Clinical Research Protocol

A Phase 2a (Proof of Concept), Randomized, Double-Blind, Placebo-Controlled Multicenter Study to Evaluate the Efficacy and Safety of AT1001 for the Treatment of COVID-19 Related Multisystem Inflammatory Syndrome in Children (MIS-C)

Protocol Number:	AT1001 MIS-C 101
Investigational Product:	AT1001 and matching placebo
IND Number:	155614
Development Phase:	Phase 2a
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Protocol Version Date:	14 January 2022

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SPONSOR APPROVAL

A Phase 2a (Proof of Concept), Randomized, Double-Blind, Placebo-Controlled Multicenter Study to Evaluate the Efficacy and Safety of AT1001 for the Treatment of COVID-19 Related Multisystem Inflammatory Syndrome in Children (MIS-C)

This study protocol has been reviewed and approved by Alessio Fasano, MD. It is confirmed that the information and guidance given in this protocol complies with scientific principles, the guidelines of Good Clinical Practices, applicable principles that have their origins in the Declaration of Helsinki, and the applicable legal and regulatory requirements.

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Date

PROTOCOL AGREEMENT

I have read the protocol specified below. In my formal capacity as Investigator, my duties include ensuring the safety of the study participants enrolled under my supervision and providing Sponsor with complete and timely information, as outlined in the protocol. It is understood that all information pertaining to the study will be held strictly confidential and that this confidentiality requirement applies to all study staff at this site. Furthermore, on behalf of the study staff and myself, I agree to maintain the procedures required to carry out the study in accordance with accepted GCP principles and to abide by the terms of this protocol.

Protocol Number: AT1001 MIS-C 101

Protocol Title: A Phase 2a (Proof of Concept), Randomized, Double-Blind, Placebo-Controlled Multicenter Study to Evaluate the Efficacy and Safety of AT1001 for the Treatment of COVID-19 Related Multisystem Inflammatory Syndrome in Children (MIS-C)

Protocol Date: 14 January 2022

Lael Yonker 14 Jan 22

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Table of Contents

Sponsor Approval 2

PROTOCOL AGREEMENT 3

List of Abbreviations 9

Protocol Synopsis 11

1 Introduction 20

1.1 Background 20
1.1.1 COVID-19 Associated with MIS-C 20

1.2 Intracellular Tight Junctions 20

1.3 Role of Zonulin in Specific Diseases 21
1.3.1 Zonulin Inhibitor AT1001 23

1.4 Overview of Non-Clinical Studies with AT1001 24
1.4.1 Nonclinical Pharmacology and Pharmacology 24
1.4.2 Toxicity Data for AT1001 25

1.5 Overview of Clinical Studies with AT1001 26
1.5.1 AT1001 in Clinical Studies 26
1.5.2 Pharmacokinetic Evaluation 27
1.5.3 Safety and Adverse Effects 27

1.6 Current Available Treatments 28

1.7 Study Rationale 28

1.8 Risk/Benefit Statement 29

2 STUDY OBJECTIVES and endpoints 30

2.1 Study Objectives 30
2.1.1 Primary Objective 30
2.1.2 Secondary Objectives 30

2.2 Study Endpoints 30
2.2.1 Primary Efficacy Endpoint 30
2.2.2 Secondary Efficacy Endpoints 30
2.2.3 Exploratory Efficacy Endpoints 31
2.2.4 Safety Evaluations 33

3 STUDY DESIGN 34

3.1 Study Overview 34

3.2 Stopping Criteria 34
3.2.1 Interruption of Dosing in an Individual Participant 34

3.2.2 Stopping Rules for Study Enrollment and Dosing....34
3.2.3 Study Treatment Discontinuation Criteria....35

4 PARTICIPANT SELECTION....36

4.1 Inclusion Criteria....36

4.2 Exclusion Criteria....37

4.3 Equitable Recruitment....37

5 Concurrent Medications....38

6 investigational product....39

6.1 Formulation and Appearance....39

6.2 Packaging and Labeling....39

6.3 Method of Assigning Participants to Treatment Groups....39

6.4 Blinding and Breaking the Blind....40

6.4.1 Dosage/Dosage Regimen....40
6.4.2 Dispensing....40
6.4.3 Drug Diary....40

6.5 Supply of Study Drug at the Site....41

6.5.1 Storage....41

6.6 Study Drug Accountability....41

6.7 Measures of Treatment Compliance....41

7 STUDY PROCEDURES AND GUIDELINES....42

7.1 Clinical Assessments....42

7.1.1 PedsQL Gastrointestinal Symptoms Scales....42
7.1.2 Assessment of MIS-C-related Symptoms....42
7.1.3 Assessment of Activity Limitations....42
7.1.4 Concomitant Medications....42
7.1.5 Demographics....43
7.1.6 Medical History....43
7.1.7 Physical Examination....43
7.1.8 Vital Signs....43
7.1.9 Electrocardiogram....43
7.1.10 Adverse Events....43

7.2 Clinical Laboratory Measurements....43

7.2.1 Hematology....43
7.2.2 Blood Chemistry Profile....44
7.2.3 Pregnancy Test....44
7.2.4 Urinalysis....44

7.3 Research Laboratory Measurements 44
7.3.1 SARS-CoV-2 44
7.3.2 Zonulin 44
7.3.3 Cytokines 44
7.3.4 Pharmacokinetic (PK) Sampling 44
8 EVALUATIONS BY VISIT 45
8.1 Visit 1 (Day 1; Screening/Baseline) 45
8.2 Visits 2-6 (Days 2-6) 45
8.3 Visits 7-9, (Week 1, Week 2 and Week 3 (Day 7 ± 3; Day 14 ± 3; Day 21 ± 3) 45
8.4 Visit 10 and 11 (Week 4 and Week 8) (Day 28 ± 3; Day 56 ± 3) 46
8.5 Visit 12 (Day 84 ± 3) 46
8.6 Visit 13 and 14 (Week 16 and Week 20) (Day 112 ± 3; Day 140 ± 3) 46
8.7 Visit 15 (Day 168 ± 3) End of Study or Early Withdrawal Visit 46
9 ADVERSE Experience REPORTING AND DOCUMENTATION 48
9.1 Adverse Events 48
9.2 AE Severity 48
9.3 AE Relationship to Study Drug 48
9.4 Serious Adverse Experiences (SAE) 49
9.4.1 Serious Adverse Experience Reporting 49
9.5 Expected Side Effects/Risks 49
9.6 Reporting Adverse Events (Including Serious Adverse Events) 50
10 DISCONTINUATION And Replacement of participants 51
10.1 Early Discontinuation of Study Drug 51
10.2 Withdrawal of Participants from the Study 51
10.3 Replacement of Participants 51
11 DATA SAFETY MONITORING 52
12 STATISTICAL METHODS AND CONSIDERATIONS 53
12.1 Data Sets Analyzed 53
12.2 Demographic and Baseline Characteristics 53
12.3 Analysis of Primary Endpoint 53
12.4 Analysis of Secondary Endpoints 53

12.5 Safety Analysis 53

12.6 Sample Size and Randomization 54

13 DATA COLLECTION, RETENTION AND MONITORING 55

13.1 Data Confidentiality 55

13.2 Data Collection Instruments 55

13.3 Data Management Procedures 56

13.4 Data Quality Control and Reporting 56

13.5 Archival of Data 56

13.6 Availability and Retention of Investigational Records 56

14 ADMINISTRATIVE, ETHICAL, REGULATORY CONSIDERATIONS 58

14.1 Protocol Amendments 58

14.2 Institutional Review Boards 58

14.3 Informed Consent Form 59

14.4 Investigator Responsibilities 59

15 References 61

LIST OF TABLES

Table 1-1 Chronic Inflammatory Diseases in which Zonulin has been Implicated 22

Table 1-2 Summary of Phase 1 / Phase 2 Clinical Studies in Celiac Disease.....26

Table 9-1 Adverse Event Severity Grading.....48

Table 9-2 Adverse Event Relationship to Study Drug 49

LIST OF FIGURES

Figure 1-1 **"March" from Mucosal Anergy to Loss Of Epithelial Barrier Function** Leading **to Inflammation**.....23

LIST OF ABBREVIATIONS

ADME	absorption, distribution, metabolism, and excretion
AE	adverse event
ALT	alanine aminotransferase
AST	aspartate aminotransferase
BUN	blood urea nitrogen
BLQ	below the limit of quantitation
Caco-2 cells	immortalized cell line of human colorectal adenocarcinoma cells
CDC	Centers for Disease, Control and Prevention
CID	chronic inflammatory disease
CFR	Code of Federal Regulations
CRF	case report form
CRP	C-reactive protein
CTCAE	Common Terminology Criteria for Adverse Events
DSMB	Data Safety Monitoring Board
EAE	experimental autoimmune encephalomyelitis
eCRF	Electronic case report form
ESR	erythrocyte sedimentation rate
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GI	gastrointestinal
GGT	gamma-glutamyl transferase
HDPE	high-density polyethylene
hERG	human ether-a-go-go related gene
HIPAA	Health Insurance Portability and Accountability Act of 1996
HIV	human immunodeficiency disease
ICF	informed consent form
ICH	International Conference on Harmonization
ICU	intensive care unit
IEC6 cells	intestinal epithelial cells
IFN-γ	interferon gamma
IgA	immunoglobulin A
IgG	immunoglobulin G
IgM	immunoglobulin M
IL-1β	interleukin-1 beta
IL-2R	interleukin-2R
IL-6	interleukin 6

IL-8	interleukin 8
IL-17	interleukin 17
INR	international normalized ratio
IRB	Institutional Review Board
ITT	intent-to-treat
IV	intravenous
LDH	lactate dehydrogenase
LLOQ	lower limit of quantitation
MDCK cells	Madin-Darby Canine Kidney cells
MIS-C	multisystem inflammatory syndrome in children
NAFLD	nonalcoholic fatty liver disease
NT proBNP	N-terminal pro-brain natriuretic peptide
PI	principal investigator
PK	pharmacokinetic
PP	per protocol
QID	four times a day
RNA	ribonucleic acid
SAE	serious adverse event
SARS-CoV-2	severe acute respiratory syndrome coronavirus 2
SAS	statistical analysis system
SGOT	serum glutamic oxaloacetic transaminase
SGPT	serum glutamate pyruvate transaminase
Spike SAg	spike superantigen
TEAEs	treatment-emergent adverse events
TID	three times a day
TJs	tight junctions
TK	toxicokinetics
TNF-α	tumor necrosis factor alpha
ULN	upper limit of normal
ZO-1	zonula occludens 1
ZOT	zonula occludens toxin

PROTOCOL SYNOPSIS

Protocol Title	A Phase 2a (Proof of Concept), Randomized, Double-Blind, Placebo-Controlled, Multi-center Study to Evaluate the Efficacy and Safety of AT1001 for the COVID-19 related Treatment of Multisystem Inflammatory Syndrome in Children (MIS-C).
Protocol Number	AT1001 MIS-C 101
Sponsor	Alessio Fasano, MD; Massachusetts General Hospital, Boston, MA
Rationale and Background	COVID-19 is a serious and potentially life-threatening disease caused by the SARS-CoV-2 virus, infecting the human host for the first time in December 2019 before igniting a global pandemic with > 306 million confirmed cases and >5.49 million deaths reported to date. Contrary to early beliefs, children are not spared from COVID-19. For example, in Massachusetts children ≤ 19 years of age have accounted for the highest number of positive COVID-19 tests by age ever since the end of January 2021. Currently, over 8.5 million children in the U.S. have tested positive for COVID-19. Most children who are acutely infected with SARS-CoV-2 develop mild upper respiratory symptoms or experience asymptomatic infection. Several days to weeks after resolution of the initial infection, some of these children will develop a severe life-threatening illness, termed Multisystem Inflammatory Syndrome in Children (MIS-C), which is an immune activation syndrome associated with prior SARS-CoV-2 infection or exposure. Pediatric patients with MIS-C present with persistent fever, marked gastrointestinal (GI) symptoms, cytokine storm, myocardial dysfunction, and cardiogenic shock with ventricular dysfunction in the setting of multisystem inflammation, reminiscent of, yet distinct from, toxic shock syndrome or Kawasaki Disease (Rowley 2020). Eighty percent (80%) of these hospitalized children develop cardiac pathology (Feldstein 2020), including coronary artery dilations, myocardial dysfunction, or ventricular failure with hypotensive shock (Belhadjer 2020). Children with MIS-C display increased monocyte recruitment and hyperphagocytosis (Bartsch 2021) with cytokine storm (Pierce 2020), T-cell activation (Carter 2020), and an inflamed expansion of immunoglobulins (Bartsch 2021, Gruber 2020). A recently discovered superantigen-like motif near the S1/S2 cleavage site on the SARS-CoV-2 Spike protein (Cheng 2020, Porritt 2020b) is hypothesized to drive this hyperinflammatory response. However, most MIS-C patients do not have SARS-CoV-2 virus detected on reverse transcription-polymerase chain reaction (RT-PCR) of the nasopharyngeal swab (Yonker 2020). In contrast, SARS-CoV-2 is detected in the majority of MIS-C stool samples, with detectable viral loads ranging $1.5x10^2$ - $2.5x10^7$ RNA copies/mL, weeks after the initial infection/exposure (Yonker 2021). Detection of SARS-CoV-2 in the stool, in addition to the predominance of severe gastrointestinal (GI) symptoms in MIS-C patients, suggest that the viral or viral antigenic source at the time of presentation of MIS-C originates from the GI system. An intact, functional intestinal mucosal barrier should prevent the passage of large antigens from the gut lumen into the bloodstream, including viral antigens derived from SARS-CoV-2 present in the GI tract (Gaebler 2021). Zonulin is a family of structurally and functionally related proteins that reversibly regulate intestinal permeability by modulating intercellular tight junctions (Fasano 2000a; Fasano 2000b; Wang 2000). Increased circulating zonulin levels signaling increased

	intestinal permeability have been reported in several diseases (Fasano 2020) including auto-immune and hyper-inflammatory diseases such as celiac disease (Drago 2006), inflammatory bowel disease (Wyatt 1993), and Kawasaki Disease (Noval Rivas 2019). Zonulin release from epithelial cells results in permissibility of paracellular trafficking of large inflammatory antigens from the gut lumen into the bloodstream. Children with MIS-C have significantly increased release of zonulin into circulation as compared to healthy controls (Yonker 2021), suggesting a breakdown of mucosal barrier function. This loss of tight junctions' competency, coupled with the identification of SARS-CoV-2 within the stool, supports the hypothesis that GI sources of SARS-CoV-2 viral components breach the mucosal barrier and enter circulation. Zonulin may play a central role in MIS-C pathogenesis as well. Based on preliminary findings that, in MIS-C, increased serum zonulin levels are associated with increased detection of the SARS-CoV2 virus in their stools and its Spike SAg in their serum, the central hypothesis is that the presence of SARS-CoV-2 in the GI tract leads to dysbiosis, increased zonulin release, and subsequent increased gut permeability allowing SARS-CoV2 Spike SAg trafficking across mucosal barriers, triggering a pathogenic, hyperinflammatory response. AT1001 is a locally acting, non-systemic, octapeptide inhibitor of the zonulin receptor that has shown efficacy in a large variety of animal models of inflammation. The effectiveness of AT1001 in controlling paracellular permeability as a tight junction regulator has been widely demonstrated in animal models both *in vitro* and *in vivo*. No signs of toxicity were observed in animal models and there were no adverse effects in any of the pharmacological safety studies. The clinical program conducted by the pharmaceutical company with AT1001 (called larazotide) for the treatment of celiac disease included seven Phase 1/2 studies and one ongoing Phase 3 study. The results of these studies, including the ongoing Phase 3 study, have shown a good safety profile with an absence of serious side effects when administered via the mucosal route. As the prevalence of SARS-CoV-2 infections among children and adolescents is increasing (MMWR 2021), MIS-C cases are increasing as well. Current treatment strategies are primarily supportive care and directed care against the underlying inflammatory process but do not address mucosal permeability or antigenemia. Given the safety and tolerability profile and the potential benefit for pediatric patients with MIS-C, it is appropriate to study AT1001 as a treatment for MIS-C. This study is intended to assess the clinical safety, tolerability, and efficacy profile of AT1001 in hospitalized pediatric patients with MIS-C.
Objective	The overall objective is to evaluate the efficacy and safety of AT1001 versus placebo in pediatric patients with SARS-CoV-2 infection who experience early signs of MIS-C and are at high risk of progression.
Study Design	Phase 2a, randomized, double-blind, placebo-controlled, multicenter study to evaluate the efficacy and safety of AT1001 for use in hospitalized pediatric patients diagnosed with MIS-C. Eligible patients (N=20) will be treated with AT1001 or matching placebo orally four times a day (QID) for 21 days as an add-on to standard of care. The study includes three phases: • Screening (inclusion / exclusion criteria; informed consent): The main purpose of this phase is to determine if the participant meets entry criterion after obtaining informed consent.

	• Treatment: Eligible participants will be treated with AT1001 at a dose of 250 or 500 mcg or matching placebo QID for 21 days as an add-on to standard of care for MIS-C. • Follow-up through 24 weeks: The participant will return for a follow-up visits during Week 1 through 3, with monthly telemedicine visits at Week 4, Week 8, Week 16, and Week 20, and clinic visits at Week 12 and Week 24. Safety monitoring, including physical examination, vitals, and clinical laboratory testing will be performed during the screening phase, periodically during treatment phase and at the follow-up phase. Adverse events and concomitant medications will be recorded during the entire study.
Stopping Criteria	**Interruption of Dosing in an Individual Participant** The dosing of an individual participant may be interrupted under any of the following circumstances: • The participant experiences a serious adverse event related to the study drug • The participant experiences an intensity Grade 3 (severe) or greater AE as defined by the current version of the Common Terminology Criteria for Adverse Events (CTCAE) • Any other apparent deterioration in the health of the participant as determined by the investigator **Stopping Rules for Study Enrollment and Dosing** Participant randomization into the study treatment phase may be stopped or paused if any of the following occur: • Death of any participant that is deemed to be probably, or definitely related to study drug • A life-threatening adverse event in any participant that is deemed to be, probably, or definitely related to study drug • A safety signal requiring further evaluation to assess the overall benefit risk of the program **Study treatment discontinuation criteria:** 1. ALT or AST >3 times baseline with direct bilirubin > 2 times upper limit of normal (ULN) or international normalized ratio (INR) increased from baseline and is >1.5 times ULN, unless the Investigator or the data safety monitoring board (DSMB) determines the abnormality is unlikely related to study drug 2. Creatinine >1.8 times ULN and >2 times baseline, or decreased creatinine clearance to < 60 mL/min, unless the Investigator or the DSMB determines the abnormality is unlikely related to study drug. 3. Clinically significant Grade 4 adverse event or confirmed Grade 4 laboratory abnormality for an individual participant that is considered by the Investigator or the DSMB to be related to the study drug.
Planned Number of Participants	Approximately 20 pediatric participants will be enrolled into the study (10 AT1001: 10 Placebo)
Planned Study Sites	Up to five study sites

Study Entry Criteria	**Inclusion Criteria:** 1. Pediatric patients with or without comorbidity 2. Age ≥ 1 month to < 21 years 3. Confirmed MIS-C by signs and symptoms as detailed by the CDC Health Advisory (https://www.cdc.gov/mis-c/hcp/; May 14, 2020) a. Persistent fever/chills (>38.0°C for ≥24 hours, or report of subjective fever lasting ≥24 hours); **AND** b. One or more laboratory parameters (evidence of inflammation); **AND,** i. elevated C-reactive protein (CRP) ii. elevated erythrocyte sedimentation rate (ESR) iii. elevated ferritin iv. elevated lactic acid dehydrogenase (LDH) v. elevated d-dimer vi. elevated fibrinogen vii. elevated procalcitonin viii. elevated interleukin 6 (IL-6) ix. increased neutrophils x. reduced lymphocytes xi. low albumin c. Evidence of clinically severe illness requiring hospitalization, with multisystem (>2) organ involvement (cardiac, renal, respiratory, hematologic, gastrointestinal, dermatologic, or neurological) -- MUST include GI symptoms, such as nausea, vomiting, diarrhea, decreased appetite, and/or abdominal pain; **AND**, d. No alternative plausible diagnoses; **AND**, e. Positive for current or recent SARS-CoV-2 infection by RT-PCR, serology, or antigen test. 4. Participant (or legal authorized representative) capable of understanding and signing an informed consent form and assent form, if appropriate **Exclusion Criteria:** 1. Female participants pregnant and/or lactating. 2. Female participant has childbearing potential and is unwilling to use an acceptable method of birth control for the duration of the study. 3. Participant has a significant co-morbid disease that by the Investigator's determination would make the participant unsuitable for enrollment, including unstable medical conditions. 4. Participation in any other clinical investigation using an experimental drug within 30 days prior to screening or intends to participate in another clinical study while participating in AT1001 MIS-C 101 study. 5. Have participated in a blood/plasma donation or blood loss greater than 400 mL within 90 days, or greater than 200 mL within 30 days prior to Screening. 6. Known hypersensitivity to any of the formulation components of AT1001.
Investigational Product, Dose and Route of Administration	AT1001 at a dose of 250 or 500 µg/dose administered orally four times a day (QID) added to standard of care for MIS-C. Investigational drug will be administered either as capsules swallowed whole (for participants who can swallow a capsule) or as beads in soft food (for participants who cannot swallow a capsule) or a liquid vehicle (for participants with an enteral tubing).

Control Product, Dose, and Route of Administration	Matching placebo at a dose of 250 or 500 µg administered orally QID added to standard of care for MIS-C. Control product will be administered either as capsules swallowed whole (for participants who can swallow a capsule) or as beads in soft food (for participants who cannot swallow a capsule) or a liquid vehicle (for participants with an enteral tubing).
Duration of Participant Participation and Duration of Study	Total duration of the participants' participation in the study is approximately 24 weeks (with 21 days treatment period). Total duration of the study is projected to be 12 months, dependent on enrollment timeline.
Criteria for Evaluation	**Efficacy Assessments** • PedsQL GI symptoms scales • Body temperature/fever • Multisystem organ signs and MIS-C-related symptoms • Activity limitations • SARS-CoV2 Spike, S1 and nucleocapsid antigens • IgM, IgG, and IgA antibodies against SARS-CoV-2 • Cytokines: IFN-γ, IL-1β, IL-17, IL-2R • Inflammatory markers (CRP, d-dimer, ferritin) • Zonulin in serum • NT-proBNP and troponin (participants with cardiac system organ involvement) **Safety Assessments:** • Frequency and severity of treatment-emergent adverse events • Vital Signs (blood pressure, pulse, temperature, respiratory rate, weight) • Physical examination findings • ECG findings • Clinical laboratory parameters

Endpoints	Primary Efficacy Endpoint: Efficacy will be evaluated by primary endpoint at Day 21, namely:
	• To determine proportion of participants with improvement in MIS-C related GI symptoms and no progression of GI system involvement.
	  ○ Improvement in GI symptoms is defined as:
	  ▪ Improvement in PedsQL GI Symptoms Scales score ≥48 hours, as determined by patient or caregiver response.
	  ▪ AND improvement in clinical manifestation of GI symptoms, as documented by complete physical exam or clinical assessment and clinical laboratory tests or imaging.
	  ○ No Progression of MIS-C is defined as:
	  ▪ No involvement of additional organ involvement, as identified by clinical assessment and clinical laboratory tests or imaging.

	▪ AND no new onset of new or worsening GI symptoms for ≥48 hours including nausea, vomiting, diarrhea, loss of appetite, and/or abdominal pain, as determined by patient or caregiver symptom report or worsening PedsQL GI Symptoms Scales scores. Secondary Efficacy Endpoints through Day 21: • Improvement in MIS-C symptoms for ≥48 hours, as determined by patient or caregiver symptom report on MIS-C Symptom Questionnaire. • Length of stay in hospital (days from baseline to readiness to discharge). • Change from baseline in additional organ system(s) involvement during acute presentation/hospitalization, as identified by clinical assessment and clinical laboratory tests. • Need for escalation of care (eg, transfer from hospital ward to ICU; supplemental oxygen; mechanical ventilation). • Re-presentation to medical care for MIS-C related symptoms after discharge • Normalization of SARS-CoV-2 Spike, S1 and nucleocapsid • Change from baseline in levels of IgM, IgG, and IgA antibodies against SARS-CoV-2 • Change from baseline in levels of Inflammatory markers (CRP, d-dimer, ferritin) • Change from baseline in levels of zonulin • Change from baseline in NT-proBNP and troponin (participants with cardiac organ system involvement) • Mortality (all causes) Exploratory Efficacy Endpoints: • Change from baseline in levels of cytokines (IFN-γ, IL-1β, IL-17, IL-2R) • Change from baseline in levels of cytokines (IFN-γ, IL-1β, IL-17, IL-2R). • Time to restoration of usual state of health ≥ 48 hours as assessed by the MIS-C Activity Limitation Questionnaire. • Time to restoration of usual activity level ≥ 48 hours as assessed by the MIS-C Activity Limitation Questionnaire. • Time to resolution of COVID-19 related symptoms ≥ 48 hours as assessed by the MIS-C Activity Limitation Questionnaire (participants with active COVID-19 related symptoms). • Time to resolution of MIS-C related organ system involvement Safety Endpoints • Incidence, severity, and type of treatment-emergent adverse events

	• Change from baseline in vital sign measurements (blood pressure, pulse, temperature, respiratory rate, weight) • Changes from baseline in physical examination findings • Changes from baseline in clinical laboratory parameters
Statistical Methodology	**Efficacy Parameters**: For the efficacy endpoints, the primary analysis will be based on the intent-to-treat (ITT) population which is defined as all randomized participants. Per Protocol (PP) population will include all ITT participants who did not have any major protocol violations. The primary endpoint will be analyzed using Fisher's exact test. Robustness analysis of the primary efficacy endpoint will include an analysis on the PP population. Binary endpoints will be analyzed by the Fisher's exact test. Change from baseline in continuous outcomes will be analyzed by fitting fixed effects linear models or non-parametric models (e.g., Wilcoxon) to the data.
	Safety Parameters: The safety analysis will include all participants from the ITT population who received at least one dose of the study drug in the double-blind treatment period. The number and percent of participants experiencing TEAEs, serious adverse events, and TEAEs leading to study discontinuation treatment-emergent adverse events (TEAEs) will be tabulated and summarized according to the current version of CTCAE. Additional summaries will include TEAE severity and relationship to study drug. Individual and summary vital signs and clinical laboratory data will be presented in tabular form with mean, median, standard deviation, and range (minimum and maximum as appropriate). For the clinical laboratory safety data and vital signs values outside the reference range will be flagged in the data listings and a list of clinically significantly abnormal values will be presented.
Planned Interim Analysis	An independent data and safety monitoring board (DSMB) will actively monitor interim data to make recommendations regarding study conduct. The DSMB will meet once the first 10 participants complete the Day 7 assessment to perform the first interim analysis of the safety data. The interim analysis will be performed by an independent un-blinded statistician, who is not otherwise associated with the conduct of this study. The DSMB will consist of three members, including two independent physicians and a statistician. At least one physician will have expertise in infectious diseases, and one will have experience in DSMB activities. The DSMB responsibilities will be further elaborated in the DSMB charter. Serious adverse events will be monitored by the committee on an ongoing basis throughout the study.

SCHEDULE OF ASSESSMENTS AND PROCEDURES

Study Schedule of Assessments and Procedures

	Screening/ Baseline	**Treatment Period**				**Follow-Up Period**			**EOS**
Day	1	2-6	7 ± 3	14 ± 3	21 ± 3	28 ± 3 56 ± 3	84±3	112 ± 3 140 ± 3	168 ± 3
Week	--	--	1	2	3	4 / 8	12	16 / 20	24
Visit	1	2-6	7	8	9	10 / 11	12	13-14	15
Assessments / Procedures	IP	IP	IP CL	CL	CL	Tel	CL	Tel	CL
Informed Consent	X								
Demographics / BL characteristics	X								
Medical History	X								
Inclusion / Exclusion Criteria	X								
Physical exam[1]	X[1]	X[1]	X[1]	X[1]	X[1]		X[1]		X[1]
Weight, height, and vital signs[2]	X[2]	X[2]	X[2]	X[2]	X[2]		X[2]		X[2]
ECG[3]	X[3]	X[3]	X[3]	X[3]	X[3]		X[3]		X[3]
Clinical laboratory tests[4]	X[4]	X[4]	X[4]	X[4]	X[4]		X[4]		X[4]
Pharmacokinetics[5]		X[5]	*	*	*				
Assessment of MIS-C Related Symptoms	X	X	X	X	X	X	X	X	X
Assessment of limitation of activities	X	X	X	X	X	X	X	X	X
PedsQL GI Scales[6]	X[6]	X[6]	X[6]	X[6]	X[6]	X[6]	X[6]	X[6]	X[6]
Study treatment & accountability[7]	X[7]	X[7]	X[7]	X[7]	X[7]				
AE Assessment	X	X	X	X	X	X	X	X	X
Concomitant Medication	X	X	X	X	X	X	X	X	X

Abbreviations: AE = adverse event; CL = clinic visit; ECG = electrocardiogram; EOS, = end of study; IP = in-patient; MIS-C = Multisystem Inflammatory Syndrome in children; Tel = telemedicine;
* = applicable only for patients still inpatient.

[1] Physical examinations may be abstracted and filed from the subject's electronic medical record during inpatient study visits. Optional, symptom directed physical examinations may be performed during the outpatient portion of the study.

[2] Vital signs: blood pressure, pulse, respirations, temperature, and body weight at Day 1, Days 2-6, clinic visits Wk 1, Wk 2, Wk 3, Wk 12 and Wk 24 (EOS); height only collected at screening

[3] ECG: Obtained at Day 1, Days 2-6, Wk 1, Wk 2, Wk 3, Wk 12, and Wk 24 (EOS). ECGs may be performed if clinically indicated for more frequent monitoring during the study.

[4] Clinical laboratory tests will be performed as clinically indicated at the following timepoints: Chemistry, hematology, urinalysis at Day 1, every 2-3 days while inpatient, Wk 1, Wk 2, Wk 3, Wk 12, and Wk 24 (EOS); Inflammatory markers (CRP, d-dimer, ferritin) at Day 1, every 3 days until discharge, Wk 1, Wk 2, Wk 3, Wk 12 and Wk 24/EOS; Cytokines (IFN-γ, IL-1β, IL-17, IL-2R), zonulin, NT-proBNP and troponin at Day 1, every 2-3 days while inpatient, Wk 1, Wk 2, Wk 3, Wk 12 and Wk 24 (EOS); SARS-CoV-2 Spike, S1 and nucleocapsid, and IgM, IgG, and IgA antibodies against SARS-CoV-2 at Day 1, every 2-3 days while inpatient, Wk 1, Wk 2, Wk 3, Wk 12, and Wk 24 (EOS); and SARS-CoV-2 viral RNA in stool at Day 1, Wk 1, Wk 2, Wk 3, Wk 12 and Wk 24 (EOS). Safety and clinical laboratory tests may be performed and recorded based on symptoms and clinical indication during the study.

[5] PK samples will be drawn after at least 24 hours of dosing and prior to discharge. Samples will be drawn pre-dose and 30 minutes, 60 minutes, 2 hours, and four hours after dosing.

[6] Surveys will be given every 48 hours for the first week while a patient is inpatient (Day 1, Day 3, Day 5, and Day 7). If a patient is discharged during this period, they will not be responsible for taking the survey until their next study visit.

[7] Dosing Days 1-21: AT1001 or placebo at a dose of 250 or 500 µg/dos administered orally QID; added to standard of care for MIS-C.

1 INTRODUCTION

1.1 Background

1.1.1 COVID-19 Associated with MIS-C

COVID-19 is a serious and potentially life-threatening disease caused by the SARS CoV-2 virus, infecting the human host for the first time in December 2019 before igniting a global pandemic with >306 million confirmed cases and >5.49 million deaths reported to date. Contrary to early beliefs, children are not spared from COVID-19. For example, in Massachusetts children ≤ 19 years of age accounted for the highest number of positive COVID-19 tests by age group since the end of January 2021. Currently, over 8.5 million children in the U.S. have tested positive for COVID-19. Most children who are acutely infected with SARS CoV-2 develop mild upper respiratory symptoms or experience asymptomatic infection. Several days to weeks after resolution of the initial infection, some of these children will develop a severe life-threatening illness, termed Multisystem Inflammatory Syndrome in Children (MIS-C), which is an immune activation syndrome associated with prior SARS-CoV-2 infection or exposure.

Pediatric patients with MIS-C present with persistent fever, marked gastrointestinal (GI) symptoms, cytokine storm, myocardial dysfunction, and cardiogenic shock with ventricular dysfunction in the setting of multisystem inflammation, reminiscent of, yet distinct from, toxic shock syndrome or Kawasaki Disease (Rowley 2020). Eighty percent (80%) of these hospitalized children develop cardiac pathology (Feldstein 2020), including coronary artery dilations, myocardial dysfunction, or ventricular failure with hypotensive shock (Belhadjer 2020).

Children with MIS-C display increased monocyte recruitment and hyperphagocytosis (Bartsch 2021) with cytokine storm (Pierce 2020), T-cell activation (Carter 2020), and an inflamed expansion of immunoglobulins (Bartsch 2021, Gruber 2020). A recently discovered superantigen-like motif near the S1/S2 cleavage site on the SARS-CoV-2 Spike protein (Cheng 2020, Porritt 2020b) is hypothesized to drive this hyperinflammatory response. However, most MIS-C patients do not have SARS-CoV-2 virus detected on reverse transcription-polymerase chain reaction (RT-PCR) of the nasopharyngeal swab (Yonker 2020). In contrast, SARS-CoV-2 is detected in the majority of MIS-C stool samples, with detectable viral loads ranging $1.5x10^2$ - $2.5x10^7$ RNA copies/mL, weeks after the initial infection/exposure. Detection of SARS-CoV-2 in the stool, in addition to the predominance of severe gastrointestinal (GI) symptoms in MIS-C patients, suggest that the viral or viral antigenic source at the time of presentation of MIS-C originates from the GI system.

1.2 Intracellular Tight Junctions

Intercellular tight junctions (TJs) are junctional complexes between endothelial cells and epithelial cells. Tight junctions regulate paracellular trafficking and fluid flow from plasma to extravascular spaces. Intercellular tight TJs are extremely dynamic structures that operate in several key functions of the intestinal epithelium under both physiological and pathological circumstances. Tight junctions, the most apical junctional complex of the paracellular pathway that segregates the apical and basolateral cellular compartment, were previously believed to be impermeable and static, so forming a sealing barrier. This paradigm was subverted in 1993 by the discovery of zonula

occludens 1 (ZO-1) as the first component of the TJ complex now being comprised of more than 150 proteins, including occludin, claudins, junctional adhesion molecules (JAMs), tricellulin, and angulins. However, despite major progress in our knowledge on the composition and function of the intercellular TJ, the mechanisms by which they are regulated are still incompletely understood. One of the major breakthroughs in understanding the role of gut permeability in health and disease has been the discovery of zonulin (Tripathi 2009), the only physiologic intestinal permeability modulator described so far.

An intact, functional intestinal mucosal barrier should prevent the passage of large antigens from the gut lumen into the bloodstream, including viral antigens derived from SARS-CoV-2 present in the GI tract (Gaebler 2021). Zonulin is a family of structurally and functionally related proteins that reversibly regulate intestinal permeability by modulating intercellular tight junctions (Fasano 2000a; Fasano 2000b; Wang 2000). Increased circulating zonulin levels signaling increased intestinal permeability have been reported in several diseases (Fasano 2020) including auto-immune and hyper-inflammatory diseases such as celiac disease (Drago 2006), inflammatory bowel disease (Wyatt 1993), and Kawasaki Disease (Noval Rivas 2019). Zonulin release from epithelial cells results in permissibility of paracellular trafficking of large inflammatory antigens from the gut lumen into the bloodstream. Children with MIS-C have significantly increased release of zonulin into circulation as compared to healthy controls (Yonker 2021), suggesting a breakdown of mucosal barrier function. This loss of tight junctions' competency, coupled with the identification of SARS-CoV-2 within the stool, supports the hypothesis that GI sources of SARS-CoV-2 viral components breach the mucosal barrier and enter circulation.

1.3 Role of Zonulin in Specific Diseases

Zonulin has been implicated in many chronic inflammatory diseases (CIDs) (Table **1-1**). Independent from the CID considered, the steps leading to the break of tolerance and subsequent development of CID seem to be similar (Figure **1-1**).

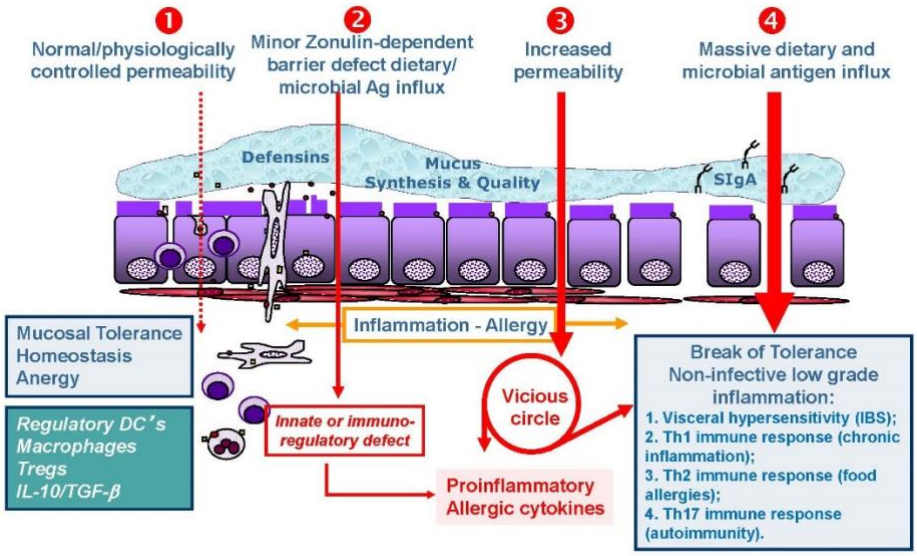
Under physiologic circumstances, there is tight control of mucosal antigen trafficking (antigen sampling) that, in concert with specific immune cell activation and production of chemokine and cytokine mediators, leads to anergy (i.e., absence of normal immune response to a particular antigen or allergen) and subsequent mucosal tolerance. The inappropriate production of increased amounts of zonulin triggered by several stimuli, including viral infections, causes a functional loss of barrier function, with subsequent inappropriate and uncontrolled antigen trafficking, instigating an innate immune response by the submucosal immune compartment. If this process continues, an adaptive immune response is mounted, causing production of pro-inflammatory cytokines (including interferon gamma [IFN-γ] and tumor necrosis factor alpha [TNF-α]) that cause further opening of the paracellular pathway to the passage of antigens, creating a vicious cycle. Ultimately, these processes lead to a break of tolerance with subsequent onset of CID (Figure **1-1**). The nature of the resultant CID is influenced by the specific host genetic background that dictates which organ or tissue will be targeted by the inflammatory process.

Table 1-1 Chronic Inflammatory Diseases in which Zonulin has been Implicated

Disease Category	Disease	Model	Disease Association
Autoimmune	Celiac Disease	Human	Specific role in pathogenesis
	Type-1-Diabetes	Human, Rat	Possible role in pathogenesis
	Inflammatory Bowel Disease / Colitis	Mouse	Possible role in pathogenesis
	Multiple sclerosis / EAE	Human, Mouse	Possible role in pathogenesis
	Ankylosing Spondylitis	Human, cell culture	Possible role in pathogenesis
Metabolic Disorders	Obesity/Insulin resistance	Human	Upregulated
	Type-2-Diabetes	Human	Upregulated
	NAFLD	Human	Upregulated
	Polycystic ovary syndrome	Human	Upregulated
Lung Disease	Acute Lung Injury	Mouse	Possible role in pathogenesis
	Asthma	Human	Upregulated
	Pneumonia	Human	Upregulated
Heart Disease	Coronary artery disease	Human	Upregulated
Neurological Disease	Glioma	Human, cell culture	Upregulated
Systemic Infectious Diseases	Septicemia	Human	Upregulated
	HIV	Human	Downregulated
Intestinal Diseases	Irritable Bowel Syndrome	Human	Upregulated
	Non-Celiac gluten sensitivity	Human	Upregulated
	Environmental Enteropathy	Human	Associated
	Necrotizing Enterocolitis	Rat, Cell Culture	Upregulated
Infectious Disease	Kawasaki Disease	Mouse models of Kawasaki disease	Upregulated
COVID-19	COVID-19 related MIS-C	Human	Upregulated

Abbreviations: COVID-19 = coronavirus disease 2019; MIS-C= multisystem inflammatory syndrome in children; EAE = experimental autoimmune encephalomyelitis; NAFLD = non-alcoholic fatty liver disease; HIV = human immunodeficiency disease

Figure 1-1 "March" from Mucosal Anergy to Loss of Epithelial Barrier Function Leading to Inflammation



Zonulin may play a central role in MIS-C pathogenesis as well. Based on preliminary findings that, in MIS-C, increased serum zonulin levels are associated with increased detection of the SARS-CoV2 virus in their stools and its Spike SAg in their serum, the central hypothesis is that the presence of SARS-CoV-2 in the GI tract leads to dysbiosis, increased zonulin release, and subsequent increased gut permeability allowing SARS-CoV2 Spike SAg trafficking across mucosal barriers, triggering a pathogenic, hyperinflammatory response.

1.3.1 Zonulin Inhibitor AT1001

AT1001 is a locally acting, non-systemic, octapeptide inhibitor of the zonulin receptor that has shown efficacy in a large variety of animal models of inflammation. The effectiveness of AT1001 in controlling paracellular permeability as a tight junction regulator has been widely demonstrated in animal models both *in vitro* and *in vivo*. No signs of toxicity were observed in animal models and there were no adverse effects in any of the pharmacological safety studies. The clinical program conducted by the pharmaceutical company with larazotide (AT1001) for the treatment of celiac disease included seven Phase 1/2 studies and one ongoing Phase 3 study. The results of these studies, including the ongoing Phase 3 study, have shown a good safety profile with an absence of serious side effects when administered via the mucosal route.

As the prevalence of SARS-CoV-2 infections among children and adolescents is increasing (MMWR, 2021), MIS-C cases are increasing as well. Current treatment strategies are primarily supportive care and directed care against the underlying inflammatory process but do not address mucosal permeability or antigenemia. Given the safety and tolerability profile and the potential benefit for pediatric patients with MIS-C, it is appropriate to study AT1001 as a treatment for MIS-C. This study is intended to assess the clinical safety, tolerability, and efficacy profile of AT1001 in hospitalized pediatric patients with MIS-C.

1.4 Overview of Non-Clinical Studies with AT1001

1.4.1 Nonclinical Pharmacology and Pharmacology

1.4.1.1 Mechanism of Action

Research into the mechanism of action indicates that AT1001 prevents the opening of intercellular TJs triggered by inflammatory cytokines and other stimuli thereby preventing potential antigens from reaching the lamina propria and stopping the inflammatory response associated with CID.

In vitro studies have demonstrated a regulatory effect of AT1001on cytoskeletal rearrangement and the opening of TJs in intestinal tissue and cell lines. AT1001 has been shown to prevent the disassembly of TJs in rabbit intestine and intestinal IEC6 and CaCo2 cells. These studies have contrasted the effect of AT1001, which prevents the opening of TJs, to the effect of zonula occludens toxin (Zot), a toxin produced by the bacterium, Vibrio cholera, that causes the opening of TJs (Fasano 1995; Fasano 2000a; Fasano 2000b; DiPierro 2001; Wang 2000).

In agonist studies with Zot, AT1001, when used to pretreat ileal tissue preparations, was shown to prevent the reduction of trans-epithelial electrical resistance (which correlates with increased permeability) induced by Zot. In a study with intestinal epithelial cells, IEC6 and CaCo-2 cells were exposed to gliadin. The result was rearrangement of the cell cytoskeleton, loss of zonula occludens-1 (ZO-1) protein-protein interaction and increased monolayer permeability. Pretreatment with AT1001 blocked these changes (Drago 2006).

In more recent studies, the effect of AT1001 on de novo tight junction assembly using two different model systems was studied. In Madin-Darby canine kidney (MDCK) cells, AT1001 promoted tight junction assembly in calcium switch assay and promoted actin rearrangement, and junctional distribution of ZO-1, occludin, claudins, and E-cadherin. AT1001 promoted tight junction maturation and decreased paracellular permeability in “leaky” Caco-2 cells. AT1001 inhibited the redistribution and rearrangement of ZO-1 and actin caused by AT-1002 (a 6-mer peptide derived from Zot) and gliadin fragments in Caco-2 and IEC6 cells. Functionally, AT1001 inhibited the AT-1002-induced transepithelial electrical resistance (TEER) reduction and tight junction opening in Caco- 2 cells. AT1001 inhibited the increase in tight junction permeability elicited by basolaterally applied cytokines (Gopalakrishnan 2012a; Gopalakrishnan 2012b). Studies using a porcine ex vivo model of intestinal ischemia demonstrated that AT1001 improved transepithelial resistance compared to untreated controls, consistent with other data indicating AT1001’s ability to regulate TJs. This effect was observed in a narrow dose range, with optimal efficacy in the range of 1-5 µM (Hernandez 2018).

Dose-volume modeling indicates that the 2 µM dose in the porcine ischemia models approximately corresponds to the 0.5 mg oral human dose that was effective in the CLIN1001-012 study (PharmaDirections 2009; data on file). The basis for this narrow dose range may be related to the generation of inhibitory peptide fragments in the intestine, which might inhibit the efficacy of AT1001 at higher doses, and which is known to occur with other peptides. Based on simulation models, such inhibition would be less likely to occur at oral doses of 0.25 or 0.5 mg (Nuventra 2018; data on file).

1.4.1.2 Nonclinical Pharmacokinetics and Safety Pharmacology

The nonclinical pharmacology and toxicology program for AT1001 includes a battery of non-clinical safety pharmacology, pharmacokinetic and toxicology studies. Collectively, these studies demonstrate that AT1001 is very poorly orally absorbed in animals and only at extremely high doses and is very well tolerated with no specific safety pharmacology or toxicology signals observed at doses ~5,000 times the proposed human dose (0.5 mg three times daily [TID]) based on the 1000 mg/kg dose in rats and ~2,000 times the highest proposed dose based on the 40 mg/kg dose in dogs.

Pharmacokinetics (PK) and toxicokinetics (TK) analyses performed with AT1001 in rat and dog show the compound to be poorly absorbed from the GI tract into the general circulation, and the small amount of AT1001 that is absorbed is rapidly cleared. The metabolic profile of AT1001 incubated in human blood, plasma, simulated gastric fluid, and simulated intestinal fluid produced a pattern of truncated peptides, indicating the primary metabolic pathway for AT1001 is likely proteolysis. A screen for inhibition activity of cytochrome P450 enzymes was negative. Overall, the PK/ absorption, distribution, metabolism, and excretion (ADME) properties of AT1001 are those expected of an 8-amino acid peptide containing all L-amino acids. Across species it is poorly absorbed from the GI tract and rapidly eliminated from both plasma and the gut, most likely by proteolysis.

Safety pharmacology studies performed to assess cardiovascular safety include two in vitro human ether-a-go-go related gene (hERG) assays at doses up to 100 μM and one cardiotoxicity study in anesthetized rabbits. In addition, a central nervous system toxicity study in rats and a cardiovascular and respiratory function study in telemetered dogs were performed. No significant effects were observed in any of the safety pharmacology studies.

1.4.2 Toxicity Data for AT1001

1.4.2.1 Single and Multiple Dose Toxicity Studies

No signals of toxicity have been observed in animal toxicology studies to date, including 28-day, 13 week, and 6-month repeat-dose studies in rats (at up to 1000 mg/kg by gavage), in repeat-dose studies in dogs including 28-days (at doses up to 9 mg/kg), 90-days (at doses up to 20 mg/kg) and 12 months (at doses up to 40 mg/kg).

1.4.2.2 Carcinogenicity

Administration of AT1001 to male and female Crl:CD (SD) rats at oral doses of up to 125 mg/kg/day for 99 weeks resulted in no test article-related macroscopic findings and no test article-related neoplastic or non-neoplastic microscopic findings.

1.4.2.3 Mutagenesis

AT1001 showed no evidence of mutagenic potential in the Ames test, the human lymphocyte chromosome aberration test, and the rat micronucleus test.

1.4.2.4 Reproductive Toxicology

Oral administration of AT1001 (50, 250, or 1,000 mg/kg/day) to male and female rats prior to and throughout mating and continuing in females through early gestation resulted in no adverse effects on fertility or pre- or post-natal development and no evidence of embryo- or feto-toxicity was observed. No evidence of maternal or embryo- or feto-toxicity was detected following oral administration of AT1001 (50, 250, or 1,000 mg/kg/day) to primiparous female rabbits.

1.5 Overview of Clinical Studies with AT1001

1.5.1 AT1001 in Clinical Studies

AT1001 has been studied in seven clinical studies (three Phase 1 [two in healthy adults and one in adults with celiac disease] and four Phase 2 studies). A summary of these studies is provided in Table 1-2. A Phase 3 study (NCT03569007) is ongoing. Results from these studies, including the ongoing Phase 3 study to date, show a good safety profile with an absence of serious or severe adverse events (AEs).

Table 1-2 Summary of Phase 1 / Phase 2 Clinical Studies in Celiac Disease

Study Identifier(s)	Description	Study Medication	Population	Reference
CLIN1001-001A	Phase 1 PK	Placebo or AT1001 3, 12 or 36 mg single dose	Healthy adults N = 24 (6 per treatment group)	---
CLIN1001-003	Phase 1 PK	Placebo or AT1001 0.25, 1 or 3 mg TID for 10 days	Healthy adults N = 24 (6 per treatment group)	---
CLIN1001-002 NCT00386490	Phase 1 PK	Placebo or AT1001 12 mg daily for 3 days	Adult Celiac patients N = 21 (14 AT1001 ; 7 placebo)	Paterson 2007
CLIN1001-004 NCT00362856	Phase 2 randomized, double-blind, placebo-controlled, dose-ranging, multicenter	Placebo or AT1001 0.25, 1, 4 or 8 mg TID for 14 days	Adult Celiac patients N = 86 (60 AT1001 ; 26 placebo)	Leffler 2012
CLIN1001-006A NCT00492960	Phase 2 randomized, double-blind, placebo-controlled, dose-ranging, multicenter	Placebo or AT1001 1, 4 or 8 mg TID for 42 days	Adult Celiac patients N = 171 (128 AT1001 ; 43 placebo	Kelly 2012
CLIN1001-006B (Extension to CLIN1001-006A) NCT00889473	Phase 2 randomized, double-blind, placebo-controlled, multicenter	Placebo or AT1001 1 mg TID for 42 days	Adult Celiac patients N = 42 (21 AT1001 ; 21 placebo)	---
AT1001-011 NCT00620451	Phase 2 randomized, , double-blind, placebo-controlled, multicenter	Placebo or AT1001 4 or 8 mg TID for 56 days	Adult Celiac patients N = 104 (70 AT1001 ; 34 placebo)	---

CLIN1001-012 NCT01396213	Phase 2 randomized, , double-blind, placebo-controlled, multicenter	Placebo or AT1001 0.5, 1.0 or 2.0 mg TID for 84 days.	Adult Celiac patients N=340 (256 AT1001 ; 84 placebo)	Leffler 2015

Abbreviations: PK = pharmacokinetics; TID = three times daily

1.5.2 Pharmacokinetic Evaluation

Two Phase 1 studies were conducted in healthy subjects.

CLIN1001-001A was a randomized, double-blind, placebo controlled, single dose escalation study to determine the safety, tolerance, and pharmacokinetics of AT1001 in healthy volunteers. Three cohorts of 8 subjects (3:1 ratio active drug to placebo) who received single escalating doses of drug (3, 12, and 36 mg) or placebo at 7-day intervals. Subjects were evaluated for safety at screening, prior to administration of study drug and following dosing. Serial blood samples were collected for pharmacokinetic determinations at baseline, 0.25, 0.75, 1.25, 1.75, 2.25, 3.25, 4.0, 5.0-, and 8-hours following study drug administration. AT1001 concentrations were determined by a validated analytical method.

CLIN1001-003 was a Phase 1, randomized, double-blind, placebo controlled multi-dose study to determine the safety, tolerance, and pharmacokinetics (including food effect) of 3 dose levels of AT1001 in healthy volunteers. Three (3) cohorts of 8 subjects, in which six (6) subjects in each cohort received AT1001 in escalating doses of 0.25 mg, 1.0 mg, or 4.0 mg TID for 10 days and two (2) subjects received placebo TID for 10 days, were studied. Subjects maintained a standard diet. Subjects were dosed three times a day 30 minutes prior to meals except for the morning dosing on Days 1, 5, and 10. For the morning dosing on Days 1, 5, and 10 subjects, fasted overnight prior to dosing and for 2 hours post dose.

Pharmacokinetic Results: In both studies, the administration of AT1001 across all dose levels resulted in plasma concentrations that were below the level of quantitation (0.5 ng/mL).

These results were confirmed in subsequent studies in subjects with celiac disease (CLIN1001-002, CLIN1001-004 and AT1001-011) where neither AT1001 nor any of its peptide metabolites was detected by sensitive LC MS/MS analysis (LLOQ = 0.5 ng/mL) in plasma following repeat oral dosing up to 8 mg TID for 56 days.

AT1001 has also been used to treat five children with COVID-19 related Multisystem Inflammatory Syndrome in Children (MIS-C) though an Emergency Investigational New Drug request. Details regarding response in a single patient have been published by Yonker 2021. In all five children, the medication was well tolerated and the children experienced improvement in clinical symptoms and inflammatory markers.

1.5.3 Safety and Adverse Effects

A total of 812 subjects participated in Phase 1/2 clinical studies of AT1001. 585 subjects (including 36 healthy subjects) received orally administered AT1001 with doses ranging from 0.25 to 12 mg TID. Approximately 475 subjects with celiac disease received AT1001 for up to 42 days or more, approximately 256 of whom received drug for up to 84 days. The most common AEs reported in all studies were mild-to-moderate GI events which were similar to placebo in frequency and nature.

There have been no deaths or dose-related AEs in any study. There was a total of five serious adverse events (SAEs), two of which occurred on placebo (appendicitis and urticaria). There were three treatment emergent SAEs reported across all studies, none of which were considered to be drug-related by the reporting investigators:

- a 61-year-old female with bipolar disorder receiving AT1001 0.5 mg TID experienced worsening depression (CLIN1001-006)
- a 21-year-old female subject experienced cholelithiasis and underwent cholecystectomy approximately 14 days after commencing AT1001 0.5 mg TID (CLIN1001-012). Operative findings were consistent with chronic cholecystitis (including scarring) that likely preceded subject's brief exposure to AT1001.
- a 32-year-old female subject who was receiving a gluten challenge as well as AT1001 1 mg TID experienced symptomatic cholelithiasis with a common bile duct stone and underwent a cholecystectomy and sphincterotomy approximately 10 days after commencing AT1001 1 mg TID.

Analysis of the population subsets in studies AT1001-011 and CLIN1001-012 showed no clinically meaningful differences between males versus females or in subjects ≥ 65 or < 65 years of age. In the CLIN1001-012 study, the largest study conducted to date, the most common GI events were diarrhea, nausea, constipation and abdominal pain. The reporting frequency of these events was less than 10% in any treatment group.

1.6 Current Available Treatments

There are currently no approved antiviral drugs to treat patients with COVID-19. According to the U.S. Centers for Disease Control (CDC; https://www.cdc.gov/coronavirus/2019-ncov/hcp/clinical-guidance-management-patients.html), the National Institutes of Health (NIH) and collaborators are working on development of candidate vaccines and therapeutics for COVID-19. Some *in vitro* or *in vivo* studies suggest potential therapeutic activity of compounds (such as remdesivir) against related coronaviruses, but there are no available data from randomized controlled trials in humans to support recommending any investigational therapeutics for patients with confirmed or suspected COVID-19 at this time.

1.7 Study Rationale

AT1001 is a locally acting, non-systemic, octapeptide inhibitor of the zonulin receptor that has shown efficacy in a large variety of animal models of inflammation. The effectiveness of AT1001 in controlling paracellular permeability as a tight junction regulator has been widely demonstrated in animal models both *in vitro* and *in vivo*. No signs of toxicity were observed in animal models and there were no adverse effects in any of the pharmacological safety studies. The clinical program conducted by the pharmaceutical company with larazotide (AT1001) for the treatment of celiac disease included seven Phase 1/2 studies and one ongoing Phase 3 study. The results of these studies, including the ongoing Phase 3 study, have shown a good safety profile with an absence of serious side effects when administered via the mucosal route.

A study conducted by Yonker 2021 showed that in MIS-C, prolonged presence of SARS-CoV-2 in the GI tract leads to release of zonulin, a biomarker of intestinal permeability, with subsequent trafficking of SARS-CoV-2 antigens into the bloodstream, leading to hyperinflammation. The MIS-C patient treated with AT1001 (through an Emergency Investigational New Drug request) displayed a coinciding decrease in plasma SARS-CoV-2 Spike antigen levels, inflammatory markers, and a resultant clinical improvement above that achieved with currently available treatments. Four other children treated with AT1001 experienced improvement in clinical symptoms and inflammatory markers.

As the prevalence of SARS-CoV-2 infections among children and adolescents is increasing (MMWR 2021), MIS-C cases are increasing as well. Current treatment strategies are primarily supportive care and directed care against the underlying inflammatory process but do not address mucosal permeability or antigenemia. Given the safety and tolerability profile and the potential benefit for pediatric patients with MIS-C, it is appropriate to study AT1001 as a treatment for MIS-C. This study is intended to assess the clinical safety, tolerability, and efficacy profile of AT1001 in hospitalized pediatric patients with MIS-C.

1.8 Risk/Benefit Statement

No specific safety concerns have been identified. The possible risks are known and expected on the basis of several studies performed over the last 15 years both in animal models and in humans. In these studies, the drug was administered orally showing good safety profile in the 561 treated patients.

In this protocol the oral route is proposed, hence the drug action is localized to the target organ, gastrointestinal, reducing systemic exposure and possibly furtherly improving the know safety profile.

The study treatment can be administered in conjunction with other therapies.

2 STUDY OBJECTIVES AND ENDPOINTS

2.1 Study Objectives

2.1.1 Primary Objective

The primary objective of this study is:

- To evaluate the efficacy and safety of AT1001 versus placebo in pediatric patients with SARS-CoV-2 infection who experience early signs of MIS-C and are at high risk of progression.

2.1.2 Secondary Objectives

The secondary objective of this study is:

- To determine impact of AT1001 on infectious and inflammatory markers of MIS-C.

2.2 Study Endpoints

2.2.1 Primary Efficacy Endpoint

Primary Efficacy Endpoint: Efficacy will be evaluated by primary endpoint at Day 21, namely:

- To determine proportion of participants with improvement in MIS-C related GI symptoms and no progression of GI system involvement.
    - Improvement in GI symptoms is defined as:
        - Improvement in PedsQL GI Symptoms Scales score ≥48 hours, as determined by patient or caregiver response.
        - AND improvement in clinical manifestation of GI symptoms, as documented by complete physical exam or clinical assessment and clinical laboratory tests or imaging.
    - No Progression of MIS-C is defined as:
        - No involvement of additional organ involvement, as identified by clinical assessment and clinical laboratory tests or imaging.
        - AND no new onset of new or worsening GI symptoms for ≥48 hours including nausea, vomiting, diarrhea, loss of appetite, and/or abdominal pain, as determined by patient or caregiver symptom report or worsening PedsQL GI Symptoms Scales scores.

2.2.2 Secondary Efficacy Endpoints

Secondary Efficacy Endpoints through Day 21:

- Improvement in MIS-C symptoms for ≥48 hours, as determined by patient or caregiver symptom report on MIS-C Symptom Questionnaire.
- Length of stay in hospital (days from baseline to readiness to discharge).

- Change from baseline in additional organ system(s) involvement during acute presentation/hospitalization, as identified by clinical assessment and clinical laboratory tests.
- Need for escalation of care (eg, transfer from hospital ward to ICU; supplemental oxygen; mechanical ventilation).
- Re-presentation to medical care for MISC-related symptoms after discharge.
- Normalization of SARS-CoV-2 Spike, S1 and nucleocapsid.
- Change from baseline in levels of IgM, IgG, and IgA antibodies against SARS CoV-2.
- Change from baseline in levels of inflammatory markers (CRP, d-dimer, ferritin).
- Change from baseline in levels of zonulin.
- Change from baseline in levels of NT proBNP and troponin (participants with cardiac organ system involvement).
- Mortality (all causes)

2.2.3 Exploratory Efficacy Endpoints

- Change from baseline in levels of cytokines (IFN-γ, IL-1β, IL-17, IL-2R).
- Time to restoration of usual state of health ≥ 48 hours as assessed by the MIS-C Activity Limitation Questionnaire.
- Time to restoration of usual activity level ≥ 48 hours as assessed by the MIS-C Activity Limitation Questionnaire.
- Time to resolution of COVID-19 related symptoms ≥ 48 hours as assessed by the MIS-C Activity Limitation Questionnaire (participants with active COVID-19 related symptoms).
- Time to resolution of MIS-C related organ system involvement, as defined by:
    - Improvement in cardiac involvement (includes improvement or resolution of ANY of the listed criteria):
        - cardiac dysrhythmia or arrythmia as assessed by ECG
        - left ventricular (LV) ejection fraction (EF) >55% as assessed by echocardiogram
        - pulmonary edema due to left heart failure as assessed by chest radiograph
        - pericarditis or pericardial effusion as assessed by echocardiogram
        - B-type natriuretic peptide (BNP) <400 pg/mL
        - troponin (to within normal ranges based on the laboratory running the assay)
        - need for vasopressor or vasoactive support
        - coronary artery dilation or aneurysm as assessed by echocardiogram

- Improvement in renal involvement (includes improvement or resolution of ANY of the listed criteria):
  - acute renal injury, as assessed by normalization of creatinine levels
- Improvement in respiratory involvement (includes improvement or resolution of ANY of the listed criteria):
  - need for mechanical ventilation
  - need for supplemental oxygen (or increased support for patients receiving respiratory support at baseline)
  - pulmonary infiltrates on chest radiograph
  - lower respiratory infection by clinical assessment
  - pleural effusion (not requiring chest tube) on chest radiograph
  - pneumonia by clinical assessment
- Improvement in gastrointestinal involvement (includes improvement or resolution of ANY of the listed criteria):
  - nausea/vomiting by patient/caregiver report
  - diarrhea by patient/caregiver report
  - abdominal pain by patient/caregiver report
  - pancreatitis by laboratory testing (amylase and/or lipase <200 U/L or radiologic findings)
  - hepatitis by laboratory testing (AST and/or ALT >500 U/L)
- Improvement in hematologic involvement (includes improvement or resolution of ANY of the listed criteria):
  - total white blood cell <4 x10^3/µL by laboratory testing
  - anemia (hemoglobin <9 g/dL) by laboratory testing
  - platelet count <150,000 /µL by laboratory testing
  - hemolysis by laboratory testing
- Improvement in mucocutaneous involvement (includes improvement or resolution of ANY of the listed criteria):
  - bilateral conjunctival injection by clinical assessment
  - oral mucosal changes by clinical assessment
  - peripheral extremity changes by clinical assessment
  - rash or skin ulcers (not related to pre-existing skin condition such as eczema or psoriasis) by clinical assessment
  - swollen red cracked lips by clinical assessment
  - erythema of palms or soles by clinical assessment
  - edema of hands or feet by clinical assessment
  - periungual (nails) desquamation by clinical assessment
  - conjunctivitis by clinical assessment
- Improvement in neurologic involvement (includes improvement or resolution of ANY of the listed criteria):
  - headache by patient/caregiver report

- irritability by patient/caregiver report
- lethargy by patient/caregiver report
- altered mental status by patient/caregiver report
- stroke or intracranial hemorrhage by clinical assessment
- seizures by clinical assessment

2.2.4 Safety Evaluations

- Incidence, severity, and type of treatment-emergent adverse events
- Change from baseline in vital sign measurements
- Change from baseline in physical examination findings
- Change from baseline in clinical laboratory parameters

3 STUDY DESIGN

3.1 Study Overview

This is a Phase 2a (Proof of Concept), randomized, double-blind, placebo-controlled study to evaluate the efficacy and safety of AT1001 for use in hospitalized pediatric patients diagnosed with MIS-C. Eligible participants (N=20) will be treated with AT1001 or matching placebo orally four times a day (QID) for up to 21 days as an add-on to standard of care.

The study includes three phases:

- Screening/Baseline: The main purpose of this phase is to determine if the participant meets entry criterion after obtaining informed consent and obtain baseline assessments.
- Treatment: Eligible participants will be treated with AT1001 at a dose of 250 or 500 mcg or matching placebo for 21 days as an add-on to standard of care for MIS-C.
- Follow-up through 24 weeks: The participant will return for follow-up visits during weekly clinic visits during Week 1 through Week 3, with monthly telemedicine visits at Week 4, Week 8, Week 16 and Week 20, and clinic visits at Week 12 and Week 24.

Safety monitoring, including physical examination, vitals, and clinical laboratory testing will be performed during the screening phase, periodically during treatment phase and at the follow-up phase. Adverse events and concomitant medications will be recorded during the entire study.

Total duration of the participants' participation in the study is approximately 24 weeks (with 21 days treatment period). Total duration of the study is projected to be 12 months, dependent on enrollment timeline.

3.2 Stopping Criteria

3.2.1 Interruption of Dosing in an Individual Participant

The dosing of an individual participant may be interrupted under any of the following circumstances:

- The participant experiences a serious adverse event related to the study drug
- The participant experiences an intensity Grade 3 (severe) or greater AE as defined by the current version of the Common Terminology Criteria for Adverse Events (CTCAE)
- Any other apparent deterioration in the health of the participant as determined by the investigator

3.2.2 Stopping Rules for Study Enrollment and Dosing

Participant randomization into the study treatment phase may be stopped or paused if any of the following occur:

- Death of any participant that is deemed to be probably, or definitely related to study drug
- A life-threatening adverse event in any participant that is deemed to be, probably, or definitely related to study drug

- A safety signal requiring further evaluation to assess the overall benefit risk of the program

3.2.3 Study Treatment Discontinuation Criteria

Study treatment should be discontinued if the following occurs:

- ALT or AST >3 times baseline with direct bilirubin > 2 times upper limit of normal (ULN) or international normalized ratio (INR) increased from baseline and is >1.5 times ULN, unless the Investigator or the data safety monitoring board (DSMB) determines the abnormality is unlikely related to study drug
- Creatinine >1.8 times ULN and >2 times baseline, or decreased creatinine clearance to < 60 mL/min, unless the Investigator or the DSMB determines the abnormality is unlikely related to study drug.
- Clinically significant Grade 4 adverse event or confirmed Grade 4 laboratory abnormality for an individual participant that is considered by the Investigator or the DSMB to be related to the study drug

4 PARTICIPANT SELECTION

Participants with a diagnosis of MIS-C who meet the inclusion and exclusion criteria will be eligible for participation in this study.

4.1 Inclusion Criteria

1) Pediatric patients with or without comorbidity
2) Age ≥ 1 month to < 21 years
3) Confirmed MIS-C by signs and symptoms as detailed by the CDC Health Advisory (https://www.cdc.gov/mis-c/hcp/; May 14, 2020)
    a) Persistent fever/chills (>38.0°C for ≥24 hours, or report of subjective fever lasting ≥24 hours); AND
    b) One or more laboratory parameters (evidence of inflammation); AND,
        i) elevated C-reactive protein (CRP)
        ii) elevated erythrocyte sedimentation rate (ESR)
        iii) elevated ferritin
        iv) elevated lactic acid dehydrogenase (LDH)
        v) elevated d-dimer
        vi) elevated fibrinogen
        vii) elevated procalcitonin
        viii) elevated interleukin 6 (IL-6)
        ix) increased neutrophils
        x) reduced lymphocytes
        xi) low albumin
    c) Evidence of clinically severe illness requiring hospitalization, with multisystem (>2) organ involvement (cardiac, renal, respiratory, hematologic, gastrointestinal, dermatologic, or neurological)—MUST include GI symptoms, such as nausea, vomiting, diarrhea, decreased appetite, and/or abdominal pain; AND,
    d) No alternative plausible diagnoses; AND
    e) Positive for current or recent SARS-CoV-2 infection by RT-PCR, serology, or antigen test

4) Subject (or legal authorized representative) capable of understanding and signing an informed consent form and assent form, when appropriate.

4.2 Exclusion Criteria

1. Female participants pregnant and/or lactating.
2. Female participant has childbearing potential and is unwilling to use an acceptable method of birth control for the duration of the study.
3. Participant has a significant co-morbid disease that by the Investigator's determination would make the participant unsuitable for enrollment, including unstable medical conditions.
4. Participation in any other clinical investigation using an experimental drug within 30 days prior to screening or intends to participate in another clinical study while participating in AT1001 MIS-C 101 study.
5. Have participated in a blood/plasma donation or blood loss greater than 400 mL within 90 days, or greater than 200 mL within 30 days prior to Screening.
6. Known hypersensitivity to any of the formulation components of AT1001.

4.3 Equitable Recruitment

The disproportionate impact and subsequently high Sars-CoV-2 infection and hospitalization rates among low-income and minority communities has been well documented. It has also been demonstrated that this impact extends throughout all age groups, including children under the age of 18, despite the overall lower incidence of severe COVID-19 infection in younger populations (Kim 2020). Of the studies investigating sociodemographic factors contributing to MIS-C, many have found subsequently high rates of MIS-C in children who are Black and Hispanic (Feldstein 2021, Lee 2020, Payne 2021). These trends are reaffirmed by the CDC's data on reported cases of MIS-C patients according to race and ethnicity (https://www.cdc.gov/mis/cases/index.html 2021). MIS-C is also slightly more common in males versus females (60% vs. 40%), however this difference is not anticipated to restrict recruitment (https://www.cdc.gov/mis/cases/index.html 2021). Thus, diverse recruitment across various racial, ethnic, socioeconomic, and gender groups is expected throughout the course of this study.

5 CONCURRENT MEDICATIONS

All participants should be maintained on the same medications throughout the entire study period, as medically feasible, with no introduction of new chronic therapies.

All concomitant medications and non-drug treatments (including prescription and over-the-counter medications, nutritional supplements, and herbal remedies) taken during the study should be recorded on the eCRF.

Standard therapy for MIS-C (e.g. IVIG and steroids) is allowed except for treatments noted in the exclusion criteria described above. Sponsor will monitor changes in clinical practice guidelines as outlined by the CDC and will note changes that may arise.

6 INVESTIGATIONAL PRODUCT

6.1 Formulation and Appearance

AT1001 product is a synthetic 8-amino acid peptide that is formulated as enteric-coated delay release beads and delivered orally in size 4, hard shell gelatin capsules. The release of AT1001 from the enteric-coated beads is pH-dependent, allowing the beads to bypass the stomach, and release AT1001 upon entry into the small intestine.

AT1001 drug substance is manufactured by Corden Pharma Brussels SA (Brussels, Belgium) under cGMP conditions. AT1001 and matching placebo capsules are manufactured and packaged by UPM Pharmaceuticals (Bristol, TN). Each AT1001 capsule will contain 0.25 or 0.50 mg of AT1001 and other ingredients: sugar spheres 25/30 mesh, Eudragit L30 D-55, triethyl citrate, hypromellose, and talc. The matching placebo capsule will contain sugar spheres 25/30 mesh, Eudragit L30 D-55, triethyl citrate, and talc.

6.2 Packaging and Labeling

AT1001 and matching placebo capsules will be provided to the clinical site in 102-count HDPE bottles with a child-resistant cap.

Each 102-count HDPE bottle of AT1001 250 or 500 µg or matching placebo capsules will be labeled to meet applicable laws for an investigational drug. This information includes, but is not limited to:

- Federal law statement: Caution New Drug-Limited by United States Law to Investigational Use
- Study number
- Manufacturing date, Re-Test date or Use by date
- Storage information
- Manufactured for

Instructions for administration (including dosages) are provided in the pharmacy manual.

6.3 Method of Assigning Participants to Treatment Groups

Up to 20 eligible participants will randomly assigned to AT1001 or placebo treatment groups. The study pharmacist(s) will create the randomization schedule according to a 1:1 randomization ratio. Participant spots will be randomized in blocks of 4. Randomization numbers will be assigned for each participant, beginning with randomization number 101. The randomization schedule will be prepared in advance of subject enrollment to allow for timely drug dispensation. Upon enrollment, each participant will be assigned a randomization number and will receive the drug which corresponds with that randomization number in the randomization schedule.

6.4 Blinding and Breaking the Blind

The study pharmacist will generate the randomization schedule for the study, which will be held under secure conditions of restricted access. To keep the double-blind design of the study it is necessary to involve unblinded person(s) at the study site for handling and dispensing of the study drug. Unblinded person(s) must not perform any assessments. Participants, sponsor, investigators, and site staff responsible for performing assessments and data analysts, will remain blinded from the time of randomization until database lock.

Only in the case of emergency, when knowledge of the study drug is essential for the clinical management or welfare of the participant, will the investigator be allowed to unblind a participant's treatment assignment. The participant must stop study treatment and be discontinued from the study prior to breaking the blind. The study blind will be broken on completion of the clinical study and after the study database has been locked.

The only un-blinded personnel will be the:

- Study pharmacist Clinical Trials Pharmacy Staff

The study blind will be broken on completion of the clinical study and after the study database has been locked.

6.4.1 Dosage/Dosage Regimen

AT1001 at a weight based dose of 250or 500 μg or matching placebo will be administered orally four times a day (QID) to the standard of care for MIS-C. All patients ≤25kg will receive a dose of 250 μg and all patients ≥25.1 will receive a dose of 500 μg. The investigational drug will be administered either as capsules swallowed whole (for participants who can swallow a capsule) or as beads in soft food (for participants who cannot swallow a capsule) or a liquid vehicle (for participants with enteral tubing).

Participants will be given all treatments by the appropriate medical personnel during their inpatient stay and will be expected to continue dosing on their own following discharge. Compliance will be assessed by documentation of drug administration in the source documents, completion of the participant drug diary, and eCRF.

6.4.2 Dispensing

AT1001 and matching placebo will be provided to the site pharmacy and dispensed in doses of 250 or 500 μg or as a matching placebo four times a day (QID) for 21 days. Three buffer days will also be included at the time of dispensation, with a total of 96 doses dispensed to subjects upon treatment initiation. Treatment groups will be assigned per the randomization schedules and all doses will be dispensed at the beginning of the study.

6.4.3 Drug Diary

Subjects will be required to track all doses of the study in a drug diary along with any potential side effects and missed doses. Drug diaries will be reviewed by MIS-C study staff at each study visit after Visit 1 when the diary is distributed.

6.5 Supply of Study Drug at the Site

AT1001 and matching placebo will be provided to the site pharmacy and dispensed per the randomization schedule. Participants who are randomized but withdraw from the study prior to treatment will be replaced.

6.5.1 Storage

All study drug, (AT1001 and matching placebo), will be kept in a locked area with limited access. Study drug is to be stored upright at approximately 20°- 25°C (68°-77°F); excursions are permitted between 15°- 30°C (59° to 86°F). Deviations in storage conditions should be noted and the Sponsor notified.

6.6 Study Drug Accountability

The clinical site pharmacy will track unblinded drug accountability and dispensation on an electronic drug accountability log. A blinded drug accountability and dispensation log will be maintained at all study visits by the research coordinator. All dispensed study drug will be recorded in the drug accountability log along with the participants' initials and/or subject number. Upon completion of dosing, all returned study drug will be counted and recorded in the drug accountability log.

6.7 Measures of Treatment Compliance

Compliance will be assessed by documentation of drug administration in the source documents, completion of the participant drug diary, and electronic CRF.

Following discharge from the hospital, participants will be instructed to bring their drug diary for review at each study visit. Participants will return all unused study drug at the day 21 visit, excepting any doses needed to complete the full 21-day course, which will depend on study visit timing. For participants with final doses that fall after the Day 21 visit, they will be sent home with their remaining required doses.

7 STUDY PROCEDURES AND GUIDELINES

A Schedule of Events representing the required testing assessments and procedures to be performed for the duration of the study.

Prior to conducting any study-related activities, written informed consent and the Health Insurance Portability and Accountability Act (HIPAA) authorization must be signed and dated by the participant or participant's legal representative. If appropriate, assent must also be obtained prior to conducting any study-related activities.

7.1 Clinical Assessments

7.1.1 PedsQL Gastrointestinal Symptoms Scales

An assessment of MIS-C-related GI symptoms will be conducted using the PedsQL Gastrointestinal Symptoms Scales every 48 hours during the first 7 days of the participant's inpatient stay (Day 1, Day 3, Day 5, Day 7) and at every subsequent visit after the Day 7 visit. Should the participant be discharged during their first 7 days, they will not be required to complete additional surveys outside of their study visits. The acute version of the PedsQL GI symptom survey will be used for visits 1-10. Visits 11-15 will use the monthly, non-acute PedsQL GI symptom survey.

The PedsQL GI symptom questionnaire will be completed independently by participants ≥ 7 years of age. To validate patient responses, caregivers will also complete an observer reported questionnaire on behalf of their child. For child < 7 years of age, only the caregiver's response will be required.

7.1.2 Assessment of MIS-C-related Symptoms

An assessment of MIS-C-related symptoms will be conducted at Day 1, Days 2-6, at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS and telephone contact visits at Week 4, Week 8, Week 16, and Week 20.

MIS-C related symptom questionnaire will be completed by patients ≥ 7 years of age. To validate patient responses, caregivers will also complete an observer MIS-C related symptom questionnaire on behalf of their child. For child < 7 years of age, only the caregiver survey will be required.

7.1.3 Assessment of Activity Limitations

An assessment of activity limitations will be conducted at Day 1, Days 2-6, at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS and telephone contact visits at Week 4, Week 8, Week 16, and Week 20.

The assessment of activity limitations questionnaire will be completed by patients ≥ 7 years of age. To validate patient responses, caregivers will also complete an observer questionnaire on behalf of their child. For child < 7 years of age, only the caregiver survey will be required.

7.1.4 Concomitant Medications

All concomitant medication and concurrent therapies will be documented at Screening/Baseline and at Study Visits 2 through 15, and at early termination when applicable. Dose, route, unit frequency of administration, and indication for administration and dates of medication will be captured.

7.1.5 Demographics

Demographic information (date of birth, gender, race) will be recorded at Screening.

7.1.6 Medical History

Relevant medical history, including history of current disease, other pertinent history, and information regarding underlying diseases will be recorded at Screening.

7.1.7 Physical Examination

Physical examinations performed by qualified clinical staff (MD, NP, and PA) -will be abstracted from the subject's electronic medical record for all inpatient study visits and filed with appropriate source documentation. Optional, symptom directed physical examinations may be performed by qualified study staff (MD, PA, NP) at outpatient visits. New abnormal physical exam findings must be documented and will be followed by a physician or other qualified staff at the next scheduled visit.

7.1.8 Vital Signs

Body temperature, blood pressure, pulse, respirations, and weight will be performed at Day 1, Days 2-6, at clinic visits Week1, Week 2, Week 3, Week 12, and Week 24/EOS. Height will be collected only at screening.

7.1.9 Electrocardiogram

An electrocardiogram will be conducted at Day 1, Days 2-6, at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS. Electrocardiograms may be performed if clinically indicated for more frequent monitoring during the study.

7.1.10 Adverse Events

Information regarding occurrence of adverse events will be captured throughout the study. Duration (start and stop dates), severity/grade, outcome, treatment, and relation to study drug will be recorded on the case report form (CRF).

7.2 Clinical Laboratory Measurements

7.2.1 Hematology

Blood will be obtained and sent to each site's clinical hematology laboratory for a complete blood count (hemoglobin, hematocrit, red blood cell count, white blood cell count, white blood cell differential, and platelet count), and erythrocyte sedimentation rate (ESR) as clinically indicated at Day 1, every 2-3 days until discharge, at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

Blood will also be obtained and sent to each site's clinical lab for determination of serum C-reactive protein (CRP), ferritin, and d-dimer for assessment of systemic evidence for infection and/or inflammation as clinically indicated at Day 1, every 2-3 days until discharge, at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

7.2.2 Blood Chemistry Profile

Blood will be obtained and sent to each site's clinical chemistry laboratory for determination of serum sodium, potassium, chloride, bicarbonate, glucose, BUN, creatinine, aspartate aminotransferase (AST/SGOT), alanine aminotransferase (ALT/SGPT), alkaline phosphatase, total bilirubin, direct bilirubin, gamma-glutamyl transferase (GGT), albumin, lactate dehydrogenase (LDH), N-terminal pro-brain natriuretic peptide (NT-proBNP), and troponin as clinically indicated at Day 1, every 2-3 days until discharge, at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

7.2.3 Pregnancy Test

A urine or serum pregnancy test will be obtained from female participants who are of childbearing age prior to their participation in the study.

7.2.4 Urinalysis

Urine will be obtained and sent to each site's clinical laboratory as clinically indicated for determination of color, specific gravity, pH, protein, glucose, ketones, and blood.

7.3 Research Laboratory Measurements

7.3.1 SARS-CoV-2

Blood will be obtained and sent to a central research laboratory for determination of plasma SARS-CoV-2 spike, S1 and nucleocapsid antigens and IgM, IgG, and IgA antibodies against SARS-CoV-2 at Day 1, every 2-3 days prior to discharge, and at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

Stool will be collected and sent to central laboratory for determination of SARS-CoV-2 RNA in stool at Day 1, Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

7.3.2 Zonulin

Blood will be obtained and sent to a central research laboratory for determination of plasma zonulin at Day 1, every 2-3 days prior to discharge, and at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

7.3.3 Cytokines

Blood will be obtained and sent to a central research laboratory for determination of plasma IFN-γ, IL-1β, IL-17, and IL-2R at Day 1, every 2-3 days prior to discharge, and at clinic visits Week 1, Week 2, Week 3, Week 12, and Week 24/EOS.

7.3.4 Pharmacokinetic (PK) Sampling

Pharmacokinetic samples will be drawn after at least 24 hours of dosing and prior to discharge. Samples must be obtained once during the 21 day dosing period. Samples will be drawn pre-dose and 30 minutes, 60 minutes, 2 hours, and four hours after dosing.

8 EVALUATIONS BY VISIT

8.1 Visit 1 (Day 1; Screening/Baseline)

1. Review the study with the participant (participant's legal representative) and obtain written informed consent and HIPAA authorization and assent, if appropriate.
2. Assign the participant a unique screening number.
3. Record demographics data.
4. Record medical history, including a history of SARS-CoV-2 symptoms
5. Record concomitant medications.
6. Perform a complete physical examination.
7. Perform and record vital signs, weight, and height.
8. Obtain an electrocardiogram
9. Complete PedsQL GI Symptoms Scale questionnaire
10. Assess MIS-C-related symptoms via questionnaire
11. Complete activity limitations questions
12. Collect biospecimens for baseline clinical laboratory and research tests (see Section 7.2 and 7.3).
13. Randomize participant
14. Initiate administration of study drug orally QID
15. Dispense drug diary.

8.2 Visits 2-6 (Days 2-6)

1. Record any adverse experiences
2. Record changes to concomitant medications.
3. Perform physical examination.
4. Perform and record vital signs and weight.
5. Obtain an electrocardiogram
6. Complete PedsQL GI Symptoms Scale questionnaire (Day 3 and 5 ONLY)
7. Assess MIS-C-related symptoms via questionnaire
8. Complete activity limitations questions
9. Collect biospecimens for clinical and research laboratory tests (see Section 7.2 and 7.3).
10. Continue to administer study drug orally QID for participants who have been discharged from the hospital dispense study medication with instructions to take one capsule orally QID.
11. For participants who have been discharged from the hospital, schedule the next clinic visit.
12. Review drug diary.
13. Complete PK sampling prior to discharge for inpatient participants.

8.3 Visits 7-9, (Week 1, Week 2 and Week 3 (Day 7 ± 3; Day 14 ± 3; Day 21 ± 3)

1. Record any adverse experiences
2. Record changes to concomitant medications.
3. Perform physical examination.
4. Perform and record vital signs and weight.
5. Obtain an electrocardiogram

6. Complete PedsQL GI Symptoms Scale questionnaire
7. Assess MIS-C-related symptoms vis questionnaire
8. Complete activity limitations questions
9. Collect biospecimens for clinical and research laboratory tests (see Section 7.2 and 7.3)
10. Continue to administer study drug orally QID; for participants who have been discharged from the hospital dispense study medication with instructions to take one capsule orally QID.
11. Conduct study drug accountability at each clinic visit
12. For participants who have been discharged from the hospital, schedule the next clinic visit or telemedicine visit.
13. Review drug diary.
14. Complete PK sampling prior to discharge for inpatient participants if not already obtained.

8.4 Visit 10 and 11 (Week 4 and Week 8) (Day 28 ± 3; Day 56 ± 3)

Conduct visits via telephone contact for those participants who have been discharged from hospital.

1. Record any adverse experiences
2. Record changes to concomitant medications
3. Complete PedsQL GI Symptoms Scale questionnaire
4. Assess MIS-C-related symptoms via questionnaire
5. Complete activity limitations questions

8.5 Visit 12 (Day 84 ± 3)

1. Record any adverse experiences
2. Record changes to concomitant medications.
3. Perform physical examination.
4. Perform and record vital signs and weight
5. Obtain an electrocardiogram
6. Complete PedsQL GI Symptoms Scale questionnaire
7. Assess MIS-C-related symptoms
8. Activity limitations via questionnaire
9. Collect biospecimens for clinical and research laboratory tests (see Section 7.2 and 7.3).
10. For participants who have been discharged from the hospital, schedule telemedicine visits.

8.6 Visit 13 and 14 (Week 16 and Week 20) (Day 112 ± 3; Day 140 ± 3)

Conduct visits via telephone contact for those participants who have been discharged from hospital.

1. Record any adverse experiences
2. Record changes to concomitant medications
3. Complete PedsQL GI Symptoms Scale questionnaire
4. Assess MIS-C-related symptoms via questionnaire
5. Complete activity limitations questions

8.7 Visit 15 (Day 168 ± 3) End of Study or Early Withdrawal Visit

1. Record any adverse experiences

2. Record changes to concomitant medications.
3. Perform complete physical examination.
4. Perform and record vital signs and weight.
5. Obtain an electrocardiogram.
6. Complete PedsQL GI Symptoms Scale questionnaire
7. Assess MIS-C-related symptoms via questionnaire
8. Complete activity limitations questions
9. Collect biospecimens for clinical and research laboratory tests (see Section 7.2 and 7.3)

9 ADVERSE EXPERIENCE REPORTING AND DOCUMENTATION

9.1 Adverse Events

An adverse event (AE) is any untoward medical occurrence in a clinical investigation of a patient administered a pharmaceutical product and that does not necessarily have a causal relationship with the treatment. An AE is therefore any unfavorable and unintended sign (including an abnormal laboratory finding), symptom or disease temporally associated with the administration of an investigational product, whether or not related to that investigational product. An unexpected AE is one of a type not identified in nature, severity, or frequency as noted in Section 9.5.

The Investigator will probe, via discussion with the participant, for the occurrence of AEs during each participant visit and record the information in the site's source documents. Adverse events will be recorded in the patient CRF. Adverse events will be described by duration (start and stop dates and times), severity, outcome, treatment, and relation to study drug, or if unrelated, the cause.

9.2 AE Severity

The current version of the National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE) should be used to assess and grade AE severity, including laboratory abnormalities judged to be clinically significant. The modified criteria can be found in the study manual. If the experience is not covered in the modified criteria, the guidelines shown in Table **9-1** should be used to grade severity. It should be pointed out that the term "severe" is a measure of intensity and that a severe AE is not necessarily serious.

Table 9-1 Adverse Event Severity Grading

Severity (Toxicity Grade)	Description
Mild (1)	Transient or mild discomfort; no limitation in activity; no medical intervention or therapy required. The participant may be aware of the sign or symptom but tolerates it reasonably well.
Moderate (2)	Mild to moderate limitation in activity, no or minimal medical intervention/therapy required.
Severe (3)	Marked limitation in activity, medical intervention/therapy required, hospitalizations possible.
Life-threatening (4)	The participant is at risk of death due to the adverse experience as it occurred. This does not refer to an experience that hypothetically might have caused death if it were more severe.

9.3 AE Relationship to Study Drug

The relationship of an AE to the study drug should be assessed using the following the guidelines in **Table 9-2**.

Table 9-2 Adverse Event Relationship to Study Drug

Relationship to Drug	Description
Definitely	Previously known toxicity of agent; or an event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to the suspected drug; that is confirmed by stopping or reducing the dosage of the drug; and that is not explained by any other reasonable hypothesis.
Probably	An event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to the suspected drug; that is confirmed by stopping or reducing the dosage of the drug; and that is unlikely to be explained by the known characteristics of the participant's clinical state or by other interventions.
Possibly	An event that follows a reasonable temporal sequence from administration of the drug; that follows a known or expected response pattern to that suspected drug; but that could readily have been produced by a number of other factors.
Unrelated	An event that can be determined with certainty to have no relationship to the study drug.

9.4 Serious Adverse Experiences (SAE)

An SAE is defined as any AE occurring at any dose that results in any of the following outcomes:

- death
- a life-threatening adverse experience
- inpatient hospitalization or prolongation of existing hospitalization
- a persistent or significant disability/incapacity
- a congenital anomaly/birth defect

Other important medical events may also be considered an SAE when, based on appropriate medical judgment, they jeopardize the participant or require intervention to prevent one of the outcomes listed.

9.4.1 Serious Adverse Experience Reporting

Study site will document all SAEs that occur (whether or not related to study drug) on an SAE Report Form. The collection period for all SAEs will begin after informed consent is obtained and end after procedures for the final study visit have been completed.

All SAE Report Forms will be reviewed by the site investigator and sent to the IRB within one business day of the site learning of the event. The site will notify the IRB of additional information or follow-up to an initial SAE Report as soon as relevant information is available. Follow-up information is reported on an SAE Report Form.

9.5 Expected Side Effects/Risks

The risks associated with the use of AT1001 are low, based on the previous experience in over 800 participants exposed to oral AT1001, both healthy and affected by an autoimmune disease (celiac disease). The most common AEs reported in studies of orally administered AT1001 were

gastrointestinal symptoms (e.g., nausea, diarrhea, constipation, and abdominal pain) and headache that were mild-to-moderate in severity. These events have been reported at similar rates in placebo-treated participants.

A positive safety profile for orally administered AT1001 has been demonstrated in a comprehensive Phase 1/Phase 2 clinical program. In these studies, AT1001 was well tolerated, and an acceptable safety profile was demonstrated. The overall incidence and type of TEAEs reported in participants treated with AT1001 were generally similar to placebo. In studies measuring PK parameters, all plasma levels were below the limit of quantification (BLQ), demonstrating that the drug is not systemically absorbed.

No signs of toxicity have been observed in animal toxicology studies and no significant effects were observed in any of the safety pharmacology studies.

9.6 Reporting Adverse Events (Including Serious Adverse Events)

Adverse events will be captured in the participants' source documents and captured retrospectively on a study specific electronic CRF (eCRF).

Each adverse clinical event will be evaluated retrospectively for duration, intensity, and whether the event may have been associated with the study drug or other causes. Start and stop dates, relationship to study drug, medical management, and alternative causality of event will be recorded.

10 DISCONTINUATION AND REPLACEMENT OF PARTICIPANTS

10.1 Early Discontinuation of Study Drug

A participant may be discontinued from study treatment at any time if the participant, the investigator, or the Principal Investigator feels that it is not in the participant's best interest to continue. The following is a list of possible reasons for study treatment discontinuation:

- Participant withdrawal of consent (or assent)
- Participant is not compliant with study procedures
- Adverse event that in the opinion of the investigator would be in the best interest of the participant to discontinue study treatment
- Protocol violation requiring discontinuation of study treatment
- Lost to follow-up
- Positive pregnancy test (females)

If a participant is withdrawn from treatment due to an adverse event, the participant will be followed and treated by the Investigator until the abnormal parameter or symptom has resolved or stabilized.

All participants who discontinue study treatment should come in for an early discontinuation visit as soon as possible and then should be encouraged to complete all remaining scheduled visits and procedures.

10.2 Withdrawal of Participants from the Study

A participant may be withdrawn from the study at any time if the participant, the investigator, or the Principal Investigator feels that it is not in the participant's best interest to continue.

All participants are free to withdraw from participation at any time, for any reason, specified or unspecified, and without prejudice.

All withdrawn participants will be strongly encouraged, even in the event of drug discontinuation, to remain in the study through the end of the double-blind period and to complete all protocol-specified safety and efficacy assessments.

Reasonable attempts will be made by the investigator to provide a reason for participant withdrawals. The reason for the participant's withdrawal from the study will be specified in the participant's source documents. As noted above, participants who discontinue study treatment early should have an early discontinuation visit.

10.3 Replacement of Participants

Participants who withdraw prior to randomization and treatment initiation will be replaced. However, if they are assigned to treatment group and initiate treatment before withdrawing, these participants will not be replaced. Participants who withdraw from study treatment will not be replaced.

11 DATA SAFETY MONITORING

An independent data and safety monitoring board (DSMB) will actively monitor interim data to make recommendations regarding study conduct. The DSMB will meet once the first 10 participants complete the Day 7 assessment to perform the first interim analysis of the safety data. The interim analysis will be performed by an independent un-blinded statistician, who is not otherwise associated with the conduct of this study. The DSMB will consist of three members, including two independent physicians and a statistician. At least one physician will have expertise in infectious diseases, and one will have experience in DSMB activities. The DSMB responsibilities will be further elaborated in the DSMB charter.

Serious adverse events will be monitored by the committee on an ongoing basis throughout the study.

12 STATISTICAL METHODS AND CONSIDERATIONS

Prior to the analysis of the final study data, a detailed Statistical Analysis Plan (SAP) will be written describing all analyses that will be performed. The SAP will contain any modifications to the analysis plan described below.

12.1 Data Sets Analyzed

For the efficacy endpoints, the primary analysis will be based on the intent-to-treat (ITT) population which is defined as all randomized participants. Per Protocol (PP) population will include all ITT participants who did not have any major protocol violations.

The safety analysis will include all participants from the ITT population who received at least one dose of the study drug in the double-blind treatment period.

12.2 Demographic and Baseline Characteristics

Demographic and baseline characteristics will be summarized descriptively by treatment and overall.

12.3 Analysis of Primary Endpoint

The primary endpoint will be analyzed using Fisher's exact test.

Robustness analysis of the primary efficacy endpoint will include an analysis on the PP population.

12.4 Analysis of Secondary Endpoints

Binary endpoints will be analyzed by the Fisher's exact test. Change from baseline in continuous outcomes will be analyzed by fitting fixed effects linear models or non-parametric models (e.g., Wilcoxon) to the data. For endpoints specific to subject subgroups, such as subjects who are discharged before the end of the study, only individuals within those subgroups will be analyzed.

12.5 Safety Analysis

The number and percent of participants experiencing TEAEs, serious adverse events, and TEAEs leading to study discontinuation treatment-emergent adverse events (TEAEs) will be tabulated by treatment group and summarized according to the current version of Common Terminology Criteria for Adverse Events (CTCAE). Additional summaries will include TEAE severity and relationship to study drug.

Individual and summary vital signs and clinical laboratory data will be presented in tabular form with mean, median, standard deviation, and range (minimum and maximum as appropriate).

For the clinical laboratory safety data and vital signs values outside the reference range will be flagged in the data listings and a list of clinically significantly abnormal values will be presented.

Concomitant medications taken from screening and during the study will be categorized by WHO classification for therapeutic class and drug name and summarized by number and percentage of participants by treatment and overall.

12.6 Sample Size and Randomization

The sample size of 20 participants (AT1001 (N=10) compared to placebo (N=10) is based on clinical judgement and deemed adequate to address study objectives**.**

13 DATA COLLECTION, RETENTION AND MONITORING

13.1 Data Confidentiality

All records identifying the participant must be kept confidential and, to the extent permitted by the applicable laws and/or regulations, not be made publicly available. The name of the participant will not be asked for nor recorded. A sequential identification number will be automatically attributed to each participant registered in the study. This number will identify the participant and must be included on all eCRFs.

Any and all participant information or documentation pertaining to a clinical trial, to the extent permitting, through a "key" kept anywhere, regardless of whether such key is supplied along with the information or documentation or not, must be considered as containing sensitive personal data of the participant, and is therefore subjected to the provisions of applicable data protection ("privacy") regulations. Breach of such regulations may result in administrative or even criminal sanctions.

Particularly, an information sheet prepared according to such regulations and a form to evidence the consent of participants to the processing of such data must therefore accompany the informed consent administered to the participant. Such information must (i) identify the roles of the holder and processor ("responsible", appointed by the holder) of the participant personal data (also if not directly identifying the participant), as well as the purposes of the personal data collection and processing (medical treatment and related/unrelated scientific research), (ii) adequately describe the flows of communication involving them, particularly if third parties should become involved, and (iii) seek the participant's prior and specific consent to such processing.

Participant information or documentation may be considered "anonymous", and as such not participant to privacy regulations, only when no key whatsoever, permitting the identification of the participant, is any longer available.

Particular attention should therefore be paid (and information/consent materials adapted accordingly) whenever participant data are supplied to third parties and may be autonomously processed, or biological samples/materials are taken and kept for future research purposes.

13.2 Data Collection Instruments

The Investigator will prepare and maintain adequate and accurate source documents designed to record all observations and other pertinent data for each participant treated with the study drug. All participant data will be captured in source documents (ie, clinic notes, laboratory data, worksheets, drug accountability records, consent forms etc.). This study will use an Electronic Data Capture (EDC) system. The designated investigator staff will enter the data required by the protocol into the eCRF. Investigator site staff will not be provided access to the EDC system until they have been trained. Automatic validation programs check for data discrepancies in the eCRFs allow modification or verification of the entered data by the investigator staff.

The Principal Investigator is responsible for assuring that the data entered in the eCRF is complete, accurate, and that entry and updates are performed in a timely manner

13.3 Data Management Procedures

The data will be entered into a validated database. The Data Management group will be responsible for data processing, in accordance with procedural documentation. Database lock will occur once quality assurance procedures have been completed.

All procedures for the handling and analysis of data will be conducted using good computing practices meeting FDA guidelines for the handling and analysis of data for clinical trials.

13.4 Data Quality Control and Reporting

After data have been entered into the study database, a system of computerized data validation checks will be implemented and applied to the database on a regular basis. Queries are entered, tracked, and resolved through the EDC system directly. The study database will be updated in accordance with the resolved queries. All changes to the study database will be documented.

13.5 Archival of Data

The database is safeguarded against unauthorized access by established security procedures; appropriate backup copies of the database and related software files will be maintained. Databases are backed up by the database administrator in conjunction with any updates or changes to the database.

At critical junctures of the protocol (e.g., production of interim reports and final reports), data for analysis is locked and cleaned per established procedures.

13.6 Availability and Retention of Investigational Records

The clinical unit will maintain appropriate medical and research records for this trial, in compliance with Section 4.9 of the ICH E6 GCP, and regulatory and institutional requirements for the protection of confidentiality of participants.

Source data are all information, original records of clinical findings, observations, or other activities in a clinical trial necessary for the reconstruction and evaluation of the trial. Examples of these original documents and data records include, but are not limited to, clinical charts, laboratory notes, memoranda, pharmacy dispensing records, recorded data from automated instruments, copies or transcriptions certified after verification as being accurate and complete, x-rays, and participant files and records kept at the pharmacy, at the laboratories, and other departments involved in the clinical trial. Data collection is the responsibility of the clinical trial staff at the site under the supervision of the site Principal Investigator.

The study eCRF is the primary data collection instrument for the study. The investigator should ensure the accuracy, completeness, legibility, and timeliness of the data reported in the eCRFs and all other required reports. Data reported on the eCRF, that are derived from source documents, should be consistent with the source documents or the discrepancies should be explained. All data requested on the eCRF must be recorded. Any missing data must be explained. For eCRFs an audit trail will be maintained by the system. The investigator/institution should maintain the trial documents as specified in Essential Documents for the Conduct of a Clinical Trial (ICH E6 Section

8) and as required by applicable regulations and/or guidelines. The investigator/institution should take measures to prevent accidental or premature destruction of these documents.

Essential documents (written and electronic) should be retained for a period of not less than fifteen (15) years from the completion of the clinical trial unless their retention for an additional period of time because of applicable laws, regulations and/or guidelines.

A file for each participant must be maintained that includes the signed Informed Consent, HIPAA Authorization and Assent Form and copies of all source documentation related to that participant. The Investigator must ensure the reliability and availability of source documents from which the information on the CRF was derived.

All study documents (patient files, signed informed consent forms, copies of CRFs, Study File Notebook, etc.) must be kept secured for a period of two years following marketing of the investigational product or for two years after centers have been notified that the IND has been discontinued.

14 ADMINISTRATIVE, ETHICAL, REGULATORY CONSIDERATIONS

The study will be conducted according to the Declaration of Helsinki, Protection of Human Volunteers (21 CFR 50), Institutional Review Boards (21 CFR 56), and Obligations of Clinical Investigators (21 CFR 312).

To maintain confidentiality, all laboratory specimens, evaluation forms, reports and other records will be identified by a coded number and initials only. All study records will be kept in a locked file cabinet and code sheets linking a patient's name to a patient identification number will be stored separately in another locked file cabinet. Clinical information will not be released without written permission of the participant, except as necessary for monitoring by the FDA. The Investigator must also comply with all applicable privacy regulations (e.g., Health Insurance Portability and Accountability Act of 1996, EU Data Protection Directive 95/46/EC).

14.1 Protocol Amendments

Any amendment to the protocol will be written by the Sponsor in collaboration with the Principal Investigator. Protocol amendments cannot be implemented without prior written IRB approval except as necessary to eliminate immediate safety hazards to patients. A protocol amendment intended to eliminate an apparent immediate hazard to patients may be implemented immediately, provided the IRBs are notified within five working days.

14.2 Institutional Review Boards

The protocol and consent form will be reviewed and approved by the IRB of each participating center prior to study initiation. Serious adverse experiences regardless of causality will be reported to the IRB in accordance with the standard operating procedures and policies of the IRB, and the Investigator will keep the IRB informed as to the progress of the study. The Investigator will obtain assurance of IRB compliance with regulations.

Any documents that the IRB may need to fulfill its responsibilities (such as protocol, protocol amendments, consent forms, information concerning patient recruitment, payment or compensation procedures, or other pertinent information) will be submitted to the IRB. The IRB written unconditional approval of the study protocol and the informed consent form will be in the possession of the Investigator before the study is initiated. This approval must refer to the study by exact protocol title and number and should identify the documents reviewed and the date of review.

Protocol and/or informed consent modifications or changes may not be initiated without prior written IRB approval except when necessary to eliminate immediate hazards to the patients or when the change(s) involves only logistical or administrative aspects of the study. Such modifications will be submitted to the IRB and written verification that the modification was submitted and subsequently approved should be obtained.

The IRB must be informed of revisions to other documents originally submitted for review; serious and/or unexpected adverse experiences occurring during the study in accordance with the standard operating procedures and policies of the IRB; new information that may affect adversely the safety of the patients of the conduct of the study; an annual update and/or request for re-approval; and when the study has been completed.

14.3 Informed Consent Form

Informed consent will be obtained in accordance with the Declaration of Helsinki, ICH GCP, US Code of Federal Regulations for Protection of Human Subjects (21 CFR 50.25[a,b], CFR 50.27, and CFR Part 56, Subpart A), the Health Insurance Portability and Accountability Act (HIPAA, if applicable), and local regulations.

The Investigator will prepare the informed consent form, assent and HIPAA authorization and submit the documents to the IRB for approval. The written consent document will embody the elements of informed consent as described in the International Conference on Harmonization and will also comply with local regulations.

A properly executed, written, informed consent will be obtained from each participant prior to entering the participant into the trial. Information should be given in both oral and written form and participants (or their legal representatives) must be given ample opportunity to inquire about details of the study. If appropriate and required by the local IRB, assent from the participant will also be obtained. If a participant is unable to sign the informed consent form (ICF) and the HIPAA authorization, a legal representative may sign for the participant. A copy of the signed consent form (and assent) will be given to the participant or legal representative of the participant and the original will be maintained with the participant's records.

14.4 Investigator Responsibilities

By signing the Agreement of Investigator form, the Investigator agrees to:

1. Conduct the study in accordance with the protocol except when to protect the safety, rights or welfare of participants.
2. Personally, conduct or supervise the study (or investigation).
3. Ensure that the requirements relating to obtaining informed consent and IRB review and approval meet federal guidelines, as stated in § 21 CFR, parts 50 and 56.
4. Report to the Principal Investigator any AEs that occur in the course of the study, in accordance with §21 CFR 312.64.
5. Ensure that all associates, colleagues, and employees assisting in the conduct of the study are informed about their obligations in meeting the above commitments.
6. Maintain adequate and accurate records in accordance with §21 CFR 312.62 and to make those records available for inspection with the Sponsor (or designee).
7. Ensure that an IRB that complies with the requirements of §21 CFR part 56 will be responsible for initial and continuing review and approval of the clinical study.
8. Promptly report to the IRB all changes in the research activity and all unanticipated problems involving risks to participants or others (to include amendments and IND safety reports).
9. Seek IRB approval before any changes are made in the research study, except when necessary to eliminate hazards to the patients/participants.

10. Comply with all other requirements regarding the obligations of clinical investigators and all other pertinent requirements listed in § 21 CFR part 312.

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