A phase II, single-arm open-label multi-center study of sirolimus in previously treated idiopathic multicentric Castleman disease

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List of Abbreviations and Definitions

Abbreviations:

iMCD	Idiopathic multicentric Castleman disease
HHV-8	Kaposi's sarcoma virus / human herpes virus-8
IL	Interleukin
PI3K	Phosphoinositide 3-kinase
mTOR	Mechanistic target of rapamycin
VEGF	Vascular endothelial growth factor
sIL2R	Soluble IL-2 receptor
CBR	Clinical benefit response
CHAP	high sensitivity **C**-reactive protein, **h**emoglobin, **a**lbumin and eastern cooperative oncology group **p**erformance score
CRP	C-reactive protein
ECOG	Eastern cooperative oncology group
S6K1	p70S6 kinase Eukaryotic translation initiation factor 4E-binding protein 1
4E-BP1	Eukaryotic translation initiation factor 4E-binding protein 1
FKBP-12	FK binding protein-12
EBV	Epstein-Barr virus
MTI	mTOR inhibitor
FDA	Food and Drug Administration
P-gp	P-glycoprotein
CYP3A4	Cytochrome P450 IIIA4
GUID	Global Unique Identifier
AE	Adverse event
SAE	Serious adverse event
CTCAE	Common terminology criteria for adverse events
eGFR	Estimated glomerular filtration rate
ANC	Absolute neutrophil count
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
HIV	Human immunodeficiency virus
CDCN	Castleman Disease Collaborative Network
IDS	Investigational Drug Service
UAMS	University of Arkansas for Medical Sciences
CHOP	Children's Hospital of Philadelphia
IP	Investigational Pharmacy
CT	Computed tomography
PET	Positron emission tomography
EDTA	Ethylenediaminetetraacetic acid
IRB	Institutional Review Board
ICF	Informed consent form
eCRF	Electronic case report form
CR	Complete response
CRu	CR/unconfirmed
PR	Partial response
SD	Stable disease
PD	Progressive disease
SPD	Sum of the products of the greatest diameters

DSMB	Data Safety Monitoring Board
HMG-CoA	3-hydroxy-3-methyl-glutaryl-CoA reductase
HIPAA	Health Insurance Portability and Accountability Act of 1996
PHI	Protected health information
LAR	legally authorized representative

Definitions:

Eligible Subject	A subject deemed to meet all inclusion criteria and not violate any exclusion criteria.
Enrolled Subject	A subject who has provided informed consent to participate in this study.
Anti-IL-6 blockade	Siltuximab or tocilizumab

Study Summary

Title	A phase II, single-arm open-label multi-center study of sirolimus in previously treated idiopathic multicentric Castleman disease
Short Title	Sirolimus in iMCD
Phase	Phase II
Methodology	Open-label
Study Duration	Each subject's participation will last up to 73 weeks. The entire study is expected to last 3 to 3.5 years.
Study Center(s)	Multi-center study with four institutions: the University of Pennsylvania, the Children's Hospital of Philadelphia, the University of Arkansas for Medical Sciences, and the University of California, San Diego.
Primary Objective	To evaluate the efficacy of sirolimus for treatment of symptomatic iMCD
Exploratory Objective	To investigate the mechanism of action by which sirolimus is efficacious in iMCD through cellular and molecular studies in the research laboratory
Number of Subjects	To achieve up to 24 evaluable subjects, up to 40 subjects may be enrolled across the three sites
Main Inclusion and Exclusion Criteria	• Males or non-pregnant females, ages 2-80 • Documented iMCD history with evidence of active disease at the time of enrollment • Failed, refractory, relapsed, or inability to tolerate anti-IL-6 or anti-IL-6 receptor therapy • Certain comorbidities and concurrent therapeutics are exclusionary
Investigational Product, dose, route of administration	Product: SIROLIMUS (AY-22989, rapamycin, Rapamune®) Dosing (rounded to the nearest mg): Adults: Loading: 5 mg/m^2 on day 1 Daily: 2.5 mg/m^2/day Children: 2.0 mg/m^2/day Target trough level: Adults: 10-15 ng/mL Children: 5-15 ng/mL Route of Administration: Oral
Duration of Administration	0 to 12 $\pm$ 1 month
Reference Therapy	N/A

Statistical Methodology	This is a study to understand the impact of sirolimus on iMCD symptomology. We will examine the changes in a variety of clinical symptoms, laboratory tests, and imaging findings pre- and post-drug treatment to determine if sirolimus is efficacious. Parallel research laboratory assays will investigate mechanisms of action or failure.
Safety Evaluations	• Complete blood count with differential • Lipid panel
Data and Safety Monitoring Plan	The licensed site investigators will be responsible for monitoring the data quality and the ongoing safety of subjects at their respective sites. A data safety monitoring board (DSMB) will provide additional guidance and will review data quality and subject safety on an ongoing basis.

1 Background and Study Rationale

This document is a clinical research protocol and the study described will be conducted in compliance with the protocol and in full accordance with all applicable University of Pennsylvania Research Policies and Procedures and all applicable Federal and state laws and regulations including, as applicable 45 CFR 46, 21 CFR Parts 50, 54, 56, and **Good Clinical Practice**: Consolidated Guidelines approved by the International Conference on Harmonisation. All episodes of noncompliance will be documented.

Human herpesvirus (HHV)-8-negative, idiopathic multicentric Castleman disease (iMCD) is a rare and deadly hematologic illness. Recent research has suggested a key role for the phosphoinositide 3-kinase(PI3K)/Akt/mechanistic target of rapamycin (mTOR) pathway in iMCD pathogenesis[1] and off-label administration of sirolimus,[2] an mTOR inhibitor (MTI), resulted in Clinical Benefit Response (CBR) (as previously used in the Phase I study of siltuximab[3]) in two iMCD patients experiencing active disease, and a prolonged and ongoing remission in one patient. Based on these experiences, we plan to evaluate the efficacy of sirolimus as a therapy for iMCD patients who are either unable to tolerate anti-IL-6 blockade therapy (siltuximab or tocilizumab), or who fail, relapse, or are refractory to such treatment.

1.1 Background and Relevant Literature

iMCD is a rare, deadly hematologic illness. Castleman disease describes a group of related diseases defined by characteristic lymph node histopathology - atrophic or hyperplastic germinal centers, prominent follicular dendritic cells, hypervascularization, polyclonal lymphoproliferation, and/or polytypic plasmacytosis. In addition, iMCD patients can experience episodic disease flares characterized by laboratory abnormalities and systemic symptoms, including constitutional symptoms, cytopenias, hepatosplenomegaly, fluid accumulation, and cytokine storm-associated multiple organ system dysfunction.[4] iMCD's heterogeneous presentation and symptoms, and its overlap with hematologic, oncologic, rheumatologic, and infectious disease makes its diagnosis and treatment difficult. iMCD is diagnosed in approximately 600-1,000 individuals annually in the USA[5] and 35% of iMCD patients die within five years of diagnosis.[6] The causes, etiological driver and pathogenic processes that give rise to iMCD are unknown.

Current therapeutic options are limited and provide benefit for only a subset of patients. Blockade of IL-6 signaling with siltuximab, the only FDA-approved iMCD treatment, or tocilizumab,[7] approved for iMCD treatment in Japan, abrogates symptoms and improves lymphadenopathy in a portion of patients. However, 66% of patients in the siltuximab Phase II clinical trial did not meet response criteria,[8] and recent studies found that IL-6 is not significantly elevated in many iMCD patients.[9] Off-label treatment options are empiric, and include corticosteroids, rituximab, and cytotoxic chemotherapy, which have varying efficacies and significant potential toxicities.[10] Identification of molecular and cellular abnormalities for therapeutic targeting is urgently needed for anti-IL-6 blockade refractory patients. Unfortunately, no new drugs are in development.

As no animal or cell models exist, our understanding of iMCD is based on analysis of patient samples. These data demonstrate a key role for the PI3K/Akt/mTOR pathway in iMCD pathogenesis: upregulation of serum vascular endothelial growth factor (VEGF) (16/20 patients),[11] activated T cells (n = 6 patients)[12] and elevated serum soluble IL-2 receptor (sIL2R) levels (20/21 patients), and elevated PI3K/Akt/mTOR signaling in lymph node tissue (n = 10 patients)[1] from iMCD patients during disease flare. Additionally, gene-set and pathway analyses of plasma proteomic data identified PI3K signaling as the most significantly enriched canonical pathway during flare compared to remission in six patients, and six of the top 20 most significant

compounds predicted to decrease expression of plasma proteins elevated in flare by enrichment analysis using the Library of Integrative Network-based Cellular Signatures 1000 database target PI3K/Akt/mTOR signaling.[13] Most notably, off-label administration of the MTI, sirolimus, to anti-IL6 blockade refractory patients resulted in CBR in two patients experiencing active disease and a prolonged and ongoing remission in one patient[2] with a history of five disease flares. Based on these experiences, we plan to evaluate the efficacy of sirolimus as a second line therapy to IL-6 blockade therapy in iMCD, and to conduct research studies aimed at better understanding the pathogenesis of iMCD and how blockade of PI3K/Akt/mTOR signaling abrogates the disease. We anticipate the results of this work will establish safety and efficacy profiles of sirolimus as a second line therapy for iMCD.

The primary endpoint of this study is efficacy, as evaluated by CBR criteria. This evaluation criteria was employed as the main efficacy end-point in the open-label Phase I clinical trial of siltuximab,[3] and is a composite of clinical (fatigue, anorexia, fever, night sweats and weight gain), radiologic (≥25% bi-dimensional decrease in size of largest lymph node, as measured using Cheson criteria[14,15] (**Appendix 15.2**), and laboratory (hemoglobin) measures relevant to the clinical management of iMCD. CBR is a well-suited and established criterion for determining efficacy in iMCD.

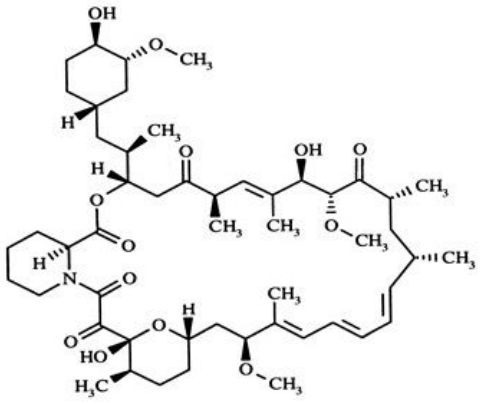
One secondary endpoint of this study is efficacy, as evaluated by modified Cheson response criteria. The Cheson response criteria provides standardized response criteria for uniform endpoints for clinical trials and was developed by a National Institutes of Cancer supported international working group. These standardized response criteria are a result of standardized methods for quantifying disease burden.

A secondary endpoint of this study is efficacy as evaluated by the CHAP criteria (**C**-reactive protein (CRP), **h**emoglobin, **a**lbumin and Eastern Cooperative Oncology Group (ECOG) **p**erformance score).[16] Each criterion in the CHAP scoring system provides a graded measure for a patient's disease activity. The sum of the four scores provides an objective scale for measuring a patient's disease activity and monitoring how it changes over time.

A secondary endpoint of this study is efficacy as evaluated by MCD-related Overall Symptom Score as measured by 34 outcome measures (Section 7.6, Efficacy Evaluations), at 3, 6, and 9 months ± 2 weeks and 12 ± 1 month. Assessments will be compared to those obtained at the Baseline Visit.

1.2 Name and Description of the Investigational Product

SIROLIMUS (AY-22989, rapamycin, Rapamune®)



Source and Pharmacology: Sirolimus is a macrocyclic lactone produced by Streptomyces hygroscopicus. The chemical name of sirolimus (also known as rapamycin) is

(3S,6R,7E,9R,10R,12R,14S,15E,17E,19E,21S,23S,26R,27R,34aS)-9,10,12,13,14,21,22,23,24,25,26,27,32,33,34, 34a-hexadecahydro-9,27-dihydroxy-3-[(1R)-2-[(1S,3R,4R)-4-hydroxy-3-methoxycyclohexyl]-1-methylethyl]-10,21-dimethoxy 6,8,12,14,20,26-hexamethyl-23,27-epoxy-3H-pyrido[2,1-c][1,4] oxaazacyclohentriacontine-1,5,11,28,29 (4H,6H,31H)-pentone. Its molecular formula is $C_{51}H_{79}NO_{13}$ and its molecular weight is 914.2 g/mol. Sirolimus is a white to off-white powder and is insoluble in water, but freely soluble in benzyl alcohol, chloroform, acetone, and acetonitrile. The structural formula of sirolimus is illustrated as follows.

1.2.1 Nonclinical Data

Sirolimus inhibits the mTOR protein.[17] mTOR is a large protein conserved throughout many species.[18,19] mTOR is a serine/threonine kinase and is a member of the PI3 kinase-related kinase family.[19] mTOR acts as a key regulator of cell growth,[20,21] protein synthesis, and cell cycle progression through interactions with other proteins, including p70S6 kinase (S6K1),[21] PI3K[22], and Eukaryotic translation initiation factor 4E-binding protein 1 (4E-BP1)[22].

By inhibiting mTOR, sirolimus mimics growth factor withdrawal by inhibition of protein synthesis and inhibition of cell cycle progression at the G_1 to S transition.[18] Sirolimus has been shown to have growth inhibitory effects in B cells and T cells.[23] In vitro inhibition of mTOR results in down-regulation of many of the pathway targets, including S6K1, S6, and 4E-BP1. This down-regulation results in decreased protein synthesis and inhibition of the cell cycle. Sirolimus inhibits T lymphocyte activation and proliferation that occurs in response to antigenic and cytokine (IL-2, IL-4, and IL-15) stimulation by a mechanism that is distinct from that of other immunosuppressants. Sirolimus also inhibits antibody production. In cells, sirolimus binds to the immunophilin, FK Binding Protein-12 (FKBP-12), to generate an immunosuppressive complex. The sirolimus:FKBP-12 complex has no effect on calcineurin activity. This complex binds to and inhibits mTOR. This inhibition suppresses cytokine-driven T cell proliferation, inhibiting the progression from the G_1 to the S phase of the cell cycle.

Sirolimus is a potent immunosuppressive agent which prolongs the survival of the host and transplanted grafts in animal transplant models of kidney, heart, skin, islet, small bowel, pancreatico-duodenal, and bone marrow. In rodent models of autoimmune disease, sirolimus suppresses immune-mediated events associated with systemic lupus erythematosus, collagen-induced arthritis, autoimmune type I diabetes, autoimmune myocarditis, experimental allergic encephalomyelitis, graft-versus-host disease, and autoimmune uveoretinitis.

1.2.2 Clinical Data to Date

MTIs are a class of signal transduction inhibitors developed as immunosuppressive agents. Sirolimus was the first MTI to be used in a clinical setting. Sirolimus has established safety data and is approved by the USA FDA for the prevention of allograft rejection in renal transplant patients ≥ 13 years of age[24,25] and for the treatment of lymphangioleiomyomatosis. Sirolimus is well tolerated in humans with minimal toxicities[26]. It has documented efficacy in patients with autoimmune diseases, including rheumatoid arthritis,[27] immunodysregulation polyendocrinopathy enteropathy X-linked syndrome[28] and thrombotic thrombocytopenic purpura.[29] It is active in Epstein-Barr virus (EBV) lymphoproliferative disease,[30] and against lymphoid malignancies.[31,32] Sirolimus also has clear anti-neoplastic activity against a variety of cancers.[33,34] Sirolimus causes little nephrotoxicity and neurotoxicity, unlike the commonly used immunophilins, cyclosporine and tacrolimus, but may cause hyperlipidemia and mild myelosuppression.[35]

Off-label administration of sirolimus to two anti-IL6 blockade refractory iMCD patients experiencing active disease resulted in CBR in both patients. Additionally, sirolimus therapy provided a prolonged and ongoing remission in one patient with a history of five disease flares.[2]

1.2.2.1 Human Pharmacokinetics

The pharmacokinetics of sirolimus have been studied in healthy subjects, pediatric dialysis patients, hepatically-impaired adult patients, and adult renal transplant patients.[36,37] Oral doses are rapidly, though variably, absorbed. Mean time-to-peak concentrations range from 1 hour in healthy subjects to 2 hours in renal transplant recipients. Half-life is upwards of 2½ days. Patients who ingested the drug after a high fat breakfast did have delayed C_{max} and it is recommended to take sirolimus consistently with or without food.

The plasma protein binding of sirolimus is approximately 92% mainly to albumin, alpha-1-acid glycoprotein and lipoproteins, and has a high level of association with erythrocytes. The systemic availability of sirolimus was estimated to be approximately 14%. Sirolimus is extensively metabolized by O-demethylation and/or hydroxylation to at least seven major metabolites. The parent drug contributes to more than 90% of the immunosuppressive activity. The main route of elimination is through the feces (91%). The mean half-life increased from 79 ± 12 hours in subjects with normal hepatic function to 113 ± 41 hours in patients with impaired hepatic function. Males have a 12% lower clearance of sirolimus than females after oral solution administration. In some studies, the immunosuppressive effect of sirolimus lasts up to 6 months after discontinuation of therapy. No differences were demonstrated between black and non-blacks. Sirolimus is a substrate for both cytochrome P450 IIIA4 (CYP3A4) and P-glycoprotein (P-gp). Sirolimus is extensively metabolized by the CYP3A4 isozyme in the intestinal wall and liver and undergoes counter-transport from enterocytes of the small intestine into the gut lumen by the P-gp drug efflux pump. Sirolimus is potentially recycled between enterocytes and the gut lumen to allow continued metabolism by CYP3A4. Therefore, absorption and subsequent elimination of systemically absorbed sirolimus may be influenced by drugs that affect these proteins. Drugs that stimulate or inhibit p-450 enzymes will alter clearance of sirolimus and close attention to potential drug interactions is crucial. Care should be exercised when drugs or other substances that are metabolized by CYP3A4 are administered concomitantly with sirolimus. Grapefruit juice reduces CYP3A4-mediated metabolism of sirolimus; subjects will be advised to avoid ingesting grapefruit.

1.2.2.2 Clinical Studies

The safety and efficacy of sirolimus in the prevention of organ rejection have been demonstrated in two randomized, double-blind, multicenter, controlled trials involving over 1000 adult patients. In these and most trials, sirolimus has been administered with cyclosporine and corticosteroids and limited pharmacokinetic data is available with sirolimus alone in this setting. The major side effects noted in these studies included thrombocytopenia, hypercholesterolemia, hypertriglyceridemia, and diarrhea. Renal function was not worse in these patients.

Sirolimus has also been studied in children, indicating a dose of 2 mg/m^2/day is required to achieve target sirolimus trough levels.[38]

1.3 Dose Rationale

Sirolimus will be dosed, rounded to the nearest mg, orally as follows: In adults, day 1, a loading dose of 5 mg/m^2 will be administered; subsequent daily dosing will be at 2.5 mg/m^2/day (rounded to the nearest mg), adjusted to achieve a target trough level of 10-15 ng/mL. The loading dose may be adjusted based on the clinical judgement of the treating physician. In children, no loading

dose will be administered, and daily dosing will be at 2 mg/m^2/day (rounded to the nearest mg), adjusted to achieve a target trough level of 5-15 ng/mL.

This dosing schedule is based on safety and efficacy data of sirolimus in diseases with similarities to iMCD. No severe toxicities were observed in any of several single dosing studies with sirolimus doses ranging from 3-21 mg/m^2. In a Phase I pharmacokinetic study conducted in renal transplant patients, doses ranging from 0.5 to 6.5 mg/m^2 were administered every 12 hours.[37] In two randomized, double-blind, multicenter, controlled trials involving over 1000 adult patients typical dosing was 2 mg or 5 mg (~1.3 – 3.3 mg/m^2) administered daily. Phase III studies to date have had concomitant use of cyclosporine, steroid, or both. At a dose of 2 mg/day (~1.3 mg/m^2) sirolimus trough concentration was 8.6 ± 4.0 ng/ml and at 5 mg/day (~3.3 mg/m^2) the trough was 17.3 ± 7.4 ng/ml. Stable renal transplant patients are dose proportional between 3 and 15 mg/m^2/day. Also, in this population a loading dose of 3 times the maintenance dose provided near steady-state concentrations within 1 day in most patients. More recently, a Phase I/II trial of sirolimus in patients with active systemic lupus erythematosus, a disease that shares overlapping clinical and laboratory abnormalities with iMCD, found efficacy with a dose of 2 mg per day with adjustment to maintain a therapeutic range of 6-15 ng/mL.[39]

2 Study Objectives

2.1 Primary Objective

To evaluate the efficacy of sirolimus for treating symptomatic iMCD.

2.2 Exploratory Objective

To investigate the mechanism of action by which sirolimus is efficacious in iMCD.

3 Investigational Plan

3.1 General Design

This study is a Phase II open label study of daily administration of sirolimus in up to 24 evaluable male or female adults. Up to 40 subjects may be enrolled, defined as providing informed consent for the trial, to obtain this target of evaluable subjects. An interim analysis will be conducted after 14 subjects have completed the Visit 9 intermediate assessment. The mechanism of action of sirolimus in clinically active iMCD will also be examined.

Adult subjects will receive a sirolimus loading dose of 5 mg/m^2, rounded to the nearest mg, on day 1. Loading dose may be lower at the discretion of the prescribing physician. Starting on day 2, adult subjects will take sirolimus daily at 2.5 mg/m^2/day (rounded to the nearest mg), adjusted to a target trough level of 10-15 ng/mL. Pediatric subjects will not receive a loading dose, and daily dosing will be at 2 mg/m^2/day (rounded to the nearest mg), adjusted to achieve a target trough level of 5-15 ng/mL. (see **Section 5.2, Intervention Regimen**). Given that sirolimus is extensively metabolized by CYP3A4, sirolimus dosing for subjects receiving strong CYP3A4 inhibitors or inducer therapy (**https://drug-interactions.medicine.iu.edu/main-table.aspx**) should be monitored carefully and adjusted accordingly. Additionally, sirolimus trough levels should be monitored if patients are started on or if their CYP3A4 therapy is altered, and sirolimus dosing adjusted to maintain trough levels of 10-15 ng/mL (5-15 ng/mL in children). The dosing of adult patients who weigh less than 40 kg should be adjusted, based on body surface area, to 3

mg/m^2 for the loading dose and 1 mg/m^2/day daily. It is recommended that the licensed site investigator reduce maintenance dose in patients with hepatic impairment.

Subjects who meet all inclusion criteria and do not violate any exclusion criteria will be determined eligible for enrollment into the study by the licensed site investigator. The Lead Principal Investigator will confirm the subject's eligibility. During the Intervention Phase (12 ± 1 month), subjects will visit their study site at regular intervals to assess any changes in health status, medication, study drug compliance, and treatment response. They will also be monitored remotely by their licensed site investigator through lab testing performed as per standard of care at local healthcare facilities. During the Follow Up Phase, 12 ± 2 weeks following the End of Treatment Visit, site study staff will contact subjects by phone. Peripheral blood will be collected at baseline and at regular intervals during the Intervention Phase for research purposes. Subjects will be asked to provide their consent to allow any biological samples collected as part of this study or collected during their enrollment period in this study, but for purposes outside of the study, to be used for laboratory research purposes, including genetic sequencing studies. Subjects will also be asked to provide their consent to allow any biological samples from their past medical procedures to be used for laboratory research purposes, including genetic sequencing studies.

3.1.1 Recruitment and Screening Phase

Candidate subjects will be informed of this study as per mechanisms described in **Section 4.3, Subject Recruitment**.

Subjects must meet all inclusion criteria and not violate any exclusion criteria to enroll into this study. Signed informed consent will be obtained from all subjects or their legally authorized representative (LAR), as appropriate, prior to the performance of any protocol-specific activities. The licensed site investigator or his or her designee will obtain consent from subjects at their respective sites. After signing the informed consent, subjects will be considered to be enrolled in the study. All subjects who have signed an informed consent for participation in the study will be assigned a Global Unique Identifier (GUID) using the National Institutes of Health Subjects Global Rare Disease Registry GUID tool available at https://grdr-guid.ncats.nih.gov. Once consented, subjects will undergo a Screening and Baseline (Visit 1, **Section 6.1.1, Baseline Visit**) including pregnancy testing of female subjects of childbearing potential. The licensed site investigator or designated staff member will complete a Patient Evaluation Form confirming all inclusion criteria are met and no exclusion criteria are violated. The Lead Principal Investigator will confirm the subject's eligibility. Subjects whose eligibility is confirmed will continue to the Intervention Phase; those found to violate any criteria will be considered screening failures.

3.1.2 Intervention Phase

Adult subjects will receive an oral sirolimus loading dose of 5 mg/m^2, rounded to the nearest mg (day 1). Loading dose may be lower at the discretion of the prescribing physician. Starting on day 2, adult subjects will receive oral sirolimus daily at 2.5 mg/m^2/day (rounded to the nearest mg), adjusted as needed to a target trough level of 10-15 ng/mL. Pediatric subjects will not receive a loading dose, and daily dosing will be at 2 mg/m^2/day (rounded to the nearest mg), adjusted to achieve a target trough level of 5-15 ng/mL. Subjects will remain on treatment for 12 ± 1 month. The period between the loading dose/first dose and the last treatment define the "Intervention Phase". Patients will continue to receive standard of care treatment during the Intervention Phase consistent with the requirements of **Section 6.6, Subject Withdrawal**. Subjects will undergo regular Intervention Phase visits (**Section 6.2, Intervention Phase**). The licensed site investigator will be ultimately responsible for all treatment decisions, including when to initiate rescue therapy (**Section 6.4, Rescue Therapy**).

3.1.3 Follow Up Phase

Subjects will be contacted via telephone 12 ± 2 weeks following the End of Treatment Visit or Early Termination Visit to document changes in health, medications and to monitor for pregnancy.

3.1.4 Allocation to Interventional Group

All subjects will receive drug; no randomization will occur.

3.2 Study Endpoints

3.2.1 Primary Study Endpoints

The primary endpoint of this study will be the proportion of patients achieving a positive CBR response (**Section 7.6, Efficacy Evaluations**) at 12 ± 1 month. Assessments at 12 ± 1 month will be compared to those obtained at the Baseline Visit.

3.2.2 Secondary Study Endpoints

The secondary endpoints of this study will be:

- The proportion of patients achieving a positive CBR response (**Section 7.6, Efficacy Evaluations**) at 3, 6, and 9 months ± 2 weeks following administration of sirolimus loading dose. Assessments will be compared to those obtained at the Baseline Visit.
- The proportion of patients that remain on study drug for the duration of the study.
- The proportion of patients that indicate that they are currently receiving sirolimus at the end of the Follow Up Phase.
- Disease activity, as measured by the CHAP scale, at 3, 6, and 9 months ± 2 weeks and 12 ± 1 month. Assessments will be compared to those obtained at the Baseline Visit.
- Disease activity, as measured by MCD-related Overall Symptom Score as measured by 34 outcome measures (**Section 7.6, Efficacy Evaluations),** at 3, 6, and 9 months ± 2 weeks and 12 ± 1 month. Assessments will be compared to those obtained at the Baseline Visit.
- The proportion of patients achieving a lymph node response, following the modified Cheson response criteria (**Section 7.6, Efficacy Evaluations**). Assessments will be compared to those obtained at the Baseline Visit.

3.2.3 Exploratory Study Endpoints

The exploratory endpoints of this study will be:

- Results of correlative studies, in the research laboratory, to investigate the mechanism of action of sirolimus in iMCD and clinical and laboratory biomarkers for treatment response. Additional correlative findings may also be assessed.
- Spleen size, as measured by PET/CT or CT.

3.2.4 Safety Endpoints

The safety of sirolimus administration will be evaluated by monitoring for by:

- All previously reported adverse events (AEs) associated with sirolimus treatment
- All possible new AEs

4 Study Population and Duration of Participation

4.1 Inclusion Criteria

To be eligible, subjects must meet all of the below inclusion criteria:

1. Male or female, age 2-80
2. Documented disease history consistent with the diagnostic criteria for iMCD (**Appendix 15.1**)
    - Confirmation of clinico-histopathological features by licensed pathologist, or a CAS grade of at least 3 in the companion registry study (ACCELERATE)
3. Failed/refractory (patient did not achieve sufficient disease control with anti-IL-6 therapy, as determined by the site investigator), relapsed (return of symptoms while on therapy), or inability to tolerate anti-IL-6 or anti-IL-6 receptor therapy
4. Evidence of active disease, defined as at least two abnormalities in the criteria comprising the CBR criteria and as defined as abnormal by CTCAE v4.0, including at least one objective measurement (hemoglobin, weight loss, or lymph node size)
    - Hemoglobin < lower limit of normal
    - Fatigue, including fatigue relieved by rest
    - Anorexia, including loss of appetite without alteration in eating habits
    - Fever or night sweats
    - Weight loss ≥ 5% from baseline
    - At least one lymph node meeting modified Cheson criteria (**Appendix 15.2**)
5. Ability to consume oral medication in the form of a tablet
6. Ability to provide, or for a LAR to provide on their behalf, informed consent prior to any study-specific activities

4.2 Exclusion Criteria

To be eligible, subjects must not violate any of the below exclusion criteria.

1. Subjects cannot be pregnant or nursing females
    - Women of childbearing potential must have a negative pregnancy test documented at the Baseline Visit. Subjects of reproductive potential may not participate unless they have agreed, as part of the informed consent, to use an effective contraceptive method for the duration of the study and for at least 12 weeks after ending treatment
2. Subjects cannot have received any systemic therapy(ies) intended to treat iMCD other than corticosteroids within 14 days of enrollment
    - Corticosteroid treatment must have been initiated more than 28 days prior to enrollment and be maintained at a stable or tapering dose (prednisone or equivalent up to 1 mg/kg/day) prior to enrollment; dose cannot be elevated for the duration of the study but can be maintained or tapered
    - For subjects who cannot or are unwilling to undergo a 14 day washout period:
        i. Anti-IL-6 therapy may be permitted if the subject has been on therapy for at least 3 months, no toxicity has been documented, and sufficient disease control is not achieved
        ii. Anti-IL-6 therapy must be discontinued within the first 6 weeks after starting sirolimus
3. Subjects cannot have previously received sirolimus monotherapy to treat iMCD

4. Subjects cannot have any of the following:
    - ECOG >3 (or Karnofsky/Lansky score ≤ 60 in children)
    - Estimated glomerular filtration rate (eGFR) < 30 mL/min/1.73 m^2 or creatinine > 3.0 mg/dL
    - Absolute neutrophil count (ANC) < 1000 x 10^9/L (< 500 x 10^9/L in children)
    - Hemoglobin ≤ 6.5 g/dL (transfusion independent, defined as not receiving a red blood cell transfusion for ≥ 7 days prior)
    - Aspartate aminotransferase (AST) or alanine aminotransferase (ALT) laboratory values greater than three times the upper limit of normal
    - Albumin < 2 g/dL (transfusion independent, defined as not receiving intravenous albumin for ≥ 7 days prior)
    - Platelet count ≤ 40 x 10^9/L (transfusion independent, defined as not receiving platelet transfusion for ≥ 7 days prior)
    - Pulmonary involvement or interstitial pneumonitis with dyspnea (adequate pulmonary function is defined as pulse oximetry > 94% on room air if there is clinical indication for determination [e.g. dyspnea at rest, history of interstitial pneumonitis, etc.])
    - Fasting cholesterol > 300 mg/dL or fasting triglyceride > 400 mg/dL
5. Subjects cannot have uncontrolled infection or infectious disease(s) that is/are exclusionary for / mimickers of iMCD (**Appendix 15.1**)
    - Fungal disease must be stable for at least two weeks before enrollment
    - Subjects with history of recent bacteremia must have a documented negative blood culture before enrollment
    - Subjects with past history of non-therapy related opportunistic infection should discuss eligibility and possible immunologic evaluation with the licensed site investigator prior to enrollment
6. Subjects cannot have rheumatologic disease(s) that is/are exclusionary for / mimickers of iMCD (**Appendix 15.1**)
7. Subjects cannot have a prior malignancy except for: (1) adequately treated basal cell or squamous cell skin cancer, (2) in situ cervical cancer, or (3) other cancer for which the subject has not received treatment within one year prior to enrollment
    - Other cancers will only be acceptable if the patient's life expectancy exceeds five years and they are not exclusionary diagnoses for iMCD (**Appendix 15.1**)
8. Subjects cannot have a documented history of human immunodeficiency virus (HIV) or HHV-8 infection, or severe combined immunodeficiency syndrome
9. Subjects cannot have a history of liver or lung transplantation
10. Subjects cannot have ongoing or planned participation in another clinical trial involving iMCD directed treatment or that involves immunomodulatory or anti-neoplastic treatment
11. Subjects cannot have prior sensitivity / allergy to any formulation of sirolimus, its components or its analogues
12. Subjects cannot have serious medical illness, or psychiatric illness or disorders that could potentially interfere with the completion of treatment according to this protocol or participation in the trial
13. Subjects cannot have psychiatric disorders that compromise the ability to provide informed consent
14. Subjects cannot have any other condition or finding that in the opinion of the investigator would make participation in this trial inappropriate

4.3 Subject Recruitment

Potential subjects will be informed of this study by:

- Verbal communication from licensed site investigators during routine patient clinical visits. Licensed site investigators may also provide a printed flier about the study to their patients
- Email or phone call, if permitted, to individuals who have consented, in the past as part of a separate research study, to being re-contacted for participation in future research studies
- Email or phone call, to individuals who have been identified via Epic Best Practice Advisories and Slicer Dicer, who have consented to be contacted for participation in research studies
- The Castleman Disease Collaborative Network (CDCN), a nonprofit organization dedicated to supporting Castleman disease patients and accelerating research.
    - The CDCN has a network of physicians located throughout the United States. These physicians have expressed interest in providing information to their patients about research studies. If a physician in the CDCN chooses to approach their patient with information regarding this study, they will provide the individual with a flier
- ClinicalTrials.gov, which will list this study along with contact information for the Site Principal Investigators and study coordinators

Subject recruitment will not use any University of Pennsylvania media services. Any interested subject will be referred to a licensed site investigator for eligibility determination.

4.4 Duration of Study Participation

The Screening/Baseline Phase will include an on-site visit and may last 0 - 28 days. The Intervention Phase will last up to 12 ± 1 month, and the Follow Up Phase will be approximately 12 ± 2 weeks. In total, each subject's participation will last up to 73 weeks.

4.5 Total Number of Subjects and Sites

Up to 40 subjects may enroll into this study to achieve the intended goal of up to 24 evaluable subjects. To determine if the trial will continue, an interim analysis will be completed after 14 subjects have completed the Visit 9 intermediate assessment. This trial involves up to four sites, which are expected to have approximately equal enrollment rates.

4.6 Vulnerable Populations:

Children 2 years of age or older may be considered for this study.
Pregnant women, fetuses, neonates, and prisoners are not included in this research study.

5 Study Intervention

5.1 Description

SIROLIMUS (AY-22989, rapamycin, Rapamune®) NSC# 226080 (092405). Sirolimus is available as a tan, triangular-shaped tablet containing 0.5 mg sirolimus; a white, triangular-shaped tablet containing 1 mg sirolimus; and a yellow to beige triangular-shaped tablet containing 2 mg sirolimus. The inactive ingredients in sirolimus tablets include sucrose, lactose, polyethylene glycol 8000, calcium sulfate, microcrystalline cellulose, pharmaceutical glaze, talc, titanium dioxide, magnesium stearate, povidone, poloxamer 188, polyethylene glycol 20,000, glyceryl

monooleate, carnauba wax, and other ingredients. The 2 mg dosage strength also contains iron oxide yellow 10 and iron oxide brown 70. Sirolimus is commercially available. The prescribing information for Rapamune can be found at:
https://www.accessdata.fda.gov/drugsatfda_docs/label/2018/021083s061,021110s080lbl.pdf.

5.2 Intervention Regimen

Adult subjects will receive sirolimus orally, a loading dose of 5 mg/m^2, rounded to the nearest mg, on day 1. Loading dose may be lower at the discretion of the prescribing physician. Starting on day 2, adult subjects will take oral sirolimus daily at 2.5 mg/m^2/day (rounded to the nearest mg), target trough level 10-15 ng/mL, based on tolerability. Pediatric subjects will not receive a loading dose, and daily dosing will be at 2 mg/m^2/day (rounded to the nearest mg), adjusted to achieve a target trough level of 5-15 ng/mL. Sirolimus trough levels will be determined at approximately Day 5, Day 14, Months 1, 3, 6, 9, and 12 (see **Appendix 15.3** for the study calendar). At the discretion of the study doctor, additional sirolimus trough levels may be obtained. Sirolimus trough levels will be determined in a clinical grade laboratory, which may be located at the study center or at a clinical facility close to the subject, if more convenient.

Frequent dose adjustments based on non-steady-state sirolimus concentrations can lead to overdosing or under dosing because sirolimus has a long half-life. Once maintenance dose is adjusted, patients should continue on the new maintenance dose for at least 7 to 14 days before further dosage adjustment with concentration monitoring. In most patients, dose adjustments can be based on simple proportion: new dose = current dose × (target concentration/current concentration). Dose adjustments are based on clinical judgment of the licensed site investigator after considering clinical toxicity, serum levels, concomitant drug use and the rate of rise or decline of the serum level. For levels < 10 ng/mL, it is suggested, but not required, that the dose of sirolimus be increased, but not to exceed a maximum daily dose of 40 mg. Conversely, for levels > 15 ng/mL, it is suggested, but not required, that the dose be decreased. Alternatively, sirolimus can be withheld entirely as long as serum levels are monitored and the drug is restarted when the level returns to the therapeutic range and the licensed site investigator feels it is appropriate to restart the agent. It is suggested, but not required, that levels of sirolimus be drawn at least weekly until steady state, then monitored thereafter unless a change in medication (e.g. use of voriconazole) or renal function might result in an acute change in level. At that point, levels should be measured as clinically indicated. The anticipated length of treatment will be 12 months. iMCD is a chronic condition and, as sirolimus is commercially available for other indications, patients who show improvement on sirolimus may remain on treatment indefinitely at the discretion of their physician. Examples of dose calculations are provided in **Appendix 15.5 Example Dose Calculations**.

5.3 Receipt

All drug will be obtained from a commercial producer (Pfizer) and be received by the investigational pharmacy at each participating site. Upon receipt of the study treatment supplies, an inventory will be performed and a drug receipt log filled out and signed by the person accepting the shipment. The designated study staff will count and verify that the shipment contains all the items noted in the shipment inventory. Any damaged or unusable study drug in a given shipment will be documented in the study files. The IDS and the IP will notify the Lead Principal Investigator of any damaged or unusable study treatments that were supplied. All drug will be supplied in bulk.

5.4 Storage

Sirolimus tablets should be stored at controlled room temperature, which is 20-25 °C with excursions permitted between 15 °C - 30 °C. The investigational pharmacist at the IDS and the IP will ensure that all study drug is stored in a secured area, under recommended storage conditions, including with regards to protection from light and sterile conditions, and in accordance with applicable regulatory requirements, and will be dispensed by qualified staff members. The pharmacist will maintain accurate records regarding study drug administration and return.

5.5 Preparation and Packaging

Sirolimus tablets will be prepared by a commercial producer (Pfizer) and will be packaged by the investigational pharmacist at the IDS and the IP in child-resistant bottles for dispensation to subjects. Bottles will be labeled to meet state and FDA requirements.

5.6 Administration and Accountability

The IDS and IP service will maintain accurate logs of study drug dispensing, and will conduct regular drug reconciliation checks to document drug assigned, drug consumed, drug remaining, product damage/destroyed and return of damaged/destroyed drug, along with dates of such damage and return. This reconciliation will be logged on the drug reconciliation form, and signed and dated by the service.

Sirolimus will be provided to patients at on-site visits or shipped to subjects as needed (e.g. if their supply is lost, or if results from remote sirolimus trough assays results in a dose adjustment). Sirolimus will be administered orally and should be taken at a consistent time of day and at consistent intervals with regard to meals. Sirolimus may be given with food as long as it is given the same way each time; however, administration with food significantly alters the rate and extent of absorption.

5.7 Subject Compliance Monitoring

At each on-site visit, compliance with study drug will be reinforced; study drug bottles will be returned and compliance will be assessed by pill count. Subjects will be provided a medication diary to complete at home, which will be reviewed at each on-site visit. Study medication dispensed or returned to IDS or the IP must be recorded for Drug Accountability and Reconciliation at each on-site visit in a standard form. Serum trough levels of sirolimus will be monitored. Patients will be informed to withhold their daily dose of sirolimus until after a clinical blood sample is drawn to assay for trough levels of sirolimus.

Non-compliance is defined as two consecutive study visits with >30% of study drug not consumed. Non-compliant patients will receive counseling from the licensed site investigator and be warned that they may be removed from the study. If there is a third study visit with >30% of study drug not consumed, then the licensed site investigator will determine if the subject should be removed from the study.

5.8 Return or Destruction of Investigational Product

At the completion of the study – following the last enrolled patient's final assessment - there will be a final reconciliation of drug shipped, drug consumed, and drug remaining by IDS and the IP. This reconciliation will be logged on the drug reconciliation form, signed and dated. Any discrepancies noted will be investigated, resolved, and documented prior to return or destruction of unused study drug. Drug destroyed on site will be documented in the study files.

6 Study Procedures

Radiological Imaging
Computed tomography (CT) with contrast or positron emission tomography/CT (PET/CT) scan images conducted as standard of care treatment for iMCD will be centrally reviewed by a radiologist at the University of Pennsylvania. PET/CT will be full body imaging, and CT will include neck, chest, abdomen and pelvis. The site physician should maintain consistency, as is medically appropriate, in imaging modality utilized for each subject to that used at the Baseline Visit.

Blood Draws
No more than 100 mL of blood will be drawn at the Baseline Visit, and no more than 55 mL at each subsequent Visit specifically for research laboratory (i.e. non-clinical health or sirolimus monitoring) purposes. Blood will be drawn into ethylenediaminetetraacetic acid (EDTA) coated tubes (Baseline Visit: 80 mL; subsequent Visits: 40 mL) and serum separator tubes (Baseline Visit: 14 mL; subsequent Visits: 7 mL).

Research samples obtained during on-site visits will be processed at the site or shipped to the University of Pennsylvania for processing. Those obtained during remote visits will be shipped to the University of Pennsylvania for processing. Subjects will be provided with appropriate shipping materials and instructions regarding how to ship research samples to the University of Pennsylvania.

Fecal Sample Collections
Fecal samples will be collected at the Baseline Visit, prior to starting sirolimus, at Week 2, and at Visit 9. Fecal sample collections are optional.

Fluid Sample Collections
Fluid samples, such as those collected during paracentesis, may be collected and stored for research analysis. Samples will be shipped to the University of Pennsylvania or the Children's Hospital of Philadelphia for processing, or may be processed at the site.

Clinical Laboratory Testing
For standard of care laboratory testing conducted by a third-party provider, the site physician should maintain consistency in the provider used for the Baseline Visit testing for each subject, as is medically appropriate.

Please see **Appendix 15.3** for the study calendar.

6.1 Screening Phase

6.1.1 Screening Visit (Visit 0)

Subjects' eligibility will be confirmed during the screening phase. The below standard of care procedures will be conducted unless they occurred within four weeks prior to the screening visit as part of standard of care treatment.

- Standard Diagnostic Laboratory Tests (**Appendix 15.4**)
- Vital signs
- Radiological imaging (PET/CT or CT)
- Lipid panel

- Review of concomitant medications

The below non-standard of care procedures will be conducted as part of the screening phase for all subjects:

- Informed consent: a signed and dated Institutional Review Board (IRB) approved informed consent form (ICF) that will be documented in the subject's source documents
- Pregnancy testing for females of childbearing potential
- Recording of demographic information (gender, date of birth, race, etc.) into electronic case report form (eCRF)
- Recording of medical history data and Laboratory Tests, as available, (**Appendix 15.4**) from medical records into eCRF

6.2 Intervention Phase

6.2.1 Baseline Visit (Visit 1)

The baseline visit should occur within 7 days of the screening visit, following confirmation of eligibility. The below standard of care procedures will be conducted unless they occurred within four weeks prior to Visit 0 as part of standard of care treatment.

- Physical exam including performance score
- Vital signs
- Lipid panel
- Review of concomitant medications

The below non-standard of care procedures will be conducted as part of Visit 1 for all subjects:

- Confirmation that subject meets inclusion criteria and does not violate any exclusion criteria, including confirmation by the Lead Principal Investigator
- GUID assigned
- Research blood draw
- Collection of fecal sample (optional)
- Recording of medical history data and Laboratory Tests, as available, (**Appendix 15.4**) from medical records into eCRF
- Dispensing of sirolimus loading dose (adults only)
- Dispensing of sirolimus daily dose

6.2.2 Visits 2 and 3

Subjects will visit the study center, or may visit their local medical provider or facility (e.g. Quest Diagnostics), 5 days $\pm$ 2 days (Visit 2) and 2 weeks $\pm$ 5 days (Visit 3) following administration of loading dose/first dose. The below non-standard of care procedures will be conducted as part of this study during Visits 2 and 3:

- Research blood draw
- Recording of medical history data and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF (at site)
- Phone assessment of AEs, study drug compliance, and review of medication changes
- Sirolimus Trough

- Collection of fecal sample (optional – Visit 3 only)

6.2.3 Visit 4

Subjects will return to the study site 1 month ± 1 week following administration of loading dose. At the physician's discretion, this visit may occur remotely and subject may visit their local medical provider or facility (e.g. Quest Diagnostics). The below procedures will be conducted as standard of care:

- Physical exam including performance score (if visit occurs on site)
- Vital signs (if visit occurs on site)
- Lipid panel
- Standard of Care Laboratory Tests (**Appendix 15.4**)
- Review of medication changes

The below non-standard of care procedures will be conducted as part of this study during Visit 4:

- Research blood draw
- Pregnancy testing for females of childbearing potential (may be self-administered by subjects if visit is conducted remotely)
- Sirolimus Trough
- Assess possible AEs
- Return of any non-consumed sirolimus (may be shipped if visit occurs remotely)
- Sirolimus pill count (if visit occurs on site)
- Dispensing of sirolimus daily dose (may be shipped if visit occurs remotely)
- Recording of medical history data, medication changes, AEs and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF

6.2.4 Visit 5

Subjects will visit the study center, or may visit their local medical provider or facility (e.g. Quest Diagnostics), 2 months ± 2 weeks following administration of loading dose. The below non-standard of care procedures will be conducted as part of this study during Visit 5:

- Research blood draw
- Recording of medical history data, AEs, and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF (at site)
- Phone assessment of AEs, study drug compliance, and review of medication changes

6.2.5 Visit 6

Subjects will return to the study site 3 months ± 2 weeks following administration of loading dose. It is highly encouraged that this visit occurs at the study site, but at the physician's discretion, this visit may occur remotely and subject may visit their local medical provider or facility (e.g. Quest Diagnostics). The below procedures will be conducted as standard of care:

- Physical exam including performance score (if visit occurs on site)
- Vital signs (if visit occurs on site)
- Radiological imaging (PET/CT or CT)
- Standard of Care Laboratory Tests (**Appendix 15.4**)
- Lipid panel

- Review of medication changes

The below non-standard of care procedures will be conducted as part of this study during Visit 6:

- Research blood draw
- Pregnancy testing for females of childbearing potential (may be self-administered by subjects if visit is conducted remotely)
- Sirolimus Trough
- Assess possible AEs
- Return of any non-consumed sirolimus (may be shipped if visit occurs remotely)
- Sirolimus pill count (if visit occurs on site)
- Dispensing of sirolimus daily dose (may be shipped if visit occurs remotely)
- Intermediate assessment of primary objective
- Recording of medical history data, medication changes, AEs and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF

6.2.6 Visit 7 and 8

Subjects will visit the study center, or may visit their local medical provider or facility (e.g. Quest Diagnostics), 4 month ± 2 week (Visit 7) and 5 month ± 2 week (Visit 8) following administration of loading dose. The below non-standard of care procedures will be conducted as part of this study during Visit 7 and 8:

- Research blood draw
- Recording of medical history data, AEs and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF (at site)
- Phone assessment of AEs, study drug compliance, and review of medication changes

6.2.7 Visit 9

Subjects will return to the study site 6 month ± 2 week following administration of loading dose. It is highly encouraged that this visit occurs at the study site, but at the physician's discretion, this visit may occur remotely and subject may visit their local medical provider or facility (e.g. Quest Diagnostics). The below procedures will be conducted as standard of care:

- Physical exam including performance score (if visit occurs on site)
- Vital signs (if visit occurs on site)
- Radiological imaging (PET/CT or CT)
- Standard of Care Laboratory Tests (**Appendix 15.4**)
- Lipid panel
- Review of medication changes

The below non-standard of care procedures will be conducted as part of this study during Visit 9:

- Research blood draw
- Collection of fecal sample (optional)
- Pregnancy testing for females of childbearing potential (may be self-administered by subjects if visit is conducted remotely)
- Sirolimus Trough
- Assess possible AEs

- Return of any non-consumed sirolimus (may be shipped if visit occurs remotely)
- Sirolimus pill count (if visit occurs on site)
- Dispensing of sirolimus daily dose (may be shipped if visit occurs remotely)
- Intermediate assessment of primary objective
- Recording of medical history data, medication changes, AEs and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF

6.2.8 Visit 10 and 11

Subjects will visit the study center, or may visit their local medical provider or facility (e.g. Quest Diagnostics), 7 month ± 2 week (Visit 10) and 8 month ± 2 week (Visit 11) following administration of loading dose. The below non-standard of care procedures will be conducted as part of this study during Visits 10 and 11:

- Research blood draw
- Recording of medical history data, AEs and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF (at site)
- Phone Assessment of AEs, study drug compliance, and review of medication changes

6.2.9 Visit 12

Subjects will return to the study site 9 month ± 2 week following administration of loading dose. It is highly encouraged that this visit occurs at the study site, but at the physician's discretion, this visit may occur remotely and subject may visit their local medical provider or facility (e.g. Quest Diagnostics). The below procedures will be conducted as standard of care:

- Physical exam including performance score (if visit occurs on site)
- Vital signs (if visit occurs on site)
- Radiological imaging (PET/CT or CT)
- Standard of Care Laboratory Tests (**Appendix 15.4**)
- Lipid panel
- Review of medication changes and updates

The below non-standard of care procedures will be conducted as part of this study during Visit 12:

- Research blood draw
- Pregnancy testing for females of childbearing potential (may be self-administered by subjects if visit is conducted remotely)
- Sirolimus Trough
- Assess possible AEs
- Return of any non-consumed sirolimus
- Sirolimus pill count (if visit occurs on site)
- Dispensing of sirolimus daily dose
- Intermediate assessment of primary objective
- Recording of medical history data, medication changes, AEs, and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF

6.2.10 Visits 13 and 14

Subjects will visit the study center, or may visit their local medical provider or facility (e.g. Quest Diagnostics), 10 month ± 2 week (Visit 13) and 11 month ± 2 week (Visit 14) following administration of loading dose. The below non-standard of care procedures will be conducted as part of this study during Visits 13 and 14:

- Research blood draw
- Recording of medical history data, AEs and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF (at site)
- Phone assessment of AEs, study drug compliance, and review of medication changes

6.2.11 End of Treatment Visit (Visit 15)

Subjects will complete the treatment at 12 ± 1 month following administration of loading dose and return to the study site for their final, End of Treatment Visit (Visit 15). Participants who choose to discontinue or who are withdrawn from the study will complete an Early Termination Visit (**Section 6.7**). It is highly encouraged that this visit occurs at the study site, but at the physician's discretion, this visit may occur remotely and subject may visit their local medical provider or facility (e.g. Quest Diagnostics). The below procedures will be conducted as standard of care:

- Physical exam including performance score (if visit occurs on site)
- Vital signs (if visit occurs on site)
- Radiological imaging (PET/CT or CT)
- Standard of Care Laboratory Tests (**Appendix 15.4**)
- Lipid panel
- Review of medication changes and updates

The below non-standard of care procedures will be conducted as part of this study during Visit 15:

- Research blood draw
- Pregnancy testing for females of childbearing potential (may be self-administered by subjects if visit is conducted remotely)
- Sirolimus Trough
- Assess possible AEs
- Return of any non-consumed sirolimus
- Sirolimus pill count (if visit occurs on site)
- Final assessment of primary objective
- Recording of medical history data, medication changes, AEs, and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF

6.3 Follow Up Phase of the Study

6.3.1 Visit 16

The subject will be contacted by phone 12 weeks ± 2 weeks after the End of Treatment Visit or Early Termination Visit and asked questions regarding their health status, if they have become pregnant (if applicable), and if they have continued on drug off-study.

6.4 Rescue Therapy

At the discretion of the licensed site investigator, a subject can be removed from the trial due to disease progression or serious AE (SAE). Decision to remove a subject from the trial and which rescue therapy(ies) to administer rest solely with the licensed site investigator.

6.5 Unscheduled Visits

An unscheduled visit may be required to repeat a laboratory assay or to conduct further safety evaluation. Enrollment into this study places no restrictions upon subjects' ability to visit their physician outside of the study's scheduled visits. Health data generated from such visits will be incorporated into the eCRF at the time of the subject's next scheduled visit.

6.6 Subject Withdrawal

The criteria for enrollment must be followed explicitly. If a subject who does not meet enrollment criteria is inadvertently enrolled, that subject should be discontinued from the study. A subject may voluntarily discontinue participation in this study at any time without impact to their care. The licensed site investigator or Lead Principal Investigator may also, at his or her discretion, discontinue the subject from participating in this study at any time. If a subject is prematurely discontinued from participation in the study for any reason, at any time, at either the licensed site investigator's discretion or the subject's request, an effort must be made to document the reason(s) why a subject failed to return to the study site for necessary visits or is discontinued from the study. Subjects withdrawn from the study may be replaced at the Lead Principal Investigator's discretion. The primary reason for discontinuing participation in the study must be stated in the CRF and may include, but is not limited to, one of the following:

- Progressive disease as determined by the licensed site investigator
- Use of unapproved concomitant medications including any systemic therapy(ies) intended to treat iMCD, as described in the eligibility criteria
- Occurrence of intolerable AEs
- Withdrawal of consent by subject
- Noncompliance with protocol, e.g., the subject fails to appear at one or more visits
- Development of an intercurrent illness, injury, or medical condition likely to interfere with subject safety, the overall assessment, or the required administration of study medication
- Pregnancy
- Development of any condition for which the licensed site investigator feels treatment withdrawal is justified
- Termination of the study

6.6.1 Data Collection and Follow-up for Withdrawn Subjects

Subjects who withdraw consent to participate in the study will be seen for one final Early Termination Visit.

6.7 Early Termination Visit

Subjects who discontinue or are withdrawn early from the study will return to the study site for a final Early Termination Visit. Unless conducted as standard of care, the below standard of care procedures will be performed:

- Physical exam including performance score
- Vital signs

- Standard of Care Laboratory Tests (**Appendix 15.4**)
- Review of medication changes and updates

The below non-standard of care procedures will be conducted as part of this study during the Early Termination Visit:

- Research blood draw
- Assess possible AEs
- Return of any non-consumed sirolimus
- Sirolimus pill count
- Recording of medical history data, medication changes, AEs, and Laboratory Test, as available, (**Appendix 15.4**) from medical records into eCRF

7 Study Evaluations and Measurements

7.1 Medical Record Review

Variables to be abstracted from medical charts (paper or electronic) and entered in the eCRF include elements that fall under the below categories, as available through routine clinical practice or obtained as part of this study.

- Contact information and socio-demographics
- Diagnoses and anthropometric
- Physical exam
- Vital signs
- Pathology
- Laboratory
- Clinical Features
- Radiographic
- Co-morbid conditions
- Medications (previous and current)
- Adverse events
- End of study information (including information on early withdrawal)

7.2 Physical Examination

Physical examination will follow standard of care for the treatment of iMCD as well as those required for assessing the Study Endpoints. Information may be obtained from the medical record.

7.3 Vital Signs

Vital signs will be assessed during each visit in line with standard of care for the treatment of iMCD. Body temperature will be assessed with a digital thermometer under the tongue or in the axilla. Pulse oximetry will be assessed digitally. Blood pressure and pulse rate will be assessed using a digitally-read cuff on individuals sitting up. Respiration rate will be determined by a medical professional.

7.4 Laboratory Evaluations

Blood sampling will be performed for this study for the following laboratory evaluations. Other laboratory tests will be collected as part of standard of care treatment and recorded in the eCRF from medical record review.

- Sirolimus trough levels
- Pregnancy testing (if applicable)
- Lipid panel (if not collected as standard of care)

7.4.1 Clinical Laboratory Tests

Appendix 15.4 contains a listing of standard of care labs that will be collected from subjects' medical records and recorded into the eCRF as part of this study.

7.5 Pregnancy Testing

Women of childbearing potential must have a negative pregnancy test documented during the Baseline Visit. Urine pregnancy testing will be conducted at each on-site visit. If a follow-up visit is conducted remotely, a subject may perform the pregnancy test at home and provide results to the study team via photo.

7.6 Efficacy Evaluations

The below measures will be used to assess the efficacy of the study intervention and determine if the subject meets criteria for a CBR. All comparisons are against those obtained during the Baseline Visit:

Positive response: Relative to baseline, improvement in at least one of the below criterion without worsening of any single criterion other than hemoglobin on two consecutive study visits.

Negative response: Relative to baseline, worsening of any single criterion other than hemoglobin on two consecutive site visits or failure to achieve improvement for any criterion.

	Hemoglobin	Fatigue	Anorexia	Fever and night sweats	Weight	Lymph node size
Improvement	≥2 g/dL increase without transfusions	≥1 grade decrease by NCI CTCAE version 4.0, May, 2009	≥1 grade decrease by NCI CTCAE version 4.0, May, 2009	≥2˚C decrease in fever or return to normal body temperature (37˚C) or improvement in night sweats, as assessed by licensed site investigator	≥5% increase not due to increase in new or existing edema	≥25% decrease bi-dimensionally in size of largest lymph node, as measured using Cheson criteria (**Appendix 15.2**)
Worsening		≥2 grade increase by NCI CTCAE version 4.0, May, 2009	≥2 grade increase by NCI CTCAE version 4.0, May, 2009	≥2˚C increase in fever or worsening in night sweats, as assessed by licensed site investigator	≥5% decrease (weight loss not due to decreased edema)	≥25% increase bi-dimensionally in size of largest lymph node, as measured using Cheson criteria (**Appendix 15.2**)

The below table provides the CHAP score, which will be used to assess the efficacy of the study intervention. All comparisons are against those obtained during the Baseline Visit:

Score:	**0**	**1**	**2**	**3**	**4**
CRP (mg/dL)	<1	≥1, <5	≥5, <10	≥10, <20	≥20
Hemoglobin (g/dL)	≥12	<12, ≥10	<10, ≥8	<8	Transfusion dependent
Albumin (g/dL)	≥3	<3, ≥2.5	<2.5, ≥2	<2, ≥1.5	<1.5
Performance Score (ECOG)	0	1	2	3	4

The below table provides the modified Cheson response criteria, which will be used to assess the efficacy of the study intervention. Unless otherwise specified, all comparisons are against those obtained during the Baseline Visit:

Complete Response (CR)	All index lesion(s) must have regressed to normal size (≤1.0 cm in their greatest transverse diameter. No new sites of lymphadenopathy >1.5 cm in longest dimension.
Partial Response (PR)	≥50% decrease in sum of the products of the greatest diameters (SPD) of index lesion(s), and no new sites of lymphadenopathy >1.5 cm in longest dimension.
Stable Disease (SD)	Failure to achieve a CR or PR (see above) without evidence of progressive disease (see below).
Progressive Disease (PD)	≥50% increase from nadir in the SPD of any index lesion, or appearance of any new sites of lymphadenopathy that measure >1.5 cm in longest dimension during or at the end of therapy.

MCD-related Overall Symptom Score: In addition to the defined CBR, 34 measures of disease activity will be prospectively evaluated and graded (as per CTCAE version 4.0, May, 2009), which will be used to assess the efficacy of the study intervention. All comparisons are against those obtained during the Baseline Visit:

General MCD-related

- Fatigue
- Malaise
- Hyperhidrosis
- Night sweats
- Fever
- Weight loss
- Anorexia
- Tumor pain
- Dyspnea
- Pruritis

Autoimmune phenomena

- Autoimmune disorder
- Immune system disorder

Fluid retention

- Generalized edema
- Edema face
- Edema limbs
- Edema trunk
- Genital edema
- Localized edema
- Neck edema
- Periorbital edema
- Capillary leak syndrome

- Ascites
- Pleural effusion
- Pericardial effusion

Neuropathy

- Peripheral motor neuropathy
- Peripheral sensory neuropathy
- Nervous system disorder, other

Skin disorders

- Rash acneiform
- Rash maculo-papular
- Papulopustular rash
- Purpura
- Skin hyperpigmentation
- Skin induration
- Skin disorder, other

7.7 Research Samples

Research samples will be collected and shipped to the University of Pennsylvania for processing and storage, or processed at the local site and shipped to the University of Pennsylvania or the Children's Hospital of Philadelphia for storage. Research samples will be coded to maintain patient confidentiality and may be stored indefinitely for use in research, including in pursuit of the exploratory endpoints described in this protocol. Subjects will be asked to provide their consent to allow any biological samples collected as part of this study or collected during their enrollment period in this study, but for purposes outside of the study, to be used for laboratory research purposes, including genetic sequencing studies. Subjects will also be given the option to consent to allow any biological samples from their past medical procedures to be used for laboratory research purposes, including genetic sequencing studies.

7.8 Genetic Testing

As part of the exploratory objective, genetic testing may be performed on subject's biological samples. Such genetic testing would be strictly exploratory in nature. Samples will be used to study the genetic information that may be involved in Castleman disease, and efficacy or failure of sirolimus. Clinically significant results of genetic testing may be disclosed to the subjects at the discretion of the treating physician.

7.9 Safety Evaluations

Enrolled subjects will be carefully monitored throughout the study by study staff, with particular attention to known sirolimus AEs (See **Section 12.1 Risks** for details). Safety will also be assessed by changes in laboratory parameters, vital signs, and physical examinations. If any safety concerns arise, the licensed site investigators will promptly inform the Lead Principal Investigator and the Data Safety Monitoring Board (DSMB). The DSMB members will meet at pre-established times and as safety concerns arise to review the safety and clinical assessment data.

7.10 Concomitant Therapies

Corticosteroids (stable dose) and best supportive care are allowed throughout the course of the Intervention Phase. No additional systemic therapies directed at treating iMCD are permitted during the Intervention Phase.

8 Statistical Plan

This trial is a Phase II study of the efficacy and mechanism of action of sirolimus in anti-IL-6 blockade refractory iMCD. The trial is designed to rule out a therapy that has little or no biological activity in the examined patient cohort, a treatment-refractory population. The minimal acceptable level of activity is ≥ 20%. A common dose and dosing schedule will be employed for all subjects. An interim analysis will be conducted after 14 subjects have completed the Visit 9 intermediate assessment. If no responses are observed in these first 14 subjects, then the probability(response probability≥20%) < 0.05, and the trial should be terminated. If at least one response is observed, additional subjects may be enrolled to increase the degree of precision of the standard error, to a total of 24 evaluable subjects.

The statistical considerations for this study were developed to answer the two overall research questions:

- Does sirolimus have activity in the treatment population? The only placebo controlled clinical trial in iMCD (NCT01024036) reported a 0% response in placebo. We have access to the historic data and plan to apply the study's endpoints, as applicable, as a benchmark for this trial, recognizing that some of the enrollment criteria were more stringent than those of this trial, and standard of care for iMCD may have changed since the first patient was enrolled into NCT01024036 (March 2010). We propose a maximum of up to 24 total evaluable patients with an interim analysis after 14 patients have completed the Visit 9 intermediate assessment. If no patients demonstrate a positive CBR response at the Visit 9 assessment, then it will be determined that sirolimus is unlikely to have activity in the study population. If one or more patients demonstrate a positive CBR, then additional patients may be enrolled in order to better estimate the response rate. Upon study completion, primary and secondary endpoints will be analyzed to determine if sirolimus has activity in the treatment population. Assessments will be based on the binary (yes/no) CBR criteria and continuation on drug, the continuous variable (0-16) CHAP disease activity scoring system, and the modified Cheson response criteria. Given the aggressive nature of the disease, historic data and preliminary nature of this trial, a placebo arm is not included in this study. The null hypothesis is that the intervention will have 0% activity.

- What is the mechanism of action by which sirolimus is efficacious in alleviating lymph node enlargement and clinical symptoms in iMCD? Cellular and molecular research assays will be performed to analyze changes in cell populations and signaling molecules between baseline and time points following sirolimus administration. Comparison will be made with paired t-tests and post-hoc corrections for multiple comparisons, as appropriate.

8.1 Primary Endpoint

The primary endpoint for this study will be the proportion of patients achieving a positive response based on the CBR criteria. The CBR criteria consist of six parameters. The CBR and its binary

analysis is described in **Section 7.6 Efficacy Evaluations**. The parameters will be measured as part of standard of care using clinically established scales (e.g. CTCAE version 4.0, May, 2009 grading and Cheson criteria). The primary assessment will occur at 12 ± 1 month. Comparisons will be made to values obtained during the patient's Baseline Visit.

8.2 Secondary Endpoints

The secondary endpoints for this study are (1) the proportion of patients achieving a positive response based on the CBR criteria during intermediate assessment time points, (2) the proportion of patients that remain on study drug for the duration of the study, (3) the proportion of patients that indicate that they are currently receiving sirolimus at the end of the Follow Up Phase, (4) changes in the patient's disease activity, as measured by the CHAP score during intermediate and final assessment time points, (5) changes in the patient's disease activity, as measured by MCD-related Overall Symptom Score as measured by 34 outcome measures during intermediate and final assessment time points and (6) lymph node response, as measured by the Cheson criteria during intermediate and final assessment time points. Comparisons will be made against values obtained during the Baseline Visit.

8.3 Exploratory Endpoints

The exploratory endpoints of this study will be results of correlative studies, in the research laboratory, to investigate the mechanism of action of sirolimus in iMCD and clinical and laboratory biomarkers for treatment response. Additional correlative findings may also be assessed. Spleen size will be measured by radiological imaging (PET/CT or CT).

8.4 Sample Size and Power Determination

Up to 24 subjects may be evaluated under this study, with an interim analysis to be conducted after 14 subjects have completed the Visit 9 intermediate assessment. Additional subjects may be enrolled to increase the degree of precision of the standard error, to a total of 24 evaluable subjects. We anticipate enrolling up to 40 subjects to reach our goal of 24 evaluable subjects.

The difference between enrollment and evaluable accounts for potential subjects that may be deemed ineligible upon screening, or who drop out of the study but are replaced.

Our expected response rate is ≥20% versus the null hypothesis of 0% response. The placebo response rate in the only placebo controlled, double blinded clinical trial in iMCD was 0%. Given these parameters, power analysis with type 1 error $\alpha = 0.05$ and power of $1-\beta = 0.8$ provides a samples size of 14 to determine if sirolimus has activity; the drug will be considered not likely to have a ≥20% activity if no responses are seen in the interim analysis.

Sample size was chosen based on (1) convenience and the logistical constraints of recruiting a rare disease population and (2) our intention to probe for a signal of response and understand sirolimus' mechanism of action.

8.5 Statistical Methods

Descriptive analyses will be performed and presented as tables and plots.

Continuous variables: (e.g., age) will be summarized using the following descriptive summary statistics: the number of subjects (n), mean, error, median, minimum value (min), and maximum value (max). For each continuous variable, the corresponding mean, median, minimum and maximum will be presented with error, as determined appropriate.

Categorical variables: (e.g., presence of an AE, gender) will be summarized using counts and percentages.

8.5.1 Efficacy Analysis

CBR will be used as the primary endpoint for assessment of treatment response. Response to drug will be summarized as CBR and its 95% confidence interval and tested using Chi-square (versus historic placebo control data). Difference in CBR in two different study sites will be summarized and compared using the Chi-square tests. Significance will be established at $\alpha=0.05$.

The baseline covariates will be tested for association with CBR using logistic regression or two-sample t-test and the unadjusted p-values will be reported. When biomarkers are evaluated for association with CBR, we will perform logistic regression analysis for each marker adjusting for possible baseline covariates. The false discovery rate procedure will be used to adjust for multiple comparisons. As an exploratory analysis, if the CBR is not too low, we will also apply the random forest method to evaluate whether such biomarkers cab be predictive to CBR.

8.5.2 Interim Analysis

An interim analysis will be completed after 14 subjects have completed the intermediate assessment for CBR at Visit 9. At this time, the safety and preliminary efficacy of the study dose will be assessed and if, deemed safe and appropriate, enrollment may continue as needed to achieve up to 24 evaluable subjects to increase the degree of precision of the standard error.

8.5.3 Safety Analysis

All subjects entered into the study will have detailed information collected on AEs for the overall study safety analysis. Treatment emergent AEs are defined as AEs with a start date or that increase in severity on or after the first study drug dose through Visit 15 of the Follow Up Phase of the study.

8.6 Subject Population(s) for Analysis

Any subject enrolled into the study that receives at least one dose of investigational product will be included for all analyses for each Visit they complete.

9 Safety and Adverse Events

9.1 Definitions

9.1.1 Adverse Event

An adverse event (AE) is any symptom, sign, illness or experience that develops or worsens in severity during the course of the study. Intercurrent illnesses or injuries should be regarded as AEs. Abnormal results of diagnostic procedures are considered to be AEs if the abnormality:

- results in study withdrawal
- is associated with a serious AE
- is associated with clinical signs or symptoms
- leads to additional treatment or to further diagnostic tests
- is considered by the licensed site investigator to be of clinical significance

9.1.2 Serious Adverse Event

AEs are classified as serious or non-serious. A serious adverse event is any AE that is:

- fatal
- life-threatening
- requires or prolongs hospital stay
- results in persistent or significant disability or incapacity
- a congenital anomaly or birth defect
- an important medical event

Important medical events are those that may not be immediately life threatening, but are clearly of major clinical significance. They may jeopardize the subject, and may require intervention to prevent one of the other serious outcomes noted above. For example, drug overdose or abuse, a seizure that did not result in in-patient hospitalization, or intensive treatment of bronchospasm in an emergency department would typically be considered serious.

All AEs that do not meet any of the criteria for serious should be regarded as non-serious AEs.

9.2 Recording of Adverse Events

At each contact with the subject, study staff will seek information on AEs by specific questioning and, as appropriate, by examination. Subjects will be monitored for all previously reported AEs associated with sirolimus treatment and possible new AEs, as well as Grade 2 or higher allergic reaction/hypersensitivity attributed to sirolimus, as judged by the site investigator. AEs will be defined by the NCI CTCAE version 5.0, November 2017. All AEs will be recorded in the source document, and also in the appropriate AE module of the eCRF. All clearly related signs, symptoms, and abnormal diagnostic procedures' results should be recorded in the source document, though should be grouped under one diagnosis.

AEs occurring during the study period (from the time of enrollment through Visit 15) will be recorded. The clinical course of each event should be followed by the licensed site investigator until resolution, stabilization, or until it has been determined that the study treatment or participation is not the cause.

9.3 Relationship of AE to Study

The relationship of each AE to sirolimus should be characterized by the licensed site investigator and recorded in the appropriate AE module of the eCRF. The licensed site investigator will classify the relationship as either definitely related, probably related, possibly related, unlikely or unrelated.

9.4 Reporting of Adverse Events and Unanticipated Problems

Licensed site investigators will conform to the AE reporting timelines, formats and requirements of the various entities to which they are responsible.

If the report is supplied as a narrative, the minimum necessary information to be provided at the time of the initial report includes:

- Study identifier
- Study center
- Subject number
- A description of the event
- Date of onset
- Current status
- Whether study intervention was discontinued
- The reason why the event is classified as serious
- Licensed site investigator's assessment of the association between the event and study intervention

Additionally all other events (unanticipated problems, adverse reactions, unanticipated adverse effects and subject complaints) will be recorded and reported with respect to institutional and federal policies as described in the Penn Manual and below.

9.4.1 Follow-up report

If an SAE has not resolved at the time of the initial report and new information arises that changes the licensed site investigator's assessment of the event, a follow-up report including all relevant new or reassessed information (e.g., concomitant medication, medical history) should be completed. The licensed site investigator is responsible for ensuring that all SAE are followed until either resolved or stable.

9.4.2 Investigator reporting: notifying the study sponsor

Licensed site investigators from all participating sites should report all unexpected and related AEs, regardless of whether they are serious or not, and all unanticipated problems to the Lead Principal Investigator.

Any study-related unanticipated problem posing risk to subjects or others, and any type of SAE, will be reported to the Lead Principal Investigator by telephone within 24 hours of the event. To report such events, a SAE form will be completed by the licensed site investigator and emailed to the Lead Principal Investigator within two business days. The licensed site investigator will keep a copy of this SAE form on file at the study site. Report SAEs by phone and email to:

David Fajgenbaum
215-614-0936
davidfa@pennmedicine.upenn.edu

Within the following 48 hours, the licensed site investigator will provide further information on the SAE or the unanticipated problem in the form of a written narrative. This should include a copy of the completed SAE form, and any other diagnostic information that will assist the understanding of the event. Significant new information on ongoing SAEs should be provided promptly to the Lead Principal Investigator.

9.4.3 Investigator Reporting: Notifying the Penn IRB

This section describes the requirements for safety reporting by investigators who are Penn faculty, affiliated with a Penn research site, or otherwise responsible for safety reporting to the Penn IRB. The University of Pennsylvania IRB (Penn IRB) requires expedited reporting of those events related to study participation that are unforeseen and indicate that subjects or others are at increased risk of harm. The Penn IRB will not acknowledge safety reports or bulk AE submissions that do not meet the criteria outlined below. The Penn IRB requires researchers to submit reports of the following problems within 10 working days from the time the investigator becomes aware of the event:

- Any AE (regardless of whether the event is serious or non-serious, on-site or off-site) that occurs any time during or after the research study, which in the opinion of the Lead Principal Investigator is:

  Unexpected (An event is "unexpected" when its specificity and severity are not accurately reflected in the protocol-related documents, such as the IRB-approved research protocol, any applicable investigator brochure, and the current IRB-approved informed consent document and other relevant sources of information, such as product labeling and package inserts.)

  AND

  Related to the research procedures (An event is "related to the research procedures" if in the opinion of the Lead Principal Investigator, the event was more likely than not to be caused by the research procedures.)

Reporting Process
Unanticipated problems posing risks to subjects or others as noted above will be reported to the Penn IRB using the form: "Unanticipated Problems Posing Risks to Subjects or Others Including Reportable Adverse Events" or as a written report of the event (including a description of the event with information regarding its fulfillment of the above criteria, follow-up/resolution and need for revision to consent form and/or other study documentation).

Copies of each report and documentation of IRB notification and receipt will be kept in the Principal Investigator's study file.

Other Reportable Events
The following events are also reportable to the Penn IRB:

- Any adverse experience that, even without detailed analysis, represents a serious unexpected AE that is rare in the absence of drug exposure (such as agranulocytosis, hepatic necrosis, Stevens-Johnson syndrome).
- Any AE that would cause the sponsor to modify the investigators brochure, protocol or informed consent form, or would prompt other action by the IRB to assure protection of human subjects.
- Information that indicates a change to the risks or potential benefits of the research, in terms of severity or frequency. For example:
  - An interim analysis indicates that subjects have a lower rate of response to treatment than initially expected.
  - Safety monitoring indicates that a particular side effect is more severe, or more frequent than initially expected.
  - A paper is published from another study that shows that an arm of your research study is of no therapeutic value.
- Change in FDA safety labeling or withdrawal from marketing of a drug, device, or biologic used in a research protocol.
- Breach of confidentiality
- Change to the protocol taken without prior IRB review to eliminate apparent immediate hazard to a research subject.
- Incarceration of a subject when the research was not previously approved under Subpart C and the investigator believes it is in the best interest of the subject to remain on the study.

- Complaint of a subject when the complaint indicates unexpected risks or the complaint cannot be resolved by the research team.
- Protocol violation (meaning an accidental or unintentional deviation from the IRB approved protocol) that in the opinion of the investigator placed one or more subjects at increased risk, or affects the rights or welfare of subjects.

9.4.4 Sponsor reporting: Notifying participating investigators

The Lead Principal Investigator will report AE or findings to all participating licensed site investigators in a timely manner.

9.4.5 Sponsor reporting: Notifying collaborating companies

The Lead Principal Investigator will report AEs to collaborating companies (e.g. Pfizer, Inc.), in accordance with contractual agreements.

9.5 Unblinding Procedures

N/A

9.6 Stopping Rules

N/A

9.7 Medical Monitoring

It is the responsibility of the licensed site investigator to oversee the safety of the study at his/her site. This safety monitoring will include careful assessment and appropriate reporting of AEs as noted above. Medical monitoring will include a regular assessment of the number and type of SAEs. Safety will be closely monitored by the licensed site investigators on an ongoing basis for all subjects. A DSMB is planned to monitor the safety of this study.

9.7.1 Data and Safety Monitoring Plan

The investigator at each site will be responsible for monitoring the data quality and the ongoing safety of subjects at their respective sites. In addition, eCRF data quality will be periodically confirmed via remote audit and review of source documents.

A DSMB will provide additional guidance and will review data quality and subject safety on an ongoing basis. The DSMB will be comprised of representatives who are independent of the investigators. A DSMB will be formed with a charter containing this information.

9.7.2 Data Safety Monitoring Board

An independent DSMB will be established to assure the safety of subjects in this trial. The DSMB will review the study for safety and overall study conduct. At the end of the study, they may adjudicate final outcome based on the clinical information. The membership of the DSMB as well as the responsibilities and procedures used to carry out these responsibilities are described separately in the DSMB charter.

10 Study Administration, Data Handling and Record Keeping

10.1 Confidentiality

Information about study subjects will be kept confidential and managed according to the requirements of the Health Insurance Portability and Accountability Act of 1996 (HIPAA). Those regulations require a signed subject authorization informing the subject of the following:

- What protected health information (PHI) will be collected from subjects in this study
- Who will have access to that information and why
- Who will use or disclose that information
- The rights of a research subject to revoke their authorization for use of their PHI.

In the event that a subject revokes authorization to collect or use PHI, the investigator, by regulation, retains the ability to use all information collected prior to the revocation of subject authorization. For subjects that have revoked authorization to collect or use PHI, attempts should be made to obtain permission to collect at least vital status (i.e. that the subject is alive) at the end of their scheduled study period.

To help protect subject confidentiality, at enrollment the licensed site investigator will assign a GUID to each study subject. The following information is needed to generate an encrypted GUID:

- Complete legal given first, middle and last names of subject at birth
- Date of birth
- Name of city/municipality in which subject was born
- Country of birth
- Physical sex of subject at birth (optional)

At each site, source documents and a master electronic list, which links the coded identifiers for subjects enrolled at that site to personal information, will be kept completely separate from the eCRF (**Section 10.2, Data Collection and Management**) on a password protected network and/or under lock and key and limited to key research team members. Research team members will have access as long as the project is still operating and they remain research qualified.

The GUID enables data from a coded subject to be integrated, tracked over time, and linked across research projects, databases and biobanks. This will allow for de-identified data to be associated between all studies that use the GUID.

10.2 Data Collection and Management

Study data, collected as part of the trial, as standard of care treatment during the trial period, and historic medical and treatment information will be entered into the eCRF. The eCRF is the primary data collection instrument for the study. All data requested on the eCRF must be recorded. All missing data must be explained. If a space on the eCRF is left blank because the procedure was not done or the question was not asked, write “N/D”. If the item is not applicable to the individual case, write “N/A”.

A shared database with common data definitions will be created using REDCap, and housed on a secure server at the University of Pennsylvania. REDCap is a secure, web-based application with the capacity for direct export to Excel and common statistical packages (SPSS, SAS, Strata,

R). REDCap has eCRFs, real-time data entry validation, audit trails, user authentication, data logging and encryption. It is HIPAA compliant with mechanisms in place to ensure confidentiality.

To ensure confidentiality of the data, subjects GUIDs will be used for identification in REDCap; personal identifiers (e.g. name, social security number, medical record number, etc.) will not be entered into the eCRF. Qualified study staff will have access to all study data collected in the eCRF.

Specific forms will be used for each component of the subject's progress. The forms and data dictionary will be available online for all individuals who perform data entry. Research personnel, trained on data definitions, will enter data via web-based data forms after abstraction from the primary medical record and source documents. Logical data checks will be used to assess data quality for mis-entry. Suspect data entries will be flagged for re-review and confirmation by the investigative team at each site. When data are complete and all suspect entries addressed for a time period, the database will be "locked" for analysis. Analysis will use only this final locked version. De-identified data will be stored indefinitely.

10.3 Records Retention

Study essential documents will be retained for at least 2 years after the last subject completes the End of Study Visit or Early Termination Visit, as the case may be.

10.4 Management of Information for Multi-Center Research

University of Pennsylvania IRB will be the single IRB of record for the University of Pennsylvania and UAMS. University of Pennsylvania is the lead site and will be responsible for ensuring all sites receive the most current version of the protocol, consent and study related documents. The lead site will be responsible for management of project timelines, management of data collected from each site, submitting progress reports, and reporting to the IRB as needed. UAMS IRB will be responsible for ensuring their local consent requirements are met without major changes to the IRB approved template. Protocol changes, consent updates and IRB continuing reviews will be initiated by the lead site in compliance with Penn's IRB policies. The Children's Hospital of Philadelphia will utilize the CHOP IRB.

Regular meetings will be held for study team members at all sites, and interim communication via phone or email will occur as needed to report urgent developments to licensed site investigators.

11 Study Monitoring, Auditing, and Inspecting

11.1 Study Monitoring Plan

The licensed site investigator at each site is responsible for assigning staff for monitoring. The Lead Principal Investigator will also ensure that the monitor or other compliance or quality assurance reviewer is given access to all the above noted study-related documents and study related facilities (e.g. pharmacy, diagnostic laboratory, etc.). Enrollment will be complete when all subjects are enrolled into the trial across sites. The following will be reviewed for all subjects after the first subject is enrolled at each site, and throughout the trial:

- Informed consent documentation
- Eligibility criteria
- Safety Monitoring (adverse event documentation and assessment)

- Regulatory documentation (IRB – amendments, continuing review and reportable events)

The Regulatory Documents will be maintained in the Regulatory Binder.

11.2 Auditing and Inspecting

The licensed site investigators will permit study-related monitoring, audits, and inspections by the IRB, government regulatory bodies, and University compliance and quality assurance groups of all study related documents (e.g. source documents, regulatory documents, data collection instruments, study data etc.). The licensed site investigators will ensure the capability for inspections of applicable study-related facilities (e.g. pharmacy, diagnostic laboratory, etc.).

Participation as a licensed site investigator in this study implies acceptance of potential inspection by government regulatory authorities and applicable University compliance and quality assurance offices.

12 Ethical Considerations

This study is to be conducted accordance with applicable US government regulations and international standards of Good Clinical Practice, and applicable institutional research policies and procedures.

This protocol and any amendments will be submitted to a properly constituted IRB, in agreement with local legal prescriptions, for formal approval of the study conduct. The decision of the IRB concerning the conduct of the study will be made in writing to the Lead Principal Investigator and a copy of this decision will be provided to the Lead Principal Investigator before commencement of this study.

12.1 Risks

The risk to patients of exposure to the treatment intervention, sirolimus, as related to AEs is relatively low. Sirolimus is an FDA-approved therapy with an established safety profile. Although uncommon, AEs are possible and will be monitored by study staff and licensed site investigators. The most common (≥20%) adverse reactions reported for sirolimus 2 mg/day at a higher incidence than reported for placebo include:

- peripheral edema
- hyperlipidemia, hypercholesterolemia, hypertriglyceridemia
- increased creatinine
- constipation
- hypertension
- abdominal pain / diarrhea
- headache
- arthralgia

Other less common serious complications include:

- angioedema
- leucopenia
- thrombocytosis / thrombocytopenia
- fluid accumulation

- impaired wound healing
- proteinuria
- interstitial lung disease/non-infectious pneumonitis
- calcineurin inhibitor-induced hemolytic uremic syndrome/thrombotic thrombocytopenic purpura/thrombotic microangiopathy

The below table summarizes the known adverse events (AEs) categorized based on timing and frequency:

Known Adverse Events (AEs)			
	Common 21-100 out of every 100	**Occasional** 5-20 out of every 100	**Rare** <5 out of every 100
Immediate: 1-2 days of receiving drug	Nausea, vomiting, oral ulcers	Rash	Allergic reaction (may be life threatening)
Prompt: 2-3 weeks on drug	Increased cholesterol, increased blood pressure, decreased kidney function, diarrhea	Infections, swelling, weight gain, muscle/joint pain, tremor, acne, rash, abdominal pain, mucositis, headache	Decreased red and white blood cells generated in bone marrow, decreased liver function
Delayed: Any time later during therapy, excluding the above conditions	Acne		Malignancy resulting from this treatment, destruction of red blood cells and decreased kidney function, interstitial lung disease, hemolytic uremic syndrome/thrombotic thrombocytopenic purpura*
Late: Any time after completion of treatment			Interstitial lung disease
Unknown Frequency and Timing:	Sirolimus was embryo/feto toxic in rats at dosages of 0.1 mg/kg and above (approximately 0.2 to 0.5 is the clinical doses adjusted for body surface area). Embryo/feto toxicity was manifested as mortality and reduced fetal weights (with associated delays in skeletal ossification). Sirolimus is excreted in trace amounts in milk of lactating rats. It is not known whether sirolimus is excreted in human milk.		
*Risk increased when sirolimus taken with other medications, including cyclosporine and tacrolimus. Increased risk of calcineurin inhibitor-induced hemolytic uremic syndrome / thrombotic thrombocytopenic purpura / thrombotic microangiopathy			

To monitor for these risks, blood pressure will be monitored from standard of care physical exams. Additionally, fasting serum lipids will be monitored from standard of care lipid panels. Treating physicians may consider use of a cholesterol-lowering agent, such as an HMG-CoA reductase inhibitor, in patients with elevated cholesterol, especially those who have responded to therapy and may be on the drug for 6 months or longer. HMG-CoA reductase inhibitors that are also

CYP3A4 inhibitors (**https://drug-interactions.medicine.iu.edu/main-table.aspx**) should be avoided. Sirolimus can sometimes impair kidney, liver and bone marrow function. Kidney, liver and bone marrow function will be monitored from standard of care metabolic, liver function and complete blood counts panels.

In general, immunosuppressive therapy may result in an increased risk of development of lymphoma and other malignancies. Such risks will be monitored for by review of subjects' complete blood counts (CBC) and imaging for evidence of malignancy. Additionally, patients receiving immunosuppressants, including sirolimus, are at increased risk for opportunistic infections, including polyoma virus infections. Polyoma virus infections in immunosuppressed patients may have serious, and sometimes fatal, outcomes. These include polyoma virus-associated nephropathy (PVAN), mostly due to BK virus infection, and JC virus-associated progressive multifocal leukoencephalopathy (PML) which have been observed in patients receiving sirolimus.

There is a risk of breach of confidentiality in that someone could trace the information in the research database back to an individual. The risk that someone will identify an individual is very small as data will be stored on a secure database on password protected computers and no personal health information will be entered into the database; subjects will be tracked using a GUID assigned at enrollment. Only designated study staff at the subject's site of enrollment will have access to the master electronic list linking personal identifiers to the subject's GUID.

12.2 Benefits

This study will enroll iMCD patients who have failed the only FDA-approved therapy for treatment of the disease. Other therapeutic approaches (e.g. corticosteroids, rituximab, and cytotoxic chemotherapy) have been used off-label, but have significant toxicities and their efficacies have not been systematically studied. Therefore, if efficacious, sirolimus therapy would provide the direct benefit of improved health, with limited toxicity, to these individuals who have few other therapeutic options. More broadly, if efficacy is found, this study could have the indirect benefit of identifying sirolimus as a standard second line therapy for treatment of iMCD.

12.3 Risk Benefit Assessment

The risks of participating in this study are outweighed by the potential benefits. Sirolimus related AEs are uncommon and generally tolerable. In contrast, iMCD is a life-threatening lymphoproliferative disorder with only a single FDA-approved therapy. For patients who do not respond, there are few therapeutic options.

12.4 Informed Consent Process / HIPAA Authorization

All subjects for this study will be provided a consent form describing this study and providing sufficient information for subjects to make an informed decision about their participation in this study. This consent form is submitted with this protocol for review and approval by the IRB for the study.

The consent process will take place during the Screening Visit. The formal consent of a subject must be obtained before that subject undergoes any study specific procedure. The licensed site investigator or their designated staff will obtain consent for each study subject. Potential subjects will be provided the opportunity to discuss any questions they have with the licensed site investigator, and will be permitted to provide consent at the time of the consent process. No subjects will be unduly influenced, encouraged, or coerced into participating in this study. The

consent form must be signed by the subject or legally acceptable surrogate, and the licensed site investigator's designated research professional obtaining the consent. Completed consent forms will be kept in a continuously locked file cabinet at the study site.

13 Study Finances

13.1 Funding Source

This work is supported by a grant from the National Heart Lung and Blood Institute of the National Institutes of Health (R01HL141408).

13.2 Conflict of Interest

All University of Pennsylvania Investigators will follow the University of Pennsylvania Policy on Conflicts of Interest Related to Research.

13.3 Subject Stipends or Payments

Due to the rarity of iMCD, subjects may need to travel to their assigned site for on-site visits. If a subject is found eligible and enrolls in this study, he or she will be provided $50 to be used for meals and travel expenses for each visit that must occur at the study center (Visits 1, 4, 6, 9, 12, and 15). Subjects traveling over 100 miles to the study site may be provided with hotel and travel arrangements (flight or train tickets), if needed, for each visit that must occur at the study center (Visits 1, 4, 6, 9, 12, and 15). Payment or reimbursement for travel expenses will be administered using a Greenphire ClinCard, by check, or similar method. Travel arrangements may also be made through third-party groups who provide transportation to patients, such as Angel Care East. Subjects may be reimbursed for research-related costs such as phlebotomy fees. Subjects will not otherwise be paid for their participation in this study. The investigational product will be provided at no cost to the subject.

14 Publication Plan

A Publication Committee will include the Principal Investigators, and will be open to site investigators and collaborators. The purpose of the Publication Committee is to effectively manage and oversee the primary, secondary and ancillary publications generated from the study while complying with all applicable guidelines and policies. This includes delivering high-quality publications that address the primary evidence needs identified and prioritized by any collaborators, site investigators, and the Publication Committee.

15 Appendix

15.1 Diagnostic Criteria for iMCD

I. Major Criteria (need both):
1. Histopathologic lymph node features consistent with the iMCD spectrum (Figure 5). Features along the iMCD spectrum include (need grade 2-3 for either regressive GCs or plasmacytosis at minimum):
Regressed/atrophic/atretic germinal centers, often with expanded mantle zones composed of concentric rings of lymphocytes in an "onion skinning" appearance
FDC prominence
Vascularity, often with prominent endothelium in the interfollicluar space and vessels penetrating into the GCs with a "lollipop" appearance
Sheetlike, polytypic plasmacytosis in the interfollicular space
Hyperplastic GCs
2. Enlarged lymph nodes (≥1 cm in short-axis diameter) in ≥2 lymph node stations
II. Minor Criteria (need at least 2 of 11 criteria with at least 1 laboratory criterion)
Laboratory*
1. Elevated CRP (>10 mg/L) or ESR (>15 mm/h)†
2. Anemia (hemoglobin <12.5 g/dL for males, hemoglobin <11.5 g/dL for females)
3. Thrombocytopenia (platelet count <150 k/μL) or thrombocytosis (platelet count >400 k/μL)
4. Hypoalbuminemia (albumin <3.5 g/dL)
5. Renal dysfunction (eGFR <60 mL/min/1.73m^2) or proteinuria (total protein 150 mg/24 h or 10 mg/100 ml)
6. Polyclonal hypergammaglobulinemia (total γ globulin or immunoglobulin G >1700 mg/dL)
Clinical
1. Constitutional symptoms: night sweats, fever (>38°C), weight loss, or fatigue (≥2 CTCAE lymphoma score for B-symptoms)
2. Large spleen and/or liver
3. Fluid accumulation: edema, anasarca, ascites, or pleural effusion
4. Eruptive cherry hemangiomatosis or violaceous papules
5. Lymphocytic interstitial pneumonitis
III. Exclusion Criteria (must rule out each of these diseases that can mimic iMCD)
Infection-related disorders
1. HHV-8 (infection can be documented by blood PCR, diagnosis of HHV-8–associated MCD requires positive LANA-1 staining by IHC, which excludes iMCD)
2. Clinical EBV-lymphoproliferative disorders such as infectious mononucleosis or chronic active EBV (detectable EBV viral load not necessarily exclusionary)
3. Inflammation and adenopathy caused by other uncontrolled infections (eg, acute or uncontrolled CMV, toxoplasmosis, HIV, active tuberculosis)
Autoimmune/autoinflammatory diseases (requires full clinical criteria, detection of autoimmune antibodies alone is not exclusionary)
1. Systemic lupus erythematosus
2. Rheumatoid arthritis
3. Adult-onset Still disease
4. Juvenile idiopathic arthritis
5. Autoimmune lymphoproliferative syndrome
Malignant/lymphoproliferative disorders (these disorders must be diagnosed before or at the same time as iMCD to be exclusionary):
1. Lymphoma (Hodgkin and non-Hodgkin)
2. Multiple myeloma
3. Primary lymph node plasmacytoma
4. FDC sarcoma
5. POEMS syndrome‡
Select additional features supportive of, but not required for diagnosis
Elevated IL-6, sIL-2R, VEGF, IgA, IgE, LDH, and/or B2M
Reticulin fibrosis of bone marrow (particularly in patients with TAFRO syndrome)
Diagnosis of disorders that have been associated with iMCD: paraneoplastic pemphigus, bronchiolitis obliterans organizing pneumonia, autoimmune cytopenias, polyneuropathy (without diagnosing POEMS‡), glomerular nephropathy, inflammatory myofibroblastic tumor

B2M, β-2-microglobulin; CMV, cytomegalovirus; CTCAE, common terminology for adverse events; eGFR, estimated glomerular filtration rate; GC, germinal center; IHC, Immunohistochemistry; LANA-1, latency-associated nuclear antigen; LDH, lactate dehydrogenase.

*We have provided laboratory cutoff thresholds as guidance, but we recognize that some laboratories have slightly different ranges. We suggest that you use the upper and lower ranges from your particular laboratory to determine if a patient meets a particular laboratory Minor Criterion.

†Evaluation of CRP is mandatory and tracking CRP levels is highly recommended, but ESR will be accepted if CRP is not available.

‡POEMS is considered to be a disease "associated" with CD. Because the monoclonal plasma cells are believed to drive the cytokine storm, we do not consider it iMCD, but rather "POEMS-associated MCD."

15.2 Modified Cheson Criteria

Lymph node selection: Up to six dominant lymph nodes ("index lesions") should be selected according to the following features:

- they should be clearly measurable in at least two perpendicular dimensions, ≥1.0 cm in the long axis, and ≥1.0 cm in the longest perpendicular transverse dimension to the long axis
- they should be from as disparate regions of the body as possible and chosen such that they are representative of the subject's disease
- they should include mediastinal and retroperitoneal areas of disease whenever these sites are involved

Lymph node measurement: The long axis and the longest perpendicular transverse dimension to the long axis of each lymph node should be measured (together, the greatest diameters).

Calculating lymph node response:

CBR response criteria: The largest of the index lesions selected at baseline should be analyzed to determine CBR response.

Positive Response: ≥25% decrease in the product of the greatest diameters, as compared to the same lymph node at baseline.

Negative Response: ≥25% increase in the product of the greatest diameters, as compared to the same lymph node at baseline.

Example: If at baseline, the long axis = 3.0 cm and the longest perpendicular transverse dimension = 2.0 cm, then the baseline value calculated for this node would be 6.0. To achieve a positive response, the node would have the shrink so that that product of the two measurements is ≤ 4.5. (e.g. 2 cm x 2.25 cm).

Cheson response criteria: all index lesions should be considered. When determining response, nodal measurements should be calculated independently and as the sum of the product of the greatest diameters, as described in (**Section 7.6 Efficacy Evaluations**).

If a single lymph node mass breaks apart into multiple discrete lymph nodes during the course of therapy, that group of nodes should continue to be considered as a single lesion on the eCRF. The measurement of the group of nodes should be recorded as the sum of the products of the greatest diameters of all of the nodes in the group. Under the "Site of Lesion" section of the Tumor Assessment CRFs please use the word "multiple" to clearly note that the measurement reported is a sum of several lesions that have broken apart. If several masses are separate at screening and in subsequent measurements appear to coalesce, this is usually an indication of PD or a technical issue with the CT scans. Please assess these lesions accordingly.

15.3 Study calendar

Study Phase	Screening	Baseline Phase	Intervention Phase														Early Termination Visit	Follow-up Phase
Study Visit Number	Screening	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15		16
Study Days**	-28 to 0**	-7 to 0**	5 ± 2 days	14 ± 5 days	1 month ± 1 week	2 months ± 2 weeks	3 months ± 2 weeks	4 months ± 2 weeks	5 months ± 2 weeks	6 months ± 2 weeks	7 months ± 2 weeks	8 months ± 2 weeks	9 months± 2 weeks	10 months ± 2 weeks	11 months ± 2 weeks	12 months ± 1 month		12 ± 2 weeks following the End of Study Visit
Diagnostic Laboratory Tests*	x																	
Physical Exam*		x			x***		x***			x***			x***			x***	x***	
Vital Signs*	x	x			x***		x***			x***			x***			x***	x***	
Radiological Imaging*	x						x			x			x			x		
Lipid Panel	x	x			x		x			x			x			x		
Review of concomitant medications*	x	x																
Confirmation subject meets enrollment criteria	x	x																
Informed Consent	x																	
Pregnancy Test (urine)	x				x		x			x			x			x	x	
GUID assigned		x																
Research blood draw		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	
Fecal sample collection (optional)		x		x						x								
Recording of Demographic information into eCRF	x																	
Recording of medical history data, medications, possible AEs and Laboratory Test, as available, from medical records into eCRF		x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	
Completion of Patient Evaluation Form and confirmation subject is eligible by Lead PI	x	x																
Dispensing of sirolimus loading dose**		x																
Dispensing of sirolimus daily dose		x			x		x			x			x					
Assess possible AEs, study drug compliance, and medication changes			x	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Sirolimus Trough			x	x	x		x			x			x			x	x	
Standard of Care Laboratory Test*					x		x			x			x			x	x	
Return of any non-consumed sirolimus					x		x			x			x			x	x	
Sirolimus pill count***					x		x			x			x			x	x	
Intermediate assessment of primary objective							x			x				x				
Final assessment of primary objective																x		
Follow Up telephone questionnaire																		x

*Standard of Care for iMCD
**Indicated times are following loading dose

15.4 Laboratory Tests

Standard Diagnostic Laboratory Tests

1. Complete blood count with differential (hemoglobin, red blood cell count, hematocrit, white blood cell count, platelet count, ANC, absolute lymphocyte count, absolute monocyte count, absolute basophil count, absolute eosinophil count)
2. Albumin
3. CRP
4. Basic metabolic panel (sodium, potassium, chloride, bicarbonate, glucose, blood urea nitrogen, creatinine/eGFR)
5. Erythrocyte sedimentation rate
6. sIL2R
7. VEGF
8. Quantitative antibodies (IgG, IgA, IgM)
9. Liver function tests (AST, ALT, gamma-glutamyl transpeptidase, alkaline phosphatase)
10. Urine protein test
11. Bilirubin, total
12. Fibrinogen
13. Ferritin
14. IgE
15. Immunofixation (serum)
16. Free light chains (Kappa)
17. Free light chains (Lambda)
18. Serum IgG4 (as clinically indicated)
19. Echo-cardiogram (as clinically indicated)
20. Genetic testing (as clinically indicated)

If required to confirm diagnosis as per diagnostic guidelines, the below (26-33) serological and autoantibody studies should be conducted as part of the Standard Diagnostic Laboratory Tests:

21. EBV PCR (quantitative)
22. HHV-8 PCR (quantitative)
23. HIV screen
24. Hepatitis B (HBsAg, HBsAb, HBcAb)
25. Hepatitis C (HCV Ab)
26. Cytomegalovirus PCR (quantitative)
27. Anti-nuclear antibody (anti-ANA) reflex panel (anti-ANA: if positive, anti-Sjogren's syndrome-related antigen (SS)A, anti-SSB, anti-double stranded DNA, anti-smith)
28. Rheumatoid factor

Standard of Care Laboratory Tests

- Standard Diagnostic Laboratory Tests 1-11 (above)

iMCD Additional Laboratory test (non-standard of care [i.e. optional])

- Absolute T cell count (CD3, CD4, CD8)
- Absolute CD19 cell count
- Absolute CD56 cell count

- Cytokines (IL-1β, IL-4, IL-5, IL-8, IL-10, IL-12, IL-13, tumor necrosis factor-α, granulocyte-macrophage colony-stimulating factor, interferon-γ)
- Prothrombin time
- Partial Thromboplastin Time
- D-Dimer

15.5 Example Dose Calculations

The table below provides examples of suggested dose corrections based on current dose and sirolimus trough levels. In all dose calculations, adjustments should ultimately be based on clinical judgment of the licensed site investigator after considering clinical toxicity, serum levels, concomitant drug use and the rate of rise or decline of the serum level.

Current dose (mg)	Trough level (ng/ml)	Target trough level (ng/ml)	Calculated new dose (mg)	New dose (rounded) (mg)
2	20	10	1.00	1
1	2	10	3.33	3
5	30	10	1.67	2
1	30	10	0.33	Withhold drug
2	15	10 - 15	N/A	No change (within target range)

In the table above, an example patient currently on a dose of 2mg per day may present with a sirolimus trough level of 20 ng/ml, above the target of 10 - 15 ng/ml. Using 10 ng/ml as a target, a new dose may be calculated using the equation New Dose = current dose × (target concentration/current concentration) In this example, new dose = 2 * (10/20) = 1. and the dose should be adjusted to 1mg per day.

16 References

1. Shilling D, Kobrin DM, Fajgenbaum DC. Pi3k/Akt/Mtor Pathway Activity Is Increased in Lymph Node Tissue from Idiopathic Multicentric Castleman Disease Patients with Tafro Syndrome. 2018;132:1121-.
2. Fajgenbaum DC, Shilling D, Partridge HL, et al. Prolonged Remission Achieved in a Relapsing Idiopathic Multicentric Castleman Disease Patient with a Novel, Targeted Treatment Approach. Blood 2017;130:3593-.
3. van Rhee F, Fayad L, Voorhees P, et al. Siltuximab, a novel anti-interleukin-6 monoclonal antibody, for Castleman's disease. J Clin Oncol 2010;28:3701-8.
4. Fajgenbaum DC, Uldrick TS, Bagg A, et al. International, evidence-based consensus diagnostic criteria for HHV-8–negative/idiopathic multicentric Castleman disease. Blood 2017;129:1646-57.
5. Munshi N, Mehra M, van de Velde H, Desai A, Potluri R, Vermeulen J. Use of a claims database to characterize and estimate the incidence rate for Castleman disease. Leuk Lymphoma 2015;56:1252-60.
6. Dispenzieri A, Armitage JO, Loe MJ, et al. The clinical spectrum of Castleman's disease. Am J Hematol 2012;87:997-1002.
7. Nishimoto N, Kanakura Y, Aozasa K, et al. Humanized anti-interleukin-6 receptor antibody treatment of multicentric Castleman disease. Blood 2005;106:2627-32.
8. van Rhee F, Wong RS, Munshi N, et al. Siltuximab for multicentric Castleman's disease: a randomised, double-blind, placebo-controlled trial. The Lancet Oncology 2014;15:966-74.
9. Iwaki N, Gion Y, Kondo E, et al. Elevated serum interferon γ-induced protein 10 kDa is associated with TAFRO syndrome. Scientific Reports 2017;7:42316.
10. van Rhee F, Voorhees P, Dispenzieri A, et al. International, Evidence-based Consensus Treatment Guidelines for Idiopathic Multicentric Castleman Disease. Blood 2018;*In Press*.
11. Liu AY, Nabel CS, Finkelman BS, et al. Idiopathic multicentric Castleman's disease: a systematic literature review. The Lancet Haematology 2016;3:e163-e75.
12. Shilling D, Stadanlick JE, Meng W, et al. Oligoclonal Expansion of Cd8+ T Cells during Idiopathic Multicentric Castleman Disease Flares Suggests an Antigen Driven Process. 2018;132:2411-.
13. Pierson SK, Stonestrom AJ, Shilling D, et al. Plasma proteomics identifies a 'chemokine storm' in idiopathic multicentric Castleman disease. Am J Hematol 2018.
14. Cheson B, Pfistner B, Juweid M. Revised response criteria for malignant lymphoma. J Clin Oncol 2007;25:57-86.
15. Cheson BD, Horning SJ, Coiffier B, et al. Report of an international workshop to standardize response criteria for non-Hodgkin's lymphomas. NCI Sponsored International Working Group. J Clin Oncol 1999;17:1244.
16. Fujimoto S, Koga T, Kawakami A, et al. Tentative diagnostic criteria and disease severity classification for Castleman disease: A report of the research group on Castleman disease in Japan. Modern rheumatology 2018;28:161-7.
17. Brown EJ, Albers MW, Shin TB, et al. A mammalian protein targeted by GI-arresting rapamycin-receptor complex. Nature 1994;369:756-8.
18. Chen Y, Chen H, Rhoad AE, et al. A putative sirolimus (rapamycin) effector protein. Biochem Biophys Res Commun 1994;203:1-7.
19. Sekulic A, Hudson CC, Homme JL, et al. A direct linkage between the phosphoinositide 3-kinase-AKT signaling pathway and the mammalian target of rapamycin in mitogen-stimulated and transformed cells. Cancer Res 2000;60:3504-13.
20. Schmelzle T, Hall MN. TOR, a central controller of cell growth. Cell 2000;103:253-62.

21. Dennis PB, Jaeschke A, Saitoh M, Fowler B, Kozma SC, Thomas G. Mammalian TOR: a homeostatic ATP sensor. Science 2001;294:1102-5.
22. Dilling MB, Germain GS, Dudkin L, et al. 4E-binding proteins, the suppressors of eukaryotic initiation factor 4E, are down-regulated in cells with acquired or intrinsic resistance to rapamycin. J Biol Chem 2002;277:13907-17.
23. Teachey DT, Axsom KM, Brown VI. Murine Ultrasonography Confirms That Rapamycin is Effective Against Autoimmune Lymphoproliferative Syndrome (ALPS). American Society Hematology Annual Meeting 2005.
24. Schreiber SL. Chemistry and biology of immunophilins and their immunosupressive ligands. Science 1991;251:283-7.
25. Saunders RN, Metcalfe MS, Nicholson ML. Rapamycin in transplantation: a review of the evidence. Kidney Int 2001;59:3-16.
26. Calne RY, Collier DS, Lim S, et al. Rapamycin for immunosuppression in organ allografting. Lancet 1989;2:227.
27. Drosos AA. Newer immunosuppressive drugs: their potential role in rheumatoid arthritis therapy. Drugs 2002;62:891-907.
28. Bindl L, Torgerson T, Perroni L, et al. Successful use of the new immune-suppressor sirolimus in IPEX (immune dysregulation, polyendocrinopathy, enteropathy, X-linked syndrome). J Pediatr 2005;147:256-9.
29. Yango A, Morrissey P, Monaco A, Butera J, Gohh RY. Successful treatment of tacrolimus-associated thrombotic microangiopathy with sirolimus conversion and plasma exchange. Clin Nephrol 2002;58:77-8.
30. Jimenez-Rivera C, Avitzur Y, Fecteau AH, Jones N, Grant D, Ng VL. Sirolimus for pediatric liver transplant recipients with post-transplant lymphoproliferative disease and hepatoblastoma. Pediatr Transplant 2004;8:243-8.
31. Teachey DT, Brown VI, Hall J, et al. mTOR Inhibitors Induce Apoptosis and Inhibit Growth of Primary Adult Human ALL in Xenograft and Tissue Culture Models. American Society Hematology Annual Meeting 2004.
32. Brown VI, Fang J, Alcorn K, et al. Rapamycin is active against B-precursor leukemia in vitro and in vivo, an effect that is modulated by IL-7-mediated signaling. Proc Natl Acad Sci U S A 2003;100:15113-8.
33. Eng CP, Sehgal SN, Vezina C. Activity of rapamycin (AY-22,989) against transplanted tumors. J Antibiot 1984;37:2131-1237.
34. Douros J, Suffness M. New antitumor substances of natural origin. Cancer Treat Rev 1981;8:63-87.
35. Ettenger RB, Grimm EM. Safety and efficacy of TOR inhibitors in pediatric renal transplant recipients. Am J Kidney Dis 2001;38:S22-8.
36. Pharmaceuticals W-A. Pakage insert for rapamune (sirolimus).
37. Vasquez EM. Sirolimus: a new agent for prevention of renal allograft rejection. Am J Health System Pharm 2000;57:437-51.
38. Scott JR, Courter JD, Saldaña SN, et al. Population pharmacokinetics of sirolimus in pediatric patients with neurofibromatosis type 1 [published correction appears in Ther Drug Monit. 2016 Jan 21;38(2):284]. Ther Drug Monit. 2013;35(3):332-337.
39. Lai ZW, Kelly R, Winans T, et al. Sirolimus in patients with clinically active systemic lupus erythematosus resistant to, or intolerant of, conventional medications: a single-arm, open-label, phase 1/2 trial. Lancet 2018;391:1186-96.