TITLE: Prospective Evaluation of Ruxolitinib Efficacy for CNL/aCML Patients with Mutation of CSF3R

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TITLE: *Prospective Evaluation of Ruxolitinib Efficacy for CNL/aCML Patients with Mutation of CSF3R*

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SCHEMA
Meets criteria for diagnosis of CNL or aCML
(see Appendix A)

Screening period
(up to 4 weeks, see Section 10.1)

Patients eligible for treatment with ruxolitinib
(up to 96 weeks, see Section 5.1)

End of treatment or early discontinuation
(within 14 days of drug discontinuation, see Section 10.3)

End of study
(end of study visit to be completed 4-6 weeks later, see Section 10.4)

Possible options at end of study:

1) Discontinuation of ruxolitinib
   - Optional taper

2) Continue ruxolitinib commercially
   - Patient responsible for obtaining drug through insurer or self-pay

3) Continue ruxolitinib through INCYTE
   - Subject to the sponsor's approval and/or study extension

1 OBJECTIVES

1.1 Primary Objectives

1.1.1 To determine the proportion of patients with chronic neutrophilic leukemia (CNL) and atypical chronic myeloid leukemia (aCML) who have a hematologic response to ruxolitinib (PR, CR, CRp). See table 6 and section 13.2 for specific parameters.

1.2 Secondary Objectives

1.2.1 To determine the frequency of grade 3 or 4 hematologic and non-hematologic adverse events experienced by subjects during therapy with ruxolitinib. See CTCAE reference in Appendix E.

1.2.2 To determine whether hematologic responses, as defined in section 13.2, correlate with certain types of mutations in CSF3R and reduction in mutant CSF3R allele burden in the peripheral blood.

1.2.3 To determine the maximum clinical responses for each subject (defined in Table 6) and the median duration of maximum clinical responses.

1.2.4 To determine the mean % reduction of spleen size, estimated by volume using the conventional prolate ellipsoid method as measured by ultrasound compare to baseline.

1.2.5 To determine the mean % reduction of total symptom score as measured by a modified MPN-SAF (see section 13.2) compared to start of study (day 1, cycle 1).

1.2.6 To determine overall survival in subjects who complete a minimum of 1 dose of study drug.

1.2.7 To determine the proportion of subjects who discontinue after completion of > 3 cycles but < 6 cycles.

1.2.8 To determine the proportion of subjects who discontinue prior to completion of cycle 3.

2 BACKGROUND

2.1 Study Disease

CNL and aCML are both hematologic neoplasms characterized by leukocytosis and hypercellular bone marrow comprised predominantly of granulocytic cells, absence of the Philadelphia chromosome (t(9;22); *BCR-ABL1*), and absence of *platelet-derived growth factor receptor A/B* (*PDGFRA/B)* or *fibroblast growth factor receptor 1* (*FGFR1*) gene rearrangements. **CNL** is diagnosed based on expansion of neutrophils in both the blood and bone marrow (segmented neutrophils and band forms > 80% of white blood cells (WBC)) and is classified as a myeloproliferative neoplasm (MPN) according to World Health Organization (WHO) diagnostic criteria. Cases of **aCML** exhibit granulocytic dysplasia and increased numbers of neutrophil precursors in both the peripheral blood and the bone marrow (typically ≥ 10% of WBCs) and are therefore classified as one subtype of the WHO category of myelodysplastic/myeloproliferative

neoplasms[1,2]. Occasional cases of CNL[3,4] and a majority of aCML cases are reported to exhibit non-specific cytogenetic abnormalities[5] or infrequently the *JAK2* V617F mutation[6,7], revealing the clonal nature of these diseases. While certain MPN subtypes have either been operationally defined by molecular abnormalities (e.g. *BCR-ABL1* in CML) or are characterized by a high frequency of specific genetic abnormalities (e.g. *JAK2* V617F in polycythemia vera (PV), essential thrombocythemia (ET), and primary myelofibrosis (PMF)[6,8-11]; *KIT* D816V in systemic mastocytosis[12,13]), the genetic basis of both CNL and aCML was unknown until recently. Standard of care is not established for CNL and aCML and the estimated median overall survival is 23.5 months.[14]

2.2 Study Agent(s)

Ruxolitinib (Jakafi, INCB018424) is a potent, and selective inhibitor of JAK1 (IC_{50} = 3.3 ± 1.2 nM) and JAK2 (IC_{50} = 2.8 ± 1.2 nM) with modest to marked selectivity against TYK2 (IC_{50} = 19 ± 3.2 nM) and JAK3 (IC_{50} = 428 ± 243 nM), respectively. Ruxolitinib was FDA-approved in November 2011 for treatment of intermediate to high-risk primary myelofibrosis or post-essential thrombocytosis (ET) or post-polycythemia vera (PV) myelofibrosis. More recently, a role for ruxolitinib for reducing disease burden in CNL and aCML was identified based on characterization of oncogenic CSF3R mutations.[15] Ruxolitinib has high solubility and permeability. It is designated as a Class I molecule in the Biopharmaceutical Classification System and exhibits moderate-to-high clearance, volume of distribution and oral bioavailability in preclinical species.

The pharmacology of ruxolitinib has been studied in both healthy volunteers and patients with hematologic and solid tumor malignancies. Ruxolitinib is more than 95% absorbed orally (regardless of food ingestion) with peak plasma concentrations occurring 1-2 hours after dosing. (Investigator's Brochure, Edition 11) The pharmacokinetics are linear and there is 97% plasma protein binding with limited penetrance across the bloodbrain barrier. The majority of drug is excreted in the urine (75%) with less than 1% excreted as unchanged parent drug. The mean terminal half-life is 3-5 hours, and it is metabolized through the cytochrome P450 isozyme CYP3A4 resulting in oxygenated and conjugative metabolites. Twice daily dosing does not result in significant accumulation of the parent compound or its metabolites. The total daily dose should be reduced by 50% when ruxolitinib is administered with moderate or potent CYP3A4 inhibitors, but not dose-adjusted when given with CYP3A4 inducers.

2.3 Other Agent(s)

Not applicable.

2.4 Study and Dose Rationale

We have identified **gain-of-function mutations in the receptor for Colony Stimulating Factor 3** (CSF3; GCSF), CSF3R, in ~60% of patients with chronic neutrophilic leukemia (CNL) and atypical (BCR-ABL-negative) chronic myeloid leukemia (aCML).[15] We have also observed these mutations in ~1% of AML patients (Table 1). CSF3R mutations predict for sensitivity of the malignant cells to JAK2 inhibition. These data suggest that JAK2 inhibitors, such as ruxolitinib, would be beneficial therapeutic agents for the majority of CNL/aCML patients. Demonstration of the clinical efficacy of this proposed therapeutic strategy will require a prospective clinical trial where CNL and aCML patients are treated with ruxolitinib, and the response of patients harboring CSF3R mutations is monitored.

Diagnosis	Mutation	Estimate of Frequency
CNL/aCML	16/27	59%
AML	3/292	~1%
T-ALL	0/8	0
ETP-T-AL	1/3	Unknown
B-ALL	0/41	0

Table 1. Summary of CSF3R mutational status by hematologic malignancy subset reveals enrichment of CSF3R mutations in patients with CNL and aCML. CNL – Chronic Neutrophilic Leukemia; aCML – atypical Chronic Myeloid Leukemia, BCR-ABL-negative; AML – Acute Myeloid Leukemia; T-ALL – T-cell Acute Lymphoblastic Leukemia; ETP-T-ALL – Early T-cell Precursor T-ALL; B-ALL – B-cell Acute Lymphoblastic Leukemia.

CSF3R is the receptor for colony stimulating factor 3 (CSF3; GCSF), and is thought to play a prominent role in the growth and differentiation of granulocytes[16,17]. *CSF3R* mutations have been described in patients with severe congenital neutropenia (SCN), which can evolve into acute myeloid leukemia (AML)[18-20]; it was recently reported that an SCN patient developed sequential acquisition of a secondary *CSF3R* mutation at the time of AML transformation[21]. These previously described nonsense/frameshift mutations truncate the cytoplasmic tail of CSF3R, impair its internalization, and alter its interactions with proteins such as SHP-1/2 and SOCS family members[22-24]. These structural and functional alterations are thought to perturb the capacity of CSF3R to regulate granulocyte differentiation and to increase granulocytic proliferative capacity[25-27]. CSF3R signals through the JAK/STAT pathway, the non-receptor tyrosine kinase SYK[28,29], and the SRC family kinase (SFK) LYN, which was recently shown to be mediated by the phosphatase SHP-2 and the adaptor protein GAB2[28-31]. With the exception of isolated case reports[32], mutations in *CSF3R* have not previously been observed in cases of *de novo* leukemia.

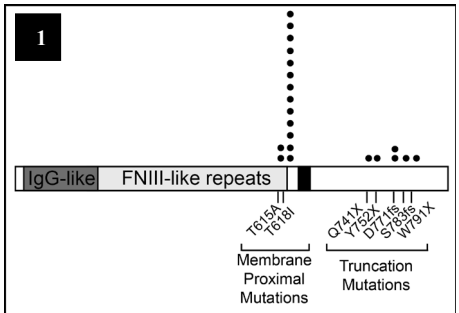

Figure 1. Distribution of CSF3R mutations in CNL/aCML
Schematic of the location and recurrence of CSF3R mutations found in human leukemia patient samples. The mutation locations and number of observations in this study are indicated by black circles. The Q741X mutation was found in an AML sample and one of the T618I mutations was found in an ETP-T-ALL. All other mutations were observed in CNL/aCML specimens. Two additional mutations (Q739X and T618I; not depicted in Figure) have been reported in AML specimens sequenced by The Cancer Genome Atlas.

The **CSF3R mutations** that we have identified cluster into two distinct regions of the receptor, with the majority occurring just extracellular of the transmembrane domain (membrane proximal mutations) and a small number resulting in truncation of the cytoplasmic tail (truncation mutations) (Figure 1). By far, the most prevalent mutations are the membrane proximal mutations (T618I and T615A), which result in ligand independent activation of CSF3R signaling. The dominant downstream substrate in this context is JAK2. As such, cells harboring membrane proximal CSF3R mutations are exquisitely sensitive to ruxolitinib, with IC50s of ~100-200 nM. An index patient with CSF3R T618I-driven CNL has been treated with ruxolitinib for ~8 months and still exhibits a dramatic and durable

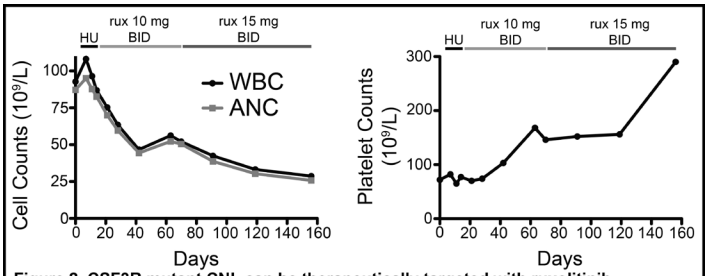

Figure 2. CSF3R mutant CNL can be therapeutically targeted with ruxolitinib
Cells from a CNL patient with CSF3R T618I were sensitive to ruxolitinib (rux) in vitro (IC50 of 127 nM, not shown). This patient was treated with 500 mg hydroxyurea (HU; hydroxy-carbamide) daily starting on day 13. Hydroxyurea was stopped on day 21 and 10 mg ruxolitinib taken orally twice daily was initiated. On day 70 the dose of ruxolitinib was escalated to 15 mg twice daily. White blood cells (black line, WBC) and absolute neutrophil count (gray line, ANC) are shown. Treatment of the CSF3R-T618I-positive CNL patient with ruxolitinib normalized platelet counts.

response with near normalization of white blood cell counts, and complete normalization of other blood parameters, including dramatic increases in platelet counts to normal levels (Figure 2).

Finally, although truncation mutations are much less common, data from primary bone marrow colony assays suggest that cells harboring truncation mutations of CSF3R may be similarly sensitive to ruxolitinib, though these cells are also sensitive to inhibitors of SRC-family kinases (Figure 3).

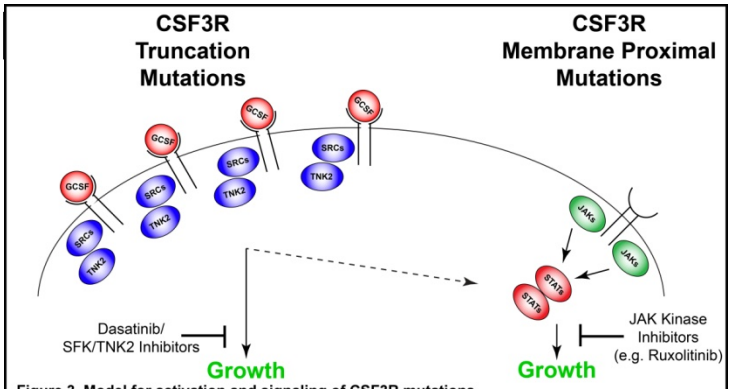

Figure 3. Model for activation and signaling of CSF3R mutations.
Truncation mutations of CSF3R result in increased expression levels. Downstream signaling mediators, SRC-family kinases (SFKs) and TNK2, are preferentially activated by truncation mutant CSF3R. Consequently, leukemic cells harboring truncation mutant CSF3R are highly sensitive to dasatinib. Truncation mutant CSF3R may also exhibit sensitivity to JAK kinase inhibitors in the context of JAK kinase stimulation downstream of high ligand concentrations. In contrast, membrane proximal mutant CSF3R exhibits complete ligand independent function. In this capacity, the dominant mode of signaling appears to operate through the JAK/STAT pathway. Hence, patients exhibiting membrane proximal mutations may be candidates for JAK kinase inhibitors, such as the JAK1/2 inhibitor ruxolitinib.

Collectively, these data argue strongly **that ruxolitinib represents a promising therapeutic for the majority of patients with CNL and aCML** and that these patients may exhibit durable, long-term responses to ruxolitinib, similar to the responses of CML patients to ABL kinase inhibitors. Currently, hydroxyurea is the only available therapy to control counts and symptoms but does little to change the natural history of the disease. In one report, the estimated median overall survival is 23.5 months with standard therapy[14]. Though CNL and aCML are not common forms of hematologic malignancy (estimate of several hundred cases per year in the U.S.), the therapeutic benefit may be long-lasting for these patients and could convert these diseases into one that patients can live with.

The clinical efficacy results of ruxolitinib in the treatment of myelofibrosis have emerged from the ongoing Phase 1 /2 and Phase 3 studies are notable, including marked reduction in splenomegaly, improvement in symptoms, performance status and activity level, and reduction in plasma levels of inflammatory, prothrombotic and angiogenic cytokines. The Phase 3 studies have been published[33] and these results led to FDA approval of ruxolitinib for intermediate to high-risk myelofibrosis in November 2011. In light of the preclinical studies and the identification of CSF3R as oncogenic drivers in CNL and aCML,[15] this study aims to test whether there is clinical benefit (defined in primary and secondary objectives) for treatment of these patients with ruxolitinib.

In this study, all subjects with a diagnosis of CNL or aCML will begin ruxolitinib at a total daily dose between 5 mg and 40 mg (range from 5 mg qd to 20 mg bid). The starting dose is based on the general dosing guideline provided in the ruxolitinib prescribing information based on platelet counts for the treatment of intermediate to high risk myelofibrosis (Appendix B). Within these parameters, we have defined specific doses based on actual start of study (day 1, cycle 1) platelet counts and on concomitant drugs categorized as moderate or potent CYP 3A4 inhibitors.

2.5 Correlative Studies Background

2.5.1 Correlative study: Correlate hematological responses (defined in Table 6 and in section 13.2) with ruxolitinib therapy to mutant CSF3R allele burden

2.5.1.1 **Rationale:** Oncogenic CSF3R drives survival and proliferation of CNL and aCML cells in preclinical studies.[15] Based on these studies, therapies that specifically inhibit CSF3R or its downstream signaling pathways are expected to reduce survival and proliferation of CNL and aCML cells. Ruxolitinib targets JAK1/2 kinase signaling, a downstream signaling pathway of CSF3R activation, and is the basis for this clinical trial concept and its predicted efficacy in patients.

2.5.1.2 **Hypothesis:** We hypothesize that in cases of CNL or aCML that harbor mutant CSF3R, the mutant allele burden is reduced compared to start of study (day 1, cycle 1) values and correlates with hematological responses to ruxolitinib.

2.5.2 Correlative study: Identification of novel alternative and cooperating mutations

2.5.2.1 **Rationale:** Some patients who meet the clinical diagnostic criteria for CNL and aCML do not harbor mutations in CSF3R. These patients may have alternative mutations downstream of CSF3R or mutations in a parallel signaling pathway that phenocopies the oncogenic potential of CSF3R. Although we expect that most patients will respond to ruxolitinib, there may be cases of primary or secondary resistance to ruxolitinib and these cases represent an opportunity to further characterize these alternative and/or cooperating mutations in CNL and aCML.

2.5.2.2 **Hypothesis:** We hypothesize that the development of CNL and aCML is limited to a few oncogenic drivers and the occurrence of primary or secondary resistance to ruxolitinib may represent alternative or cooperating mutations that could enhance our effort to study early and late steps in the pathogenesis of CNL and aCML.

3 STUDY POPULATION

We plan to enroll approximately 50 subjects in order to achieve an overall of 25 evaluable subjects.

3.1 Inclusion Criteria

All subjects must meet the following inclusion criteria:

3.1.1 Subjects must be newly diagnosed or previously diagnosed with CNL or aCML (please refer to Appendix A). All patients must have a bone marrow biopsy completed during the screening or baseline period if one has not been done within 90 days of day 1, cycle one.

3.1.2 Subjects must have platelet count greater than 25,000 per microliter at baseline and at the start of study (day 1, cycle 1) visit.

3.1.3 Subjects must be able to discontinue any drug treatment aimed at lowering disease burden in CNL or aCML. Subjects should discontinue hydroxyurea to treat underlying CNL or aCML disease no later than Day -7 (one week before starting ruxolitinib). For drugs that have more long-lasting effects on the marrow, such as thalidomide and its analogs, and interferon, subjects should discontinue these no later than Day -28.

3.1.4 Subjects must be willing to accept/continue transfusions to treat low hemoglobin levels.

3.1.5 Subjects must be 18 years or older, male or female.

3.1.6 Subjects must have a life expectancy of > 6 months.

3.2 Exclusion Criteria

Any of the following are causes for exclusion from the study, and must be demonstrated not to be present on the day of first dose, C1D1 (historical data for measurements or labs may not be used).

3.2.1 Subjects unable to review and sign informed consent form.

3.2.2 Females who are pregnant or breastfeeding, and males and females who cannot comply with requirements to avoid fathering a child or becoming pregnant. See Appendix G.

3.2.3 Subjects with known diagnosis of HIV or chronic active Hepatitis B or C. Viral testing is not required. Subjects with a history of Hepatitis B and/or C are allowed on trial if the virus is undetected at the time of enrollment.

3.2.4 Subjects with inadequate liver (ALT/SGPT above 4X ULN or direct bilirubin 4X ULN AND the lab abnormalities are felt to be due to underlying liver dysfunction).

3.2.5 Subjects with end stage renal function (CrCl<15 mL/min or GFR <15 mL/min) regardless of whether hemodialysis is required.

3.2.6 Subjects with clinically serious infections requiring ongoing antibiotic therapy.

3.2.7 Subjects with severe (immediately life threatening) and recent (occurring within the last 3 months) cardiac dysfunction, pulmonary dysfunction, esophageal variceal bleeding, hemorrhagic strokes, or intracranial hemorrhage are not eligible for study participation.

3.2.8 Subjects requiring therapeutic doses of anticoagulation or anti-platelet therapies (aspirin above 81mg daily, plavix or similar agents) AND platelet counts are below 50,000 on two different laboratory evaluations, separated by minimum of two weeks.

3.2.9 Taking investigational or commercial agents or therapies with the intent to treat the subject's malignancy other than those therapies permitted as described in section 5.2

3.2.10 Subjects with invasive malignancy over the previous 2 years except treated early stage carcinomas of the skin, completely resected intraepithelial carcinoma of the cervix, and completely resected papillary thyroid and follicular thyroid cancers

3.2.11 Previous allergic reactions to JAK inhibitors or excipients.

3.2.12 Prior therapy with ruxolitinib or other JAK inhibitors.

3.2.13 Subjects who have had major surgery within 4 weeks prior to entering the study.

3.2.14 Subjects who are anticipated to receive a transplant within the first 6 months of treatment on trial.

4 REGISTRATION PROCEDURES

4.1 Subject Registration

Detailed instructions for subject registration can be found in the separate Study Operations Manual.

There is no randomization for treatment. This is a phase II trial with an intention-to-treat all patients who are enrolled in this clinical trial. Potential subjects will be seen by investigators of this study as a new patient or consult visit or as a follow-up visit. Referral of potential subjects to co-investigators of this study is made as part of standard of care, with the referring physician seeking advice on the diagnosis, evaluation, and/or treatment of CNL or aCML

4.1.1 Local registration.

Registrations from all consented subjects must be entered into the Knight

Clinical Research Management System (CRMS). Registration of OHSU patients will include the minimum of the following:

- A completed Subject Enrollment Form
- A completed Eligibility Checklist signed by the investigator
- Signed copies of the most recently IRB-approved, informed consent form and HIPAA authorization

4.1.2 Multicenter Registration

The OHSU coordinating center study team will manage subject registration. Investigators at participating sites will identify eligible subjects and send screening materials with source documents that support eligibility to OHSU in real time and in accordance with study protocol. Designated Knight clinical staff must review and verify eligibility before the participating site may enroll and treat its subject. The OHSU coordinating center team will verify completeness of documents, enter registration information into the Knight CRMS, and assign a study number/identifier. The coordinating center will send an email to the participating site indicating whether or not the subject is eligible, verify registration, and assign a participant number/identifier.

Registration will include a minimum of the following:

- A completed Subject Enrollment Form
- A completed Eligibility Checklist signed by the investigator
- Signed copies of the most recently IRB-approved, informed consent form and HIPAA authorization

arise.

5 TREATMENT PLAN

5.1 Agent Administration

We propose that all subjects with a diagnosis of CNL or aCML (please refer to Appendix A) will be eligible for treatment with ruxolitinib on this study. Initial doses of 5, 10, 15, or 20 mg BID will be administered based on platelet counts at the start of study (day 1, cycle 1). The FDA-approved prescribing information for ruxolitinib dosing based on platelet counts is provided as a reference (Appendix B). However, we have made additional suggestions where no specific guidance is provided but maintained the suggestions within the parameters outlined in the prescribing information. We highly suggest adjusting the starting dose if the subject is on concomitant CYP 3A4 inhibitors, according to the table below. However, whenever medically possible, we suggest avoiding potent CYP3A4 inhibitors and to consult with the main study site for advice. Table 2 (below) serves to advise investigators on the initiation of ruxolitinib on study, but the sponsor and site principal investigator may choose to modify the dose based on clinical judgment and consideration of any safety issues.

Table 2: Recommendations for Initiation of Ruxolitinib Dosing Based on Platelet Count

Platelet count at start of study (day 1, cycle 1; per microliter)	No CYP 3A4 inhibitors†‡	Moderate CYP 3A4 inhibitors†	Potent CYP 3A4 inhibitors†
>200,000	20 mg twice daily	15 mg twice daily	10 mg twice daily
150,000-199,000	15 mg twice daily	10 mg twice daily	5 mg twice daily
100,000-149,000	10 mg twice daily	5 mg twice daily	5 mg daily
50,000-99,000	5 mg twice daily	5 mg daily	5 mg every other day
<50,000 (but more than 25,000)	5 mg twice daily	5 mg daily	5 mg every other day

† Common drugs listed as CYP 3A4 inhibitors and inducers are provided in Appendix D. Drugs that are weak inhibitors of CYP3A4 or are CYP 3A4 inducers require no dose adjustment for the starting dose but the investigators may choose to adjust the dose at their discretion after observing platelet counts.

‡ The suggested starting doses comply within the guideline provided in the ruxolitinib prescribing information. However, the starting doses are further defined for patients who have platelet counts of less than 150,000 per microliter.

For patients already on a stable dose of ruxolitinib, when a moderate or potent CYP3A4 inhibitor is to be co-administered during the clinical trial, the dose of ruxolitinib should be reduced by approximately 50%.

Treatment will be administered on a planned outpatient basis. Reported adverse events and potential risks associated with ruxolitinib are described in Section 7. Appropriate dose modifications for ruxolitinib are described in Section 6. Doses should be taken by mouth in the morning and evening, approximately 12 hours apart, and without regards to food.

Subjects will bring all bottles of unopened, empty, and opened/partially used study drug with them to each study visit. Investigative site staff will perform a count of returned pills to assess compliance. Study drug, including all bottles of unopened, partially opened or empty bottles will then be returned to research pharmacy for storage.

5.2 General Concomitant Medication and Supportive Care Guidelines

CYP3A4 Inhibitors

The principal investigator should be alerted if the subject is taking any agent categorized as a moderate or potent **CYP3A4 inhibitor** (please see Appendix D). Dose adjustment is not required when ruxolitinib is co-administered with rifampin or other CYP3A4 inducers, but these agents should be used with caution in combination with ruxolitinib and alternative therapy used if available. Use of moderate or potent CYP3A4 inhibitors (Appendix D) is discouraged and investigators should seek other options where possible. For patients already on a stable dose of ruxolitinib, when a moderate or potent CYP3A4 inhibitor is to be co-administered during the clinical trial, the dose of ruxolitinib should be reduced by approximately 50% and extra hematology assessments should be instituted for close monitoring of platelet levels.

Anticoagulation/Antiplatelet Medications

Anticoagulation is allowed provided that platelet counts are maintained above 50,000 per microliter. Medications that are anticoagulants or inhibit platelet function are generally not recommended with the exception of a maximal dose of 81 mg a day of aspirin given per physician discretion per standard of care and over-the-counter doses of NSAIDs and acetaminophen. Subjects receiving over-the-counter NSAIDs should not exceed the recommended dose and should be encouraged to use gastroprotective agents (antacids, H2 antagonists or proton pump inhibitors).

- Prohibited Anticoagulation/Antiplatelet:
  Subjects requiring therapeutic doses of anticoagulation or anti-platelet therapies (aspirin above 81mg daily, plavix or similar agents) AND platelet counts are below 50,000 on two different laboratory evaluations, separated by minimum of two weeks. Patients who meet these criteria are excluded from the study and are withdrawn from the study if other drug or dosing alternatives are not possible or if reduction of ruxolitinib dosing does not improve platelets to over 50,000 per microliter.

Hematopoietic Growth Factor Receptor Agonists

Hematopoietic growth factor receptor agonists (e.g., erythropoietin (Epo) may be used at Investigator's discretion in certain circumstances based on the clinical indications and risks/benefit considerations as described in the NCCN ESA guideline (http://www.nccn.org/professionals/physician_gls/pdf/anemia.pdf).

- Prohibited Growth Factors
  Romiplostim or eltrombopag are not permitted beginning with the Baseline Visit (Day -7) through the final dose of ruxolitinib.

Other Prohibited Medications and Measures

- Any prior or concomitant use of a JAK inhibitor other than ruxolitinib.
- Any investigational medication other than the study drugs or any investigational agent
- Use of interferon, thalidomide, busulfan, lenalidomide or anagrelide, or any investigational medication used to treat CNL or aCML is not permitted at any time beginning on Day -28 up until

the time that ruxolitinib therapy is permanently discontinued.

- Hydroxyurea should be discontinued before Day -7 up until the time that ruxolitinib therapy is permanently discontinued.

5.3 Duration of Treatment

In the absence of treatment delays due to adverse events, treatment may continue for 96 weeks or until one of the following criteria listed in section 5.6 applies**.** If there is benefit from the study drug, the patient may be eligible to continue on treatment past 96 weeks. If the study drug becomes commercially available for the disease indication during the duration of treatment, the study team will help transition the patient to commercial drug supply.

Table 3: Indices of Disease Progression

Parameter	Signs of disease progression
White blood count	More than doubling of white blood cell count in the absence of a concurrent acute or subacute medical illness. The white blood cell count is compared to the two most recent prior lab values and the white blood cell count change is confirmed on another lab draw minimum of 2 weeks apart.
Blasts	Evolution to AML, defined by >20% blasts in the peripheral blood or bone marrow in the absence of a concurrent acute or subacute medical illness. The peripheral blood or bone marrow findings are confirmed on another lab draw minimum of 2 weeks apart.
Splenomegaly	>25% spleen enlargement compared to baseline. This can sometimes be appreciated by palpation (from the mid-costochondral line) but can also be confirmed by ultrasound if spleen growth is suspected.

5.4 Duration of Follow-up

Subjects will be in the treatment phase of the study for maximum of 96 weeks. Subjects will have an end of treatment visit (within 14 days of stopping study drug) and an end of study visit (within 4-6 weeks of stopping study drug). Subjects removed from study for unacceptable adverse event(s) will be followed until resolution or stabilization of the adverse event whichever is longer.

5.5 Criteria for Removal of a Subject from Study

Subjects may choose to withdraw from the study at any time without penalty of jeopardizing their health care or loss of benefits to which the subject is otherwise entitled. Every reasonable effort should be made to determine the reason a subject withdraws prematurely, and this information should be recorded in the eCRF.

A subject may be withdrawn from the study, if, in the investigator's expert medical judgment, the subject is non-compliant with the study requirements. Subjects may be withdrawn at the discretion of the Food and Drug Administration (FDA) or the investigator.

Subjects **must** discontinue study drug for the following reasons (investigators will review with the main study site appropriateness for continuing or discontinuing subject from study):

- 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 for continued study drug treatment is withdrawn by the subject
- Termination of the study by the Sponsor Investigator, local health authority, or IRB
- The subjects exhibits leukemic transformation (as evidenced by bone marrow or peripheral blasts >20% persisting for at least 2 consecutive laboratory studies at least 2 weeks apart in the absence of clinical circumstances suggesting an exaggerated leukoerythroblastic reaction e.g., sepsis, major surgery, trauma, or stress).
- The subjects who meet WBC and spleen criteria for disease progression (Table 3), but do not have leukemic transformation, AND is on the maximum dose of ruxolitinib 25mg twice daily for more than 2 consecutive months. For patients who meet the WBC and spleen criteria for disease progression but are not on ruxolitinib 25mg twice daily, see section 6, dose increases section. Investigator can also assert discretionary clinical benefit not addressing these circumstances. For particular situations not addressed here, drug can continue if there is ongoing clinical benefit.
- The subject cannot maintain platelet counts of at least 20,000 per microliter over a given 4 week interval at the lowest possible dose of 5 mg every other day
- The subject experiences a recurrence of a Grade 3 or Grade 4 bleeding, and believed to be from the same source or etiology AND the subject has a platelet count of less than 50,000 per microliter AND subject is on the lowest dose possible of 5mg every other day. Please see Appendix E for grading of hemorrhagic events.
- Subjects requiring therapeutic doses of anticoagulation or anti-platelet therapies (aspirin above 81mg daily, plavix or similar agents) AND platelet counts are below 50,000 on two different laboratory evaluations, separated by minimum of two weeks.

5.6 Study Discontinuation

If a subject is withdrawn from the study:

- The Coordinating Site must be notified.
- The reason(s) for withdrawal must be documented in the subject's medical record and CRF
- The End-of-treatment/Early Termination visit should be performed within 2 weeks of last dose.
- All subjects must be followed for safety until the time of the follow-up evaluation or until study drug related toxicities until resolution or stabilization of the adverse event whichever is longer whichever is longer.

If the sponsor investigator or regulatory authority discovers conditions during the study that indicate that the study or study site should be terminated, this action may be taken after appropriate consultation with the investigators. The sponsor investigator has the right to terminate the participation of either an individual site or the study at any time. Reasons for terminating the study may include the following:

- Incidence or severity of adverse events in this or other studies indicates a potential health hazard to subjects.
- Subject enrollment is unsatisfactory.
- Data recording is inaccurate or incomplete.
- Investigator(s) do not adhere to the protocol or applicable regulatory guidelines in conducting the study.

6 DOSING DELAYS/DOSE MODIFICATIONS

Dose Adjustments

If subjects experience thrombocytopenia with platelet **counts less than 20,000 per microliter**, ruxolitinib dosing should be held until the platelet counts are above 25,000 per microliter. The restarting dose should be as follows (Table 4):

Table 4: Restarting doses in subjects who experienced toxicity that required ruxolitinib hold

Current Platelet Count	Maximum Dose When Restarting ruxolitinib Treatment
Greater than or equal to 125 X 10^9/L	20 mg twice daily
Between and including 100 and 124 X 10^9/L	20 mg twice daily
Between and including 75 and 99 X 10^9/L	up to 15 mg twice daily
Between and including 50 and 74 X 10^9/L	up to 10 mg twice daily
Between and including 25 and 49 X 10^9/L	up to 5 mg twice daily
24 X 10^9/L or less	DO NOT RESTART WITHOUT CONSULTING WITH SPONSOR-INVESTIGATOR.

Subjects with a grade 3 or 4 hemorrhage event must HOLD dosing until further evaluation. Ruxolitinib may be continued without hold if bleeding event is deemed ongoing from baseline or determined by site PI in consultation with Sponsor to be well controlled and unrelated to administration of ruxolitinib. Subjects will have the option to restart or re-escalate the dose with improving platelet counts and the source of the bleeding has resolved.

Table 5 outlines the SUGGESTED approach for any grade 3 or 4 non-hematologic toxicity (refer to CTCAE for grading, reference in Appendix E). The investigators' assessment of unrelated, possible, and definite causality to ruxolitinib guides the degree of dose reduction. The investigators may choose to deviate from this approach or HOLD the study drug, based on safety and other medical considerations.

Table 5: Dose Adjustment for Ruxolitinib based on Adverse Event Relationship

AE, is it related to ruxolitinib?	Dose at time of AE	Dose adjustment
Unrelated	5 mg twice daily	No adjustment
	10 mg twice daily	No adjustment
	15 mg twice daily	No adjustment
	20 mg twice daily	No adjustment
Possible	5 mg twice daily	5mg daily or HOLD
	10 mg twice daily	5 mg twice daily
	15 mg twice daily	5 mg in AM; 10mg in PM
	20 mg twice daily	10 mg twice daily
Definite[§, §§]	5 mg twice daily	HOLD
	10 mg twice daily	HOLD
	15 mg twice daily	HOLD
	20 mg twice daily	HOLD

[§] If the toxicity was determined to be "definitely" related to ruxolitinib, but at a later time point, the AE is NOT determined to be "definitely" related to ruxolitinib, then the patient may restart as if the AE has been determined to be "possibly" related or "unrelated" in consultation with Study Principal Investigator or designee.

[§§] If the toxicity was determined to be "definitely" related to ruxolitnib, and the AE has been resolved completely or has resolved to a grade 1 or 2 AE or has resolved to baseline, the investigator, after consultation with the Study Principal Investigator or designee, may restart the subject on the study drug at a reduced dose or the prior dosing level depending on the variables of the AE and clinical circumstances, keeping subject safety at the forefront of all considerations. The AE is not expected to recur based on the investigators' best clinical judgment and in consultation with the Study Principal Investigator or designee.

If there has been a new concomitant medication categorized as **a moderate or potent CYP3A4 inhibitor or there has been an increase in doses of ruxolitinib**, the investigator is encouraged to monitor platelet counts more frequently around the time of the change or dose adjustment. Investigators should refer to Tables 2 and 4.

Returned oral medication will be evaluated and compared to the amount dispensed and the amount that should have been taken by the subject to assess compliance with regimen. If the subject is <90% compliant, education on proper dosing will be provided to the subject and documented in source.

Investigators may also decrease the dose for any reason based on their clinical judgment, including a more conservative approach based on hematological abnormalities or non-hematologic safety findings.

Dosing must be held if any of the following occur:

- Platelet counts decline below 20,000 per microliter (dosing must be interrupted immediately).
- ANC declines to less than 500 per microliter.
- Ongoing Grade 3 or higher hemorrhage event. Ruxolitinib may be continued without hold if bleeding event is deemed ongoing from baseline or determined by site PI in consultation with Sponsor to be well controlled and unrelated to administration of ruxolitinib.

Dose Increases

Dose increases are optional and are at the discretion of the investigator. The investigator should refer to the prescribing information for ruxolitinib (Appendix B) and Table 2 (Section 5.1) as a general guideline based on platelet counts and concomitant moderate and potent CYP3A4 inhibitors. Dose increases of 5mg daily or twice daily increments and no more frequent than every 4 weeks are suggested. Subjects who have had a sub-therapeutic response (i.e., less than 25% reduction in white blood cells counts and absolute neutrophil counts) can have their dose increased to 25mg BID. For most patients, the maximal dose will be 20 mg BID, however, in some instances patients may receive 25 mg BID if the patient has demonstrated a positive, but partial response at 20 mg BID in the absence of a dose-limiting toxicity. The investigator will monitor platelet counts around the time of the change or dose adjustment per standard of care.

Note that when a 5 mg qd increment is added, creating a regimen such as 5 mg/10 mg, it is suggested that the higher dose should be taken in the evening and the lower dose in the morning.

No dose adjustment is recommended when ruxolitinib is co-administered with a CYP3A4 inducer. Patients should be closely monitored and the dose titrated based on safety and efficacy.

For subjects not on the maximum dose allowed for the study (ruxolitinib 25mg twice daily) but meets the WBC and splenomegaly criteria (table 3) for disease progression, the investigator may increase the dose of ruxolitinib up to the maximum tolerated dose or maximum dose allowed for the study (subjects must maintain platelet counts greater than 20,000 per microliter, section 5.5). The coordinating center will review the source data and provide suggestions to subsite investigators on dosing.

The subject may stay on study if the WBC (>50% reduction) OR spleen size (>25% reduction by ultrasound) is reduced after the subject is either on the maximum tolerated dose or the maximum dose allowed for the study for 2 consecutive months. There is still a perceived clinical benefit to stay on study and stay on drug if only the WBC or the spleen size is controlled.

Restarting ruxolitinib after Recovery of Platelet or ANC Levels or Resolution of Hemorrhage

- Note that the restarting doses should be reduced by at least 5mg twice daily from what the subject was on at the time of the platelet or ANC level or hemorrhagic event that warranted dose interruption.
- When dosing is held for ANC decline to less than 500 per microliter, dosing may be restarted when ANC is greater than 1,500 per microliter.
- For Grade 2 hemorrhage events of any causality (Appendix E), dosing may be restarted at the prior level once the event is resolved to Grade 0, and its underlying cause has been alleviated or resolved, as long as platelet count and ANC do not preclude a restart at that dose level.
- For Grade 3 or 4 hemorrhage events of any causality (Appendix E, requires a mandatory dose interruption), dosing may only be restarted provided: platelet count >20,000 per microliter and ANC >1,000 per microliter, the event has resolved to Grade 0, and the underlying cause has been alleviated or resolved, and following discussion with the Study Principal Investigator or designee taking into account causality, resolution of the bleeding and underlying etiology and likelihood of recurrence, in order to reach agreement that the potential benefits of a restart at an appropriate dose outweigh risks of recurrent hemorrhage. Re-escalation by 5mg daily or twice daily increments and no more frequent than every 4 weeks are suggested. Investigators should exercise caution and closely monitor platelet counts around the time of the change or dose adjustment.

Use of Platelet Transfusions

Platelet transfusions may be used at Investigator's discretion to treat thrombocytopenia. Dosing restart or increase decisions may not be based on platelet count values observed within 7 days of a platelet transfusion.

Management of Anemia

Subjects with a hemoglobin level < 6.5 g/dL must receive red cell transfusion(s) to maintain a level of ≥ 6.5 g/dL. Erythropoietin (EPO) use is strongly discouraged, but is permitted at the Investigator's discretion in consultation with the Study Principal Investigator or designee, although use is not permitted in lieu of transfusions for hemoglobin below 6.5 g/dL.

Optional Dose Tapering Strategy for Any Reason (medical indication, withdrawal from study, end of study, etc.)

In ongoing clinical studies with ruxolitinib, rapid return of constitutional symptoms and spleen growth has been observed in subjects after drug discontinuation. When a decision is made to permanently discontinue ruxolitinib therapy for reasons other than low platelet counts or ANC levels, tapering of ruxolitinib dosage may be considered, based on evaluation of the condition of the subject, current dose regimen and the clinical judgment of the investigator. Investigators may institute a tapering regimen according to what seems medically appropriate. Doses of ruxolitinib may be very slowly lowered until the subject is completely off drug. If medically indicated, doses of ruxolitinib may be temporarily restarted after they have been discontinued. If considered to be medically necessary, the investigator may use any treatment to manage withdrawal from ruxolitinib including, but not limited to, the management of events which may be secondary to discontinuation, and interruption or reduction of dose administered of ruxolitinib. Short-term courses of high-dose corticosteroids, equivalent to doses of > 10 mg/day of prednisolone have been used to moderate the withdrawal of ruxolitinib and may be considered as part of a tapering strategy. Corticosteroids may be started prior to, or concurrent with, ruxolitinib tapering in anticipation of the possibility of occurrence of withdrawal symptoms. When a decision has been made to discontinue the subject with utilization of a tapering strategy, regardless of the use of concomitant medications, safety data will continue to be assessed in accordance with the protocol. An End of treatment and subsequent Follow-up Visit must be scheduled.

Procedures for Interruption of Study medication
In some circumstances, it may be necessary to temporarily interrupt treatment as a result of adverse experiences that may have an unclear relationship to study drug. Refer to Table 5. The investigator is encouraged to consult with the Study Principal Investigator or designee for reasons other than protocol mandated medication holds for platelet or ANC counts. Any AE grade 3 or higher and RELATED to ruxolitinib requires mandatory reporting to the OHSU coordinating site within 24 hours of the site's knowledge of the event. Additionally, the investigator must notify the OHSU coordinating site via email before restarting study drug that was temporarily discontinued for an AE.

Procedures for Permanent Discontinuation of Study Drug
In the event that any subject discontinues the study drug and subsequently withdraws from 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 10.

The date the subject discontinued the study drug and the specific reason for discontinuation will be recorded. This will include reasons such as discontinuation 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.

7 ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of AEs (Section 7.1) and the characteristics of an observed AE (Section 7.2) will determine whether the event requires expedited reporting.

All AEs grade 3 and greater and all SAEs, regardless of grade, will be recorded in the CRF.

7.1 Adverse Events and Potential Risks List(s)

Potential Risks:
No specific findings in nonclinical repeat dose toxicity studies identify clinical risks 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. Appendix F lists the most common adverse effects. Dose-dependent, reversible thrombocytopenia has been observed in subjects with MF and represents the DLT. The risk of spontaneous hemorrhage in patients as a result of thrombocytopenia generally does not become manifest until platelet counts fall below 10,000 per microliter (Slichter, 2004); platelet counts below 25,000 per microliter (Grade 4) were not observed in any subjects initiating ruxolitinib at starting doses of 10 mg bid or 15 mg bid in the ongoing phase 1 /2 study. In the current study, dosing must be interrupted for platelet count below 20,000 per microliter, or for grade 3 or 4 hemorrhage event.

Anemia and, rarely, neutropenia have also been observed in patients with MF treated with ruxolitinib. 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.

7.2 Adverse Event Characteristics

CTCAE term (AE description) and grade: The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 4.0. A copy of the CTCAE version 4.0 can be downloaded from the CTEP web site: http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

Expectedness: Adverse Events can be 'Unexpected' or 'Expected' (see Section 7.1 above for expected AEs) based on available safety data.

Attribution of the AE:

- Definite – the AE *is clearly related* to the study treatment.
- Probable – the AE is *likely related* to the study treatment.
- Possible – the AE *may be related* to the study treatment.
- Unlikely – the AE *is unlikely related* to the study treatment, but a causal relation cannot be completely excluded.
- Unrelated – the AE *is clearly NOT related* to the study treatment.

7.2.1 Adverse Event (AE)

An adverse event (AE) is any undesirable sign, symptom or medical condition or experience that develops or worsens in severity after starting the first dose of study treatment or any procedure specified in the protocol, even if the event is not considered to be related to the study. Abnormal laboratory values or diagnostic test results constitute adverse events only if they induce clinical signs or symptoms or require treatment or further diagnostic tests.

7.2.2 Serious adverse event (SAE)

A serious adverse event (SAE) is any adverse event, occurring at any dose and regardless of causality that:

- Results in death
- Is life-threatening. Life-threatening means that the person was at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form.
- Requires or prolongs inpatient hospitalization (i.e., the event required at least a 24-hour hospitalization or prolonged a hospitalization beyond the expected length of stay). Hospitalization admissions and/or surgical operations scheduled to occur during the study period, but planned prior to study entry are not considered SAEs if the illness or disease existed before the person was enrolled in the trial, provided that it did not deteriorate in an unexpected manner during the trial (e.g., surgery performed earlier than planned).
- Results in persistent or significant disability/incapacity. Disability is defined as a substantial disruption of a person's ability to conduct normal life functions.
- Is a congenital anomaly or birth defect; or
- Is an important medical event when, based upon appropriate medical judgment, it may jeopardize the participant and require medical or surgical intervention to prevent one of the outcomes listed above. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home; blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

Events **not** considered to be serious adverse events are hospitalizations for:

- Routine treatment or monitoring of the studied indication, not associated with any deterioration in condition, or for elective procedures.
- Elective or pre-planned treatment for a pre-existing condition that did not worsen.
- Emergency outpatient treatment for an event not fulfilling the serious criteria outlined above and not resulting in inpatient admission.
- Respite care.

7.3 OHSU IRB Reporting of Unanticipated Problems and Adverse Events

Unanticipated Problems (UP) and Adverse Events (AE) will be reported to OHSU IRB according to the policies, procedures and guidelines posted on the OHSU IRB web site: http://www.ohsu.edu/research/rda/irb/policies.shtml.

Fatal and life-threatening events must be reported to OHSU IRB within 7 calendar days after the OHSU Study PI learns of the event. If any of these require a change (as determined by the PI or the IRB) to the protocol or consent form, the PI will make those changes promptly and submit the revised documents to the OHSU IRB.

All other UP reports will be submitted to OHSU IRB no later than 15 calendar days of notification of the event. If the event requires changes as determined by the PI or the IRB) to the protocol or consent form, the PI will make the changes promptly and submit the revised documents to the IRB. UP and AE reports are submitted through OHSU e-IRB and will be reviewed by OHSU IRB.

7.4 Central Reporting of Adverse Events for Multicenter Studies

The SAE/UP reporting for multicenter investigator initiated clinical trials will follow the guidelines outlined in the OHSU Knight Cancer Institute Multi-Center Investigator Initiated Trials Coordinating Center Operations Manual.

A participating site must report an SAE to the to the institution's local IRB for action as required, as well as to the OHSU coordinating center study team by phone, fax, or email within 24 hours of learning of the event. The participating center will send the coordinating center materials regarding the SAE including pertinent progress notes, hospital admission and discharge notes, laboratory and radiographic studies, and the investigators' assessment and report of the SAE.

The OHSU coordinating center study team will review and submit SAEs to the FDA, OHSU IRB, and any other required contacts as required by the Knight Data Safety Monitoring Plan, OHSU IRB policy and/or federal regulations. The principal investigator at the OHSU Coordinating Center is responsible for distributing IND Action Letters or Safety Reports, as applicable, to participating institutions for review and submission to their institution's local IRB.

7.5 MedWatch Reporting

For this investigator-initiated study, the OHSU Principal Investigator is the study sponsor. The sponsor-investigator is required to report adverse experiences to the FDA through the MedWatch reporting program, even if the trial involves a commercially available agent. Adverse experiences to be reported include any unexpected (not listed in the package label), serious adverse experiences with a suspected association to the study drug. Adverse events that occur during clinical studies are to be reported to FDA as specified in the investigational new drug/biologic regulations using the FDA 3500 Voluntary Reporting form, which is available for submission online at: http://www.fda.gov/Safety/MedWatch/HowToReport/ucm085568.htm and is also available as a .pdf.

Sub sites will report all serious adverse events to the study sponsor within 24 hours of knowledge of the event via the OHSU study-specific Safety Reporting Form, along with supporting materials. The sponsor will review the submitted SAE report and supporting documentation and submit to the FDA after review. Copies of the MedWatch 3500 form and formal safety report notification will be submitted to each sub site and OHSU IRB, when appropriate. A copy of the MedWatch 3500 form, supporting materials and documentation of site PI review will be kept on file in the study regulatory binder. The OHSU Coordinating Center is responsible for meeting the MedWatch reporting requirements for the study.

7.6 Sponsor or additional reporting requirements

Unexpected fatal or life-threatening experiences associated with the use of the study treatment will be reported to FDA as soon as possible but in no event later than 7 calendar days after initial receipt of the information.

All other serious unexpected experiences associated with the use of the study treatment will be reported to FDA as soon as possible but in no event later than 15 calendar days after initial receipt of the information.

Events will be reported to the FDA by submission online using FDA Form 3500 (Voluntary Reporting Form for commercial agents).

All events reported to the FDA will also be reported to Incyte Pharmacovigilance representative as provided in the Research Agreement within 24 hours of reporting. A copy of the FDA report will also be faxed to the Incyte Pharmacovigilance representative.

8 PHARMACEUTICAL AND/OR IMAGING AGENT INFORMATION

A list of the adverse events and potential risks associated with the investigational or commercial agents administered in this study can be found in Section 7.1 and in Appendix F.

8.1 Agent Accountability

The Investigator, or a responsible party designated by the Investigator at each site, must maintain a careful record of the inventory and disposition of the study agent. (See the NCI Investigator's Handbook for Procedures for Drug Accountability and Storage).
http://ctep.cancer.gov/investigatorResources/investigators_handbook.htm

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
- return 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 correct study medication specified.

Completed accountability records will be archived by the site. At the completion of the study, the Investigator or designee will oversee shipment of any remaining study drug back to Incyte for destruction according to institutional standard operating procedures. If local procedures mandate site destruction of investigational supply, prior written approval must be obtained from Incyte or Incyte's study drug distributor.

Please refer to the Pharmacy Manual located in the separate Study Operations Manual for details on drug supply, ordering and shipment process.

8.2 Study Agent(s)

Availability: Ruxolitinib will be provided by Incyte Corporation and shipped directly to each site by Incyte or its distributor. Each site will be responsible for contacting Incyte or its distributor for study drug supply.

Route of administration: Doses should be taken by mouth in the morning and evening, approximately 12 hours apart, and without regards to food.

Study Drug Packaging, Labeling and Preparation: Ruxolitinib Phosphate tablets will be provided as 5 mg tablets packaged as 60 count in high-density polyethylene bottles. All bottles of Incyte investigational product contain the following language: "Caution: New Drug—Limited by Federal law to investigational use."

Incyte will be responsible for quality and stability of the drug for shipment to sites. Incyte will also communicate any information related to manufacturing issues and expiration dates for each site's shipment.

Study Drug Storage and 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 clinical batches to support the clinical trial.

8.3 Commercial Agent(s) (if applicable)

Not applicable.

9 BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

We will determine the *mutational status* of CSF3R, the *mutation type* of CSF3R, and the CSF3R mutant *allele frequency*. We will correlate these findings with patient clinical responses.

Refer to the lab manual located in the separate Study Operations Manual

9.1 Biomarker Studies

Not applicable.

9.2 Laboratory Correlative Studies

Whole exome sequencing will be performed on genomic DNA isolated from patient peripheral blood, bone marrow specimens as well as germline skin biopsy specimens that are collected upon enrolment of patients on this trial. This will provide both qualitative and quantitative assessment of CSF3R and JAK2 mutational status as well as identification of other cooperating mutations such as SETBP1[34]. In addition, specimens

from any patients exhibiting a clinical response, followed by relapse will be re-sequenced (whole exome) to understand genetic contributions to drug resistance. In this manner, we will be able to study possible mechanisms of primary resistance (from initial whole exome sequencing) as well as secondary resistance (whole exome sequencing of relapse specimens). Overall, this will involve whole exome sequencing of both pre-treatment and post-treatment specimens in subjects with a response to treatment and only pre-treatment sequencing in non-respondents who discontinue study prior to primary endpoint (estimate for relapse based on experience of ABL inhibitors in CML; germline from enrolment specimen can be used to filter relapse specimens). Finally, genomic DNA from all CSF3R-mutant enrolment and follow-up bone marrow specimens will be analyzed to assess CSF3R mutation allele frequency using a customized ion torrent CSF3R assay, as discussed below in Section 9.3. Special Studies.

After testing exome sequencing is performed and the subject has consented to the storage of samples for this study, remaining materials will be stored in a biorepository for future research. The excess sample will be processed and stored in the OHSU laboratory, which is an NCI-approved biorepository (OHSU eIRB# 7267). The repository, which includes banked samples from participants with acute leukemia (AL), lymphoproliferative disorders (LPD), myelodysplastic syndrome (MDS), and myeloproliferative diseases (MPD). A primary goal of this repository is to store samples from patients with hematologic malignancies that can be made available to researchers at OHSU, the collaborating institutions, and commercial laboratories.

The biorepository will include de-identified, coded samples from subjects who consented to have their samples banked on this study; de-identified samples such as those from other institutions and cord blood samples, and "medical waste" samples such as left-over samples from the OHSU Clinical Flow Cytometry Laboratory and Clinical Leukapheresis Unit.

Coded, de-identified samples will be processed and frozen in liquid nitrogen freezers. Currently, the repository has a capacity of 17,000 samples. The number of stored vials per sample, the type of processing they received, and their location in the freezer will be included in the AL, LPD, MDS, and MPD database. When a sample is removed, the person who removed it and the destination are recorded in the database. The freezer will remain locked when not being accessed.

Stored patient samples are a limited and valuable resource. To assure their best possible use, requests for samples will be reviewed for scientific merit and IRB compliance. The group assembled to review sample requests will include the study PI as well as OHSU pathologists/oncologists and co-investigators. Coded samples and the key linking patient identifiers to clinical data will be released to OHSU investigators following group review and IRB approval from OHSU and the institution from which they were received. Only coded clinical information will be released.

Samples may also be released to commercial laboratories. These samples will be de-identified and protected information will not be released.

9.3 Special Studies

9.3.1 Determination of CSF3R Mutation Allele Frequency

9.3.1.1 We will measure the percentage of CSF3R ion torrent reads that are wild-type versus mutant.

9.3.1.2 Assessment

9.3.1.2.1 Method of Assessment

9.3.1.2.2 Allelic burden will be assessed on bone marrow specimens taken at study enrolment and again on bone marrow specimens taken at the 6-month interval.

9.3.1.3 Data Recording

9.3.1.3.1 Data will be recorded in the relational database that will be used to track specimens and all correlative data (see Section 12).

9.3.1.3.2 Data will be recorded upon performance of the assay.

10 STUDY PROCEDURES AND SCHEDULE OF EVENTS

10.1 Screening/Baseline

Screening and Baseline visit procedures can be combined in one visit or performed on more than one visit and will be completed within 4 weeks of Cycle 1 Day 1. Refer to Section 10.5 and 10.7 for more detailed description of study procedures. The coordinating center must review subject source data for verification of inclusion/exclusion criteria prior to subjects receiving drug on trial. Refer to the Study Operations Manual for specific enrollment instructions.

10.2 Study Treatment Visits

Each patient will be followed for a maximum of 96 weeks (24 cycles, 1 cycle is 4 weeks long). If the study drug continues to be effective, the patient may be eligible to continue on study drug past 24 cycles. If a patient continues on study drug past 24 cycles, standard of care visits and standard of care labs will be collected every 8 weeks. In addition, research data will be collected for the post-24 cycles extension period. If the study drug becomes commercially available for the disease indication during the duration of treatment, the study team will help transition the patient to commercial drug supply. There is no established standard of care for CNL and aCML. The schedule lists the mandatory data collected at each time point for the clinical trial but it is expected that additional tests/procedures or visits may occur as standard of care, which is entirely at the discretion of the investigator. Most patients will have additional laboratory studies and clinical evaluations outside of this schedule as part of their standard of care. Frequent monitoring is suggested for patients who have severe anemia or thrombocytopenia (for example, Hgb <7 g/dL and platelet counts <30,000 per microliter). The labs or studies outlined in section 10.8 schedule of events may be completed within 7 days of the target week (7 days before or 7 days after). However, every effort will be made to adhere to this schedule as close as possible. This is the minimum schedule of laboratory studies and follow-up visits required for the study

10.3 End of Treatment Visit

Subjects will be evaluated during Week 4 of Cycle 24 for end of treatment or within 2 weeks after stopping study drug. Every attempt will be made to devise a treatment plan weeks before the patient stops

the study drug. In those patients that withdraw from the study abruptly for any reason, a standard of care treatment plan will be formulated as soon as possible. Subject should be encouraged to return for the follow-up visit. Subjects will have the option to taper off ruxolitinib or go on commercial ruxolitinib therapy especially if there is a documented clinical benefit. See section 6.0 regarding approach for tapering ruxolitinib.

10.4 End of Study Visit

Subjects will be evaluated 4-6 weeks after stopping study drug for a safety evaluation. Reasonable efforts should be made to have the subject return for the follow up visit. Every attempt will be made to devise a treatment plan weeks before the patient stops the study drug. In those patients that withdraw from the study abruptly for any reason, a standard of care treatment plan will be formulated as soon as possible. Subjects will have the option to taper off ruxolitinib or go on commercial ruxolitinib therapy especially if there is a documented clinical benefit. See section 6.0 regarding approach for tapering ruxolitinib.

10.5 Study Assessments and Procedures

Dose adjustments or the occurrence of hematologic or non-hematologic grade 3 or 4 adverse events will require additional clinical evaluations and/or laboratory studies but should not be any less frequent than the schedule outlined.

- **Demographics and Medical History:** Demographic data and a complete medical history will be collected at visits prior to starting study treatment. Medical history in between visits should be documented at each study visit once study treatment has started. All transfusions including date, product transfused, number of units, and reason for transfusion should also be collected as part of the interval medical history.

- **Concomitant Medications:** All concomitant medications and treatments must be recorded in the case report form (CRF). Any prior medication received up to 30 days prior to the Baseline visit will be recorded in the CRF. Concomitant treatments that are required to manage a subject's medical condition during the study will also be recorded in the CRF. Prior and/ or ongoing medications will be reviewed during screening to determine subject eligibility. The medication record will be maintained following enrollment including any changes to the dose or regimen. Prior and concomitant medication including any prescription, over the counter or natural/herbal/multivitamin preparations taken will be recorded.

- **Physical Exam:** Physical exams must be performed by a medially qualified individual such as a licensed physician, Physician's Assistant or advanced Registered Nurse Practitioner as local law permits. The physical exam at baseline should include the following organ or body systems per institutional standards: skin, head, eyes, ears, nose, throat, thyroid, lungs, cardiovascular, abdomen, extremities, lymph nodes. All other physical exams will include an evaluation of any AEs or any previously reported symptoms or prior physical examination findings.

  Physical Exam must include an IPSS score at each time point as described in the Schedule of Events (one point for each criteria, maximum of 5 points total):

  - Age >65 y/o
  - Hgb <10 g/dL
  - Leukocytosis >25,000 per microliter
  - Peripheral blasts > or = 1%
  - Weight loss of >10% of average adult weight prior to leukemia diagnosis, or presence of disease-related night sweats or fevers

- **Performance Status:** ECOG will be determined and performed at the visits indicated in the schedule of events in section 10.8. Refer to Appendix C for ECOG scale.

- **Pregnancy Test:** A urine pregnancy test is required for all female subjects during screening for women of childbearing potential. If the urine pregnancy test is positive, serum pregnancy tests must be performed per institutional standards.

- **Weight:** Must be collected at each study visit

- **Height:** Must be collected prior to day of study treatment.

- **Vital Signs:** Vital sign measurements should be taken per institutional standards at each study visit.

- **Complete Blood Count:** Local hematology will be collected per institutional standard of care and must include an extended differential, hematocrit and MCV. The exact values of the extended differential profile is per local institutional standard of care.

- **Coagulation panel:** INR/PT and aPTT are required.

- **Chemistry:** Local chemistry will be collected per institutional standards and must include LDH and uric acid.

- **Spleen size:** In this study, spleen size is only estimated by ultrasound (limited ultrasound or full ultrasound per standard of care) at the mandatory time points as indicated in the schedule of events. Spleen volume will be calculated by the conventional prolate ellipsoid method. Measure spleen width, thickness and maximum length in centimetres. Multiply width by thickness by max length by 0.524 to get the total spleen volume in cm3. The longest dimension (centimetres) in the cranio-caudal axis is also recorded. A percentage of change will be calculated at the mandatory time points as indicated in the schedule of events in cases when only 2 dimensions are available. Studies completed within 2 months of start of drug date are acceptable for baseline evaluation. Additional ultrasounds may be completed during the study per investigator discretion. Beyond the mandatory ultrasounds to estimate spleen size, Investigators should estimate spleen response by palpation as part of the physical exam throughout the study and can be recorded in the clinical evaluation and in Medidata.. If the clinical assessment suggests disease progression (refer to table 3), the investigator will perform repeat labs, bone marrow evaluation, and ultrasound of the spleen as part of standard of care to confirm the presence or absence of suspected disease progression.

- **Bone Marrow Exam:** A bone marrow exam aspirate must be collected at the visits outlined in the Schedule of Events and include the elements listed below per standard of care. Additional bone marrow evaluations may occur if clinically indicated at the discretion of the investigator, e.g., at the time of suspected disease progression or disease remission. An independent hematopathologist(s) not involved in the study will adjudicate availablebone marrow biopsy slides to enhance the assignment of the diagnostic category for each subject given some existing arbitrary or ill-defined WHO criteria.

  If a bone marrow exam has been done within the last 90 days, the actual slides and the hematopathology report may be reviewed and used in place of repeating a bone marrow evaluation so long as the studies described below have been performed and the sample is of sufficient quality for interpretation. Investigators from other sites may consult with the coordinating center (OHSU Coordinating Center) if necessary to help make this determination.

  Bone Marrow requirements:

- Hematopathology
- Cellularity %
- Blasts %, by morphology
- CD34+ blasts by immunohistochemistry staining %, if collected as standard of care per institutional standards
- Degree of granulocytic hyperplasia %, if collected as standard of care per institutional standards
- Degree of granulocytic dysplasia %, if collected as standard of care per institutional standards
- Degree of reticulin fibrosis (reticulin staining) WHO grade 0-3, if collected as standard of care per institutional standards
- Degree of collagen fibrosis (trichome staining) ELN/international consensus grade 0-3, if collected as standard of care per institutional standards
- Cytogenetics and MDS/AML FISH panel: can be performed if collected as standard of care per institutional standards
- Flow cytometry: can be performed if collected as standard of care per institutional standards
- Central lab aspirate as described in the central lab manual located in the separate Study Operations Manual

- **Symptom Score:** Symptom score assessment using a modified MPN-SAF a tool developed by Dr. Ruben Mesa to assess total symptom score[35]. A simplified version is based on the 10 most common symptoms associated with MPNs. The total score is the sum of each symptom scored on a 0 to 10 scale (0 describes no symptoms and 10 is maximal symptoms). The maximum total score is 100. If subject does not circle a line within the survey (skipped question), the data is considered missing and then the denominator of total symptom score will be changed to the total number of questions answered. If more than one value is circled by the subject then values will be averaged and entered as a single value.

- **Adverse Event assessments:** Toxicities and adverse experiences will be assessed at each visit using the NCI Common Toxicity Criteria for Adverse Events 4.0. See Appendix F for grading of the most common adverse events and side effects with ruxolitinib. Unexpected grade 3 or 4 adverse events (according to CTCAE term (AE description)) categorized as definitely or possibly attributed to ruxolitnib will also be recorded and reported as indicated in section 7. http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm

  All AEs grade 3 and greater and all SAEs, regardless of grade, will be recorded in the CRF.

- **Central Samples:** A clinical specimen is sent via overnight shipping to the main site (OHSU) at the following time points:

  For detailed instructions on collection and shipping refer to the Lab Manual located in the separate Study Operations Manual

  - Peripheral Blood (10ml in sodium heparin-coated tubes): Collected at the time points listed in the Schedule of event Calendar. In addition, a sample is sent at the time of confirmed or suspected disease progression, if applicable.

  - Bone Marrow Aspirate: 5-8 ml in sodium heparin-coated tubes collected at the time of the bone marrow evaluations as listed in the schedule of events calendar. In lieu of repeating a

baseline bone marrow evaluation, slides and hematopathology reports of sufficient quality will be accepted for subjects who had a bone marrow exam within 90 days of cycle #1, day one.

- Skin Biopsy (3.5 mm punch biopsy, one time during the entire study): A skin sample will be excised at the site of the bone marrow biopsy incision, preferably collected at baseline. However, if a subject does not have a bone marrow biopsy collected at baseline, the skin biopsy may be collected beginning of Cycle 7 or at EOT/early termination bone marrow exam.

10.6 Clinical Response

Clinical Response will be evaluated at the time points listed in the schedule of events calendar:

Table 6: Assessment occurs at every clinical evaluation visit and compared to start of study (day 1, cycle 1) evaluation (see Schedule of Events, Section 10.3)[§]

Parameter	Progression	Stable disease	Partial response	Complete response
WBC and ANC, blood	See Table 3	Less than 50% reduction	>50% reduction	Normal range
Morphologic evaluation of granulocytic pre-cursors, bone marrow	See Table 3	No major reduction in granulocytic hyperplasia (CNL) or dyspoiesis (aCML)	>50% reduction in granulocytic hyperplasia (CNL) or granulocytic dyspoiesis (aCML)	No evidence of granulocytic hyperplasia (CNL) or dyspoiesis (aCML)
Spleen size estimated by palpation or ultrasound[†]	See Table 3	No change	> 25% reduction	Normal spleen size

[§]Some subjects will not perfectly meet all the criteria (WBC/ANC, marrow findings, or spleen size) in any one of these categories (CR, PR, SD, or progression). The coordinating center will confirm each parameter separately and will choose the best category that reflects the overall hematologic response based on source documentation. Three or more reviewers (co-PIs) will review a summarized table of these parameters with redacted data and also assign the category that best fits the hematologic response in evaluable subjects at cycle 7, week 1. The overall consensus and rationale for each response assessment will be documented. Subjects not reaching cycle 7, week 1 are considered non-responders unless otherwise indicated. In addition to overall response, the change in each parameter (WBC/ANC, marrow findings, or spleen size) at cycle 7, week 1 and at end of study compared to baseline will be reported and evaluated separately.

[†]In this study, spleen size is estimated in centimeters by ultrasound (longest dimension in the cranio-caudal axis) at baseline, cycle 7, week 1 and end of treatment when spleen size must be measure by ultrasound. Additional ultrasounds will be performed if disease progression is suspected at any time during the study (table 3).

[‡]Complete response, partial (CRp) is defined as responses that meet CR criteria as outlined in this table but platelet counts remain below the laboratory's normal range.

10.7 Schedule of Events

Please note that this is the **mandatory (minimum) schedule of events** required for data collection in this study in tables. In most cases, patients will require additional laboratory or clinical evaluations as part of the patients' standard of care.

Table 7: Screening, Baseline, and Cycles 1 through 3

Visit or Cycle #	**Screening/Baseline**	**C1**				**C2**		**C3**	
Cycle Week		**1**	**2**	**3**	**4**	**1**	**3**	**1**	**3**
Study Windows	**Within 4 weeks of Cycle 1, Day 1**	**+/- 7 days (every effort should be made to adhere to actual target dates)**							
Consent	X								
Study registration	X								
Study Drug Ruxolitinib dispensation PO		X				X		X	
Study Drug Ruxolitinib administration PO		X-DAILY DOSING							
Pregnancy Test- Urine (if applicable)	X								
Physical Exam, medical history, ECOG, IPSS	X	X				X		X	
Height	X								
Weight	X	X				X		X	
Vital Signs (BP, HR, RR, O2 Sat, Temp)	X	X				X		X	
CBC w/ extended differential[1]	X	X	X	X	X	X	X	X	X
Coagulation Panel: INR/PT and aPTT	X								
Chemistry panel with LDH and Uric Acid	X	X	X	X	X	X	X	X	X
Spleen Measurement- Ultrasound	X								
Bone marrow Exam	X[2]								
Central Lab: Research Bone marrow Aspirate Sample[3]	X[2]								
Central Lab: Research Blood sample[3]	X	X						X	
Central Lab: Research Skin Punch Biopsy Sample at time of Bone marrow Biopsy (performed only once during the entire study)[4]	X[4]								
QOL Questionnaire- MPN-SAF	X							X	
AE assessment		X	X	X	X	X	X	X	X

[1]The exact values of the extended differential profile is per local institutional standard of care.
[2]A bone marrow evaluation completed within 90 days prior to Cycle 1, Day 1 may be used as the screening/baseline evaluation.
[3]Please send blood and marrow samples also at time of confirmed disease progression.
[4]If skin punch biopsy was not done at the screening/baseline period the sample may be collected at the time of the bone marrow evaluation at Cycle 7, early termination, or end of treatment.

Table 8: On Study Treatment, Cycles 4 through 12

Cycle #	C4	C5	C6	C7	C8	C9	C10	C11	C12
Cycle Week	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**
Study Windows	**+/- 7 days (every effort should be made to adhere to actual target dates)**								
Study Drug Ruxolitinib dispensation PO	X	X	X	X	X	X	X	X	X
Study Drug Ruxolitinib administration PO	X-DAILY DOSING								
Physical Exam, medical history, ECOG, IPSS		X		X		X		X	
Weight		X		X		X		X	
Vital Signs (BP, HR, RR, O2 Sat, Temp)		X		X		X		X	
CBC w/ extended differential[1]	X	X	X	X	X	X	X	X	X
Coagulation Panel: INR/PT and aPTT				X					
Chemistry panel with LDH and Uric Acid	X	X	X	X	X	X	X	X	X
Spleen Measurement- Ultrasound				X					
Bone marrow Exam				X[2]					
Central Lab: Research Bone marrow Aspirate Sample[2, 3]				X					
Central Lab: Research Blood sample[3]				X					
QOL Questionnaire- MPN-SAF				X					
AE assessment	X	X	X	X	X	X	X	X	X

[1]The exact values of the extended differential profile is per local institutional standard of care.
[2]If skin punch biopsy was not done at the screening/baseline period the sample may be collected at the time of this bone marrow evaluation. Performed only once during the entire study.
[3]Please send blood and marrow samples also at time of confirmed disease progression.

Table 9: On Study Treatment, Cycles 13 through 24[1]

Cycle #	C 13	C 14	C 15	C 16	C 17	C 18	C 19	C 20	C 21	C 22	C 23	C 24[1]
Cycle Week	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**	**1**
Study Windows	**+/- 7 days (every effort should be made to adhere to actual target dates)**											
Study Drug Ruxolitinib dispensation PO	X	X	X	X	X	X	X	X	X	X	X	X
Study Drug Ruxolitinib administration PO	X-DAILY DOSING											
Physical Exam, medical history, ECOG, IPSS	X		X		X		X		X		X	
Weight	X		X		X		X		X		X	
Vital Signs (BP, HR, RR, O2 Sat, Temp)	X		X		X		X		X		X	
CBC w/ extended differential[1,2]	X	X	X	X	X	X	X	X	X	X	X	X
Coagulation Panel: INR/PT and aPTT[1]							X					
Chemistry panel with LDH and Uric Acid[1]	X	X	X	X	X	X	X	X	X	X	X	X
Spleen Measurement- Ultrasound												
Bone marrow Exam												
Central Lab: Research Bone marrow Aspirate Sample[3]												
Central Lab: Research Blood sample[3]	X						X					
QOL Questionnaire- MPN-SAF	X						X					
AE assessment	X	X	X	X	X	X	X	X	X	X	X	X

[1]If a patient continues on study drug past 24 cycles, standard of care visits and standard of care labs will be collected every 8 weeks. Research data will be collected for the extension period.
[2]The exact values of the extended differential profile is per local institutional standard of care.
[3]Please send blood and marrow samples also at time of confirmed disease progression.

Table 10: Early termination, End of Treatment, and End of Study Visits

Visit **Cycle Week**	**Early termination (ET) or End of treatment (EOT)**	**End of study safety visit**
Study Windows	**Within 14 days of ET or During week 4 of Cycle 24[1] for EOT**	**Within 4-6 weeks after last dose of study drug**
Physical Exam, medical history, ECOG, IPSS	X	X
Height		
Weight	X	X
Vital Signs (BP, HR, RR, O2 Sat, Temp)	X	X
CBC w/ extended differential[1, 2]	X	X
Coagulation Panel: INR/PT and aPTT[1]	X	
Chemistry panel with LDH and Uric Acid[1]	X	X
Spleen Measurement- Ultrasound	X	
Bone marrow Exam	X[3]	
Central Lab: Research Bone marrow Aspirate Sample[3, 4]	X	
Central Lab: Research Blood sample[4]	X	
QOL Questionnaire- MPN-SAF	X	
AE assessment	X	X

[1]If a patient continues on study drug past 24 cycles, subjects will be evaluated for EOT within 2 weeks after stopping study drug.
[2]The exact values of the extended differential profile is per local institutional standard of care.
[3]If skin punch biopsy was not done at the screening/baseline period the sample may be collected at the time of this bone marrow evaluation. Performed only once during the entire study.
[4]Please send blood and marrow samples also at time of confirmed disease progression.

MEASUREMENT OF EFFECT

10.6 Antitumor Effect – Solid Tumors
Not applicable.

10.7 Antitumor Effect – Hematologic Tumors
To measure the antitumor effects of ruxolitinib on CNL or aCML cells, subjects' white blood cell count and absolute neutrophil count will be determined according to the suggested schedule. All of these laboratory parameters are expected to be elevated above the standard cut-off for normal control patients. The mutant CSF3R allele burden is expected to correlate with tumor burden.

10.8 Other Response Parameters
Other response parameters measured include durability of responses, clinical outcome, effects on spleen size, and change in total symptom score modified MPN-SAF.

11 DATA REPORTING/REGULATORY REQUIREMENTS

11.1 Data Collection and Storage
Information for each participant will be collected and entered directly into an existing electronic relational database. Each site will be responsible for entry of data for patients accrued at that site with all correlative data entered by personnel at OHSU, as detailed below.

Medidata Rave, the database that will be used for this trial, was built to house voluminous, diverse data sets in a three-dimensional, relational setting whereby all information for a given patient can be viewed and correlated with all other pieces of information. This database was launched in October 2011. It is a web-based interface that references data, which is stored on a secure server such that all information collected for any patient can be traced through a hierarchy to show the exact lineage of that information stream. In addition, a sophisticated query/report function allows for complex analyses of data that has been collected and can thus facilitate integration of diverse data types. This database is supported by a web-portal login, and team members currently teams at all trial sites will have login credentials and have been trained in procedures for entering, accessing and storing data. This will facilitate information being stored in a unified format and location, which will be important for the long-distance collaborations of this trial.

This database utilizes a tiered user format whereby each user is defined based on access to read/edit each field as well as privileges of viewing protected health information (PHI). Since every data field is strictly defined as to whether PHI is present in that field, users lacking PHI privileges will be completely shielded from viewing such information. In addition, all information is stored on an encrypted server and access to the database is password protected. Finally, this database undergoes a rigorous back-up and audit trail such that information is never in danger of being permanently lost. Hence, the security and integrity of this database far surpasses requirements for data storage.

Clinical and demographic information about each patient (i.e. gender, race, ethnicity, date of birth, white blood cell count, extended differentials, karyotype/cytogenetics, etc.) will be entered directly into this database by each site at which a new patient is enrolled onto the trial. Data from correlative studies will be entered by laboratory personnel at OHSU.

11.2 Quality assurance will be conducted as outlined in section 12.7 under data safety and monitoring. Data, including PHI, will be exchanged with Incyte through a secure portal. PHI is limited to date of birth and disease diagnosis. No further patient identifiers will be included. Multicenter Guidelines

This protocol will adhere to the guidelines of the OHSU Knight Multi-Center Investigator Initiated Trials Coordinating Center Operations Manual http://ozone.ohsu.edu/cancer/sharedres/kctoresdocs.cfm

OHSU Coordinating Center will manage trial data in the following ways:

- Confirm that all sites have received and are using the most recent version of the protocol. The protocol must not be rewritten modified by anyone other than the OHSU Knight Principal Investigator. Documentation of the version that was sent to the site must be kept in the regulatory binders.
- Confirm that the protocol and informed consent form have local IRB approval at each site prior to registration of the first subject. Documentation of IRB approval from other sites for continuing review must be submitted and kept in the binder.
- Provide centralized subject registration in the clinical research management system (eCRIS)
- Ensure collection and review of applicable source documents and case report by the OHSU PI to ensure protocol compliance
- Maintain documentation for all SAE reports and submit a summary of all AE, SAE and unanticipated problems (UP) to the Knight DSMP
- Prepare quarterly summary reports of SAEs and UPs from all sites
- Ensure that relevant IRB correspondence and study status changes are communicated to all participating sites within five business days. Any changes that affect patient safety of study enrollments will be communicated immediately
- Submit documentation to the FDA such as protocol amendments, annual reports, SAE reports for unexpected, fatal or life-threatening events that are associated with the use of the study drug

Participating sites must submit regulatory documents including, but not limited to the following:

- Current CV (signed and dated) for each investigator
- Current medical license number for physician investigators
- Current signed FDA Form 1572
- Certificate of completion of institution-required human subject training course, the NIH online training in the protection of human research subjects or other appropriate training
- Documentation of institutional Conflict of Interest
- IRB approved site-specific ICF (must be reviewed and approved by OHSU PI and study team prior to submission to the local IRB
- All IRB approved documents and approval memos
- Delegation log
- Data Safety and Monitoring Plan (DSMP)
- Completed Case Report Forms within 10 business days of study visit

11.3 Protocol Review

The protocol and informed consent form for this study must be reviewed and approved in writing by the OHSU Knight Cancer Institute Clinical Research Review Committee (CRRC) and the appropriate Institutional Review Board (IRB) prior to any subject being consented on this study. All sites must have IRB approval of protocol by the IRB of record before consenting any subjects.

11.4 Informed Consent

Written informed consent will be obtained from all subjects, or the legally authorized representative of the subject, participating in this trial, as stated in the Informed Consent section of the case of Federal Regulations, Title 21, Part 50. If a subject's signature cannot be obtained, and for all subjects under the age of 18, the investigator must ensure that the informed consent is signed by the subject's legally authorized representative. Documentation of the consent process and a copy of the signed consent shall be maintained in the subject's medical record. All sites must have IRB approval of ICF by the IRB of records before consenting any subjects.

11.5 Changes to Protocol

Any modification of this protocol must be documented in the form of a protocol revision or amendment signed by the principal investigator and approved by the CRRC and IRB, before the revision or amendment may be implemented. The only circumstance in which the amendment may be initiated without regulatory approval is for a change necessary to eliminate an apparent and immediate hazard to the subject. In that event, the investigator must notify the CRRC and IRB in writing within 10 working days after the implementation. Investigators holding the IND must notify FDA of substantive changes to the protocol. Participating site must submit proposed changes to protocol to the OHSU Coordinating Center for review and endorsement before participating site may implement changes.

11.6 Maintenance of Records

Records and documents pertaining to the conduct of this study, source documents, consent forms, laboratory test results and medication inventory records, must be retained by the investigator for a period of 2 years following the date a marketing application is approved for the drug for the indication for which it is being investigated; or, if no application is to be filed or if the application is not approved for such indicate, until 2 years after the investigation is discontinued and FDA is notified. If the investigator relocates or for any reason is unable to retain the records, the study records must be transferred to an agreed upon designee, such as another institution, another investigator, or to OHSU Knight Cancer Institute Clinical Trials Office. Records must be maintained according to sponsor or FDA requirements.

11.7 OHSU IRB Reporting of Unanticipated Problems and Adverse Events

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 7.0 (Adverse Events: List and Reporting Requirements).

11.8 OHSU Knight Cancer Institute Data and Safety Monitoring Plan

In addition to complete study and pharmacy files, complete records must be maintained on each subject treated on this protocol. OHSU Knight Cancer Institute, through the auditing function of the Knight Data and Safety Monitoring Committee, is responsible for ensuring that all member investigators and affiliate investigators conduct clinical research studies in compliance with local IRB standards, FDA regulations and NIH policies and in accordance with the Data and Safety Monitoring Plan policies and procedures: http://ozone.ohsu.edu/cancer/sharedres/kctoresdocs.cfm

Locally initiated studies will be audited by OHSU Knight Data and Safety Monitoring Committee Auditor. Newly approved studies may be audited any time after enrollment has been initiated. Each OHSU Knight approved treatment protocol will be audited on an annual basis in accordance with the Knight Data and Safety Monitoring Plan. The OHSU Coordinating Center will audit records maintained at all participating centers and if necessary, visit the center to review records and provide training.

It is the responsibility of each participating site's principal investigator to ensure that the study is conducted in compliance with local IRB standards, FDA regulations, and NIH policies. It is also the responsibility of each site's principal investigator to ensure that quality assurance audits at their site are

conducted according to their institution's policies and procedures. The quality assurance audit process provides assurance that reported data accurately reflects the data in the primary subject record.

11.9 Inclusion of Women, Minorities and Children

11.9.1 Inclusion of Women and Minorities

No OHSU Knight Cancer Institute study will focus on any particular gender, racial or ethnic subset. No subject will be excluded from the study on the basis of gender, racial or ethnic origin. Male, female and minority volunteers will be recruited for this study from the general population and approximately 50% men and 50% women will be studied. The projected gender, racial, and ethnic composition of the study will represent that of the state of Oregon.

Table 11: Population Demographics - Oregon (%)

Ethnic Category	**Sex/Gender**		
	Females	Males	Total
Hispanic or Latino	5.9	5.8	11.7
Not Hispanic or Latino	44.5	43.8	88.3
Ethnic Category: Total of all subjects*	50.4	49.3	100*
Racial Category			
American Indian or Alaskan Native	0.7	0.7	1.4
Asian	1.9	1.8	3.7
Black or African American	0.9	0.9	1.8
Native Hawaiian or other Pacific Islander	0.2	0.1	0.3
White	42.1	41.5	83.6
More than one race	1.9	1.9	3.8
Unknown/Other	2.7	2.6	5.3
Racial Category: Total of all subjects*	50.4	49.5	100*
TOTALS	50.4	49.6	100*

Table 12: Projected Accrual for the Present Study

Ethnic Category	**Sex/Gender**			
	Females	Males	Unknown	Total
Hispanic or Latino	3	3	0-1	6
Not Hispanic or Latino	22	22	0-1	44
Unknown	0-1	0-1	0-1	0-1
Ethnic Category: Total of all subjects*	25	25	0-1	50*
Racial Category				
American Indian or Alaskan Native	0-1	0-1	0-1	0-1

Ethnic Category	Sex/Gender			
	Females	Males	Unknown	Total
Asian	1	1	0-1	2
Black or African American	0-1	0-1	0-1	1
Native Hawaiian or other Pacific Islander	0-1	0-1	0-1	0-1
White	21	21	0-1	42
More than one race	1	1	0-1	2
Unknown	1	1	0-1	3
Racial Category: Total of all subjects*	25	25	0-1	50*

Source: Adapted from U.S. Census Bureau, 2010 *Totals may not equal 100 due to rounding.

11.9.2 Inclusion of Children

In accordance with NIH guidelines on the inclusion of children as participants in research involving human subjects, children under the age of 18 years must be included in all human subjects' research, conducted or supported by the NIH, unless there are clear and compelling reasons not to include them. Therefore, proposals for research involving human subjects must include a description of plans for the inclusion of children. This protocol does not include children for the following reason because the number of children with this type of cancer is limited; therefore, children are excluded from this study but will be eligible for future pediatric trials with this study agent.

12 STATISTICAL CONSIDERATIONS

12.1 Study Design

This is an open label single-arm phase II clinical trial of ruxolitinib in patients with CNL and aCML. The study is comprised of 4 stages:

1) Screening: up to 4 weeks.
2) Baseline: up to 2 weeks.
3) Treatment: up to 96 weeks(extended if benefit to the patient).
4) Follow-up: and end of treatment visit will occur within 2 weeks of the last dose of ruxolitinib. An end of study visit will occur 4-6 weeks after the last dose of ruxolitinib while on study.

12.2 Primary and Secondary Endpoints

The *CSF3R* mutations represent a biologically unifying feature of CNL and atypical CML and define a new molecular subset of hematologic cancers.[15] CSF3R has been shown to signal through downstream SRC family and JAK-kinase pathways. The two types of *CSF3R* mutations may have differential susceptibility to classes of tyrosine kinase inhibitors, with *CSF3R* truncation mutations showing activation of SRC family-TNK2 kinase signaling and sensitivity to dasatinib, and *CSF3R* membrane proximal mutations showing preferential activation of the JAK signaling pathway. However, these are based on *in vitro* studies and we predict that the majority of CNL and atypical CML will have some degree of hematologic response with ruxolitinib treatment. Our observation that a patient with membrane proximal mutation had an excellent clinical response to the JAK inhibitor ruxolitinib, resulting in a marked decrease in the numbers of white cells and neutrophils and an increased platelet count, constitutes a proof of concept. Although anecdotal,

this observation provides an impetus for further investigation of tyrosine kinase inhibitors for the treatment of patients with CNL and atypical CML.[15]

Primary Endpoints:

The primary objective of this study is to determine the proportion of subjects with a hematologic response to ruxolitinib (PR, CR, CRp, as defined in Table 6). The primary endpoint is the indicator variable of whether a patient has achieved.

- A subject is defined as being responsive if he or she has achieved PR, CR, or CRp, determined at the clinical and laboratory evaluations performed at either the beginning of cycle #7 and/or at the end of treatment visit compare to start of study (day 1, cycle 1).
- A subject is defined as a non-responder if he or she does not reach cycle 7, week 1 unless otherwise specified.

Secondary Endpoints:

We will evaluate safety endpoints, including:

- The indicator variable for any hematologic grade III or IV adverse effects for thrombocytopenia, anemia, and neutropenia at any time point.
- The indicator variable for any grade III or IV adverse events or any effects/toxicities directly attributed to study drug requiring permanent cessation of drug (refer to Appendix F).

We will evaluate these other endpoints, including:

- The indicator variable for whether a patient has achieved clinical responses of PR or better at the clinical and laboratory evaluations performed at the beginning of cycle #7 This will be used to compute the proportion of such patients among all patients who carry a mutant CSF3R and have a more than 25% reduction in mutant CSF3R allele burden with ruxolitinib therapy compare to start of study (day 1, cycle 1).
- The indicator variable for drop-off and the time of drop-off since treatment.
- The indicator variable of dropping off before cycle 3.
- The indicator variable of dropping off between cycle 3 and 6.
- The indicator variable that subjects has reached cycle 7.
- Maximum clinical responses
- Duration of maximum clinical responses.
- Reduction of spleen size evaluated by ultrasound at the beginning of cycle #7 compare to start of study (day 1, cycle 1).
- Reduction of total symptom score as measured by a modified MPN-SAF at the beginning of cycle #7 compare to start of study (day 1, cycle 1).
- Overall survival in subjects who complete minimum of 1 dose of study drug. Verification of status (living or deceased) and cause of death will be attempted, by mail or phone call, up to 5 years after enrollment in the study.
- Clinical benefit and response analysis will be evaluated by applicable IWG criteria Blood. 2015;125(12):1857-1865

12.3 Analysis Populations

Safety and efficacy analysis will be conducted on every patient who consents and takes study drug, regardless of how long they stayed on study drug. The end of treatment and early termination visit (see schedule) includes an evaluation of response at that visit. Reason(s) for going off study will be collected

on every patient.

12.4 Statistical Analysis Plan

Descriptive statistical analysis will be conducted for all primary and secondary endpoints. In particular, we will compute proportions with 95% exact confidence intervals for categorical variables, and report the summary statistics (mean, standard deviation, median, interquartile range) for continuous variables. An empirical plot of trend will be made to graphically illustrate the change over time for peripheral blood WBC and ANC, CSF3R allele burden, spleen size, and total symptom score. Chi-square tests will be used to assess the association between hematologic response and mutant CSF3R type, and "≥ 50% reduction mutant CSF3R allele burden". Odds ratios and the corresponding 95% confidence intervals will be reported. Kaplan-Meier methods will be used to illustrate and summarize overall survival in subjects who complete the study.

Safety and tolerability will be assessed by monitoring the frequency, duration and severity of adverse events, performing physical examinations, collecting vital signs, collecting laboratory data for hematology, serum chemistry, and coagulation parameters. Proportion of subjects with new onset of Grade 4 thrombocytopenia events, and proportion of subjects with new onset of Grade 3 or higher hemorrhage, as measured by CTCAE v 4.03 will be tabulated with summary statistics. The hazard functions of time to onset of the above two safety measures will be estimated using life table method.

Clinical Laboratory Tests

The clinical laboratory data will be analyzed using summary statistics (e.g., means and frequencies), and no formal statistical comparisons among the treatments are planned.

12.5 Sample Size and Power

This study has an accrual goal of 25 evaluable subjects who reach cycle 7, week 1. Using the criteria that has been generally accepted in general oncology practice, we consider the therapy promising if it has an estimated 30% or more response rate (including CR, CRp, or PR, defined in Table 6, reference 36). The null hypothesis to be tested is 10% response rate[36]. CNL and aCML are rare diseases with no established standard of care or effective drug therapies. As a result, there is no best available therapy to compare to. Based on Simon's two-stage Minimax design, a total of 25 subject will achieve 80% power at 5% significance level, with an interim look planned upon the available of primary endpoint for the first 15 evaluable subjects (i.e., 15 patients have reached the beginning of cycle #7). The trial will be stopped for futility if 1 or 0 subjects responded (response is defined as PR or better) for the first 15 subjects. Under the null hypothesis, the probability of stopping the trial early is 0.549. The expected sample size is 19.50 for the two-stage design. By the end of the trial (assuming the trial did not get stopped early for futility), if 6 or more subjects responded (out of a total of 25), then the drug will be claimed as effective. We expect that each site will on average accrue 5 patients over 2 years. We expect that the majority of subjects will be enrolled by year 3. Considering the drop-off rate of ~ 50%, we may end up accruing 50 subjects for the trial (with only 25 evaluable for the primary endpoint).

12.6 Randomization Method

Not applicable.

12.7 Handling of Missing Data

Every attempt will be made to obtain data at the defined time points as described in the primary and secondary endpoints. The labs or studies outlined in section 10.3, schedule of events, may be completed within 7 days of the target week (7 days before or 7 days after). However, every effort will be made to adhere to this schedule as close as possible. For time points that have no data, we will evaluate whether or not the other time points can be used to fulfill the primary and secondary data. If the data is not sufficient

to analyze specific endpoints, the subject's data may be excluded entirely or partially, depending on the specific endpoints in question and in consultation with the biostatistician.

APPENDIX A CRITERIA FOR DIAGNOSIS OF CNL AND aCML

Chronic neutrophilic leukemia

- Peripheral blood
  - Leukocytosis ≥25 x 10^9/L
  - Segmented neutrophils and bands >80% of leukocytes
  - Immature granulocytes <10% of leukocytes*
  - Myeloblasts <1% of leukocytes
- Bone marrow
  - Hypercellular bone marrow
  - Increased number and percentage of neutrophilic granulocytes
  - Nucleated marrow cells, with <5% myeloblasts
  - Normal neutrophilic maturation pattern
- No Philadelphia chromosome or *bcr/abl* fusion gene
- No other cause for neutrophilia
  - No infectious or inflammatory process
  - No evidence of another myeloproliferative disease
  - No evidence of a myelodysplastic or myelodysplatic, myeloproliferative disease
  - No evidence of a tumor or, if present, myeloid cells must show clonality

  Note: For this study, leukocytosis should persist for at least 2 months, either by historical laboratory results or subsequent follow-up laboratory results.

Atypical chronic myeloid leukemia

- Peripheral blood
  - WBC count > 13 x 10^9/L
  - >10% neutrophil precursors (promyelocytes, myelocytes, metamyelocytes*), basophils <2% of leukocytes, monocytes <10% of leukocytes, blasts <20% of nucleated cells
- Bone marrow
  - Hypercellular marrow with granulocytic proliferation and dysplasia**
  - Blasts <20% of nucleated cells
- No BCR/ABL1 gene fusion, no rearrangement of PDGFRA, PDGRB or FGFR1

  Note: For this study, leukocytosis should persist for at least 2 months, either by historical laboratory results or subsequent follow-up laboratory results.

* Day to day fluctuation of immature granulocytes frequently occurs in patients with CNL or aCML, therefore the intent is for the patient to meet diagnostic criteria based on their typical % immature granulocytes.

**Patients who don't meet the dysplasia or immature granulocytes criteria are eligible if they have oncogenic CSF3R mutations.

APPENDIX B SUMMARY OF STARTING DOSE AND DOSE ADJUSTMENTS FROM PRESCRIBING INFORMATION FOR RUXOLITINIB

The information in this appendix is for reference only from the prescribing information. See Table 2 in the main protocol for dosing SUGGESTIONS for this clinical trial.

Proposed ruxolitinib Starting Doses

Platelet Count	**Starting Dose**
Greater than 200 X 10^9/L	20 mg orally twice daily
100 X 10^9/L to 200 X 10^9/L	15 mg orally twice daily

Maximum Restarting Doses for ruxolitinib After Safety Interruption

Current Platelet Count	**Maximum Dose When Restarting ruxolitinib Treatment**
Greater than or equal to 125 X 10^9/L	20 mg twice daily
100 to less than 125 X 10^9/L	15 mg twice daily
75 to less than 100 X 10^9/L	10 mg twice daily for at least 2 weeks; if stable, may increase to 15 mg twice daily
50 to less than 75 X 10^9/L	5 mg twice daily for at least 2 weeks; if stable, may increase to 10 mg twice daily
Less than 50 X 10^9/L	Continue hold

For the most current prescribing information, go to:
http://www.incyte.com/sites/default/files/Jakafi_PI.pdf

APPENDIX C ECOG PERFORMANCE STATUS*

Grade	ECOG
0	Fully active, able to carry on all pre-disease performance without restriction
1	Restricted in physically strenuous activity but ambulatory and able to carry out work of a light or sedentary nature, e.g., light house work, office work
2	Ambulatory and capable of all selfcare but unable to carry out any work activities. Up and about more than 50% of waking hours
3	Capable of only limited selfcare, confined to bed or chair more than 50% of waking hours
4	Completely disabled. Cannot carry on any selfcare. Totally confined to bed or chair
5	Dead

* As published in Am. J. Clin. Oncol.:
Oken, M.M., Creech, R.H., Tormey, D.C., Horton, J., Davis, T.E., McFadden, E.T., Carbone, P.P.: Toxicity And Response Criteria Of The Eastern Cooperative Oncology Group. Am J Clin Oncol 5:649-655, 1982.

APPENDIX D TABLE OF MODERATE AND POTENT CYP3A4 INHIBITORS

Use of all CYP3A4 inhibitors is discouraged as they may have effects on ruxolitinib levels, and alternative therapies should be sought if available.

Strong Inhibitors	Moderate Inhibitors	Weak Inhibitors	All other inhibitors
indinavir[1]	aprepitant[2]	cimetidine	amiodarone
nelfinavir[1]	erythromycin[2]		chloramphenicol
ritonavir[1]	fluconazole[2]		delaviridine
saquinavir[1]	grapefruit juice[2]		diethyl-dithiocarbamate
clarithromycin[2]	verapamil[2]		fluvoxamine
itraconazole[2]	diltiazem[2]		gestodene
ketoconazole[2]			imatinib
nefazodone[2]			mibefradil
posaconazole[2]			mifepristone
voriconazole[2]			norfloxacin
telithromycin[2]			norfluoxetine
			starfruit

1 Subjects receiving these medications should not be allowed in the study as HIV+ patients are excluded.

2 Close monitoring of platelet counts and dose reductions are required with the addition of moderate to potent CYP 3A4 inhibitors. See Section 6.

APPENDIX E GRADING OF HEMORRHAGIC EVENTS

	Grade I	Grade II	Grade III	Grade IV	Grade V
Blood and lymphatic system disorders - Other, specify	Asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated	Moderate; minimal, local or noninvasive intervention indicated; limiting age appropriate instrumental ADL	Severe or medically significant but not immediately life threatening; hospitalization or prolongation of existing hospitalization indicated; disabling; limiting self-care ADL	Life-threatening consequences; urgent intervention indicated	Death

Refer to Common Terminology Criteria for Adverse Events (CTCAE) Version 4.0; Published: May 28, 2009 (v4.03: June 14, 2010).

For the latest version, go to:

http://evs.nci.nih.gov/ftp1/CTCAE/About.html

APPENDIX F ADVERSE EVENTS AND SIDE EFFECTS WITH RUXOLITINIB

Relevant sections from prescribing information provided here. Adverse events highlighted in BOLD will be recorded and summarized based on the indicated cut-off values. Unexpected grade 3 or 4 adverse events (according to CTCAE term (AE description)) categorized as definitely or possibly attributed to ruxolitnib will also be recorded and reported as indicated in section 7.

5.1 Thrombocytopenia[a], Anemia[b], and Neutropenia[c] Treatment with **ruxolitinib** can cause thrombocytopenia, anemia and neutropenia. [*see Dosage and Administration (2.1)*]. Thrombocytopenia was generally reversible and was usually managed by reducing the dose or temporarily interrupting **ruxolitinib**. Platelet transfusions may be necessary [*see Dosage and Administration (2.2), and Adverse Reactions (6.1)*]. Patients developing anemia may require blood transfusions and/or dose modifications of Jakafi. Severe neutropenia (ANC less than 0.5 X 109/L) was generally reversible. Withhold **ruxolitinib** until recovery [*see Adverse Reactions (6.1)*]. Perform a pre-treatment complete blood count (CBC) and monitor CBCs every 2 to 4 weeks until doses are stabilized, and then as clinically indicated. [*see Dosage and Administration (2.2), and Adverse Reactions (6.1)*].

5.2 Risk of Infection[d] Serious bacterial, mycobacterial, fungal and viral infections may occur. Active serious infections should have resolved before starting therapy with **ruxolitinib**. Observe patients receiving **ruxolitinib** for signs and symptoms of infection and initiate appropriate treatment promptly.

[a] less than 50,000 per microliter
[b] less than Hgb 7 g/dL
[c] less than ANC 500 cells per microliter
[d] requiring hospitalization or intravenous antibiotics

APPENDIX G EFFECTIVE CONTRACEPTIVE PRACTICES

For Males Subjects Participating in the Study:
The following methods have been determined to be more than 99% effective (failure rate less than 1% per year, when used consistently and correctly) (Trussell, 2004), by the male subject and his partner and are permitted under this protocol:

- Complete abstinence from sexual intercourse
- Double barrier methods
    - Condom with spermicide in conjunction with use of an intrauterine device (IUD)
    - Condom with spermicide in conjunction with use of a diaphragm
- Oral, injectable, or implanted contraceptives
- Tubal ligation or vasectomy (surgical sterilization)

For Female Subjects Participating in the Study:
The following methods have been determined to be more than 99% effective (failure rate less than 1% per year, when used consistently and correctly) (Trussell, 2004), by the female subject and her partner and are permitted under this protocol:

- Complete abstinence from sexual intercourse
- Double barrier methods
    - Condom with spermicide in conjunction with use of an IUD
    - Condom with spermicide in conjunction with use of a diaphragm
- Tubal ligation or vasectomy (surgical sterilization)
- Oral, injectable, or implanted contraceptives

Subjects >55 y/o and without menses for at least 3 years are considered in menopause.

LIST OF ABBREVIATIONS

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

Term	Explanation
AE	Adverse event
ALT	Alanine aminotransferase
ANC	Absolute neutrophil count
bid	Twice daily
CBC	Complete blood count
C_{max}	Maximum observed plasma concentration
CRF	Case report form
CT	Computed Tomography
CTCAE	Common Terminology Criteria for Adverse Events
dL	Deciliter
DLT	Dose limiting Toxicity
DMC	Data Monitoring Committee
ECG	Electrocardiogram
eCRF	Electronic Case Report Forms
ECOG	Eastern Cooperative Oncology Group
ET	Essential throbocythemia
FDA	Food and Drug Administration
GCP	Good Clinical Practice
IB	Investigator's Brochure
ICF	Informed consent form
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
INR	International Normalized Ratio
IPSS	International Prognostic Scoring System
IRB	Institutional Review Board
IVRS	Interactive Voice Response System
IWG-MRT	International Working Group for Myelofibrosis Research and Therapy
JAK	Janus kinase
MF	Myelofibrosis
MPN-SAF	Myeloproliferative neoplasm symptom assessment form v2.0
mg	Milligram
MPN	Myeloproliferative neoplasm

MRI	Magnetic Resonance Imaging
PD	Pharmacodynamic
PET-MF	Post essential thrombocythemia myelofibrosis
PGIC	Patient Global Impression of Change
PI	Principal Investigator
PK	Pharmacokinetics
PMF	Primary Myelofibrosis
PV	Polycythemia vera
PPV-MF	Post polycythemia vera myelofibrosis
PR	Partial remission
PTT	Partial Thromboplastin time
qd	Once daily
ruxolitinib	ruxolitinib phosphate or ruxolitinib phosphate tablets, based on context
SAE	Serious adverse event
STAT	Signal transduction and activator of transduction
ULN	Upper limit of normal
WBC	White blood cells

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