopeia (USP) Guidelines.

2.6 Proposed Clinical Population and Dosing Regimen

JOENJA® (leniolisib) is proposed for the treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adults and adolescents aged 12 or older. The recommended dosage of JOENJA® (leniolisib) tablet is 70 mg orally twice daily approximately 12 hours apart, with or without food.

2.7 Regulatory Background

The original IND (b) (4) and IND 124,045 for APDS/PASLI (activated PI3Kδ syndrome/p110δ-activating mutation causing senescent T cells, lymphadenopathy and immunodeficiency) were submitted by Novartis Pharmaceuticals Corporation in 2012 and 2014, respectively. Pharming Technologies B.V. acquired the compound in 2019. Provided below is a summary of the regulatory background related to the nonclinical development program for leniolisib (under IND (b) (4) IND 124045, and NDA 217759).

- June 8, 2017: Type C meeting. “1. The microscopic findings of decreased germinal cell epithelium in the testes of male rats at CDZ173 doses ≥ 40

mg/kg/day in the 26-week rat toxicology study (Study 1470444) are considered to be adverse. CDZ173 exposure in rats at the 40 mg/kg/day dose is lower than the projected clinical AUC_{0-24} at the 70 mg BID dose. These findings are not clinically monitorable and did not appear to reverse after a 4-week recovery period. The findings should be described in the Informed Consent document for your proposed clinical study as well as in your updated Investigator's Brochure. 2. QTc prolongation was observed in male and female monkeys at all CDZ173 doses tested (20 mg/kg/day and above) in your 39-week toxicology study (Study 1470445). These observations are consistent with findings in previously completed toxicology studies of shorter duration as well as the *in vitro* hERG assay and cardiovascular safety pharmacology studies. The findings should be described in the Informed Consent document for your proposed clinical study as well as in your updated Investigator's Brochure."

- November 17, 2020: Type C meeting WRO issued. Based on the indication for APDS/PASLI, the Division tentatively agreed to post-approval submission of the 2-year carcinogenicity study with rats and a pre- and postnatal development studies with rats.
- March 9, 2022: Type C meeting WRO issued. (b) (4) .
- April 29,2022: PreNDA meeting issued. Nonclinical package is adequate for NDA filing and based on the indication for APDS/PASLI, agreed that the 6-month transgenic (rasH2) mouse carcinogenicity study reports will be provided post approval.
- July 29, 2022: The NDA for JOENJA® (leniolisib) was submitted on July 29, 2022, and the application was filed on September 27, 2022, for priority review.

3 Studies Submitted

3.1 Studies Reviewed

Study #	Title
PHARMACOLOGY	
Primary pharmacology	
RD-2011-00084	In vitro pharmacology of the PI3K delta inhibitors NVPCDZ173 and NVP-CFL375
RD-2014-00588	Expression of gain-of-function mutants of PI3Kδ 110kDa and its inhibition by CDZ173
RD-2011-00469	The effect of PI3K delta inhibitors on antibody formation specific for sheep red blood cells using an ex vivo plaque forming cell assay
RD-2011-00419	Effect of the PI3-kinase delta inhibitor, NVP-CDZ173, in an Ozone model of acute lung inflammation
RD-2011-00108	Anti-IgM/IL-4-stimulated B cell activation (CD69, CD86) in 90% human whole blood: Inhibition by PI3K delta-specific compounds
Secondary pharmacology	

RD-2011-00084	In vitro pharmacology of the PI3K delta inhibitors NVPCDZ173 and NVP-CFL375
PK/ADME	
1100674	In vitro blood distribution and plasma protein binding of [14C]CDZ173 including stability in blood and plasma of mouse, rat, dog, cynomolgus and human
1300432	Assessment of CDZ173 hepatobiliary disposition in vitro using sandwich-cultured human hepatocytes
1500236	Assessment of melanin binding parameters of CDZ173
1600175	Investigation of covalent protein binding of CDZ173 or metabolites in monkey ADME plasma samples
1100672	In vitro metabolism of [14C]CDZ173 in mouse, rat, dog, monkey and human hepatocytes and in human liver microsomes
1100170	In vitro assessment of cytochrome P450 enzyme inhibition by CDZ173
1100152	Assessment of efflux transporter (P-gp, BCRP) inhibition by CDZ173
1100153	Assessment of uptake transporter (OATP1B1, OATP1B3) inhibition by CDZ173
1100154 and 1100154-01a	Assessment of uptake transporter (OAT1, OAT3) inhibition by CDZ173
1100155	Assessment of uptake transporter (OCT1, OCT2) inhibition by CDZ173
1400476	Assessment of SLC transporter (MATE1, MATE2K) inhibition by CDZ173
1500167	QWBA in male pigmented rats after a single oral dose of [14C]CDZ173 phosphate (10 mg base/kg)
1100673	Absorption, distribution, metabolism, and excretion in male rats after a single intravenous (3 mg/kg) or oral (10 mg/kg) dose of [14C]CDZ173
1500290	Absorption, metabolism and excretion following an intravenous (3 mg/kg) or oral dose (20 mg/kg) of [14C]CDZ173 in the monkey
1100476	
SPECIAL TOXICOLOGY STUDIES	
1670143	CDZ173: In Vitro Balb/c 3T3 Neutral Red Uptake Phototoxicity Assay
1770661	A 1-Day topical ocular tolerability study in male rabbits
Non-GLP in vitro impurity studies including 1112543, 1412577, 1419399, 1419436 are discussed in the impurities section 2.5 above. Most of the dose range-finding repeat dose studies were reviewed previously under IND 124045 and IND (b) (4) The pivotal 39-week oral toxicity study in monkeys and 26-week oral toxicity study in rats were reviewed by Wei Sun, PhD/David Klein, PhD/Matthew Whittaker, PhD under IND 124045 and will be discussed in the unireview instead of this NDA review.	

3.2 Studies Not Reviewed

RD-2011-00135	PI3Kdelta selective inhibitor NVP-CDZ173-NX-8 is efficacious in a rat collagen-induced arthritis model
RD-2014-00785	Activity of PI3Kdelta selective inhibitor NVP-CDZ173 in a therapeutic rat collagen-induced arthritis model
RD-2011-00087	Relationship of PK and ex vivo-stimulated B cell activation in rats after a single oral dose of the PI3K delta-inhibitor NVP-CDZ173
RD-2011-00088	Relationship of PK and ex vivo-stimulated B cell activation in cynomolgus monkeys after a single oral dose of the PI3K delta-inhibitor NVP-CDZ173

1070424	Glucose and insulin tolerance test in C57/BL/6 mice
1200813	Structure identification of CDZ173 metabolites synthesized via bioreactions
1700820	Ex Vivo stability and metabolism of CDZ173 in feces
1200021	Identification of cytochrome P450 enzymes involved in the primary oxidative metabolism of CDZ173 in HLM
1100733	In vitro metabolism of [14C]CDZ173: Assessment of (b) (4) formation
1600611-01	Stability assessment of N-oxide metabolite LNB838 (M9) in human blood and plasma
1700820	Ex Vivo stability and metabolism of CDZ173 in feces
1800370	Metabolite structure alignment across CDZ173 biotransformation studies
1100169	Evaluation of CDZ173 to activate human PXR and human AhR in CYP3A4 and CYP1A2 reporter gene assays
1300388	Evaluation of CDZ173 as inducer of drug metabolizing enzymes in human hepatocytes
1100380	Salt selection support in rats after oral administration of 30 mg/kg CDZ173 as free base, as (b) (4) and as phosphate salt
1015544	in vitro 3T3 NRU Phototoxicity Profiling Assay (non-GLP)
GLPG0634-TX-020	Investigative Testicular Toxicity Study in Rats (with Genomics) by Oral (Gavage) Route
1270476	Investigation of thyroid liabilities with PI3 Kinase δ inhibitor CDZ173 using an in vitro model employing rat FRTL-5 cells
Additional PK/ADME method validation reports are not discussed as well.	

3.3 Previous Reviews Referenced

None.

4 Pharmacology

4.1 Primary Pharmacology

Leniolisib Biochemical Assays

Study No. RD-2011-00084: In vitro pharmacology of the PI3K delta inhibitors NVPCDZ173 and NVP-CFL375

Key Study findings:

- In transscreener time-resolved fluorescence resonance energy transfer (TR-FRET) kinase assays for monitoring ADP formation, leniolisib inhibited recombinant enzymes PI3Kδ and PI3Kγ with IC_{50}s of 0.011 and 2.57 μM, respectively. In another luminescence assays for monitoring ATP consumption, leniolisib inhibited recombinant enzymes PI3Kα and PI3Kβ with IC_{50}s of 0.28 and 0.48 μM, respectively. Leniolisib selectively inhibited the lipid kinase PI3Kδ with an IC_{50} at least 28-fold over other Class I PI3K isoforms.

Table 5 Human PI3K Inhibition IC_{50} Values for Leniolisib in Biochemical Assays

Enzyme isoform	IC_{50} (μM)	Selectivity of PI3Kδ over other Class I isoforms
PI3Kδ a	0.011 ± 0.004	-
PI3Kα b	0.28 ± 0.11	28-fold
PI3Kβ b	0.48 ± 0.28	43-fold
PI3Kγ a	2.57 ± 2.13	257-fold

a = ADAPTA format; b = KGlo format.

Leniolisib Cell-based Assays

Study No. RD-2011-00084: In vitro pharmacology of the PI3K delta inhibitors NVPCDZ173 and NVP-CFL375
Study No. RD-2014-00588: Expression of gain-of-function mutants of PI3Kδ 110kDa and its inhibition by CDZ173
Study No. RD-2011-00108: Anti-IgM/IL-4-stimulated B cell activation (CD69, CD86) in 90% human whole blood: Inhibition by PI3K delta-specific compounds

Key Study findings:

- In cell-based kinase assays for monitoring PI3K-dependent phosphorylation of Akt in Rat-1 cells transfected with human myr-p110 PI3K isoforms, leniolisib inhibited PI3Kδ, PI3Kα, and PI3Kβ with IC_{50}s of 0.056, 1.62, and 2.37 μM, respectively.
- In cell-based kinase assays for monitoring PI3Kγ dependent activation of U937 monocytes or activation of mouse bone marrow-derived mast cells (BMMCs), leniolisib inhibited PI3Kγ with IC_{50}s of 7.76 or >7.42 μM, respectively.
- In the Rat-1 cell line expressing mutant PI3Kδ (i.e., N334K, C416R, E525K, or E1021K) with elevated levels of pAkt, leniolisib inhibited enhanced activity of the mutant enzymes with IC_{50} values between 0.205 μM for the wt and 0.05 μM for the C416R mutant.
- In whole blood assays in rats, monkeys, and humans and splenocytes in mice, leniolisib inhibited both activation (i.e., phosphorylation of Akt, expression of CD69 and CD 86) and proliferation of B cells from nanomolar to sub micromolar range.
- In mouse splenocytes and human PBMCs, leniolisib inhibited T cell proliferation and differentiation in the sub micromolar range.
- In human PBMC and whole blood and mouse bone marrow-derived mast cells, leniolisib inhibited the oxidative burst of human neutrophils and monocytes and activation of mouse mast cells in the sub micromolar range.

Table 6 Human PI3K Inhibition IC_{50} Values for Leniolisib in Cell-based Assays

Enzyme isoform	IC_{50} (μM)	Selectivity of PI3Kδ over other Class I isoforms

PI3Kδ a	0.056 ± 0.02	-
PI3Kα a	1.62 ± 0.29	29-fold
PI3Kβ a	2.37 ± 0.66	42-fold
PI3Kγ b	7.76 ± 3.08	138-fold
PI3Kγ c	>7.42 ± 4.4	>132-fold

a = Rat-1 fibroblasts transfected with human myr-p110 PI3K isoforms; b = Adenosine-induced mouse bone marrow-derived masT-Cells (PI3Kγ dependent); c = MIP-1α induced U937 monocytes;

Table 7 Human mutant PI3K Inhibition IC_{50} Values for Leniolisib in Cell-based Assays

Enzyme isoform	IC_{50} (μM)
wtPI3Kδ	0.205 ± 0.064
N334K	0.087 ± 0.034
E525K	0.128 ± 0.054
E1021K	0.064 ± 0.015
C416R	0.05 ± 0.015
RAT-1 cells (control)	0.891 ± 0.267

Table 8 Human, Monkey, Rat, and murine B-cell Function Inhibition IC_{50} Values for Leniolisib in Whole Blood or Spenocytes Assays

Species	Mouse spleen		Rat Blood		Monkey Blood	Human Blood		
	CD86	Proliferation of B cell	CD86	pAkt	pAkt	CD69	CD86	pAkt
IC_{50} (μM)	0.048 ± 0.008	0.008 ± 0.006	0.099 ± 0.021	0.150 ± 0.059	0.084 ± 0.028	0.193 ± 0.117	0.202 ± 0.053	0.144± 0.013

Table 9 Human and murine T-cell Function Inhibition IC_{50} Values for Leniolisib in PBMCs or Spenocytes

Species	Mouse				Human		
	IFNγ	IL-13	IL-17	Proliferation	IL-13	IL-17	Proliferation
IC_{50} (μM)	0.130	0.010	0.101	0.033 ± 0.01	0.095 ± 0.084	0.073 ±0.034a	0.079 ± 0.052

*Mixed lymphocyte reaction (MLR) is a model for allogeneic (Allo) T cell activation *in vitro*.

Table 10 Human and murine innate immune system Inhibition IC_{50} Values for Leniolisib in PBMCs or bone marrow-derived mast cells

Species	Mouse	Human PBMC	Human Whole Blood		
	mast cell	Isolated pDC	Basophil	Neutrophil	Monocytes
	pAkt	IFNα	CD63	Oxidative burst	
IC_{50} (μM)	0.042 ± 0.048	0.033 ± 0.003	0.424 ± 0.081	0.061 ± 0.073	<0.100

Leniolisib In-vivo Assays

Currently, there is no validated APDS disease model in animals. The Sponsor explored antibody response to sheep red blood cells (SRBC) in rat and ozone-induced neutrophil infiltration in mouse bronchoalveolar lavage (BAL).

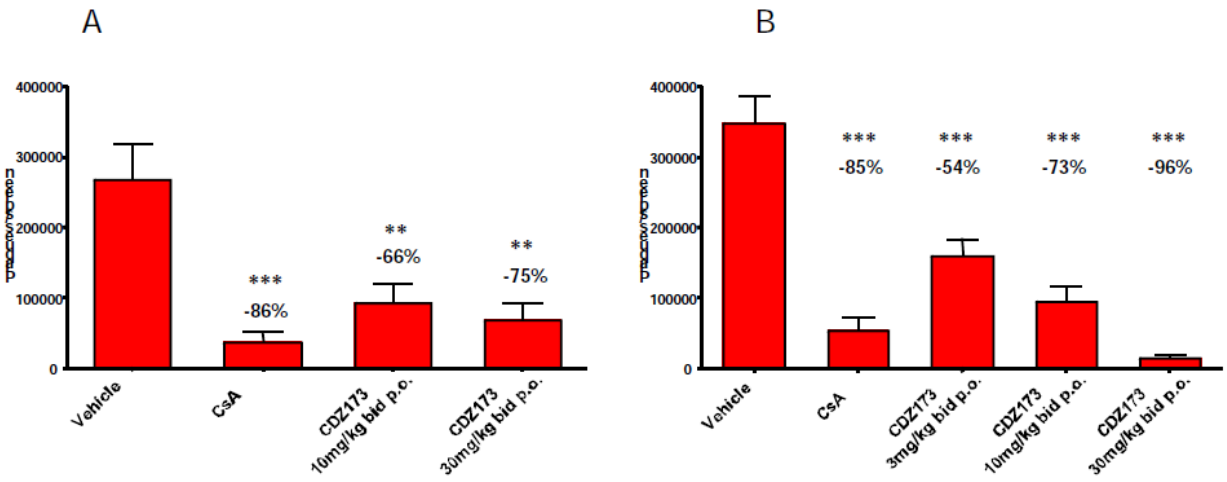
Study No. RD-2011-00469: The effect of PI3K delta inhibitors on antibody formation specific for sheep red blood cells using an ex vivo plaque forming cell assay.

Key Study findings:

- In a 5-day Antibody response to sheep red blood cells (SRBC) model, immunized rats received orally BID doses of 0, 3, 10, and 30 mg/kg leniolisib from Day 0 to day 4. Leniolisib inhibited T-cell dependent specific antibody response (i.e., anti-SRBC producing plaque-forming cells (PFC)) in a dose-dependent manner.

Figure 1 Inhibition of leniolisib on antibody response to sheep red blood cells (SRBC) in rat

Figure 3-1 Effect of CDZ173 on antibody formation to SRBC in rats



OFA rats were treated in 2 independent experiments with CDZ173-NX-3 (A) and CDZ173-NX-5 (B) as described in Section 2.5 and an *ex-vivo* assay described in Section 2.6 was carried out to assess antigen-specific antibody responses. Bars denote mean plaque number/group ± SEM and inhibition levels are denoted. Data was analyzed using the Anova/Dunnett using exact p values where *: $p<0.05$, **: $p<0.01$, ***: $p<0.001$, ns: not significant.

(Excerpted from Applicant's submission, SD1-M4-4211- rd-2011-00469, pg9)

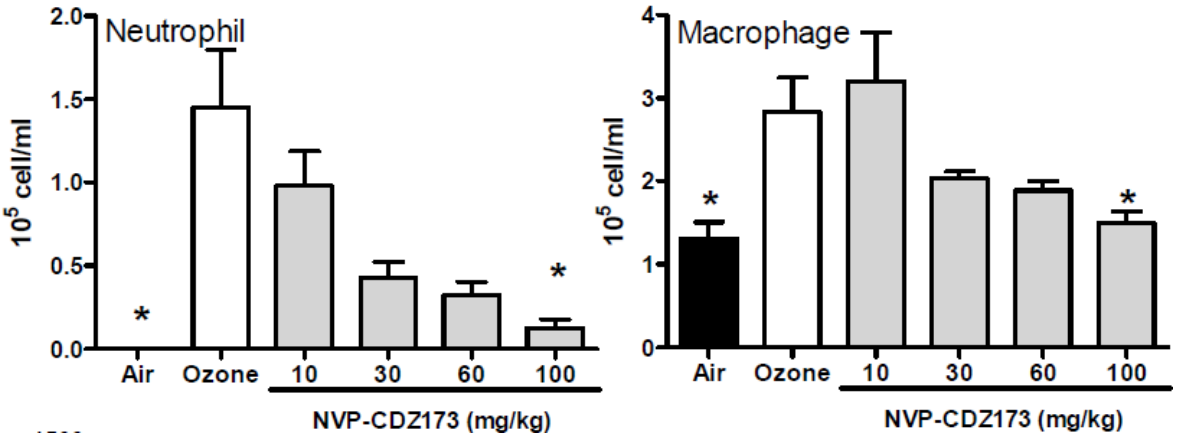
Study No. RD-2011-00419: Effect of the PI3-kinase delta inhibitor, NVP-CDZ173, in an Ozone model of acute lung inflammation.

Key Study findings:

- In a C57BL/6 model of ozone-induced acute lung inflammation, mice received orally doses of 0, 10, 30, 60, and 100 mg/kg leniolisib at one hour before and four hours after ozone exposure. Leniolisib inhibited ozone induced increase in BAL neutrophil and macrophage numbers in a dose-dependent manner.

Figure 2 Inhibition of leniolisib on airway inflammation in Ozone model in mice

Figure 3-1 NVP-CDZ173 dose-response against ozone-induced airway inflammation in C57BL/6J mice



(Excerpted from Applicant's submission, SD1-M4-4211- rd-2011-00419, pg7)

4.2 Secondary Pharmacology

Study No. RD-2011-00084: In vitro pharmacology of the PI3K delta inhibitors NVPCDZ173 and NVP-CFL375

Key Study findings:

- In ATP consumption (KinaseGlo) and antibody-dependent TR-FRET recombinant enzyme assays, CDZ173 did not show inhibitory activity on a panel of 4 other enzymes in the PI3K family (Vps34, mTOR, DNA-PK, PI4K).

- In KINOMEScan against a total of 442 serine/threonine, tyrosine and lipid kinases and a Novartis Protease panel (a panel of proteases and 47 serine/threonine and tyrosine kinases), 10 μM CDZ173 did not inhibit any kinases or proteases, with the exception of Class I PI3K isoform of RPS6KA5 (76% inhibition).

- In the cardiac ion channels Nav1.5 and Cav1.2a and against the P5 receptor panel and Ricerca/MDS panel against a panel of 82 receptors and ion channels, 10 μM leniolisib showed inhibition of the G-protein coupled receptor GPR8 (72% inhibition). In the Novartis P5 receptor profile, CDZ173 had an IC50 of 4.7 μM for the hPDE4D receptor and 7.7 μM for the 5HT2B receptor.

Table 11 Human PI3K family Inhibition IC_{50} Values for Leniolisib in biochemical Assays

Enzyme isoform	IC_{50} (μM)	Selectivity of PI3Kδ
Vps34	>9.1	>910-fold

mTOR	>9.1	>910-fold
DNA-PK	0.88	88-fold
PI4K	>9.1	>910-fold

4.3 Safety Pharmacology

Safety pharmacology studies were reviewed previously under IND 124045.

5 Pharmacokinetics/ADME/Toxicokinetics

5.1 PK/ADME

Study No. 1100674, 1300432, 1500236, 1600175, 1100672, 1200813, 1100733, 1200021, 1600611-01, 1700820, 1800370, 1100169, 1100170-01, 1300388, 1100152, 1100153, 1100154, 1100154-01a, 1100155, 1400476, 1500167, 1100673, 1100380.

Key Study findings:

- Leniolisib is rapidly absorbed after oral gavage with a mean T_{max} of 0.25-0.5 hour in mice, 0.25 hour in rats and 2.6 hours in monkeys. In rats, terminal plasma $T_{1/2}$ of total radioactivity was approximately 33.4 hours after oral gavage dosing. The plasma clearance was approximately 15 mL/min/kg in rats and monkeys, while the blood clearance was moderate to high compared to the hepatic blood flow (i.e., 23%-44%).

- Leniolisib has a low first-pass effect and a moderate to strong binding to plasma proteins (i.e., fraction unbound: 12.4% in mice, 8.2% in rats, 3.8% in dogs, 9.9% in monkeys and 5.5% in humans) in a concentration independent manner.

- The oral bioavailability was 59% in rats and ~100% in monkeys.

- In radioactive studies, leniolisib was moderately distributed into tissues (i.e., Vss of 1.06 L/kg in rats and 0.657 L/kg in monkeys) with a high skin affinity. After a single oral dose of 10 mg/kg dose to male rats, the highest exposures were found in the bile, hair follicle, liver, intestinal wall, kidney as well as in melanin-containing structures.

- In mouse, rat, dog, monkey, and human hepatocytes, the biotransformation of leniolisib occurred mainly via phase I metabolism (i.e., at the tetrahydropyridopyrimidine and the pyrrolidine parts of the molecule).

- In rats and monkeys, the major route of excretion of [14C] leniolisib was via the feces (approximately 91% and 51-58% of the radioactive dose by oral gavage).

- Characterization and structural elucidation of metabolites in plasma, urine or bile from nonclinical species have demonstrated the similarities in the metabolic

profiles between these species and humans. It is noted that no major human metabolites were identified in completed clinical studies.

- Leniolisib did not inhibit efflux transporter Pgp, but inhibited BCRP (IC_{50} = 18.9 µM) *in vitro*. Leniolisib inhibited OATPB1 (IC_{50} = 3.0 µM), OATPB3 (IC_{50} = 13.4 µM), OAT3 (IC_{50} = 15.2 µM), and OCT2 (IC_{50} = 3.4 µM), but not OCT1 and OAT1 transporters. Leniolisib inhibited the human multidrug and toxin extrusion protein family MATE1 and MATE2K *in vitro* (MATE1 IC_{50}=6.70 µM and MATE2K IC_{50}=0.85 µM).

5.2 Toxicokinetics

Toxicokinetic studies were discussed in general toxicology studies.

6 General Toxicology

General toxicity studies were discussed in previous PT reviews and will be summarized in the unireview.

7 Genetic Toxicology

Genetic toxicity studies were discussed in previous PT reviews and will be summarized in the unireview.

8 Carcinogenicity

No carcinogenicity studies were conducted yet.

9 Reproductive and Developmental Toxicology

Reproductive and developmental toxicology studies were discussed in previous PT reviews and will be summarized in the unireview.

10 Special Toxicology Studies

Study No. 1500236: Assessment of melanin binding parameters of CDZ173

In an *in vitro* melanin binding assay, [^{3}H]leniolisib bound to synthetic melanin at two binding sites with Kd1 of 36.6 µM and Kd2 of 1.8 µM, respectively, suggesting prolonged retention in the posterior eye cup is to be expected.

Table 4-1 Melanin binding parameters of CDZ173

	Bmax1 nmol/mg	Kd1 µM	Bmax2 nmol/mg	Kd2 µM	CR1 Bmax1/Kd1	CR2 Bmax2/Kd2	50% rule
	122.7	36.6	15.1	1.8	3.4	8.6	yes

Excerpted from Applicant's submission, SD1-M4-4223-1500236, pg12.

Study No. 1770661: A 1-Day topical ocular tolerability study in male rabbits

In a one-day ocular tolerability study, male rabbits (3/group) received ophthalmology suspension of leniolisib at concentrations of 0.5%, 2% or 5% by dropper into the right eye. One animal in the 2% group and two animals in the 5% group showed a minimal and transient conjunctival hyperemia which resolved by 24 hours post last dose. All fluorescein staining results at the 24-hour examination were negative.

Phototoxicity:
Study No. 1670143: CDZ173: In Vitro Balb/c 3T3 Neutral Red Uptake Phototoxicity Assay (GLP)

The UV spectrum of leniolisib shows absorption at a 290 nm wavelength and a secondary absorption maximum at a 330 nm wavelength. The molar extinction coefficients at these wavelengths were 2,261 and 2,042 L mol-1 cm-1, respectively. In the follow-up GLP compliant *in vitro* Balb/c 3T3 neutral red uptake phototoxicity assay, leniolisib was not phototoxic at up to 100 µg/mL. The risk of phototoxicity of leniolisib is considered minimum according to ICH S10.

11 Integrated Summary and Safety Evaluation

Pharming Technologies B.V. submitted an Original 1 (type 1-NME) NDA submission NDA on July 29, 2022, for JOENJA® (leniolisib), which is proposed for the treatment of activated phosphoinositide 3-kinase delta (PI3Kδ) syndrome (APDS) in adults and adolescents aged 12 or older. This NDA review evaluates the impurity, primary pharmacology, secondary pharmacology, PK/ADME, and phototoxicity studies to support the safety of leniolisib for approval. Summaries of all relevant Pharmacology and Toxicology studies will be provided in the unireview.

Impurities:
The Sponsor followed the recommendations of the ICH M7 Guideline and evaluated the mutagenic potential of impurities based on literature, internal databases, and in silico assessment including both expert rule-based (i.e., Derek Nexus (v.4.0.5, V4.1.0 or v.5.0.2)) and statistical-based systems (i.e., Case Ultra (v.1.5.0.1, v.1.5.2.0 or v.1.6.0.3) and/or Sarah Nexus (v.1.1.2, v1.2.0 or v.2.0.1)). A total of six mutagenic or potentially mutagenic impurities were identified: (b) (4) The Sponsor conducted Ames assays for (b) (4) Both impurities were negative in the Ames test. The other four impurities were either likely eliminated (b) (4) or controlled according to ICH M7.

(b) (4)

Other impurities that were identified are appropriately controlled per ICH Q3A, ICH Q3B, or United States Pharmacopeia (USP) Guidelines.

Overall, the proposed impurity specifications are acceptable and there are no outstanding nonclinical safety concerns.

Pharmacology:
Primary Pharmacology:
In biochemical assays, leniolisib inhibited recombinant enzymes PI3Kα and PI3Kβ with IC50s of 0.28 and 0.48 μM, respectively. Leniolisib selectively inhibits the lipid kinase PI3Kδ with at least 28 over other Class I PI3K isoforms.

In cell-based assays, leniolisib inhibited PI3Kδ, PI3Kα, and PI3Kβ with IC50s of 0.056, 1.62, and 2.37 μM, respectively. Leniolisib inhibited PI3Kγ with IC50s of 7.76 or >7.42 μM, respectively. In the Rat-1 cell line expressing mutant PI3Kδ (i.e., N334K, C416R, E525K, or E1021K) with elevated levels of pAkt, leniolisib inhibited enhanced activity of the mutant enzymes with IC50 values between 0.205 μM for the wt and 0.05 μM for the C416R mutant. In whole blood assays in rats, monkeys, and humans and splenocytes in mice, leniolisib inhibited both activation (i.e., phosphorylation of Akt, expression of CD69 and CD 86) and proliferation of B cells from nanomolar to sub micromolar range. In mouse splenocytes and human PBMCs, leniolisib inhibited T cell proliferation and differentiation in the sub micromolar range. In human PBMC and whole blood and mouse bone marrow-derived mast cells, leniolisib inhibited oxidative burst of human neutrophils and monocytes and activation of mouse mast cells in the sub micromolar range.

Currently, there is no validated APDS disease model in animals. The Sponsor explored antibody response to sheep red blood cells (SRBC) in rat and ozone-induced neutrophil infiltration in mouse bronchoalveolar lavage (BAL). In a 5-day Antibody response to sheep red blood cells (SRBC) model, leniolisib inhibited T-cell dependent specific antibody response (i.e., anti-SRBC producing plaque-forming cells (PFC)) in a dose-dependent manner. In a C57BL/6 model of ozone-induced acute lung inflammation, leniolisib inhibited ozone induced increase in BAL neutrophil and macrophage numbers in a dose-dependent manner.

Secondary Pharmacology:
CDZ173 did not show inhibitory activity on a panel of 4 other enzymes in the PI3K family (Vps34, mTOR, DNA-PK, PI4K). In KINOMEScan against a total of 442 serine/threonine, tyrosine and lipid kinases and a Novartis Protease panel (a panel of proteases and 47 serine/threonine and tyrosine kinases), 10 μM CDZ173 did not inhibit any kinases or proteases, with the exception of Class I PI3K isoform of RPS6KA5 (i.e., 76% inhibition). In the cardiac ion channels Nav1.5 and Cav1.2a and against the P5 receptor panel and (b) (4)/MDS panel against a panel of 82 receptors and ion channels, 10 μM leniolisib showed inhibition of the G-protein coupled receptor GPR8 (72% inhibition). In the Novartis P5 receptor profile, CDZ173 had an IC50 of 4.7 μM for the hPDE4D receptor and 7.7 μM for the 5HT2B receptor.

PK/ADME:
Leniolisib is rapidly absorbed after oral gavage with a mean T_{max} of 0.25-0.5 hour in mice, 0.25 hour in rats and 2.6 hours in monkeys. In rats, terminal plasma $T_{1/2}$ of total radioactivity was approximately 33.4 hours after oral gavage dosing. The plasma clearance was approximately 15 mL/min/kg in rats and monkeys, while the blood clearance was moderate to high compared to the hepatic blood flow (i.e., 23%-44%). Leniolisib has a low first-pass effect and a moderate to strong binding to plasma proteins (i.e., fraction unbound: 12.4% in mice, 8.2% in rats, 3.8% in dogs, 9.9% in monkeys and 5.5% in humans) in a concentration independent manner. The oral bioavailability was 59% in rats and ~100% in monkeys.

In radioactive studies, leniolisib was moderately distributed into tissues (i.e., Vss of 1.06 L/kg in rats and 0.657 L/kg in monkeys) with a high skin affinity. After a single oral dose of 10 mg/kg dose to male rats, the highest exposures were found in the bile, hair follicle, liver, intestinal wall, kidney as well as in melanin-containing structures.

In mouse, rat, dog, monkey, and human hepatocytes, the biotransformation of leniolisib occurred mainly via phase I metabolism (i.e., at the tetrahydropyridopyrimidine and the pyrrolidine parts of the molecule). Characterization and structural elucidation of metabolites in plasma, urine or bile from nonclinical species have demonstrated the similarities in the metabolic profiles between these species and humans. It is noted that no major human metabolites were identified in completed clinical studies.

In rats and monkeys, the major route of excretion of [14C] leniolisib was via the feces (approximately 91% and 51-58% of the radioactive dose by oral gavage).

In in vitro assays, leniolisib inhibited BCRP with an IC_{50} of 18.9 µM. Leniolisib did not inhibit efflux transporter Pgp. Leniolisib inhibited OATPB1 (IC_{50} = 3.0 µM), OATPB3 (IC_{50} = 13.4 µM), OAT3 (IC_{50} = 15.2 µM), and OCT2 (IC_{50} = 3.4 µM). Leniolisib did not OCT1 and OAT1. Leniolisib inhibited the human multidrug and toxin extrusion protein family MATE1 and MATE2K (MATE1 IC_{50}=6.70 µM and MATE2K IC_{50}=0.85 µM).

Special studies and Phototoxicity:
In an *in vitro* melanin binding assay, [^{3}H]leniolisib bound to synthetic melanin at two binding sites with Kd1 of 36.6 µM and Kd2 of 1.8 µM, respectively, suggesting prolonged retention in the posterior eye cup is to be expected. In a one-day ocular tolerability study, male rabbits (3/group) received ophthalmology suspension of leniolisib at concentrations of 0.5%, 2% or 5% by dropper into the right eye. One animal in the 2% group and two animals in the 5% group showed a minimal and transient conjunctival hyperemia which resolved by 24 hours post last dose. All fluorescein staining results at the 24-hour examination were negative. In addition, there were no test article-related ophthalmic or histopathologic findings in the eyes of treated animals in the 39-week repeat dose toxicity study in monkeys or in the 26-week repeat dose toxicity study in rats.

The UV spectrum of leniolisib shows absorption at a 290 nm wavelength and a secondary absorption maximum at a 330 nm wavelength. The molar extinction coefficients at these wavelengths were 2,261 and 2,042 L mol-1 cm-1, respectively. In the follow-up GLP compliant *in vitro* Balb/c 3T3 neutral red uptake phototoxicity assay, leniolisib was not phototoxic at up to 100 µg/mL. The risk of phototoxicity of leniolisib is considered minimum according to ICH S10.

12 Appendix/Attachments

None

This is a representation of an electronic record that was signed electronically. Following this are manifestations of any and all electronic signatures for this electronic record.

/s/

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12/02/2022 03:48:28 PM

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I concur