Clinical Study Protocol

Title:	**A Phase 2 Multicenter, investigator initiated study of Oral Ruxolitinib Phosphate for the Treatment of Relapsed or Refractory Diffuse Large B-cell and Peripheral T-cell Non-Hodgkin Lymphoma**
IND Number:	**112445**
UNMC IRB Number:	**283-11**
NCT#	**01431209**
Product:	**Ruxolitinib Phosphate (also known as INCB018424 and, INC424)**
Phase of Study:	**Phase 2**
Sponsor-Investigator:	**Julie Vose, M.D.** **987680 Nebraska Medical Center** **Omaha, NE 68198-7680** **United States**
Study Drug provided by:	**Incyte Corporation** **Route 141 & Henry Clay Road** **Building E336** **Wilmington, DE 19880** **United States**
Date of Protocol:	**5 May 2011**
Date(s) Revised:	**Amendment 2, date 5 December 2011** **Amendment 3, date 27 June 2012** **Amendment 4, date 23 January 2013** **Amendment 5, date 06 February 2014** **Amendment 6, date 16 February 2015** **Amendment 7, date 29 February 2016**

STUDY CONTACT INFORMATION

Lead Institution	**Participating Institution**
PI and Study Director: Julie M. Vose, M.D. Ph: 402-559-3848 Fax: 402-559-8101 jmvose@unmc.edu University of Nebraska Medical Center 987680 Nebraska Medical Center Omaha, NE 68198-7680 Sub-Investigators: James Armitage, M.D., Philip Bierman, M.D., R. Gregory Bociek, M.D. Matthew Lunning, D.O. Study Coordinator: Susan Blumel, R.N. Ph: 402-559-9183 Fax: 402-559-8101 sblumel@unmc.edu	PI: Thomas Witzig, M.D. Ph: 507-266-2040 Fax: 507-266-9277 Witzig.thomas@mayo.edu Mayo Clinic Hem 200 First St. SW Rochester, MN 55905-0001
	Participating Institution PI: Wyndam Wilson, M.D. Ph: 301-435-2415 Fax: 301-480-4087 wilsonw@mail.nih.gov National Cancer Institute 9000 Rockville Pike, Rm 4n115 Bldg 10 Bethesda, MD 20892-0001

TABLE OF CONTENTS

1.0 PROTOCOL SYNOPSIS 6

2.0 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS 10

3.0 INTRODUCTION 11
- 3.1 Background 11
- 3.2 Trial Rationale 14
- 3.3 Potential Risks and Benefits 16
- 3.4 Justification of Route, Dose Regimen and Treatment Period 17

4.0 STUDY OBJECTIVES 17
- 4.1 Primary Objectives 17
- 4.2 Secondary Objectives 17
- 4.3 Exploratory Objectives 17

5.0 INVESTIGATIONAL PLAN 18
- 5.1 Trial Endpoints 18
- 5.2 Other Exploratory Evaluations 18

6.0 SUBJECT ELIGIBILITY 20
- 6.1 Inclusion Criteria 20
- 6.2 Exclusion Criteria 21

7.0 SCHEDULE OF OBSERVATIONS 23
- 7.1 Schedule of Observations Flowchart 23
- 7.2 Screening and Enrollment 25
- 7.3 On-Treatment Evaluations 26
- 7.4 End-of-treatment Evaluations 27
- 7.5 Follow-up Evaluations 28
- 7.6 Duration of Participation 28

8.0 STUDY ASSESSMENTS 29
- 8.1 Demographic and Other Pre-Treatment Assessments 29
- 8.2 Safety Assessments 29
  - 8.2.1 Adverse Events 29
  - 8.2.2 Physical Examinations 29
  - 8.2.3 Vital Signs 29
  - 8.2.4 12-Lead ECG Interpretation 30
  - 8.2.5 Clinical Safety Laboratory Assessments 30

9.0 TREATMENT OF SUBJECTS 28
- 9.1 Investigational Product(s) Description 30
  - 9.1.1 Dosage and Dose Regimen 30
  - 9.1.2 Packaging and Labeling 31
  - 9.1.3 Storage/Stability 31
  - 9.1.4 Drug Accountability 31

9.2 Administration of Study Medication(s) 32
9.2.1 Starting Dose Assignment 32
9.2.2 Dose Increases for Inadequate Efficacy 32
9.2.3 Dose Adjustments for Safety 33
9.3 Concomitant Medications / Measures 36
9.4 Restricted and / or Prohibited Medications 37
9.4.1 Restricted Therapies 37
9.4.2 Prohibited Therapies 38
9.5 Discontinuation of Study Medication(s) 38

10.0 WITHDRAWAL OF STUDY SUBJECTS 38
10.1 Withdrawal Criteria and Procedures 38
10.1.1 Withdrawal of Subjects from Study 38
10.2 Replacement Subjects 39
10.3 Protocol Deviations 39

11.0 ADVERSE EVENTS 40
11.1 Definitions and Reporting 40
11.2 Laboratory Test Abnormalities 41
11.3 Serious Adverse Events 42
11.3.1 Definitions 42
11.3.2 Serious Adverse Events Reporting 43
11.4 Pregnancies 45
11.5 Warnings and Precautions 46

12.0 STATISTICS 46
12.1 Study Populations 46
12.2 Efficacy Analysis 46
12.2.1 Primary Efficacy Analyses 46
12.2.2 Secondary Efficacy Analyses 48
12.2.3 Exploratory Analyses 49
12.3 Safety Analysis 49
12.3.1 Adverse Events 49

13.0 DATA MANAGEMENT ....
13.1 Data Collection 50
13.1 Data Management 50

14.0 STUDY ADMINISTRATION ....
14.1 Access to Source Documents 51
14.2 Statement of Good Clinical Practices 52
14.3 Protocol Adherence 52
14.4 Study Termination 52
14.5 Financial Disclosure 53
14.6 Quality Control and Assurance 53
14.6.1 Sponsor Audits 53
14.6.2 Inspection by Regulatory Authorities 54

15.0 ETHICS.....
15.1 Institutional Review Board or Independent Ethics Committee 54
15.2 Informed Consent.....55
15.2.1 Data Privacy 56

16.0 RECORD KEEPING / RETENTION OF RECORDS .....
16.1 Confidentiality .....

17.0 REFERENCES .....

18.0 INVESTIGATOR'S SIGNATURE PAGE.....61

19.0 LIST OF APPENDICES.....62

APPENDIX 1 – RESPONSE CRITERIA 63

APPENDIX 2 – LABORATORY METHODS.....66

APPENDIX 3 – GRADING OF ADVERSE EVENTS 75

1.0 PROTOCOL SYNOPSIS

Title	A Phase 2 study of Oral Ruxolitinib Phosphate for the Treatment of Relapsed or Refractory Diffuse Large B-cell and Peripheral T-cell Non-Hodgkin Lymphoma
Protocol IND Number	112445
Study Phase	II
Study Design	This is an open-label, multicenter, Phase II study
Planned Number of Study Sites and Planned Number of Subjects	Four study sites will participate. Approximately 90 subjects will be enrolled
Coordinating Investigator	Julie Vose, M.D.
Study Objectives	**Primary Objective:** Assess the overall response rate (ORR) of subjects with diffuse large B-cell BCL) and peripheral T-cell non-Hodgkin lymphoma (PTCL) who are relapsed or refractory to front-line treatment and ineligible for stem cell transplantation or have recurrent disease after stem cell transplantation to oral ruxolitinib. **Secondary Objectives:** Evaluate safety of oral ruxolitinib therapy in subjects with DLBCL and PTCL Determine progression free survival (PFS), duration of response, and overall survival (OS) in subjects with DLBCL and PTCL **Exploratory Objectives:** Explore relationship between responses to oral ruxolitinib and alterations in GEP signatures as well as biomarker immunophenotypic changes related to JAK2/STAT3, NF-κB, BCR, PI3K/AKT, and mTOR pathways. Evaluate potential effect of oral ruxolitinib exposure on JAK2/STAT3 pathway inhibition in serial tumor samples.
Dose and Mode of Administration and Duration of Treatment	Subjects will receive continuous dosing with oral ruxolitinib for 28 day cycles. The study is comprised of 3 phases: **Screening:** up to 28 days **On-Treatment Phase:** participation may continue for subjects receiving benefit and who have not met any reason for study discontinuation **Follow-up:** Subjects will be followed after discontinuation from the study to assess disease status, subsequent therapies and survival. Follow-up will occur by a clinic visit or telephone contact every 3 months for the 1st year and every 6 months thereafter.

	Evaluable subjects will include subjects who have received at least 2 cycles of therapy, or subjects with documented progression of disease prior to completion of 2 cycles of therapy.
Correlative Studies	An initial mandatory tumor sample will be obtained for PY-STAT3 activation, NF-kB activation (nuclear staining for p50/52, CARD 11 mutation status, AKT pathway activations (pAKT IHC) An additional two tumor samples will also be obtained in subjects who consent to voluntary second biopsy to evaluate JAK2/STAT3 pathway inhibition and changes of other STAT3 targets after 2 cycles of therapy and at progression. An archived tumor sample collected at subjects' initial diagnosis will be obtained for correlation between current and initial tumor samples.
Summary of Entrance Criteria	Subjects may be included in the study if they are: 1. Subjects must have histologically documented relapsed, or refractory disease, with a diagnosis of one of the following lymphoid malignancies: Diffuse Large B-cell Lymphoma, Peripheral T-cell Lymphoma (any subtype). Subjects must have received at least one prior systemic chemotherapy and must have either received an autologous stem cell transplant, refused or been deemed ineligible for an autologous stem cell transplant; 2. Subjects must be willing and able to have a fresh tumor biopsy prior to start of study treatment for research evaluations and cohort categorizing *Note: if insufficient fresh tissue is obtained to provide sub-classification for cohorts, then tissue material from a previous relapse biopsy and/or original diagnostic block may be requested to meet this criterion*. 3. Subjects must have measurable lesions (at least one target lesion measuring 2 cm in diameter) by computerized tomography (CT) scan, and/or measurable lymphoma cutaneous lesions of any size; 4. Adult Age as defined by the State where the study site is located ($\geq$ 19 years old at UNMC; $\geq$ 18 years at MDACC, Mayo, NCI) 5. Eastern Cooperative Oncology Group (ECOG) performance status 0-2; 6. Adequate bone marrow, renal, and hepatic function, per local reference laboratory ranges (values must not be achieved with transfusions or growth factors) as follows: a. Absolute neutrophil count (ANC) $\geq$ 1,000/mm^3; b. Platelet count $\geq$ 75,000/mm^3; c. Hemoglobin $\geq$ 8.0 g/dL; d. Serum creatinine $\leq$ 2.0 g/dL or calculated creatinine clearance $\geq$ 60mL/min (Cockcroft-Gault Method); e. AST and ALT $\leq$ 2.5 x institutional upper limit of normal (ULN) or $\leq$ 5 x ULN if liver involved by lymphoma f. Bilirubin < 2.0 x ULN unless subject has Gilbert's disease, low-grade hemolysis, or liver involvement with lymphoma.

	7. At least 2 weeks since prior chemotherapy, biological therapy, radiation therapy, major surgery, other investigational, or anti-cancer therapy that is considered disease-directed and recovered from prior toxicities to Grade 0-1 at least 2 weeks prior to investigational therapy; 8. Females will be either postmenopausal for at least 1 year or surgically sterile for at least 3 months OR Females of child-bearing potential must have a negative pregnancy test at screening and agree to take appropriate precautions to avoid pregnancy from screening Until 3 months after their last dose of study medication. 9. Males must agree to take appropriate precautions to avoid fathering a child from screening until 3 months after their last dose of study medication. 10. Able to comprehend and willing to sign an Informed Consent Form (ICF). Subjects will be excluded from the study if they have: 1. History of or active central nervous system (CNS) malignancy 2. Allogeneic stem cell transplant within the last 6 months, or active graft versus host disease following allogeneic transplant, or subjects currently on immunosuppressive therapy following allogeneic transplant. 3. Uncontrolled intercurrent illness including, but not limited to, ongoing active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situation that would limit compliance with study requirements as judged by treating physician; subjects receiving antibiotics that are under control may be included in the study 4. Pregnant or breastfeeding women. 5. Clinically symptomatic and uncontrolled cardiovascular disease 6. History of myocardial infarction, severe/unstable angina, or symptomatic congestive heart failure, within the 6 months prior to study drug administration 7. Current or recent history (< 21 days prior to start of treatment) of a clinically significant bacterial, viral, fungal, parasitic or mycobacterial infection. 8. History of other malignancy, with the exception of squamous cell carcinoma of the skin, basal cell carcinoma of the skin, cervical intraepithelial neoplasia, or other malignancies that have been in remission for at least 3 years 9. Presence of a malabsorption syndrome possibly affecting drug absorption (eg, Crohn's disease or chronic pancreatitis). 10. Any prior or concomitant use of another JAK inhibitor. 11. Known active hepatitis B or C, or HIV infection. 12. Subjects who, in the opinion of the Investigator, are unable or unlikely to comply with the dosing schedule and study evaluations.
Statistical Methods	The study is 3-step design. There will be 3 cohorts of subjects: Cohort 1 - DLBCL ABC Cohort 2 - DLBCL GCB and Cohort 3 - PTCL. Each

	cohort will have a minimum of 10 and a maximum of 30 subjects enrolled. There will be a maximum of 90 subjects enrolled in the study. Enrolled subjects who do not fit into the cohorts described above (i.e. non-GCB, primary mediastinal large b-cell, or other) will be placed into a 4th cohort.

2.0 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

AE	Adverse event
BMI	Body Mass Index
CFR	Code of Federal Regulations
eCRF	Electronic Case Report Form
CRO	Contract Research Organization
ECG	Electrocardiogram
HIPAA	Health Insurance Portability and Accountability Act (in relation to the Privacy Rule implemented April 2003)
ICH	International Conference on Harmonization of Pharmaceuticals for Technical Use
IRB/IEC	Institutional Review Board or Independent Ethics Committee
mcg	Microgram
mg	milligram
mL	milliliter
mmol	millimol
PK	Pharmacokinetics
SAE	Serious Adverse Event

3.0 INTRODUCTION

3.1 Background

Non Hodgkin Lymphomas (NHLs) are the most common hematologic malignancy with annual US incidence of approximately 66,000 in the US [1]. NHLs are a heterogeneous group of lymphoproliferative malignancies with differing patterns of behavior and responses to treatment [2]. NHL is much less predictable than Hodgkin Lymphoma and has a far greater predilection to disseminate to extranodal sites. NHLs can be divided into 2 prognostic groups: indolent and aggressive lymphomas. Despite progress in lymphoma therapy, many patients with these diseases still die from relapse and disease progression or from complications associated with treatment; therefore, more selective and less toxic treatments are needed. Constitutive activation of the JAK-STAT pathway has been reported in various types of leukemias and lymphomas, suggesting that targeted therapies directed at this pathway may show clinical benefit in these malignancies. More recent reports have also identified activating mutations or gene amplification of JAK1 and JAK2 in acute myeloid and lymphoblastic leukemias, lymphoma and in myelodysplastic syndrome [3-13]. As with many therapeutics, selective JAK inhibitors may be most beneficial when combined with other targeted or cytotoxic therapies.

Ruxolitinib phosphate (also known as INCB018424 phosphate or INC424) is an inhibitor of the Janus kinase family of protein tyrosine kinases and will be referred to as ruxolitinib throughout this document. Ruxolitinib is an investigational product that is in development for treatment of myeloproliferative neoplasms (MPNs), hematologic malignancies and solid tumors. JAKs play an important role in signal transduction following cytokine and growth factor binding to their receptors. Aberrant activation of JAKs has been associated with increased malignant cell proliferation and survival. In particular, a pathologic role for JAK2 has recently been suggested for the majority of patients with Philadelphia chromosome negative MPNs. Therefore, JAK inhibitors represent potential therapeutic agents for these disease states.

<u>Clinical Summary</u>

Ruxolitinibhas been administered to over 180 healthy volunteers as single, repeat single, or multiple doses of up to 10 days duration. Ithas also been administered to approximately 450 subjects with MF for periods of up to > 24 months, and over 100 subjects with prostate cancer, multiple myeloma, polycythemia vera or essential thrombocythemia for periods of up to > 24 months.

In healthy volunteer studies, a transient, reversible decrease in neutrophil count has frequently been seen following dosing, which reverses after 12 to 24 hours off drug, suggestive that the neutropenia may reflect an effect of ruxolitininb blocking IL-6 signaling and causing neutrophil margination on blood vessel walls. In a repeat dose healthy volunteer study, neutropenia of any severity grade was seen in 22% of placebo subjects, 11% of subjects receiving 50 mg qd, 67% of subjects receiving 100 mg qd, 13% of subjects receiving 15 mg bid, 33% of subjects receiving 25 mg bid and 67% of subjects receiving 50 mg bid. Importantly, these neutropenia events were of Grade 1 or Grade 2 severity with a single instance of severe Grade 4 neutropenia that led to discontinuation of ruxolitinib in 1 subject receiving a 50 mg bid dose (NOTE: This was the highest dose administered during any clinical study). The maximum tolerated dose in healthy volunteers was determined to be 25 mg bid and no dose limiting toxicities (DLT) were seen at 100 mg qd. (See the Investigator's Brochure for additional details). Ruxolitinib was tolerated in patients with rheumatoid arthritis dosed up to 28 days. Adverse events were seen in similar frequency in patients receiving ruxolitinib as in patients receiving placebo. One patient dosed with 25 mg bid exhibited Grade 3 neutropenia, which improved with continued dosing, and one patient who had a prior history of thrombocytopenia developed Grade 3 platelet decline. One subject dosed with 25 mg bid was withdrawn due to nausea and vomiting. (See the Investigator's Brochure for additional details).

In a study of patients with advanced prostate cancer, ruxolitinib was generally well tolerated. Serious adverse events seen were generally related to the expected progression of the underlying disease of prostate cancer. One patient, with a history of congestive

heart failure, hypertension, hypercholesterolemia and T-wave abnormality on ECG who was severely ill with progressive disease, was reported to have experienced sudden death while on study. This death was attributed to cardiac arrest, which was felt by the investigator to be possibly related to ruxolitinib. (See the Investigator's Brochure for additional details).

In a study of patients with advanced multiple myeloma, ruxolitinib was generally well tolerated. Observed toxicities included thrombocytopenia and anemia. SAE's reported included disease progression, pneumonia, urinary tract infection, anemia, pyrexia and other cardiac and pulmonary complications. Safety results were consistent with those observed in other ruxolitinib studies. (See the Investigator's Brochure for additional details).

In a study of patients with Advanced Polycythemia Vera (PV) or Essential Thrombocythemia (ET), ruxolitinib was generally well tolerated. The most commonly observed AEs were anemia in ET subjects and anemia and thrombocytopenia in PV subjects, and these were reversible with dose modifications. Serious adverse events deemed at least possibly related to study medication included a case of atrial flutter in a PV subject which resolved; acute renal failure in an ET subject which resolved; and a renal tumor in a PV subject which was surgically resected and the subject recovered. (See the Investigator's Brochure for additional details).

In an ongoing study in subjects with myelofibrosis (MF) where median time on drug is ~ 15 months (N=154), ruxolitinib was well tolerated by this elderly population (median age 65) with advanced disease. The DLT was thrombocytopenia. Initial dose ranging established an MTD of 25 mg bid and 100 mg qd in the MF population. Most adverse events were mild to moderate in severity and considered unrelated to administration of study drug. Related adverse events occurring in at least 5% (8 subjects) of the 154 subjects included in the safety database through December 31, 2009 were restricted to anemia (45 subjects), thrombocytopenia (66 subjects), weight increased (11 subjects),

diarrhea (10 subjects) and fatigue (8 subjects). Both anemia and thrombocytopenia represent JAK-inhibitor induced myelosuppression. Forty (40) subjects (26% of study population) had a Grade 3 or Grade 4 decline in platelet count during the study (31 Grade 3 events, 9 Grade 4 events). Subjects with Grade 3 or 4 thrombocytopenia entered the study, in general, with platelet counts less than 200 K/μL, although there are exceptions to this trend. Twenty percent of Grade 3 + 4 thrombocytopenia events occurred in the first 4 weeks of dosing, just under half (48%) of Grade 3 + 4 events occurred in the first 16 weeks of treatment. The incidence of thrombocytopenia was dose dependent, as anticipated. For most subjects, thrombocytopenia was rapidly reversible and manageable with ruxolitinib dose interruption and/or dose reduction. Anemia, in general, reflected the low hemoglobin status at baseline in this disease population.

Ruxolitinib has demonstrated marked reduction in spleen size in the ongoing study in patients with MF, and without regard to presence of the JAK2 V617F mutation. In addition to spleen size reduction, Eastern Cooperative Oncology Group (ECOG) scores and symptoms thought to be related to splenomegaly, as well as symptoms related to elevated cytokine levels, all show improvement with ruxolitinib treatment. As a surrogate marker for functional benefit, exercise capacity was assessed with a standardized six minute walk test (6MWT). ruxolitinib therapy resulted in improved 6MWT performance after 1, 3 or 6 months of therapy. After an initial small weight loss likely due to resolution of ascites and/or reduction in splenomegaly, there is an increase in total body weight; importantly, there is weight gain in subjects with low body mass index at entry, i.e., cachectic subjects. See the Investigator's Brochure for additional details.

In Study INCB 18424-256 in patients with advanced PV or ET, and after a median follow-up of 15 months (range 8-21), 97% of enrolled PV subjects achieved Hct control to < 45% in the absence of phlebotomy, and all continued to maintain phlebotomy-independence at the time of last follow-up visit. Prior to treatment with ruxolitinib, twenty-six subjects were phlebotomy-dependent with the majority being maintained on

cytoreductive therapy. After treatment, all but one of the 26 have maintained phlebotomy-independence as of the third week of the study. Rapid and sustained reductions in mean Hct %, WBC count, and platelet counts have been noted (Table 18). Leukocytosis > 15 x 10^9/L was present in 44% of subjects at baseline and improved (≤ 15 x 10^9/L) or normalized (≤ upper limit of normal, ULN) in 87% and 67% of these subjects, respectively. Thrombocytosis > 600 x 10^9/L was present in 38% of subjects at baseline and improved (≤ 600 x 10^9/L) or normalized (< ULN) in 92% and 69%, respectively. See the Investigator's Brochure for additional details.

Two Phase 3 registrations studies are ongoing with subject participation but the protocol-specified criteria to allow the database to be locked and data analyzed has been met. Study INCB 18424-351 is a double blind, placebo controlled study of ruxolitinib in subjects with MF, and Study INCB 18424-352 is an open label comparative study of ruxolitinib versus best available therapy in subjects with MF. In both Phase 3 studies, subjects begin dosing at 20 mg bid if their platelet count at study entry is > 200,000/μL and at 15 mg bid for a platelet count at study entry of 100,000 to 200,000/μL, inclusive.

3.2 Trial Rationale

This study is designed to investigate potential benefits of ruxolitinib in patients with Non-Hodgkin's Lymphoma, DLBC subtype and PTCL. This will be the first study of ruxolitinib in patients with lymphoma. The pre-clinical data provide support for the use of ruxolitinib in targeting the JAK-STAT pathway for the treatment of lymphomas.

For the potential treatment of neoplastic disease ruxolitinib was evaluated in two mouse models where either a cytokine-dependent multiple myeloma cell line, INA-6, or a BaF3 cell line engineered to express JAK2V617F was inoculated. Ruxolitinib administration (oral, bid) inhibited JAK-STAT signaling and INA-6 tumor growth in a dose-dependent manner as demonstrated by the percent tumor growth inhibition (TGI) and reduction in pSTAT3 levels determined by ELISA. Based on the data obtained from this study, it appears that > 80% suppression of JAK-STAT signaling for a minimum of 8 h each day

(4 h following each of two daily doses), and not continuous JAK inhibition, is required for maximal suppression of tumor growth in this model.

Ruxolitinib is an inhibitor of the Janus kinase family of protein tyrosine kinases (JAKs). Subject eligibility criteria are consistent with those used in studies of this population. The multi-center nature of this study provides greater likelihood that the results will have general applicability.

3.3 Potential Risks and Benefits

Potential Risks

No specific findings in nonclinical repeat dose toxicity studies identify clinical risk other than noting that consequences of immunosuppression may occur. Hypotension and increases in heart rate were noted at a high dose in a cardiovascular preclinical study. However, these findings have not been recapitulated in a clinical setting.

The primary clinical risks with ruxolitinib treatment are the potential sequelae of decreased hematopoietic proliferation secondary to the inhibition of growth factor pathways by JAK2 inhibition. Dose-dependent, reversible thrombocytopenia has been observed in the ongoing study in subjects with MF and represents the DLT. Anemia and, less frequently, neutropenia have also been observed in the ongoing study in patients with MF. Increased rates of infection and anemia are potential risks of myelosuppression, and there are multiple sequelae of anemia including the burden and risks of transfusion. A few subjects have had an apparent worsening of their pre-morbid disease symptoms following rapid cessation of ruxolitinib therapy and a gradual tapering and use of steroids in fragile subjects may be considered when stopping ruxolitinib therapy. In healthy volunteers and patients with RA with greater bone marrow reserve, the effects on hematopoietic proliferation appear to be less pronounced.

Pneumonias and herpes zoster infections have been observed in MF patients treated with ruxolitinib. These infections have, in general, ruxolitinib (See Investigator's Brochure) been assessed as unrelated to study medication, and may reflect in large part the

heightened incidence of these infections in this diseased and elderly study population. In ongoing phase 2 and 3 studies, other opportunistic infections have been seen in MF patients, including a serious adverse event of extra-pulmonary tuberculosis.

3.4 Justification of Route, Dose Regimen and Treatment Period

In prior studies evaluating treatment in patients with myelofibrosis, the maximum tolerated oral dose was 25 mg po BID. Therefore, this dose will be the upper reference point in the current study. Starting doses explored in polycythemia vera and myelofibrosis range from 10 mg po BID to 20 mg po BID. Since patients on this study may have limited marrow reserves from prior therapies and since thrombocytopenia is the most prominent toxicity seen in earlier studies, the baseline platelet count will be used to guide the initial dose, either 15 mg po BID or 10 mg po BID (see section 9.2). Doses may be decreased in the presence of toxicities, or may be increased in the absence of toxicity and lack of objective response to a maximum dose of 25 mg po BID.

4.0 STUDY OBJECTIVES

4.1 Primary Objective

- Assess the overall response rate (ORR) of subjects with relapsed DLBCL and PTCL who are relapsed or refractory to front-line treatment and ineligible for stem cell transplantation or have recurrent disease after stem cell transplantation to oral ruxolitinib.

4.2 Secondary Objectives

- Evaluate safety of oral ruxolitinib in subjects with DLBCL and PTCL
- Determine progression free survival (PFS), duration of response, and overall survival (OS) in subjects with DLBCL and PTCL

4.3 Exploratory Objectives

- Explore the relationship between responses to oral ruxolitinib and alterations in GEP signatures as well as biomarker immunophenotypic changes related to JAK2/STAT3, NF-κB, BCR, PI3K/AKT, and mTOR pathways.
- Evaluate potential effect of oral ruxolitinib exposure on JAK2/STAT3 pathway inhibition in serial tumor samples

5.0 INVESTIGATIONAL PLAN

5.1 Study Endpoints

- Determination of response rates according to revised Cheson criteria.
- Evaluate safety of oral ruxolitinib in subjects with DLBCL and PTCL according to NCI CTC v. 4.0 criteria.
- Determine progression free survival (PFS), duration of response, and overall survival (OS) in subjects with DLBCL and PTCL
- Explore relationship between responses to oral ruxolitinib (and alterations in GEP signatures as well as biomarker immunophenotypic changes related to JAK2/STAT3, NF-κB, BCR, PI3K/AKT, and mTOR pathways.
- Evaluate potential effect of oral ruxolitinib exposure on JAK2/STAT3 pathway inhibition in serial tumor samples

5.2 Other Exploratory Evaluations

5.2.1. Tumor Genotyping

A mandatory tumor sample will be obtained from all subjects at screening for gene expression profiling studies to assess pathway activation (PY-STAT3 activation, NF-κB activation (nuclear staining for p50/52, CARD 11 mutation status, AKT pathway activations (pAKT IHC). Either excisional or two 14 gauge or larger cores will be submitted to the central lab. In addition, all diffuse large B-cell non-Hodgkin lymphoma tumor samples will be evaluated with immunohistochemistry to determine subtype (activated B-cell versus germinal center B-cell subtype versus other subtypes of DLBC). The central pathology review will be used to identify subject cohort assignment. (See appendix 2 for details).

Should there not be sufficient tumor samples to evaluate and identify subject cohort assignment, then an archived tumor sample (paraffin blocks) from a relapsed or initial diagnostic biopsy may be requested. These specimens would be submitted to the central lab for review.

Archived tumor sample from the time of initial diagnosis will be obtained from all subjects for RNA sequencing and comparison of gene expression between initial and current tumor tissue. A minimum of 5 unstained 10 μm slides will be submitted to the central lab.

Additional, optional fresh tumor samples will also be evaluated in subjects who consent to voluntary biopsies to evaluate JAK2/STAT3 pathway inhibition and changes of other STAT3 targets while on therapy and post-treatment for subjects who have documented disease progression. Either excisional biopsy or two 14 gauge or larger, $\geq$ 2cm length cores will be obtained sometime after the completion of 2 cycles for the on therapy biopsy and within 6 weeks post treatment for the post treatment biopsy. (See appendix 2).

5.2.2. Blood Sampling for Cytokine Profiling

Serial blood samples will be collected before the first dose of study drug, at three hours after the first dose of study drug, and then weekly on Days 8, 15, and 22 during cycle 1 and on Day 1 of Cycle 2 for a total of 5 time points. Samples will be collected and shipped to the central laboratory at the University of Nebraska Medical Center for analysis to measure cytokines and chemokines using plasma and PBMC. (See appendix 2).

5.2.3 Blood Sampling for C-reactive protein

Blood samples will be collected within 7 days prior to the first dose of study drug, and on Day 1 of Cycle 2, and at time of relapse. Samples will be collected and shipped to the central laboratory at the University of Nebraska Medical Center. (See appendix 2).

6.0 SUBJECT ELIGIBILITY

6.1 Inclusion Criteria

Subjects may be included in the study if they are:

1. Subjects must have histologically documented relapsed or refractory disease, with a diagnosis of one of the following lymphoid malignancies: Diffuse Large B-cell Lymphoma, Peripheral T-cell Lymphoma (any subtype). Subjects must have received at least one prior systemic chemotherapy and must have either received an autologous stem cell transplant, refused or been deemed ineligible for an autologous stem cell transplant;
2. Subjects must be willing and able to have a fresh tumor biopsy prior to start of study treatment for research evaluations and cohort categorizing. *Note: if insufficient fresh tissue is obtained to provide sub-classification for cohorts, then tissue material from a previous relapse biopsy and/or original diagnostic block may be requested to meet this criterion*;
3. Subjects must have measurable lesions (at least one target lesion measuring 2 cm in diameter) by computerized tomography (CT) scan, and/or measurable lymphoma cutaneous lesions of any size;
4. Adult Age as defined by the State where the study site is located ($\geq$ 19 years old at UNMC; $\geq$ 18 years at MDACC, Mayo, NCI)
5. Eastern Cooperative Oncology Group (ECOG) performance status 0-2;
6. Adequate bone marrow, renal, and hepatic function, per local reference laboratory ranges (values must not be achieved with transfusions or growth factors) as follows:

   a. Absolute neutrophil count (ANC) $\geq$ 1,000/mm^3;

   b. Platelet count $\geq$ 75,000/mm^3;

   c. Hemoglobin $\geq$ 8.0 g/dL;

   d. Serum creatinine $\leq$ 2.0 g/dL or calculated creatinine clearance $\geq$ 60mL/min (Cockcroft-Gault Method);

   e. AST and ALT $\leq$ 2.5 x institutional upper limit of normal (ULN) or $\leq$ 5 x ULN

if liver involved by lymphoma

f. Bilirubin < 2.0 x ULN unless subject has Gilbert's disease, low-grade hemolysis, or liver involvement with lymphoma.

7. At least 2 weeks since prior chemotherapy, biological therapy, radiation therapy, major surgery, other investigational, or anti-cancer therapy that is considered disease-directed and recovered from prior toxicities to Grade 0-1 at least 2 weeks prior to investigational therapy;
8. Females will be either postmenopausal for at least 1 year or surgically sterile for at least 3 months;

   OR

   Females of child-bearing potential must have a negative pregnancy test at screening and agree to take appropriate precautions to avoid pregnancy from screening until 3 months after their last dose of study medication.
9. Males must agree to take appropriate precautions to avoid fathering a child from screening until 3 months after their last dose of study medication.
10. Able to comprehend and willing to sign an Informed Consent Form (ICF).

6.2 Exclusion Criteria

Subjects will be excluded from the study if they have:

1. History of or active central nervous system (CNS) malignancy
2. Allogeneic stem cell transplant within the last 6 months, or active graft versus host disease following allogeneic transplant, or subjects currently on immunosuppressive therapy following allogeneic transplant.
3. Uncontrolled intercurrent illness including, but not limited to, ongoing active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situation that would limit compliance with study requirements as judged by treating physician; subjects receiving antibiotics that are under control may be included in the study
4. Pregnant or breastfeeding women.
5. Clinically symptomatic and uncontrolled cardiovascular disease

6. History of myocardial infarction, severe/unstable angina, or symptomatic congestive heart failure, within the 6 months prior to study drug administration
7. Current or recent history (< 21 days prior to start of treatment) of a clinically significant bacterial, viral, fungal, parasitic or mycobacterial infection.
8. History of other malignancy, with the exception of squamous cell carcinoma of the skin, basal cell carcinoma of the skin, cervical intraepithelial neoplasia, or other malignancies that have been in remission for at least 3 years
9. Presence of a malabsorption syndrome possibly affecting drug absorption (eg, Crohn's disease or chronic pancreatitis).
10. Any prior or concomitant use of another JAK inhibitor.
11. Known active hepatitis B or C, or HIV infection.
12. Subjects who, in the opinion of the Investigator, are unable or unlikely to comply with the dosing schedule and study evaluations.

7.0 SCHEDULE OF OBSERVATIONS

7.1 Schedule of Observations Flowchart

	Screening		**Cycle 1 (+/- 2 days each time point)**				**Cycle 2**	**Every Cycle thereafter**	**End of Treatment**	**Follow-up**
	≤28 days	**≤14 days**	**Day 1**	**Day 8**	**Day 15**	**Day 22**	**Day 1** (+/- 7 days)	**Day 1** (+/- 7 days)	**(+/- 7 days)**	**(each time point +/- 4 weeks)**
Evaluation										
Informed consent	**X**									
Medical history	**X**									
Physical examination		**X**					**X**	**X**	**X**	
Vital Signs		**X**					**X**	**X**		
ECOG performance score		**X**					**X**	**X**		
Concomitant medications		**X**	**X**				**X**	**X**	**X**	
12-lead ECG		**X**[a]								
Serum Pregnancy test		**X**[a]								
Hematology Lab		**X**	**X**[b]	**X**	**X**	**X**	**X**	**X**	**X**	
Chemistry Lab		**X**	**X**[b]	**X**	**X**	**X**	**X**	**X**	**X**	
PET scan	**X**[c]									
CT scan	**X**[c]							**X**[c]	**X**[c]	
Bone Marrow Biopsy								**(X)**[d]		
Tumor Tissue collection for GEP,IHC or other subtype analysis	**X**[e]							(X)[e]	**(X)**[e]	
Archive Tissue collection	**X**									
Blood Sampling for Cytokine Profiling[f]			**X**[b,f]	**X**[f]	**X**[f]	**X**[f]	**X**			
Blood Sampling for CRP[g]			**X**				**X**		**X**	
Study med dispensed			**X**				**X**	**X**		
Adverse event reporting	**X**		**X**	**X**	**X**	**X**	**X**	**X**	**X**[h]	
Compliance assessment – study med.				**X**	**X**	**X**	**X**	**X**	**X**	
Subject Status										**X**[i]

a. Repeat ECG's and pregnancy testing are not required but may be repeated throughout the study at the discretion of the investigator.

b. Cycle 1, day 1 Hematology and Chemistry labwork may be collected within 7 days prior to C1, D1 study drug dosing.

c. Baseline staging should include measurable disease by CT scan and a PET scan. If sites can accomplish tumor measurements with one PET/CT scan, this will be acceptable. CT scans will be done for restaging purposes. In subjects with cutaneous lesions only, bidimensional measurements and photographs will be used for response assessment. PET scan may be repeated to confirm PR or CR at the discretion of the investigator. Disease restaging will occur after 2 cycles concurrent with the start of Cycle 3 and then every 2 cycles thereafter. In subjects who are beyond the post cycle 6 response assessment AND have demonstrated objective resonse (PR or CR) restaging may be done every 3 cycles thereafter.

d. Bone marrow aspirate and biopsy will be performed to confirm complete remission unless the subject had a documented negative bone marrow biopsy evaluated with 45 days prior to treatment initiation.

e. Tumor excisional biopsy or 2 core biopsies are required at baseline and are optional during treatment at time of disease progression (post Cycle 2) and End of treatment. See appendix 2 for tumor tissue collection, processing and shipping instructions.

f. The Cycle 1, day 1 blood may be collected within 7 days prior to initiation of ruxolitinib. On Cycle 1, Day 1 only, an additional blood for cytokine analysis will be drawn 3 hours after the first dose of ruxolitinib. See appendix 2 for cytokine collection, processing and shipping instructions. Blood for cytokine analysis will be drawn prior to the morning dose of ruxolitinib on days 8, 15, 22 in Cycle 1 and on Cycle 2, day 1.

g. Blood for C-reactive protein (CRP) will be analyzed from blood collected for cytokine profling at pre-treatment (within 7 days prior to initiation of ruxolitinib) and at cycle 2, day 1. An additional blood sample for CRP will be collect at time of progression or relapse.

h. All subjects must be followed for safety for at least 30 days after study treatment has ended, or until study drug related toxicities resolve, return to Baseline or are deemed irreversible, whichever is longer.

i. Subjects will be followed every 3 months for the first year and every 6 months thereafter via clinic contact or telephone to obtain disease status and survival information (see section 7.6).

7.2 Screening Evaluations and Subject Enrollment

7.2.1 The following procedures will be performed at the Screening visit within 28 days prior to start of treatment unless noted otherwise:

- Informed consent must be obtained before study-specific screening evaluations are performed, unless performed as standard of care. The informed consent process should be documented in the subject's medical chart.
- Medical history;
- Physical examination, including height, weight, and clinical signs and symptoms (within 14 days prior to cycle 1 day 1);
- Vital signs, including pulse, systolic and diastolic blood pressure (BP), respiration rate, and body temperature(within 14 days prior to cycle 1 day 1);
- ECOG performance status(within 14 days prior to cycle 1 day 1);
- Single 12-lead ECG;
- Hematology: CBC with differential within 14 days prior to cycle 1 day 1 (see section 8.2.5);
- Blood chemistry within 14 days prior to cycle 1 day 1(see section8.2.5);
- Serum pregnancy test for women of child-bearing potential (within 14 days prior to cycle 1 day 1);
- Concomitant medications(within 14 days prior to cycle 1 day 1);
- Whole body PET scan and CT of chest, abdomen and pelvis (may include neck at the discretion of the investigator).
- Tumor tissue collection must be obtained on all subjects prior to start of treatment (see section 5.2.1 and appendix 2 for details) for research evaluation.
- Archived tumor tissue from time of initial diagnosis of DLBC or PTCL must be obtained for research evaluation and correlation with the current tissue sample.

7.2.2 Subject Enrollment

The results from the Screening visit evaluations will be reviewed to determine if the subject continues to meet the eligibility requirements as specified in the protocol. If a subject does not meet eligibility requirements or withdraws consent prior to treatment initiation, then that subject will be determined a screen failure and will not be eligible to receive treatment in the study.

Subjects who have signed the informed consent and meet all the entry criteria (see Inclusion/Exclusion Criteria) will be enrolled in the study. At the time of enrollment a subject study identifier will be assigned by the sponsor.

7.3 On-Treatment Evaluations

Cycle 1, Day 1 Assessments:

- Hematology and Chemistry laboratory assessments unless completed within 7 days prior (see section 8.2.5) to treatment. If the screening laboratory assessments were collected within 7 days prior to treatment, these results may be also used for the Cycle 1, day 1 assessment.
- Vital Signs within 30 minutes prior to first dose
- AE assessment
- Con Meds
- Subjects will be prescribed ruxolitinib orally twice daily. A cycle consists of 28 consecutive days of BID dosing. (See section 9.0)
- Blood Samples for Cytokine Profiling and C reactive protein analysis will be collected within 7 days before first dose and blood for cytokine profiling will be collected at 3 hours after the first dose of ruxolitinib. (see appendix 2)
- Vital Signs one hour after first dose.
- Subjects will be instructed to take doses approximately 12 hours apart.
- Subjects will be asked to complete a study drug dosing diary and to return all empty study drug bottles and completed diaries to the study center.

Cycle 1, weekly assessments on Day 1, Day 8, Day 15, and Day 22:

- Hematology and chemistry laboratory assessments (see section 8.2.5)
- Blood Sample for Cytokine Profiling will be collected prior to the morning dose of ruxolitinib (see appendix 2)

Cycle 2, and each subsequent Cycle assessments:

- Hematology and Chemistry laboratory assessments (see section 8.2.5)

- Cycle 2, Day 1 only: Blood Sample for Cytokine Profiling and C-reactive protein analysis will be collected prior to initiation of cycle 2 dosing (see appendix 2).
- Vital Signs
- AE assessment
- Concomitant Medications
- Physical Exam
- ECOG performance status
- Optional fresh tumor sample collection: Either excisional biopsy or two 14 gauge or larger, $\geq$ 2cm length cores will be obtained sometime after the completion of 2 cycles of treatment (see appendix 2).

Disease restaging will occur after 2 cycles concurrent with the start of Cycle 3 and then every 2 cycles thereafter. In subjects who are beyond the post cycle 6 response assessment AND have demonstrated objective resonse (PR or CR) restaging may be done every 3 cycles thereafter. The following tests will be completed:

- CT scan chest, abdomen, pelvis. However, in subjects with measureable cutaneous lesions only, photographs and bidimensional measurements of skin lesions may be utilized for response assessment. The Investigator should take care to ensure consistency in response assessment evaluation throughout the study.
- PET scan may be performed at the discretion of the investigator to confirm CR or PR
- If subject has complete remission by CT or CT/PET scan perform a bone marrow biopsy for confirmation of complete response unless the patient had a documented negative bone marrow biopsy within 45 days prior to treatment initiation.

7.4 Duration of Participation

Subjects who are demonstrating benefit may continue to receive treatment with ruxolitinib unless a criterion for discontinuation from the study has been met. The criteria for study discontinuation are listed below:

- unacceptable toxicity;
- disease progression or relapse
- investigator discretion
- withdrawal of consent
- pregnancy

If a subject is withdrawn from the study treatment for any reason, the study sponsor-investigator must be notified and an end-of-treatment visit should be performed. Follow-up evaluations will also be performed in accordance with the protocol requirements.

7.5 End-of-treatment Evaluations

Subjects who are withdrawn from the study treatment for any reason will have evaluations performed at End-of-Treatment. The following evaluations and procedures will be performed at this visit.

- AE assessment
- Concomitant Medications
- Physical Exam
- ECOG performance status
- Hematology and Chemistry laboratory assessments
- Documentation of response
- All subjects must be followed for safety for at least 30 days after ruxolitinib treatment has ended, or ruxolitinib related toxicities resolve or return to Baseline or are deemed irreversible, whichever is the longer duration.
- At time of relapse subjects will have a blood sample collected for C-reactive protein evaluation (see appendix 2. II. C)
- Optional fresh tumor sample collection: subjects who have documented disease progression will have either excisional biopsy or two 14 gauge or larger, $\geq$ 2cm length cores within 6 weeks post treatment (see appendix 2).

7.6 Follow-up Evaluations

Subjects will be followed every 3 months for the first year and every 6 months thereafter via a clinic visit or contact by telephone by research staff personnel to obtain the following information:

- Disease Status will be collected until first documented relapse or progression of the disease under study;
- Date last alive or date of death

8.0 STUDY ASSESSMENTS

8.1 Demographic and Other Pre-Treatment Assessments

After written informed consent is obtained, demographic data and a complete medical and medication history will be collected at Screening. Height, body weight and BMI measurements will be done. Medical history will be updated up through the first dose of study medication, after which adverse events will be recorded. Disease staging, laboratory assessments, physical exam, performance status will be completed as outlined in the study flowchart. In addition, all subjects will undergo tumor biopsy for exploratory tumor GEP and IHC studies.

8.2 Safety Assessments

8.2.1 Adverse Events

Adverse events will be monitored continuously during the study after the first dose of study medication. Subjects will be instructed to report all AEs during the study and subjects will be assessed for the occurrence of AEs throughout the study. All AEs (serious and nonserious) must be recorded on the source documents and case report forms regardless of the assumption of a causal relationship with the study medication. The definition, reporting, and recording requirements for AEs are described in Section 11.

8.2.2 Physical Examinations

A comprehensive physical examination will be performed at the times indicated on the Schedule of Observations. The comprehensive physical examination will include the following organ or body system assessments: skin; head, eyes, ears, nose, and throat; thyroid; lungs; cardiovascular; abdomen (liver, spleen); extremities, lymph nodes, and a brief neurological exam.

8.2.3 Vital Signs

Vital sign measurements (blood pressure, heart rate, and body temperature) will be collected on the days and times noted in the Schedule of Observations.

8.2.4 12-Lead ECG Interpretation

The baseline 12-lead ECG will be interpreted by the Investigator at the site and will be used for immediate subject management. Subsequent ECG evaluations may be performed at the discretion of the investigator. The decision to include/exclude a subject or discontinue a subject's participation in the study based on an ECG flagged as "Abnormal, Clinically Significant" is the responsibility of the Investigator.

8.2.5 Clinical Safety Laboratory Assessments

All subjects will have samples of blood collected on the days and times noted in the Schedule of Observations for analysis of serum chemistry, hematology, and pregnancy status. The hematology panel will include: hematocrit, hemoglobin, platelet count, red blood cell count, white blood cell count, absolute basophils, absolute eosinophils, absolute lymphocytes, absolute monocytes, absolute neutrophils, blasts. The chemistry panel will include: sodium, potassium, calcium, blood urea nitrogen (BUN), creatinine, total (unconjugated) bilirubin, glucose, total protein, albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), lactic dehydrogenase (LDH), alkaline phosphate.

9.0 TREATMENT OF SUBJECTS

9.1 Investigational Product(s) Description

9.1.1 Dosage and Dose Regimen

Open label ruxolitinib will be supplied by Incyte Corporation. Ruxolitinib tablets (5 mg) will be administered as oral doses without regard to food in an outpatient setting. Starting doses will be either 15 mg BID or 10 mg BID as described in Section 9.2. Maximum dose will not exceed 25 mg BID. A cycle consists of 28 consecutive days of BID dosing. Dosing is continuous without planned interruptions between cycles. Dosing may be interrupted or modified due to toxicities (see section 9.2).

9.1.2 Packaging and Labeling

Ruxolitinib 5 mg tablets are packaged as 60-count in high-density polyethylene (HDPE) bottles. The bottles will include labeling "New Drug - Limited by Federal (USA) Law to Investigational Use".

9.1.3 Storage/Stability

The bottles of tablets should be stored at room temperature, 15°C to 30°C (59°F to 86°F). Stability studies will be conducted on all clinical batches to support the clinical trial.

9.1.4 Drug Accountability

Responsibility for drug accountability at the study site rests with the Investigator; however, the Investigator may assign some of the drug accountability duties to an appropriate pharmacist or designee. Inventory and accountability records must be maintained and readily available for inspection by the study monitor and are open to inspection at any time by any applicable regulatory authorities.

The Investigator or designee will be expected to collect and retain all used, unused, and partially used containers of study medication until the end of the study. The Investigator or designee must maintain records that document:

- investigational product delivery to the study site
- the inventory at the site
- use by each subject including pill/unit counts from each supply dispensed
- product returned to the Investigator or designee.

These records should include dates, quantities, batch/serial numbers (if available), and the unique code numbers (if available) assigned to the investigational product and study subjects.

The investigational product must be used only in accordance with the protocol. The Investigator will also maintain records adequately documenting that the subjects were provided the study medication specified.

Completed accountability records will be archived by the site. If any unused ruxolitinib bottles remain at the end of the study, they will be accounted for at the site close-out visit. If a site is able to destroy ruxolitinib at their premises, or through a certified vendor, they may do so upon authorization by the Sponsor, provided their destruction policy is made available to the Sponsor.

9.2 Administration of Study Medication

9.2.1 Starting Dose Assignment

Continuous BID dosing is planned. A cycle is 28 days of BID dosing. The starting dose of ruxolitinib tablets will be determined based on Baseline platelet count as follows:

• Subjects with Baseline platelet count $\geq$ 100,000/μL will begin dosing at 15 mg BID (three 5 mg tablets BID).

• Subjects with Baseline platelet count of 75,000/μL to <100,000/μL will begin dosing at 10 mg BID (two 5 mg tablets BID).

A standardized dosing paradigm will be used to determine dose adjustments for safety and efficacy so that each subject is titrated to their most appropriate dose. Doses will not exceed 25 mg BID.

9.2.2 Dose Increases

The initial dose of ruxolitinib will be determined by the Baseline platelet count value. After every 2 cycles (post cycle 2, 4, 6, etc.) of therapy, at the discretion of the

investigator, the dose may be increased in 5 mg BID increments, to a maximum dose of 25 mg BID for subjects who meet all three of the following conditions:

1. Inadequate efficacy is demonstrated by no response or stable disease relative to baseline.

2. Platelet count is ≥ 50,000/μL and platelet count has never been below 50,000/μL at a prior laboratory evaluation since Baseline.

3. Absolute neutrophil count levels have remained at or above 1000/μL since baseline.

4. No grade 4 anemia has been observed since baseline.

9.2.3 Dose Adjustments for Safety

Dosing must be held for toxicities according to Table 1. In order to provide sufficient data to make the dose adjustment decisions, it is recommended that hematology parameters be obtained at least weekly for platelet count < 50,000/μL or ANC < 1000/μL and at least two times weekly for platelet count < 25,000/μL or ANC < 500/μL. Dosing may be restarted or increased following recovery of toxicities to acceptable levels. Subjects with hematologic toxicities requiring dose holds for 3 weeks or longer will be removed from the study.

The objective for restarting or escalating after a reduction for safety is to find the highest safe dose of ruxolitinib for each subject, with increases in dose not more than in increments of 5 mg BID and not more often than every 2 cycles.

Table 1 Dose Modification Guidelines

NCI CTCAE Toxicity Grade	Action Required
Thrombocytopenia: Platelets < 35,000 /uL	• Hold (interrupt dosing) • Follow CBC at least twice weekly • If thrombocytopenia resolved to < Grade 2 (50,000/uL, may restart at next lower dose level) • Subjects in whom benefit is seen and who recover to at least 40,000/uL platelets, may be considered for retreatment with a dose reduction after consultation with the study PI on a case by case basis.
Grade 3 or 4 Neutropenia (ANC <1000/uL)	• Hold (interrupt dosing) • Follow CBC at least weekly • If neutropenia resolves to ≤ Grade 2, may restart at next lower dose level • Use of growth factors is permitted at the discretion of the investigator • Subjects in whom benefit is seen and who recover to at least grade 3 ANC, may be considered for retreatment with a dose reduction after consultation with the study PI on a case by case basis.
Grade 4 Anemia (life-threatening, urgent intervention indicated)	• Hold (interrupt dosing) • Follow CBC at least weekly • If anemia resolves to ≤ Grade 2, may restart at next lower dose level • Use of transfusions and/or erythropoietin is permitted at the discretion of the investigator
Related Grade 3 non-hematologic Toxicities	• Hold (interrupt dosing) • Manage toxicity according to institutional guidelines • If toxicity resolves to ≤ Grade 2 within 2 weeks, the subject may restart at next lower dose level or maintain per the investigator discretion
Related Grade 4 non-hematologic toxicities	• Discontinue dosing • Remove subject from the study

NOTE: Whether the dose interruption occurred because of neutropenia, thrombocytopenia, anemia or any combination of cytopenias, all cytopenias must be considered to determine the 'new' dose that the subject will receive. The lowest calculated dose will be used and in all cases the maximum dose must not exceed 5 mg BID LESS than the dose that resulted in the toxicity that required the dose interruption. Subjects with hematologic toxicities requiring dose holds for 3 weeks or longer will be removed from the study, unless permission is granted from the study PI on a case by case basis.

Table 2 Optional Dose Increase or Mandatory Reduction Levels for Subjects initiating treatment at 15 mg daily

+2 Dose Level	25 mg BID daily for 28 days per cycle
+1 Dose Level	20 mg BID daily for 28 days per cycle
Starting Dose	**15 mg BID daily for 28 days per cycle**
-1 Dose Level	10 mg BID daily for 28 days per cycle
-2 Dose Level	5 mg BID daily for 28 days per cycle

Table 3 Optional Dose Increase or Mandatory Reduction Levels for Subjects initiating treatment at 10 mg daily

+3 Dose Level	25 mg BID daily for 28 days per cycle
+2 Dose Level	20 mg BID daily for 28 days per cycle
+1 Dose Level	15 mg BID daily for 28 days per cycle
Starting Dose	**10 mg BID daily for 28 days per cycle**
-1 Dose Level	5 mg BID daily for 28 days per cycle

In summary, it is the intention to identify the optimal dose for each individual subject over the first several weeks of therapy as follows:

• Starting dose determined by baseline platelet count level.

• Optional dose increase after every 2 cycles if an objective response (PR or CR) was not achieved and no significant thrombocytopenia or neutropenia was observed.

• Frequent monitoring of hematology parameters, with mandatory dose decreases for toxicities.

9.3 Concomitant Medications / Measures

All concomitant medications and treatments must be recorded in the eCRF. Any prior medication received up to 30 days prior to randomization will be recorded in the eCRF. Concomitant treatments that are required to manage a subject's medical condition during the trial will also be recorded in the eCRF.

9.4 Restricted and / or Prohibited Medications and Therapies

9.4.1 Restricted Therapies

The following medications have restrictions on use or doses or require changes to the way in which study drug is administered during the study:

· Systemic corticosteroid doses should not be given except when necessary for non-lymphoma related conditions and must not exceed a daily dose of prednisolone 10mg/day;

· Aspirin in doses exceeding 150 mg per day is not permitted. Low dose aspirin (≤ 150 mg/day) and non steroidal anti-inflammatory agents (acetaminophen, ibuprofen) may be used;

· Potent systemic inhibitors of CYP3A4 metabolizing enzymes (ketoconazole, clarithromycin, itraconazole, nefazodone and telithromycin should not be given. When concomitant administration of a potent systemic inhibitor of CYP3A4 is required for subject management, and no alternative therapies are available, the sponsor must be contacted for discussion on a case by case basis. Subjects may not receive ruxolitinib concurrently with potent systemic inhibitors of CYP3A4 without prior approval by the sponsor, and in most cases ruxolitinib dose adjustments will be necessary.

· When concomitant administration of an anticoagulant/antiplatelet medication is required for subject management, the platelet count history, and any observations of thrombocytopenia during the study while on ruxolitinib should be considered.

· During the study, use of CYP3A4 inducers is discouraged, and investigators should consider alternative therapies wherever possible. No dose adjustment will be used when moderate CYP3A4 inducers are co-administered with study drug. Any concomitant use of moderate CYP3A4 inducers (rifabutin, carbamazepine, phenytoin) must be documented.

9.4.2 Prohibited Therapies

The following medications are prohibited during the study:

· Concurrent anti-cancer medications or therapies, including radiation therapy;

· Any prior or concomitant use of another JAK inhibitor.

· Any investigational medication other than the study drug. Use of such medications within 14 days or 6 half-lives, whichever is longer, prior to the first dose of study drug and during the study through the Follow-up Visit is prohibited.

9.5 Discontinuation of Study Medication(s)

In the event that any subject discontinues the study medication and subsequently the study prior to completion, regardless of reason, reasonable efforts should be made to have the subject return for an early termination visit and have the End-of-treatment procedures completed as described in Section 7.4.

The date the subject discontinued the study medication and the specific reason for discontinuation will be recorded in the eCRF. This will include reasons such as discontinued due to treatment failure or withdrawn due to adverse event. This information will be used to summarize the reasons for study discontinuation and treatment failure.

10.0 WITHDRAWAL OF STUDY SUBJECTS

10.1 Withdrawal Criteria and Procedures

10.1.1 Withdrawal of Subjects from Study

Subjects will be withdrawn from the study for any of the following reasons:

- In the Investigator's medical judgment, further participation would be injurious to the subject's health or well being
- Positive urine pregnancy test, confirmed by positive serum pregnancy (serum human chorionic gonadotropin) test results

- Consent is withdrawn
- Termination of the study by the Sponsor.
- Termination of the study by the local health authority or IRB/IEC

If a subject is withdrawn from the study:

- The study monitor or Sponsor must be notified.
- The reason(s) for withdrawal must be documented in the subject's medical record and eCRF.
- An End-of-treatment visit should be performed.
- All subjects must be followed for safety for at least 30 days after study treatment has ended, or until study drug related toxicities resolve, return to Baseline or are deemed irreversible, whichever is longer.

If a subject is non-compliant in the opinion of the Investigator, the Sponsor-Investigator should be consulted for instruction on handling the subject.

Criteria for discontinuation of study therapy are provided in Section 7.4.

10.2 Replacement Subjects

Non-evaluable subjects will be replaced, at the Sponsor-Investigator's discretion, to assure that a statistically appropriate number of subjects complete the study.

10.3 Protocol Deviations

Subjects must meet eligibility requirements to be enrolled in the study. Major deviations must be reported to the IRB/IEC in accordance with the IRB/IEC requirements. During the course of the study the monitor must notify the Sponsor-Investigator of subjects found not to have met eligibility criteria. The Sponsor-Investigator in collaboration with the Investigator will determine if the subject should be withdrawn from the study.

11.0 ADVERSE EVENTS

11.1 Definitions and Reporting

An Adverse Event for the purposes of this protocol is defined as the appearance of (or worsening of any pre-existing) undesirable sign(s), symptom(s), or medical condition(s) that occur after subject's signed informed consent has been obtained. Abnormal laboratory values or test results occurring after informed consent constitute Adverse Events only if they induce clinical signs or symptoms, are considered clinically significant, require therapy (e.g., hematologic abnormality that requires transfusion), or require changes in study medication(s).

Adverse Events that begin or worsen after informed consent should be recorded in the Adverse Events section of the eCRF. Conditions that were already present at the time of informed consent should be recorded in the Medical History section of the eCRF. Adverse Event monitoring should be continued for at least 30 days following the last dose of study treatment. Adverse Events (including lab abnormalities that constitute AEs) should be described using a diagnosis whenever possible, rather than individual underlying signs and symptoms. When a clear diagnosis cannot be identified, each sign or symptom should be reported as a separate Adverse Event.

Adverse Events will be assessed according to the Common Terminology Criteria for Adverse Events (CTCAE) version 4.0). If CTCAE grading does not exist for an Adverse Event, the severity of mild, moderate, severe, and life-threatening, or grades 1 - 4, will be used. CTCAE grade 5 (death) will not be used in this study; rather, information about deaths will be collected as an outcome of the event. The occurrence of Adverse Events should be sought by non-directive questioning of the subject during the screening process after signing informed consent and at each visit during the study. Adverse Events also may be detected when they are volunteered by the subject during the screening process or between visits, or through physical examination, laboratory test, or other assessments. As far as possible, each Adverse Event should be evaluated to determine:

- The severity grade (CTCAE grade 1-4)
- Reasonable possibility that AE is related to the study treatment: (no, yes)

- Start and end dates, unless unresolved at final exam
- Action taken with respect to study drug (none, dose adjusted, temporarily interrupted, permanently discontinued, unknown, not applicable)
- Outcome (not recovered/not resolved, recovered/resolved, recovering/resolving, recovered/resolved with sequelae, fatal, unknown)
- Whether it is serious, as per SAE definition provided in Section 11.3.1.

Unlike routine safety assessments, SAEs are monitored continuously and have special reporting requirements, see Section 11.3.2.

All Adverse Events should be treated appropriately. If a concomitant medication or non-drug therapy is given, this action should be recorded on the Adverse Event eCRF.

Once an Adverse Event is detected, it should be followed until its resolution or until it is judged to be permanent, and assessment should be made at each visit (or more frequently, if necessary) of any changes in severity, the suspected relationship to the study drug, the interventions required to treat it, and the outcome.

Disease progression should not be regarded or reported as an Adverse Event itself unless associated with a separate Adverse Event.

Participating sites must agree to comply by UNMC Data and Safety Monitoring Committee (DSMC) procedures. The DSMC will monitor the protocol on at least a quarterly basis and as requested. Sites must also comply with their local IRB requirements. All grade 3 and grade 4 adverse events and all Serious Adverse Events will be reported to the UNMC Eppley Cancer Center Data and Safety Monitoring Committee (DSMC) via the UNMC study coordinator. Sites will be provided with applicable policies and are required to enter all adverse events into the eCRF. Adverse Event information will be collected from the eCRF database and reported to the UNMC DSMC.

11.2 Laboratory Test Abnormalities

Definitions and Reporting

Laboratory abnormalities that constitute an Adverse Event in their own right (are considered clinically significant, induce clinical signs or symptoms, require concomitant therapy or require changes in study treatment), should be recorded on the Adverse Events

eCRF. Whenever possible, a diagnosis, rather than a symptom should be provided (e.g. anemia instead of low hemoglobin). Laboratory abnormalities that meet the criteria for Adverse Events should be followed until they have returned to normal or an adequate explanation of the abnormality is found. When an abnormal laboratory or test result corresponds to a sign/symptom of an already reported Adverse Event, it is not necessary to separately record the lab/test result as an additional event.

Laboratory abnormalities, that do not meet the definition of an Adverse Event, should not be reported as Adverse Events. A grade 3 or 4 event (severe) as per CTCAE does not automatically indicate a SAE unless it meets the definition of serious as defined below and/or as per investigator's discretion. A dose hold or medication for the lab abnormality may be required by the protocol in Section 8.6 and should not contribute to designation of a lab parameter abnormality as a SAE:

11.3 Serious Adverse Events

11.3.1 Definitions

Serious Adverse Event (SAE) is defined as one of the following:

- Is fatal or life-threatening (that is, immediate risk of dying)
- Results in persistent or significant disability/incapacity
- Constitutes a congenital anomaly/birth defect
- Is medically significant, i.e., defined as an event that jeopardizes the subject or may require medical or surgical intervention to prevent one of the outcomes listed above. Considered significant by the investigator as an important medical event (s) that may not result in death, be life-threatening, or require hospitalization but may be considered a serious adverse event (s) when, based upon appropriate medical judgment, they may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.
- Requires inpatient hospitalization or prolongation of existing hospitalization, unless hospitalization is for:

- Routine treatment or monitoring of the studied indication, not associated with any deterioration in condition (specify what this includes)
- Elective or pre-planned treatment for a pre-existing condition that is unrelated to the indication under study and has not worsened since signing the informed consent
- Treatment on an emergency outpatient basis for an event not fulfilling any of the definitions of a SAE given above and not resulting in hospital admission
- Social reasons and respite care in the absence of any deterioration in the subject's general condition
- Any SAEs that are expected due to the condition being treated, including if the SAE is a primary outcome measure, and whether there has been a clear agreement with regulators not to consider these as SAEs, provided the information is collected elsewhere

11.3.2 Reporting

To ensure subject safety, every SAE, regardless of suspected causality, occurring after the subject has provided informed consent and until at least 30 days after the subject has stopped study treatment must be reported to the Sponsor-Investigator or designee within 24 hours of learning of its occurrence. Any SAEs experienced after this at least 30 day period should only be reported to the Sponsor-Investigator or designee if the investigator suspects a causal relationship to the study drug. Recurrent episodes, complications, or progression of the initial SAE must be reported as follow-up to the original episode within 24 hours of the investigator receiving the follow-up information. An SAE occurring at a different time interval or otherwise considered completely unrelated to a previously reported one should be reported separately as a new event. Serious adverse event collection begins after the subject has signed informed consent and has received study drug. If a subject experiences a SAE after signing informed consent, but prior to receiving ruxolitinib, the event will NOT be collected unless the investigator feels the event may have been caused by a protocol procedure. Previously planned (prior to signing the informed consent document) surgeries should not be reported as SAEs unless the underlying medical condition has worsened during the course of the study.

Information about all SAEs is collected and recorded on the Serious Adverse Event Report Form. The investigator must assess and record the relationship of each SAE to the

study drug, complete the SAE Report Form in English, and send the completed, signed form by fax within 24 hours to the Sponsor-Investigator via a Sponsor-approved method. The investigator must assess if there is a Reasonable possibility that SAE is related to the study treatment: (no, yes)

All serious adverse events regardless of severity or relationship must be reported to the sponsor-investigator within 24 hours of the investigational staff's knowledge. Please send a completed SAE form to:

UNMC, ATTN: Susan Blumel		Sentryx
Fax: 402-559-8101	AND	Fax: 1-866-726-9234
Phone: 402-559-9183		Helpdesk: 1-866-278-6759, ext 250

The Sponsor-Investigator will follow UNMC IRB and DSMC safety reporting requirements. Participating sites must follow their local IRB and safety reporting requirements.

The original copy of the SAE Report Form and the fax confirmation sheet must be kept with the case report form documentation at the study site.

Follow-up information is sent to the same person to whom the original SAE Report Form was sent, using a new SAE Report Form stating that this is a follow-up to the previously reported SAE and giving the date of the original report. Each re-occurrence, complication, or progression of the original event should be reported as a follow-up to that event regardless of when it occurs. The follow-up information should describe whether the event has resolved or continues, if and how it was treated and whether the subject continued or withdrew from study participation.

If the SAE is not previously documented in the Investigator's Brochure for study medication (new occurrence) and is thought to be related to the study drug, a Sponsor's associate may urgently require further information from the investigator for Health Authority reporting.

The Sponsor-Investigator may need to issue an Investigator Notification (IN), to inform all investigators involved in any study with the same drug that this SAE has been reported. Suspected Unexpected Serious Adverse Reactions (SUSARs) will be collected and reported to the competent authorities and relevant ethics committees in accordance regulatory requirements.

11.4 Pregnancies

Pregnancy, in and of itself, is not regarded as an adverse event, unless there is suspicion that study medication may have interfered with the effectiveness of a contraceptive medication or method. The procedures that will be followed based on whether a pregnancy is confirmed by a positive serum test result are listed below:

- Investigator and subject must notify each other immediately
- Investigator must notify the Sponsor immediately
- Discontinue study medication immediately
- Perform the required End-of-treatment visit study evaluations
- Investigator must complete and submit the Pregnancy Initial and Follow-up report forms to the Sponsor

To ensure subject safety, each pregnancy in a subject during maternal or paternal exposures to study drug must be reported within 24 hours of learning of its occurrence. Data on fetal outcome and breast-feeding are collected for regulatory reporting and drug safety evaluation. The pregnancy should be followed up to determine outcome, including spontaneous or voluntary termination, details of the birth, and the presence or absence of any birth defects, congenital abnormalities, or maternal and/or newborn complications by following until the first well-baby visit. Pregnancy should be recorded on a Clinical Study Pregnancy Form and reported by the investigator to the Sponsor-Investigator. Pregnancy follow-up should be recorded on the same form and should include an assessment of the possible relationship to the study drug of any pregnancy outcome and follow-up to the first well-baby visit. **Any SAE experienced**

during pregnancy must be reported on the SAE Report Form and needs to be reported to the Sponsor.

11.5 Warnings and Precautions

No evidence available at the time of the approval of this study protocol indicated that special warnings or precautions were appropriate, other than those noted in the provided Investigator Brochure. Additional safety information collected between IB updates will be communicated in the form of Investigator Notifications. This information will be included in the subject informed consent and should be discussed with the subject during the study as needed.

12.0 STATISTICS

12.1 Study Populations

The populations to be analyzed include the following:

Intent-to-treat (ITT) population - Subjects enrolled in the study.

Evaluable population – Subjects who had completed 2 cycles of therapy with response assessments or progressed before the evaluation of the primary endpoint.

Safety evaluable population. - Subjects who received at least one dose of study medication.

12.2 Efficacy Analysis

Primary efficacy analyses will be based on both the ITT and evaluable populations. Secondary efficacy analyses will be based on ITT population only.

12.2.1 Primary Efficacy Analyses

Primary endpoint

The primary objective of the study is to evaluate subject overall response rate (ORR) after 6 cycles for each of the subject cohorts. The description provided in this section includes the primary analyses for the primary objective.

Statistical hypothesis and sample size justification

Subjects meeting the stated eligibility requirements will be enrolled onto the study. Subjects will be grouped into cohorts 1) DLBCL ABC 2) DLBCL GCB 3) PTCL and 4) OTHER SUBTYPES (subjects who are not able to be placed into cohorts 1, 2, and 3), based upon a central determination of immunohistochemistry and gene-expression profile. A target sample size of 30 is planned for cohorts 1, 2, and 3. Cohorts 1, 2, and 3 will have a minimum of 10 and a maximum of 30 subjects. There is no minimum or maximum requirement for Cohort 4, since there is a very low number of subjects anticipated for this cohort. (Maximum in all cohorts is 90 subjects).

These sample sizes are based on a 3-stage design of alpha=0.07 with power of 77% (beta=0.23) to test the null hypothesis that ORR after 6 cycles will be ≤ 10% (not considered clinically compelling) versus the alternative hypothesis that ORR will be ≥ 25% (considered clinically meaningful in this patient population) within each cohort. Based on the simulation study, to maintain the above type I and II errors, two interim analyses for futility will be performed for each cohort separately according to the following stopping rule:

1) The first interim analysis will occur when 10 subjects have been enrolled in the cohort. If there are any responders (CR+PR) observed among the 10 subjects, enrollment will continue; otherwise, enrollment will be suspended and these 10 subjects will be followed to further assess their responses. During this follow-up period, as soon as one response is observed, enrollment will resume. If no response is observed after the 10 subjects have been fully assessed after 6 cycles, the trial will be terminated for futility.

2) The second interim analysis will occur when 20 subjects have been enrolled in the cohort. If there are 3 or more responders (CR+PR) observed among the 20 subjects, enrollment will continue; otherwise, enrollment will be suspended and these 20 subjects will be followed to further assess their responses. During this follow-up, as soon as 3 or more responders are observed, enrollment will resume. If there are < 3 responders observed after the 20 subjects have been fully assessed, the trial will be terminated for futility.

3) At the end of the trial, if there are ≥ 6 responders (CR+PR) observed among the 30 subjects, the null hypothesis will be rejected and the drug is valuable to pursue further research; otherwise we fail to reject the null hypothesis and futility of the drug will be claimed.

12.2.2 Secondary Efficacy Analyses

Secondary endpoints

Safety endpoint

All adverse events recorded during the study will be summarized by each subject cohort. The incidence of treatment-emergent adverse events (new or worsening from baseline) will be summarized by severity and type of adverse event. Listings of deaths, SAEs, and AEs leading to early termination of study treatment or premature withdrawal from study will also be provided.

Duration of response (DOR)

Duration of response is defined as the interval from the date of complete or partial remission documented to date of recurrence/progression, death due to any cause, or lost to follow-up. The Kaplan-Meier method will be used to estimate the median duration of response and its 95% confidence interval (CI).

Progression-free survival (PFS)

PFS is defined as the time from the date of start of treatment to the date of event defined as the first documented progression or death due to any cause. If a subject has not had an event, progression-free survival is censored at the date of last adequate assessment or start of another therapy, whichever occurs first. The Kaplan-Meier method will be used to estimate PFS and its 95% CI.

Overall survival (OS)

OS is defined as the time from date of start of treatment to date of death due to any cause. If a subject is not known to have died, survival will be censored at the date of last contact. The Kaplan-Meier method will be used to estimate the median OS time and its 95% CI.

12.2.3 Exploratory Analyses

Exploratory endpoints

The exploratory aspect of the study is to explore relationship between responses to oral ruxolitinib and alterations in GEP signatures as well as biomarker immunophenotypic changes related to JAK2/STAT3, NF-κB, BCR, PI3K/AKT, and mTOR pathways, and to evaluate potential effect of oral ruxolitinib exposure on JAK2/STAT3 pathway inhibition in serial tumor samples. Descriptive statistics will be used. Mean, standard deviation (STD), median and range will be reported for markers pre- and post-treatment as well as for the marker changes between pre- and post-treatment. Paired t-test will be used to evaluate the marker changes between pre- and post-treatment. Spearman or Pearson correlation coefficients will be calculated to evaluate the correlations among markers. Logistic regression will be utilized to assess the effects of markers at baseline on response to therapy.

12.3 Safety Analysis

12.3.1 Adverse Events

Adverse events will be tabulated by the Medical Dictionary for Regulatory Activities (MedDRA®) preferred term and by body system. Severity of adverse events will be based on the scale as indicated in Section 11.2.2.

The subset of adverse events that are considered by the Investigator to have a possible or probable relationship to study medication will be considered to be treatment-related adverse events. If the Investigator does not specify the relationship of the adverse event to study medication, the adverse event will be considered to be treatment-related. The incidence of adverse events and treatment-related adverse events will be tabulated.

13.0 DATA MANAGEMENT

13.1 Data Collection

The Investigator will be provided with a eCRF for each subject.

Entries made in the eCRF must be verifiable against source documents, or, as directed by the Sponsor, directly entered into the eCRF, in which case the entry in the eCRF will be considered source documentation. Data reported in the eCRF derived from source documents should be consistent with the source documents. The investigator must explain any discrepancies between the eCRF and the source documents.
The Investigator will be responsible for reviewing all data and eCRF entries in each subject's eCRF, verifying the information is true and correct.

13.2 Data Management

Data management will be performed using electronic case report forms (eCRFs).

All eCRF data will be entered into a validated database.

All data entry, verification and validation will be performed in accordance with the current standard operating procedures of the Data Management Department at the sponsor or its designee. The sponsor will authorize the database for lock once all data queries are complete, all study data are considered clean, and all defined procedures completed by the investigator.

14.0 STUDY ADMINISTRATION

14.1 Access to Source Documents

14.1.1 Scientific Review Committee Audit Procedures

The UNMC Eppley Cancer Center Scientific Review Committee will review this protocol on at least an annual basis. This study will not be audited by the UNMC Audit Committee, however monitoring updates will be provided to the Audit Committee as required.

14.1.2 Monitoring Visits

Monitoring visits provide the Sponsor-Investigator with the opportunity to:

- Evaluate the progress of the study
- Verify the accuracy and completeness of eCRFs

- Assure that all protocol requirements, applicable laws and/or regulations, and Investigator's obligations are being fulfilled
- Resolve any inconsistencies in the study records.

The Investigator must allow study monitors to periodically review, all eCRFs and office, hospital, and laboratory records supporting the participation of each subject in the study, at mutually convenient times, during the study and after the study has been completed. The eCRFs and other documentation supporting the study must be kept up-to-date by the Investigator and the research staff at the investigative site. These study materials must be available for review by the study monitor, and/or other qualified representatives of the Sponsor, at each monitoring visit.

The study monitor will review the various records of the study (eCRFs, subject medical and laboratory records, and other pertinent data). The study monitor will verify the eCRF data against original source documentation for accuracy and completeness. The study monitor will identify data discrepancies and collaborate with

the Investigator and research staff to resolve the discrepancies in a timely manner. Protocol deviations will also be identified and recorded on a "Protocol Deviation Log." The study monitor will follow an "Issue Escalation" plan in order to ensure each issue identified during a monitoring visit is appropriately documented, reported, and resolved in a timely manner in accordance with the plan's requirements.

14.2 Statement of Good Clinical Practices

This trial will be conducted in adherence to the study protocol, Good Clinical Practices as defined in Title 21 of the US Code of Federal Regulations Parts 50, 54 56, 312 and Part 11 as well as ICH GCP consolidated guidelines (E6) and applicable regulatory requirements. http://www.fda.gov/cder/guidance/index.htm

14.3 Protocol Adherence

Each Investigator must adhere to the protocol as described in this document and agree that deviations to the protocol, with the exception of medical emergencies, must be discussed and approved by the Sponsor-Investigator prior to seeking approval from the IRB/IEC. Each Investigator is responsible for enrolling subjects who have met the protocol inclusion and exclusion criteria or must have obtained prior documented approval from the Sponsor prior to enrollment in the study. The IRB/IEC that granted original approval, or the IRB/IEC currently responsible for overseeing the conduct of the study, must be notified of all changes in and deviations from the protocol that may increase risk to the subject, and/or that may adversely affect the rights of the subject or validity of the investigation. The Investigator must send a copy of the approval letter from the IRB/IEC to the Sponsor or CRO and retain the original in the site study regulatory file.

14.4 Study Termination

Both the Sponsor-Investigator and the Investigator reserve the right to terminate the study, according to the terms specified in the study contract. The Investigator is to notify the IRB/IEC in writing of the study's completion or early termination, and send a copy of

the notification to the Sponsor or CRO and retain one copy for the site study regulatory file.

14.5 Financial Disclosure

All clinical Investigators participating in clinical studies subject to FDA Regulation Title 21 Code of Federal Regulations (CFR) Part 54 – Financial Disclosure by Clinical Investigators are required prior to study initiation to submit a completed Clinical Investigator Financial Certification/Disclosure Request Form that sufficiently details any financial interests and arrangements that apply. For the purpose of this regulation, clinical Investigator is defined as any Investigator or Sub-Investigator who is directly involved in the treatment or evaluation of research subjects, including the spouse and any dependent child of the Investigator, but not that of any Sub-Investigators. These requirements apply to both US and foreign clinical Investigators conducting covered clinical studies.

Any new Investigators or Sub-Investigators added to the covered clinical study during its conduct must also submit a completed Clinical Investigator Financial Disclosure Request Form. At the conclusion of the covered clinical study, the Investigators will be reminded of their obligation to report to the Sponsor/designee any changes to the financial disclosure information previously reported. The clinical Investigators will also be reminded that they must report any changes in their financial information regarding significant equity interests and significant payments for a period of 1 year after completion of their participation in the covered clinical study.

14.6 Quality Control and Assurance

14.6.1 Sponsor Audits

At some point during the study, individuals from the Sponsor's Quality Assurance department and/or their authorized representative may visit the Investigator's site to conduct an audit of the study. The purpose of this visit will be to determine the Investigator's adherence to the protocol, applicable regulations, and the Sponsor's

procedures, in addition to assessing the accuracy of the study data. Prior to initiating this audit, the Investigator will be contacted by the Sponsor to arrange a convenient time for this visit. The Investigator and staff are expected to cooperate with the auditors and allow access to all subject records supporting the eCRFs and other study-related documents.

14.6.2 Inspection by Regulatory Authorities

At some point during the investigational product's development program, a regulatory authority may visit the Investigator to conduct an inspection of the study and the site. The Investigator and staff are expected to cooperate with the inspectors and allow access to all source documents supporting the eCRFs and other study-related documents. The Investigator must immediately notify the Sponsor when contacted by any regulatory authority for purposes of conducting an inspection.

15.0 ETHICS

15.1 Institutional Review Board or Independent Ethics Committee

It is the responsibility of the Investigator to assure that all aspects of the ethics review are conducted in accordance with the Declaration of Helsinki as described in the International Conference on Harmonisation (ICH) E6: Guideline for Good Clinical Practice (GCP), and/or local laws, whichever provides the greatest level of protection for the study participants [14]. The protocol and any information supplied to the subject to obtain informed consent, including written informed consent form(s), subject recruitment procedures (eg, advertisements), and written information to be provided to subjects (information leaflets), must be reviewed and approved by a qualified IRB/IEC prior to enrollment of participants in the study. Prior to initiation of the study, the Sponsor-Investigator must receive documentation of the IRB/IEC approval, which specifically identifies the study/protocol, and a list of the committee members.

Amendments to the protocol and revisions to the informed consent must also be submitted to and, if required, approved by the IRB/IEC.

Investigators must submit progress reports to the IRB/IEC in accordance with the IRB/IEC requirements. Annual re-approval of the study must be obtained. Copies of progress reports and Annual re-approvals must be sent to the Sponsor-Investigator.

When the Sponsor-Investigator provides the Investigator with a safety report, the Investigator must promptly forward a copy to the IRB/IEC, according to site IRB/IEC reporting requirements.

After completion or termination of the study, the Investigator must submit a final report to the IRB/IEC and to the Sponsor-Investigator.

The Investigator, as part of the records retention requirements for the study, must maintain documentation of all submissions, correspondence, and approvals to and from the IRB/IEC.

Each clinical Investigator is responsible to conduct the study in accordance with the protocol, all applicable laws, regulations, and GCP according to ICH guidelines.

15.2 Informed Consent

Preparation of the consent form is the responsibility of the Investigator and the Sponsor-Investigator and must include all elements required by the ICH, GCP, and applicable regulatory requirements, and must adhere to GCP and to the ethical principles that have their origin in the Declaration of Helsinki.

A template will be provided by the Sponsor-Investigator. The Sponsor-Investigator or designee must review and approve all changes to site-specific Informed Consent forms.

The consent form must include a statement that the Sponsor-Investigator or designee and regulatory authorities have direct access to subject records. Prior to the beginning of the

study, the Investigator must have the IRB/IEC's written approval/favorable opinion of the written informed consent form and any other information to be provided to the subjects. Before being enrolled in the clinical study, subjects must consent to participate after the nature, scope, and possible consequences of the study have been explained in a form understandable to them.

An informed consent document that includes both information about the study and the consent form will be prepared and given to the subject. This document will contain all the elements required by the ICH E6 Guideline for Good Clinical Practice and any additional elements required by local regulations. The document must be in a language understandable to the subject and must specify who informed the subject. Where required by local law, the person who informs the subject must be a physician.
A copy of the signed consent document must be given to the subject. The original signed consent document will be retained by the Investigator.

The Investigator will not undertake any measures specifically required only for the clinical study until valid consent has been obtained.

The Investigator must inform the subject's primary physician about the subject's participation in the trial if the subject has a primary physician and if the subject agrees to the primary physician being informed.

15.2.1 Data Privacy

Applicable data privacy laws and regulations must be adhered to. The Investigator and the Sponsor are responsible for ensuring that sensitive information is handled in accordance with local requirements (eg, HIPAA). Appropriate consent and authorizations for use and disclosure and/or transfer (if applicable) of protected information must be obtained.

16.0 RECORD KEEPING / RETENTION OF RECORDS

The Investigator must ensure that all records pertaining to the conduct of the clinical study, informed consent forms, drug accountability records, source documents, and other study documentation are adequately maintained for a period of 2 years after the last approval of a marketing application in an ICH region and until there are no pending or contemplated marketing applications in an ICH region or at least 2 years have elapsed since the formal discontinuation of clinical development of the investigational product.

The Investigator must not destroy any records associated with the study without receiving approval from the Sponsor-Investigator. The Investigator must notify the Sponsor-Investigator in the event of accidental loss or destruction of any study records. If the Investigator leaves the institution where the study was conducted, the Sponsor-Investigator must be contacted to arrange alternative record storage options.

Whenever possible, an original recording of an observation must be retained as the source document. However, a photocopy of a record is acceptable provided it is legible and is a verified copy of the original document.

All eCRF data entered by the site (including audit trail), as well as computer hardware and software (for accessing the data), will be maintained or made available at the site in compliance with applicable record retention regulations. The Sponsor-Investigator will retain the original eCRF data and audit trail.

16.1 Confidentiality

Subject names will not be supplied to the Sponsor. Only the subject number and subject initials will be recorded in the eCRF, and if the subject name appears on any other document (eg, laboratory report), it must be obliterated on the copy of the document to be supplied to the Sponsor-Investigator. Study findings stored on a computer will be stored in accordance with local data protection laws. The subjects will be informed that representatives of the Sponsor-Investigator, IRB/IEC, or regulatory authorities may

inspect their medical records to verify the information collected, and that all personal information made available for inspection will be handled in strictest confidence and in accordance with local data protection laws.

17.0 REFERENCES

1. Jemal, A., et al., *Cancer statistics, 2010.* CA Cancer J Clin, 2010. **60**(5): p. 277-300.

2. World Health Organization, *WHO Classification of Tumours of Haematopoietic and Lymphoid Tissues.* Geneva, Switzerland: WHO Press, 2008.

3. Rosenwald, A., et al., *The use of molecular profiling to predict survival after chemotherapy for diffuse large-B-cell lymphoma.* N Engl J Med, 2002. **346**(25): p. 1937-47.

4. Mottok, A., et al., *Inactivating SOCS1 mutations are caused by aberrant somatic hypermutation and restricted to a subset of B-cell lymphoma entities.* Blood, 2009. **114**(20): p. 4503-6.

5. Ritz, O., et al., *Recurrent mutations of the STAT6 DNA binding domain in primary mediastinal B-cell lymphoma.* Blood, 2009. **114**(6): p. 1236-42.

6. Lam, L.T., et al., *Cooperative signaling through the signal transducer and activator of transcription 3 and nuclear factor-{kappa}B pathways in subtypes of diffuse large B-cell lymphoma.* Blood, 2008. **111**(7): p. 3701-13.

7. Chiarle, R., et al., *Stat3 is required for ALK-mediated lymphomagenesis and provides a possible therapeutic target.* Nat Med, 2005. **11**(6): p. 623-9.

8. van Kester, M.S., et al., *Cucurbitacin I inhibits Stat3 and induces apoptosis in Sezary cells.* J Invest Dermatol, 2008. **128**(7): p. 1691-5.

9. Zhang, C., et al., *Curcumin selectively induces apoptosis in cutaneous T-cell lymphoma cell lines and patients' PBMCs: potential role for STAT-3 and NF-kappaB signaling.* J Invest Dermatol, 2010. **130**(8): p. 2110-9.

10. Verma, N.K., et al., *STAT3 knockdown by siRNA induces apoptosis in human cutaneous T-cell lymphoma line Hut78 via downregulation of Bcl-xL.* Cell Mol Biol Lett, 2010. **15**(2): p. 342-55.

11. Burger, R., et al., *Janus kinase inhibitor INCB20 has antiproliferative and apoptotic effects on human myeloma cells in vitro and in vivo.* Mol Cancer Ther, 2009. **8**(1): p. 26-35.

12. Ding, B.B., et al., *Constitutively activated STAT3 promotes cell proliferation and survival in the activated B-cell subtype of diffuse large B-cell lymphomas.* Blood, 2008. **111**(3): p. 1515-23.

13. Verstovsek, S., et al., *Safety and efficacy of Ruxolitinib, a JAK1 and JAK2 inhibitor, in myelofibrosis.* N Engl J Med, 2010. **363**(12): p. 1117-27.

14. WORLD MEDICAL ASSOCIATION DECLARATION OF HELSINKI Ethical Principles for Medical Research Involving Human Subjects. Adopted by the 18th WMA General Assembly, Helsinki, Finland, June 1964, and amended by the 29th WMA General Assembly, Tokyo, Japan, October 1975, 35th WMA General Assembly, Venice, Italy, October 1983, 41st WMA General Assembly, Hong Kong, September 1989, 48th WMA General Assembly, Somerset West, Republic of South Africa, October 1996, and the 52nd WMA General Assembly, Edinburgh, Scotland, October 2000. Note of Clarification on Paragraph 29 added by the WMA General Assembly, Washington 2002. Note of Clarification on Paragraph 30 added by the WMA General Assembly, Tokyo 2004. Available at http://www.wma.net/e/policy/b3.htm.

18.0 INVESTIGATOR'S SIGNATURE PAGE

A Phase 2 Multicenter, investigator initiated study of Oral Ruxolitinib Phosphate for the Treatment of Relapsed or Refractory Diffuse Large B-cell and Peripheral T-cell Non-Hodgkin Lymphoma

Protocol IND number : **112445**

Protocol Version/Date: **Amendment 7, version date 29 February 2016**

I have read, understand, and agree to follow the attached Protocol.

_____ _____

(Investigator Signature) (Date)

_____

(Printed Name)

19.0 APPENDICES

List of Appendices

1. **RESPONSE CRITERIA**
2. **LABORATORY METHODS**
3. **NCI CTC VERSION 4.0**

APPENDIX 1 – RESPONSE CRITERIA

Adapted from *Journal of Clinical Oncology*, Vol 25, No 5 (February 10), 2007: pp. 579-586

Revised Response Criteria for Malignant Lymphoma by Bruce D. Cheson,

CR
The designation of CR requires the following

1. Complete disappearance of all detectable clinical evidence of disease and disease-related symptoms if present before therapy.

2a. Typically FDG-avid lymphoma: in patients with no pretreatment PET scan or when the PET scan was positive before therapy, a post-treatment residual mass of any size is permitted as long as it is PET negative.

2b. Variably FDG-avid lymphomas/FDG avidity unknown: in patients without a pretreatment PET scan, or if a pretreatment PET scan was negative, all lymph nodes and nodal masses must have regressed on CT to normal size (≤ 1.5 cm in their greatest transverse diameter for nodes > 1.5 cm before therapy). Previously involved nodes that were 1.1 to 1.5 cm in their long axis and more than 1.0 cm in their short axis before treatment must have decreased to ≤ 1.0 cm in their short axis after treatment.

3. The spleen and/or liver, if considered enlarged before therapy on the basis of a physical examination or CT scan, should not be palpable on physical examination and should be considered normal size by imaging studies, and nodules related to lymphoma should disappear. However, determination of splenic involvement is not always reliable because a spleen considered normal in size may still contain lymphoma, whereas an enlarged spleen may reflect variations in anatomy, blood volume, the use of hematopoietic growth factors, or causes other than lymphoma.

4. If the bone marrow was involved by lymphoma before treatment, the infiltrate must have cleared on repeat bone marrow biopsy. The biopsy sample on which this determination is made must be adequate (with a goal of > 20 mm unilateral core). If the sample is indeterminate by morphology, it should be negative by immunohistochemistry. A sample that is negative by immunohistochemistry but that demonstrates a small population of clonal lymphocytes by flow cytometry will be considered a CR until data become available demonstrating a clear difference in patient outcome.

CRu
The use of the above definition for CR and that below for PR eliminates the category of CRu.

PR
The designation of PR requires all of the following:

1. At least a 50% decrease in sum of the product of the diameters (SPD) of up to six of the largest dominant nodes or nodal masses. These nodes or masses should be selected according to all of the following: they should be clearly measurable in at least 2 perpendicular dimensions; if possible they should be from disparate regions of the body; and they should include mediastinal and retroperitoneal areas of disease whenever these sites are involved.
2. No increase should be observed in the size of other nodes, liver, or spleen.
3. Splenic and hepatic nodules must regress by ≥ 50% in their SPD or, for single nodules, in the greatest transverse diameter.
4. With the exception of splenic and hepatic nodules, involvement of other organs is usually assessable and no measurable disease should be present.
5. Bone marrow assessment is irrelevant for determination of a PR if the sample was positive before treatment. However, if positive, the cell type should be specified (eg, large-cell lymphoma or small neoplastic B cells). Patients who achieve a CR by the above criteria, but who have persistent morphologic bone marrow involvement will be considered partial responders.
   When the bone marrow was involved before therapy and a clinical CR was achieved, but with no bone marrow assessment after treatment, patients should be considered partial responders.
6. No new sites of disease should be observed.
7. Typically FDG-avid lymphoma: for patients with no pretreatment PET scan or if the PET scan was positive before therapy, the post-treatment PET should be positive in at least one previously involved site.
8. Variably FDG-avid lymphomas/FDG-avidity unknown: for patients without a pretreatment PET scan, or if a pretreatment PET scan was negative, CT criteria should be used.

Stable Disease
Stable disease (SD) is defined as the following:

1. A patient is considered to have SD when he or she fails to attain the criteria needed for a CR or PR, but does not fulfill those for progressive disease (see Relapsed Disease [after CR]/Progressive Disease [after PR, SD]).
2. Typically FGD-avid lymphomas: the PET should be positive at prior sites of disease with no new areas of involvement on the post-treatment CT or PET.
3. Variably FDG-avid lymphomas/FDG-avidity unknown: for patients without a pretreatment PET scan or if the pretreatment PET was negative, there must be no change in the size of the previous lesions on the post-treatment CT scan.

Relapsed Disease (after CR)/Progressive Disease (after PR, SD)
Lymph nodes should be considered abnormal if the long axis is more than 1.5 cm regardless of the short axis. If a lymph node has a long axis of 1.1 to 1.5 cm, it should

only be considered abnormal if its short axis is more than 1.0. Lymph nodes ≤ 1.0 x ≤ 1.0 cm will not be considered as abnormal for relapse or progressive disease.

1. Appearance of any new lesion more than 1.5 cm in any axis during or at the end of therapy, even if other lesions are decreasing in size. Increased FDG uptake in a previously unaffected site should only be considered relapsed or progressive disease after confirmation with other modalities. In patients with no prior history of pulmonary lymphoma, new lung nodules identified by CT are mostly benign. Thus, a therapeutic decision should not be made solely on the basis of the PET without histologic confirmation.
2. At least a 50% increase from nadir in the SPD of any previously involved nodes, or in a single involved node, or the size of other lesions (eg, splenic or hepatic nodules). To be considered progressive disease, a lymph node with a diameter of the short axis of less than 1.0 cm must increase by ≥ 50% and to a size of 1.5 x 1.5 cm or more than 1.5 cm in the long axis.
3. At least a 50% increase in the longest diameter of any single previously identified node more than 1 cm in its short axis.
4. Lesions should be PET positive if observed in a typical FDG-avid lymphoma or the lesion was PET positive before therapy unless the lesion is too small to be detected with current PET systems (< 1.5 cm in its long axis by CT).

Measurable extranodal disease should be assessed in a manner similar to that for nodal disease. For these recommendations, the spleen is considered nodal disease. Disease that is only assessable (eg, pleural effusions, bone lesions) will be recorded as present or absent only, unless, while an abnormality is still noted by imaging studies or physical examination, it is found to be histologically negative.

Appendix 2 – Laboratory methods

SCHEDULE OF LABORATORY ASSESSMENTS

	Pre-treatment	Cycle 1 Day 1	Cycle 1 Day 8	Cycle 1 Day 15	Cycle 1 Day 22	Cycle 2 Day 1	Post Cycle 2	Disease Progression
Archived Tumor slides (required)	X							
Fresh or Archived Tumor Biopsy (required)	X							
Fresh Tumor Biopsy (optional)							X	X
Blood for cytokine profiling [a]	X	X	X	X	X	X		
Blood for C-Reactive Protein (CRP) [b]	X					X		X

a: Collect blood for cytokine profiling within 7 days prior to first dose of ruxolitinib and at 3 hours post first dose on Cycle 1 Day 1. Weekly samples will be collected prior to the morning dose of ruxolitinib on Cycle 1, days 8, 15, 22, and on Cycle 2, day 1. Analysis of C-Reactive Protein (CRP) will be done on samples collected for cytokine profiling at the pre-treatment and Cycle 2 Day 1 timepoints. .

b: C-Reactive Protein (CRP) will be tested from cytokine blood collection at Pre-treatment and at Cycle 2, day 1 and no additional blood is required for these two timepoints. An additional separate sample for CRP will be collected at time of relapse or progression.

I. TUMOR TISSUE REQUIREMENTS

A. ARCHIVED TUMOR TISSUE

Archived tumor tissue obtained at the time of initial diagnosis of DLBC or PTCL is required for all subjects for RNA sequencing and comparison of gene expression between initial and current tumor tissue. A minimum of 5 unstained 10 µm slides will be submitted to the central lab.

Complete and fax the completed Tumor Tissue Requisition and Shipment Notification Form on the day of shipment and include a copy of the form in each shipment.

Include a copy of the site tumor pathology report in the shipment. The report must have subject identifiers removed and the patient study ID and study protocol number must be written at the top of the report.

Ship to:

Dr. Kai Fu
University of Nebraska Medical Center
601 South Saddle Creek Rd.
Omaha, NE 68105- 31356
402-559-3135
Phone: 402-559-7526
Fax: 402-559-6018
Pager: 402-888-2528

B. PRE-TREATMENT BIOPSY

A pre-treatment biopsy of tumor tissue collected since the most recent prior therapy is required for all subjects. Either excisional tumor biopsy or two 14 gauge (or larger) cores will be collected and submitted to the Central lab. The central lab will perform morphologic review and gene expression profiling. No reports from the central lab will be provided back to the investigator and site but the tumor content and degree of necrosis and degeneration will be recorded in the database. If a formal pathology review is required for clinical use, please collect **additional tissue** for review by your local pathologist.

Preparing tissue for transport to the Central Labs:

1. Collect either an excisional biopsy or two 14 gauge or larger cores of tumor tissue. If cores are collected, each core must be at least 2 cm in length to ensure enough tissue for evaluation. If long cores cannot be obtained, additional cores should be collected to provide an equivalent amount of tissues. Transport tissue in saline soaked non-stick pads to the hematopathologist immediately (within 15 minutes).

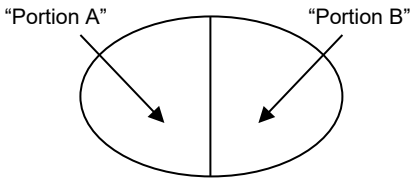
2. The tissue will be examined by a hematopathologist within 60 minutes of tissue collection, and each of the core biopsies or the excisional biopsy is divided into two halves. Label the halves of each biopsy "A" and "B". Label according to the illustration:

If two cores are collected, label as follows:

"Biopsy 1, portion A"	**"Biopsy 1, portion B"**

"Biopsy 2, portion A"	**"Biopsy 2, portion B"**

If excisional tissue is collected, label as follows:



*If the excisional tissue is large it can be divided into "Biopsy 1" and "Biopsy 2" before separation to portion "A" and "B".

3. All portion "A" halves are to be processed individually and fixed in neutral buffered formalin (If two core biopsies are collected, two separate portion A samples will be processed and shipped). Tissue must be processed into a paraffin block for submission to the Central Pathologist. Whole block(s) must be submitted, and partially cut blocks will not be acceptable as insufficient tissues may be left for subsequent studies. Label each block as illustrated above and include the study number, subject number, and date of biopsy. Ship to the Central Lab at ambient temperature.

4. All portion "B" halves are to be snap-frozen individually in separate plastic containers and stored at -80^{o}C until shipment to the Central Lab. Label each specimen as illustrated above and include the study number, subject number, and date of biopsy. Ship to the Central Lab frozen in sufficient dry ice to ensure that the tissue will remain frozen for over 24 hours **(See SOP for shipment**).

5. Complete and fax the completed Tumor Tissue Requisition and Shipment Notification Form on the day of shipment and include a copy of the form in each shipment.

6. Include a copy of the site tumor pathology report in the shipment. If the confirmation of relapse biopsy is obtained concurrent with this research biopsy, please provide the report by fax to Dr. Fu at 402-559-6018 when it becomes available. The report must have subject identifiers removed and the patient study ID and study protocol number must be written at the top of the report.

6. Ship to the Central Lab on day of collection or as soon as possible. If a delayed shipment is necessary please contact the Sponsor for approval. Please ensure that

shipments can be delivered within 24 hours after shipment (do not ship on Thursday and Fridays, or within 2 days before holidays).

Ship to:

Dr. Kai Fu
University of Nebraska Medical Center
601 South Saddle Creek Rd.
Omaha, NE 68106
402-559-3135
Phone: 402-559-7526
Fax: 402-559-6018
Pager: 402-888-2528

7. Should there not be sufficient fresh tissue the Central lab to complete the gene profiling requirements, then archive tissue from a previous relapse biopsy will be requested. If both the fresh and relapse tissue are not available, then analysis will be perform using an archived block from the initial diagnostic biopsy specimen. Submission of this tissue would be handled as indicated above for the Archived tissue submissions.

C. OPTIONAL ON-STUDY TUMOR BIOPSIES

Two additional tumor biopsies will be collected for those subjects who provide consent. A biopsy will be collected On-therapy sometime after the completion of 2 cycles of therapy, and/or a biopsy will be collected at 6 weeks post-treatment in patients with disease progression.. Samples will be collected and shipped according to the instructions provided for the Pretreatment Tumor Tissue Biopsy.

II. BLOOD SAMPLES

A. CYTOKINE Plasma Sample Blood Collection and preparation (includes C-reactive protein analysis at pretreatment and at cycle 2, day 1)

1. Collect 7cc to 10cc whole blood into EDTA tubes (Purple top). * At Cycle 1, day 1 predose timepoint only: Make two to three **blood smear slides** and air dry, and save them at room temperature until shipment to the central lab.

2. Process within 2 hours of collection. Cells are removed from plasma by centrifugation for 10 minutes at 1,000-2,000 rpm using a refrigerated centrifuge. Centrifugation for 15 minutes at 2,000 rpm depletes platelets in the plasma sample. The resulting supernatant is designated plasma.

3. Following centrifugation, immediately transfer the plasma apportioned into two clean polypropylene aliquot tubes using a pipette. The samples should be maintained at 2-8°C while handling. Store and at –20°C, or lower until shipment to the central lab. It is important to avoid freeze/thaw cycles. Samples which are hemolyzed, icteric, or lipemic can invalidate certain tests.

4. Transfer the cell pellet to a 50 ml Falcon tube which contains 40 ml ice cold ddH2O. Mix it gently by inverting tube for several times to lyse the red cells (about 10 sec). Then divide the mixture into two 50mL Falcon tubes in equal volume, add 25 ml 2 X PBS in each tube and mix it gently by inverting the tube several times. Centrifuge the tubes at 1000 rpm (250g) for 10 min, remove supernatant and re-suspended the cell pellets in 500µL 1XPBS of each. Transfer both into a clean polypropylene aliquot tube, centrifuge at 2000g for 5 min. Discard the supernatant, and save the cell pellet in -80°C until shipment to the central lab. Label it with **whole cell pellet.**

5. Send blood smear, plasma sample and whole cell pellet to the central lab according to the shipment instruction below.

B. CYTOKINE Peripheral Blood Mononuclear Cell (PBMC) Sample Collection and preparation

1. Collect 10cc whole blood into EDTA tubes (**purple** top). Process within 2 hours of collection according to Ficoll-Hypaque method:

2. Place fresh heparinized blood into 15- or 50-ml conical centrifuge tubes. Using a sterile pipet, add an equal volume of room-temperature 1 × PBS. Mix well.

3. Pellet the leukocyte/RBC fraction by centrifuging the cells 15 min at 200 × g, room temperature. The platelet cell fraction will remain in the upper suspension. Using a sterile pipet, remove the supernatant suspension containing this platelet cell fraction. The blood should immediately be gently mixed, and should be examined when it reaches the laboratory for small blood clots. As the clotting reaction releases proteins that can affect lymphocyte phenotype and function, samples containing clots should be discarded.

4. Using a sterile pipet, add an equal volume of room-temperature 1× PBS suspension (if utilizing a 15-ml conical tube) or bring the leukocyte/RBC cell suspension to a final volume of 40 ml with 1× PBS (if utilizing a 50-ml conical tube).

5. Slowly layer the Ficoll-Hypaque solution underneath the leukocyte/RBC/PBS mixture by placing the tip of the pipet containing the Ficoll-Hypaque at the bottom of the sample tube. Use 3ml Ficoll-Hypaque per 10ml blood/PBS mixture. Use a maximum of 10 ml Ficoll-Hypaque per 40 ml leukocyte/RBC/PBS mixture.

To maintain the Ficoll-Hypaque/blood interface, it is helpful to hold the centrifuge tube at a 45◦ angle. Alternatively, the leukocyte/RBC/PBS mixture may be slowly layered over the Ficoll-Hypaque solution. *See Figure below.*

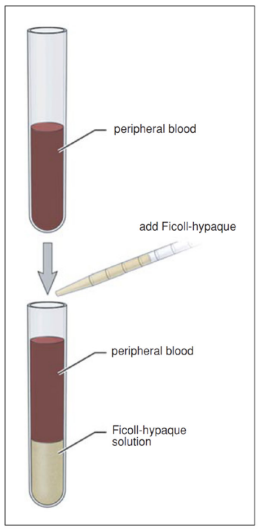


Figure 7.1.1 Ficoll-Hypaque density isolation of mononuclear cells. Ficoll-Hypaque is underlayered onto whole blood or cord blood. The sample is centrifuged allowing the separation of lymphocyte cell population from other blood elements based upon density gradient.

6. Centrifuge 20 to 30 min at 2000 rpm (900 × *g*), 18◦ to 20◦C, with no brake.

After a centrifugation with Ficoll-Hypaque, platelets (specific gravity, 1.040) and plasma (specific gravity, 1.025 to 1.029) are located above the Ficoll-Hypaque, lymphocytes (specific gravity, 1.070), and some platelets are found at the plasma-

Ficoll-Hypaque interface. Granulocytes (specific gravity, 1.087 to 1.092) and RBCs (specific gravity, 1.093 to 1.096) form a cell pellet at the bottom of the tube (see Fig. below)

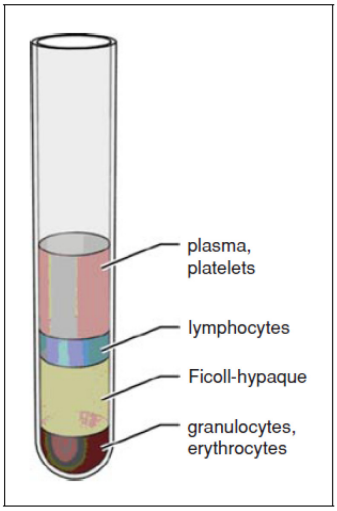


Figure 7.1.2 Separation of blood components on a Ficoll-Hypaque gradient.

7. Using a sterile pipet, remove the upper layer that contains the plasma and most of the remaining cell platelet fraction. Using another pipet, transfer the mononuclear lymphocyte cell layer to another centrifuge tube. This will appear as a white, cloudy band between the plasma and the Ficoll-Hypaque layers. Wash cells by adding HBSS (~3 times the volume of the mononuclear cell layer) and centrifuging 10 min at 450 to 600×*g*, 18◦ to 20◦C. Remove supernatant, re-suspend cells in HBSS, and repeat the wash once to remove any remaining platelets.

The washing steps described above usually remove most of the platelets from the mononuclear cell suspension. There are certain disease states associated with increased platelet concentrations (mononuclear cell to platelet cell ratio >10:1) in the peripheral blood, and additional steps are needed to remove the extra platelets in these cases. Add 3 ml FBS to a centrifuge tube for each milliliter of mononuclear cells. Layer the cell suspension (1-2 × 107 cells/ml) over the FBS (alternatively, carefully layer the FBS under the cell suspension, which will rise as FBS is added). Centrifuge 15 min at 150 × g, 18◦ to 20◦C. Discard the supernatant containing the platelets. Re-suspend cell pellet in complete RPMI-1640 and proceed as in step 8.

8. Re-suspend mononuclear lymphocyte cells in complete RPMI-1640. Count cells and determine viability by trypan blue exclusion. If desired, purity of PBMC population can be determined by flow cytometry.

9. Centrifuge 5 min at 2000 × g, 18° to 20°C. Discard the supernatant and save the PBMC in -80°C until shipment to the central lab. Label it clearly with "PBMC" and estimate cell number.

10. Freeze sample in one aliquot tube at -80C until shipment to the central lab. See shipping instructions.

Shipment of Cytokine Plasma and PBMC samples (includes C-reactive protein at Pre-treatment and at Cycle 2, day 1):

1. Samples may be batched and shipped on a monthly basis.

2. Package samples according to IATA regulations.

2. Label specimens appropriately with subject study ID, date and time of collection, and type of collection (plasma or PBMC). Complete and fax the completed Blood Sample Requisition and Shipment Notification Form on the day of shipment and include a copy of the form in the shipment.

3. Slides made from the C1, D1 predose timepoint will be transported at ambient temperature using overnight express service.

4. The plasma, pellet, PBMC tubes will be shipped frozen and on dry ice to Dr. Fu. Transportation will be through overnight express service in sufficient dry ice to ensure that the samples will remain frozen for over 24 hours. Please ensure that shipments can be delivered within 24 hours after shipment (do not ship on Fridays, or days before holidays).

Ship to:
Dr. Kai Fu
University of Nebraska Medical Center
601 South Saddle Creek Rd.
Omaha, NE 68106
402-559-3135
Phone: 402-559-7526
Fax: 402-559-6018
Pager: 402-888-2528

5. Dr. Fu's laboratory will be informed of the shipment and tracking number immediately and the status of the shipment will be tracked. A detailed log of the specimen will be kept.

C. C-REACTIVE PROTEIN Blood Collection (use these instructions when collecting blood only for CRP and not for cytokine profiling) at time of progression or relapse

1. Collect 7cc to 10cc whole blood and centrifuge within 2 hours of collection.
Stability: For serum and plasma:
Ambient: 15 days
Refrigerated: 2 months
Frozen: 3 years

Plasma
Collect 7cc to 10cc blood in a light green-top (lithium heparin) gel tube(s). Spin down and send 1 mL of lithium heparin plasma. Store at -80.
Note: Indicate plasma on request form.

Serum
Collect 7cc to 10cc blood in a gold-top serum gel tube(s) or a plain, red-top tube(s). Spin down and send 1 mL of serum.
Note: Indicate serum on request form.

2. Label specimen appropriately with subject study ID, date and time of collection, and type of collection (plasma or serum). Complete and fax the completed Blood Sample Requisition and Shipment Notification Form on the day of shipment and include a copy of the form in the shipment.

3. Ship either ambient, refrigerated, or frozen according to the stability chart above. Package samples according to IATA regulations. Transportation will be through overnight express service to ensure that shipments can be delivered within 24 hours after shipment (do not ship on Fridays, or days before holidays).

Ship to:

Dr. Kai Fu
University of Nebraska Medical Center
601 South Saddle Creek Rd.
Omaha, NE 68106
402-559-3135
Phone: 402-559-7526
Fax: 402-559-6018
Pager: 402-888-2528

4. Dr. Fu's laboratory will be informed of the shipment and tracking number immediately and the status of the shipment will be tracked. A detailed log of the specimen will be kept.

APPENDIX 3 – GRADING OF ADVERSE EVENTS

Adverse Events will be graded according to the National Cancer Institute Common Toxicity Criteria for Adverse Events (CTCAE), Version 4, which may be found at the following website:

http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm#ctc_40