THE UNIVERSITY OF TEXAS
MD Anderson Cancer Center

Protocol Page

PHASE II STUDY OF PACRITINIB FOR PATIENTS WITH LOWER-RISK MYELODYSPLASTIC SYNDROMES
2013-0224

Core Protocol Information

Short Title	PACRITINIB FOR PATIENTS WITH LOWER-RISK MDS
Study Chair:	Courtney DiNardo
Additional Contact:	Tawana Heiskell Vicky H. Zoeller Leukemia Protocol Review Group
Department:	Leukemia
Phone:	713-794-1141
Unit:	428
Full Title:	PHASE II STUDY OF PACRITINIB FOR PATIENTS WITH LOWER-RISK MYELODYSPLASTIC SYNDROMES
Protocol Type:	Standard Protocol
Protocol Phase:	Phase II
Version Status:	Terminated 06/05/2017
Version:	07
Submitted by:	Tawana Heiskell--1/29/2016 10:24:00 AM
OPR Action:	Accepted by: Debbie D. Stroughter -- 2/12/2016 4:00:56 PM

Which Committee will review this protocol?

(●) The Clinical Research Committee - (CRC)

Protocol Body

Pracritinib 2013-0224 (final) 11-17-2015 rsvd 11-30-2015.pdf

PHASE II STUDY OF PACRITINIB FOR PATIENTS WITH LOWER-RISK MYELODYSPLASTIC SYNDROMES

STUDY PRINCIPAL INVESTIGATOR
Courtney DiNardo, M.D.

CO-PRINCIPAL INVESTIGATOR
Guillermo Garcia-Manero, M.D.

CO- INVESTIGATORS
Xueling Huang Ph.D.
Hagop Kantarjian M.D.
Srdan Verstovsek, M.D., PhD
Elias Jabbour, M.D.
Gautham Borthakur M.D.
Tapan Kadia, M.D.
Naval Daver, M.D.
Naveen Pemmaraju, M.D.
Farhad Ravandi M.D.
Graciela M. Nogueras Gonzalez

From the Department of Leukemia, Division of Cancer Medicine, The University of Texas, M.D. Anderson Cancer Center, 1515 Holcombe Blvd., Houston, Texas 77030. Telephone: (713) 794-1141

Table of Contents

PHASE II STUDY OF PACRITINIB FOR PATIENTS WITH LOWER-RISK MYELODYSPLASTIC SYNDROMES

1.0 OBJECTIVES

To determine the clinical activity, safety and tolerability of Pacritinib in patients with lower-risk myelodysplastic syndromes (MDS).

2.0 BACKGROUND

2.1 Myelodysplastic syndrome

Myelodysplastic syndromes (MDS) are malignant clonal disorders characterized by ineffective hematopoiesis, bone marrow dysplasia, peripheral cytopenias, and a propensity to transform into acute myeloid leukemia (AML). Over 10,000 new cases of MDS are diagnosed annually in the United States alone, with a median age at diagnosis of 70 years. MDS thus most frequently affects patients that are generally not candidates for intensive chemotherapy or bone marrow transplantation strategies.

MDS can be classified using different systems, such as the FAB, or the WHO classifications[1]. The International Prognostic Score System (IPSS)[2], is a prognostic classification of MDS that allows calculation, based on the age of the patient, the expected median survival and probability of progression to AML. With this scoring system, patients are divided into low, intermediate (Int-1),Iint-2 and high-risk disease. This is based on percentage of marrow blasts, cytogenetic alterations and number of cytopenias (Table 1).

Table1. IPSS for MDS[2]

	Score Value				
Variable	**0**	**0.5**	**1.0**	**1.5**	**2.0**
% BM blasts	<5	5-10	--	11-20	21-30
Karyotype	Good	Intermediate	Poor		
Cytopenias	0/1	2/3			

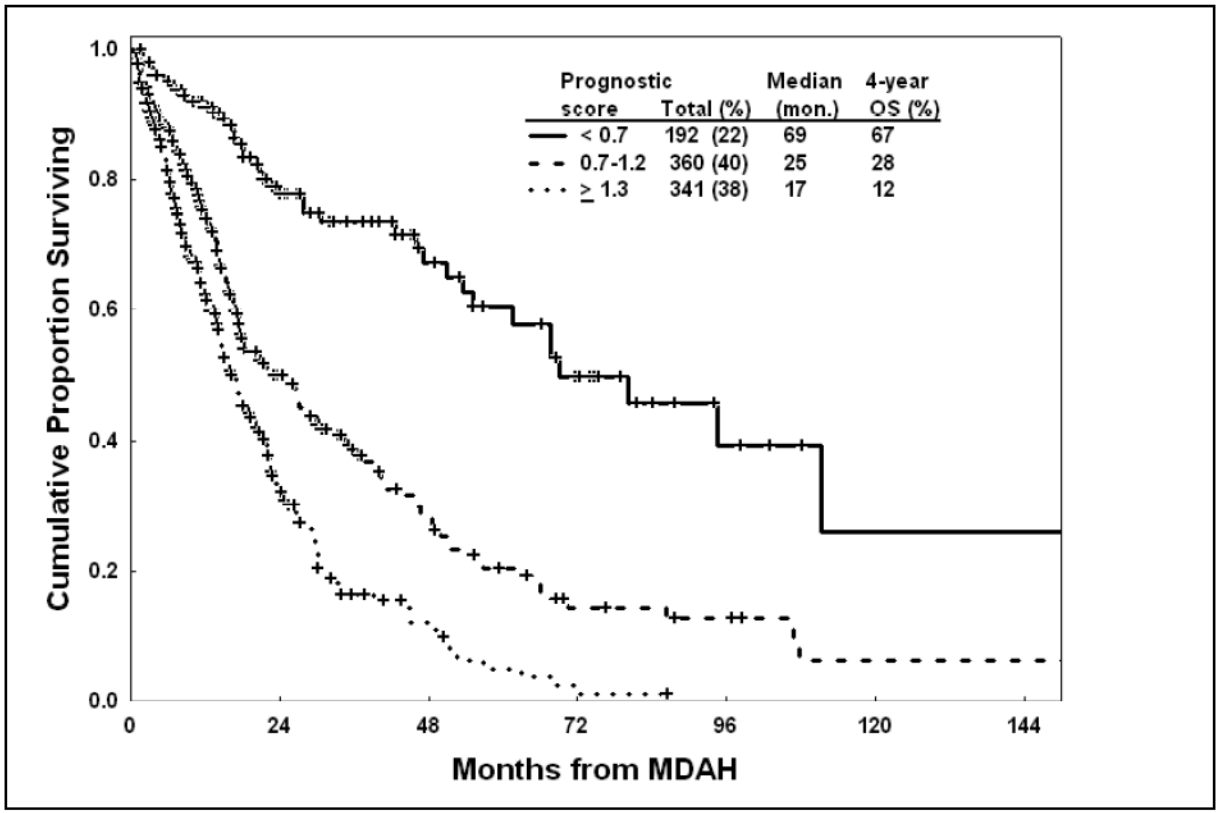
Scores are as follows: Low: 0; INT-1: 0.5-1.0; INT-2: 1.5-2.0 and high risk ≥ 2.5 points. Good karyotype includes: diploid, -Y, del(5q), and del(20q).
Poor karyotype includes: complex (>3 abnormalities) or chromosome 7 abnormalities. Intermediate karyotype refers to all others. Cytopenias are defined as an absolute neutrophil count < 1500 K/uL; hemoglobin less than 10 g/dl and platelets are less than 10^5 K/uL.Table 2 summarizes the expected overall survival based on age and IPSS score. Based on this data, the median survival of patients with low or Int-1 MDS ranges from 3.5 to 5.7 years. This has led to the practice followed by many physicians in the community of “observing” these patients with supportive care options such as growth factors and transfusions as necessary. There are no accepted or standard agents for use in this lower-risk MDS population.

Table 2. Estimated survival and time to AML progression based on age and IPSS score[2]

	No. of Patients	Low	Int-1	Int-2	High
A. Median Survival (yr)					
Total no. of patients (%)	816	267 (33)	314 (38)	176 (22)	59 (7)
Median (yr)		5.7	3.5	1.2	0.4
Age (yr)					
≤60	205(25)	11.8	5.2	1.8	0.3
>60	611	4.8	2.7	1.1	0.5
≤70	445(54)	9.0	4.4	1.3	0.4
>70	371	3.9	2.4	1.2	0.4
B. 25% AML Evolution (yr)					
Total no. of patients (%)	759	235 (31)	295 (39)	171 (22)	58 (8)
Median (yr)		9.4	3.3	1.1	0.2
Age (yr)					
≤60	187 (25)	>9.4 (NR)	6.9	0.7	0.2
>60	572	9.4	2.7	1.3	0.2
≤70	414 (55)	>9.4 (NR)	5.5	1.0	0.2
>70	345	>5.8 (NR)	2.2	1.4	0.4

Importantly, we have recently performed an analysis of the prognosis of patients with lower-risk MDS (henceforth defined as Low and Int-1 MDS) referred to MDACC over the last 25 years[3]. The outcomes of 898 previously untreated patients referred to our center were analyzed, and a prognostic score was developed which was based on clinical characteristics including cytogenetics, age, percentage of blasts, hemoglobin and platelet counts. Based on this model, patients with lower-risk MDS can be divided in 3 groups (Figure 1), and notably over 60% of patients with lower risk MDS had poor prognosis. Of great importance, it should be noted that only 10% of the patients transformed to AML. This information suggests that a majority of patients with lower risk MDS die as a consequence of MDS and not from AML transformation, and therefore the traditional strategy of "watch and wait" used by many physicians in patients with lower-risk MDS may not be indicated. Therefore new therapies are needed for this subgroup of patients.

Figure 1. Prognosis of patients with lower-risk MDS based on new



MDACC low-risk model.

At the present time, 3 agents are approved for treatment of MDS in the United States. These include 5-azacitidine (Vidaza)[4], lenalidomide[5] (Revlimid), and 5-aza-2'-deoxycitidine (decitabine or Dacogen)[6]. Lenalidomide is a thalidomide derivative indicated for a specific subset of patients with MDS, in particular those with alterations of chromosome 5 and anemia with low-risk disease[5]. Clinical use of the two approved hypomethylating agents (HMAs), Vidaza and Dacogen have been shown to improve quality of life, decrease transfusion requirements, and improve outcome parameters in MDS patients, and are now standard of care for MDS patients requiring therapy. In a Phase III trial of high-risk MDS, treatment with 5-azacitadine was associated with a significant survival benefit (OS 24.4 months compared to 15 months with conventional therapy, p <0.001)[7]. With decitabine, an overall

response rate (ORR) of 30% versus 7% in favor of decitabine ($p < 0.001$) with a non-significant improvement in progression to AML and overall survival was similarly noted[6]. In is however important to note that the clinical benefit of the hypomethylating agents, at this time, is primarily limited to patients with more advanced disease, such as those with Int-2 or High-risk MDS[4,6,8,9].

2.2 Rationale for the use of Pacritinib in MDS

The JAK2 inhibitors are a relatively new class of therapeutic agents in leukemia. Pacritinib (SB1518) is an oral JAK2-FLT3 inhibitor with the capacity to modulate cytokine function in patients with myeloid malignancies. Furthermore, cytokine signaling is known to be abnormal in MDS. Work from our lab (Wei et al, ASH 2010) using whole genome CHIP-seq in primary CD34+ cells has indicated that cytokine pathways mediated by JAK/STAT signaling are overexpressed in MDS. Furthermore, inhibition of TREM, a key mediator of this pathway, resulted in ex vivo differentiation changes in MDS colonies. Recent data in myeloproliferative disorders indicate that changes in cytokine profile are associated with clinical responses in these related disorders[10]. These data, taken together with their excellent toxicity profile, are the basis for the proposed study of pacritinib in MDS.

2.3 Pacritinib

Pacritinib (SB1518) is a novel Janus kinase 2-fms-like receptor kinase 3 (JAK2-FLT3 inhibitor) that has shown promising anti-tumor activity. Six clinical studies of pacritnib have been completed to date.The drug is being evaluated in two ongoing phase 3 studies, PAC325 (PERSIST-1) and PAC326 (PERSIST-2). Preclinical and clinical studies have established the oral dose of pacritinib. Kinase screening has shown that pacritinib is selective for JAK2 and FLT3. In vitro pharmacology studies have indicated that pacritinib exerts antiproliferative activity on a variety of tumor cell lines, with and without JAK2 and FLT3 mutations. In vivo activity has been shown in two mouse models of human myeloproliferative malignancies; one model is driven by a JAK2 mutation, the other by FLT3 mutation.

Pacritinib has shown antitumor activity in 2 mouse models of human malignancies and in 4 completed clinical studies. Two clinical studies have characterized the pharmacokinetic (PK) profile of pacritinib and the effect of food on pacritinib administration. The indications of interest include (a) myelofibrosis (primary, post-polycythemia vera and post-essential thrombocythemia), polycythemia vera, and essential thrombocythemia, all of which are myeloproliferative neoplasms (MPNs) with a high frequency of the JAK2V617F mutation, and other mutations that result in overactivation of the JAK-STAT pathway; (b) certain leukemias and lymphomas in which other forms of JAK aberrations have been reported; and (c) acute myeloid leukemia (AML), in which FLT3 inhibitors have shown preliminary clinical promise. Pacritinib is believed to be active in MF, whether or not the JAK2V617F mutation is present.

The preclinical pharmacological and toxicological profiles and current status of pacritinib in the clinic are presented in this document.

Investigational Product

Laboratory name: SB1518
Pharmacological class: JAK2-FLT3 inhibitor

Summary of Nonclinical Studies
Results from non-clinical studies have shown the very favorable pharmacological properties (including efficacy, pharmacokinetics, pharmacodynamics and toxicology) of pacritinib and support its continued clinical development.

Nonclinical Pharmacology
Pacritinib is a potent inhibitor of JAK2 and FLT3 kinase activities (50% inhibitory concentration (IC50) = 23 nM and 22 nM, respectively), as well as a potent inhibitor of cellular proliferation in human leukemia and lymphoma cell lines selected for their dependence on either of the target kinases (cellular IC50 ranges from 0.03 to 0.46 μM). Consistent with these activities, exposure to pacritinib resulted in the reduction of phos-JAK2, phos-STAT3 or phos-STAT5 in the relevant cell lines. Unlike some JAK2 inhibitors, pacritinib does not inhibit JAK1.

The antitumor activity of pacritinib was demonstrated in two tumor models driven by FLT3 or JAK2 mutations. In nude mice bearing MV4-11, a FLT3-dependent acute myeloid leukemia cell line, pacritinib treatment caused a dose-dependent inhibition of tumor growth, with complete regression at the highest dose tested (100 mg/kg). In nude mice bearing BaF3-JAK2V617F, a JAK2-dependent leukemia, untreated mice developed the hallmark symptoms of myeloproliferative disease, including spleen/liver hyperplasia and severe leukocytosis. Pacritinib treatment (300 mg/kg daily) alleviated clinical symptoms including leukocytosis and splenomegaly. The higher dosage required to achieve comparable therapeutic effects in the BaF3 model relative to the MV4-11 model (300 mg/kg vs 100 mg/kg daily) was consistent with the antiproliferative effects of SB1518 on these cells in vitro (IC50=461 nM and 32 nM, respectively).

Pacritinib had no activity in two xenograft models which were not dependent on FLT3 or JAK2 mutations, namely HCT116 colorectal carcinoma and HL60 acute myeloid leukemia. The lack of efficacy in these models shows the specificity of pacritinib for JAK2 or FLT3 driven disease.

For in vitro safety pharmacology evaluation, pacritinib was tested at 10 μagainst a panel of 56 pharmacologically important receptors and enzymes. In these screens, Pacritinib caused >50% inhibition against 4 enzymes and 12 receptors. Based on these findings, cardiovascular safety pharmacology studies were conducted.

Cardiovascular safety testing in conscious dogs showed no treatment-related changes in cardiac electrophysiology (P, R-R, PQ, QRS, QTR, and QTc intervals), heart rate, respiratory rate, body temperature, blood pressure or activity, following a single oral dose of 30 mg/kg, despite gastrointestinal effects (vomiting) that were observed clinically. In the enzyme screen, pacritinib was shown to bind to the human ether-a-go-go-related gene (hERG) at an IC50 of 3.51 μM. With respect to the in vitro hERG bining result, given the high plasma-protein binding observed for pacritinib in humans (98.3%), plasma concentrations of free pacritinib that might cause cardiac safety issues are unlikely to be achieved at the top clinical dose (400mg QD) of pacritinib.

Nonclinical Pharmacokinetics

The PK of pacritinib was evaluated in mice, rats, and dogs following single intravenous (IV) and PO dose administration. Pacritinib was administered both IV and PO in the same dogs, whereas different sets of rats and mice were given either IV or PO doses of pacritinib.

Following oral administration, pacritinib showed rapid absorption in mice (time to reach maximum concentration [T_{max}] = 1.3 h) and moderately fast absorption in rats and dogs (T_{max} = 4 h). The oral terminal elimination half-lives were 2.2, 5.7, and 4.4 h in mice, rats, and dogs, respectively. Pacritinib showed a high volume of distribution (V_{ss}) in mice (V_{ss} = ~ 65 L/kg), rats (V_{ss} = 13 L/kg), and dogs (V_{ss} = 8.5 L/kg). The systemic clearance of pacritinib was high in mice (8 L/h/kg) and dogs (1.6 L/h/kg) and moderate in rats (1.6 L/h/kg), relative to liver blood flow. The terminal elimination half-lives following IV administration were 5.6, 6.0, and 4.6 h in mice, rats, and dogs, respectively. The oral bioavailabilities of pacritinib were 39%, 10%, and 24% in mice, rats, and dogs, respectively.

In toxicokinetic studies, pacritinib was absorbed rapidly into the systemic circulation (T_{max} ranged from 0.5 to 2 h in mice and 2 to 3 h in dogs). Systemic exposure levels of pacritinib were roughly proportional to dose over the range of doses studied and slight (< 2-fold) accumulation was observed. At all tested doses, pacritinib plasma levels reached concentrations for in vitro JAK2 and FLT3 enzyme inhibition.

Preliminary assessments of tissue distribution to plasma, brain, and lungs following administration of a single PO dose in nude mice showed that pacritinib levels were higher in lungs (tissue:plasma ratio of 19) than in brain (tissue:plasma ratio of 1.8). Plasma protein binding studies have shown > 99% binding by pacritinib in mouse, dog, and human plasma.

In a radiolabeled mass balance and tissue distribution ADME study in mice, following a single oral dose of ^{14}C-SB1518 (~100 mg/kg, 2.8 μCi/animal), the recovery of dosed radioactivity was 92.97% (90.98% in the feces). The highest concentrations of radioactivity were in the contents of the alimentary canal and bile. The data suggested that pacritinib was primarily eliminated in feces by biliary clearance in mice.

Biotransformation in vitro was examined by incubating pacritinib in liver microsomes of mouse, rat, dog, and human in the presence

of an NADPH regeneration system. Seven metabolites were identified by LC-MS/MS analysis of incubation mixtures. The distribution of metabolites varied with species, with the dog (M4>M1>M2>>M6=M7) being most similar to humans (M4>M1=M2>M5>>M6=M7) and the mouse slightly different in terms of the degrees of formation (M1>M2>M3>M4>M5>>M6). Most of the metabolites formed in human in vitro and in vivo were observed in mice and dogs, showing that the mouse and dog are relevant toxicological species.

Findings from in vitro metabolic studies suggest that pacritinib is mainly metabolized by CYP3A4. Pacritinib showed no significant induction of CYP1A2 and CYP3A4 isozymes and did not inhibit the catalytic activity of the CYP isozymes tested (3A4, 1A2, 2C9, 2D6, 2C19) in the human liver microsome assay. Results from a bi-directional Caco-2 permeability assay showed that pacritinib has high permeability and low efflux ratio, so an active p-glycoprotein transport mechanism is unlikely to be involved in the disposition of pacritinib.

Overall, there is no dispositional or metabolic property of pacritinib that precludes its development in humans.

To support the switch from the hydrochloride (HCl) to citrate salt form of pacritinib in the clinic, retrospective analyses of PK and toxicokinetic data were performed to demonstrate bioequivalence of the two salt forms in the preclinical species (mouse and dog). A retrospective, comparative analysis of the oral PK of pacritinib HCl and citrate salts did not show any significant differences between the PK parameters in the salt forms at equivalent doses.

Nonclinical Toxicology

The adverse effects of pacritinib were evaluated in 30-day repeated oral dose toxicity studies with 14-day recovery in both mice and dogs and in 26- and 39-week chronic toxicity studies in mice and dogs, respectively.

In both 30-day studies, interpretation of findings was complicated by severe toxicities observed in the first week of treatment that led to substantial dose reductions for the remaining three weeks of treatment. For the last three weeks of the 30-day studies, animals were given fixed doses twice daily, with mice administered doses of 0, 50, 150, and 200 mg/kg bid by oral gavage and dogs administered doses of 0, 5, 10, and 20 mg/kg bid as hard gelatin capsules. In the 26-week study, BALB/c mice were administered doses of 0, 30, 100, and 200 mg/kg bid by oral gavage. Due to toxicity, the 200 mg/kg bid dose was reduced to 150 mg/kg bid by study Week 6. In the 39-week study, three groups of beagle dogs were administered doses of 0, 3, and 10 mg/kg bid. A fourth group was administered doses of 25 mg/kg bid on Days 1 through 7 and then 20 mg/kg bid throughout the remainder of the study.

The no observable adverse effect level (NOAEL) in mice was 100 mg/kg bid when administered by oral gavage for up to 26 weeks. The NOAEL in dogs was 20 mg/kg/day when administered as hard gelatin capsules for up to 39 weeks. The maximum tolerated dose (MTD) of 80 mg/kg/day from the 30-day study in dogs was used for calculation of the clinical starting dose.

In a phototoxicity study in mice, no skin reactions indicative of phototoxicity were observed following PO administration of pacritinib at 150 mg/kg BID for 4 consecutive days and subsequent exposure to an ultraviolet radiation (UVR) dose equivalent to 0.5 minimal erythema dose.

The standard genotoxicity panel showed no mutagenic or clastogenic activity for pacritinib by in vitro and in vivo testing.

In a male fertility toxicity study in mice (pacritinib dosed at 15, 50, or 150 mg/kg BID), no effects at any dose level were seen from uterine implantation data, macroscopic findings, reproductive organ weights, or sperm analyses. The NOAEL of pacritinib for male reproductive performance and fertility was 100 mg/kg/day.

Summary of Clinical Studies
Pacritinib has been studied in 6 completed clinical trials as of May 31, 2014; the drug is being studied in two ongoing phase 3 trials.

Study SB1518-001 -This was an open-label phase 1/2 study to determine the MTD and DLTs of pacritinib when given as a single agent PO once daily in patients with advanced myeloid malignancies.

This study has been completed. A clinical study report (CSR) is pending for Study SB1518-2007-001; thus, data are preliminary.

Study Phase 1

The primary objective of study Phase 1 was to establish the maximum tolerated dose (MTD) of pacritinib as a single agent when administered orally daily in subjects with advanced myeloid malignancies. Secondary objectives were to (1) assess the safety and tolerability of pacritinib, administered orally once daily in subjects with advanced myeloid malignancies; (2) assess the pharmacokinetic (PK) profile of pacritinib; and (3) evaluate pharmacodynamic (PD) activity of pacritinib.

In the phase 1 part of the study, cohorts of 3 to 6 patients received one of a series of escalating doses of pacritinib, ranging from 100 to 600 mg/day for 25 days. Six (6) additional patients were dosed with the citrate salt formulation at the 200 mg dose to assess tolerability and PK of that formulation. A total of 43 patients were enrolled and treated at 7 dose levels.

Study Phase 2

The objectives in study Phase 2 were to (1) assess the spleen response rate in subjects with [chronic idiopathic myelofibrosis] CIMF who are treated with pacritinib at the [recommended dose] RD; (2) assess the duration of spleen response at the RD; and (3) assess the safety and tolerability of pacritinib, administered orally once daily at the RD in subjects with CIMF. The exploratory objective was to assess quality of life and disease-related symptoms in subjects with CIMF who are treated with pacritinib at the RD.

In the phase 2 part, 31 patients with primary myelofibrosis (PMF), post-polycythemia vera myelofibrosis (PPV-MF), or post-essential thrombocythemia myelofibrosis (PET-MF) were treated with 400 mg PO daily.

Study SB1518-2007-002 was an open-label phase 1 dose-escalation study to determine the MTD and the DLTs of pacritinib when given as a single agent orally once daily in patients with advanced lymphoid malignancies. The primary study objective was to establish the maximum tolerated dose (MTD) of pacritinib as a single agent when administered orally daily in subjects with advanced lymphoid malignancies. The secondary objectives were to (1) assess the safety and tolerability of pacritinib, administered orally once daily in subjects with advanced lymphoid malignancies; (2) assess the pharmacokinetic (PK) profile of pacritinib; and (3) evaluate pharmacodynamic (PD) activity of pacritinib.

This study has been completed. A CSR is pending for Study SB1518-2007-002; thus, data are preliminary.

Three to 6 patients received one of a series of escalating doses of pacritinib, ranging from 100 to 600 mg per day. Thirty-five patients were enrolled: 15 with Hodgkin lymphoma (HL), 10 with follicular lymphoma (FL), 4 with diffuse large B-cell lymphoma (DLBCL), 5 with mantle cell lymphoma (MCL), and 1 with small lymphocytic lymphoma (SLL). Thirty-four patients were treated at 5 dose levels.

Study SB1518-2008-03 This was an open-label phase 1/2 study in patients with MF. Study Phase 1 was a dose-escalation study to determine the MTD and the DLT of pacritinib when given as a single agent orally once daily in subjects with CIMF, regardless of their JAK2 mutational status. Phase 1 objectives were to: (1) assess the safety and tolerability of pacritinib, administered orally once daily in subjects with chronic idiopathic myelofibrosis (CIMF); (2) establish the Maximum Tolerated Dose (MTD) of pacritinib as a single agent when administered orally daily; (3) characterize the pharmacokinetic (PK) profile of pacritinib; (4) evaluate pharmacodynamic (PD) activity of pacritinib; and (5) determine a dose for Phase 2 development (recommended dose) based on pharmacodynamic exposure (PK), safety and clinical benefit data. Following the identification of the MTD, the Phase 2 portion of the study evaluated the efficacy and safety profile of single-agent pacritinib at the RD in subjects with CIMF. Phase 2 objectives were to: (1) assess the spleen response rate in subjects with CIMF who are treated with pacritinib at the recommended dose (RD); (2) assess the duration of spleen response; and (3) assess the safety and tolerability of pacritinib, administered orally once daily at the RD in subjects with CIMF.
Exploratory objective was to assess Quality of Life and disease-related symptoms in subjects with CIMF who are treated with pacritinib at the RD.

This study has been completed. A CSR is pending for Study SB1518-2008-003; thus, data are preliminary.

Study SB1518-2010-004- This was an open-label, randomized, single-dose, three-treatment, six-sequence, three-period, crossover study to characterize the PK and inter- and intra-individual variability of pacritinib PK after a single oral administration (100, 200, or 400 mg) to healthy volunteers under fasted conditions. The primary objective was to assess the pharmacokinetics of pacritinib after single oral doses. The secondary objectives were to assess the linearity or dose proportionality of pacritinib and to evaluate the between- and within-subject variability.

This study has been completed, and a CSR is available.

Pacritinib systemic exposure increased in a linear, dose-related, but less than dose-proportional, manner over the dose range of 100 to 400 mg. Single-dose PO administration of pacritinib at 100 and 200 mg was well tolerated in healthy adult volunteers. At 400 mg, the AEs were mainly GI and nervous system related and were mild in intensity.

Study SB1518-2010-005

This was an open-label, phase 2 study of single-agent pacritinib at 400 mg administered orally once daily in patients with selected advanced lymphoid malignancies. The primary objective was to assess the efficacy of pacritinib in the treatment of patients with advanced myeloid malignancies, including Hodgkin lymphoma [HL], mantle cell lymphoma [MCL], and indolent lymphoma (follicular lymphoma [FL], lymphoplasmacytic lymphoma, marginal zone lymphoma, and small lymphocytic lymphoma). Secondary objectives were to (1) assess the durability of response and PFS following pacritinib treatment in patients with advanced lymphoid malignancies; and (2) assess the safety and tolerability of pacritinib administered orally once daily at 400 mg in patients with advanced lymphoid malignancies.

This study has been completed. A CSR is pending for Study SB1518-2010-005; thus, data are preliminary.

The study's primary efficacy endpoint was the overall response rate (ORR). Secondary efficacy endpoints included duration of response and progression-free survival (PFS).

The study enrolled 10 patients with HL, 8 with MCL, and 10 with indolent FL.

Study SB1518-2010-006

Study Title: A Randomized, Single-Dose, Two-Treatment, Two-Sequence, Two-Period Crossover Study of the Effect of Food on the Bioavailability and Pharmacokinetics of SB1518 in Healthy Volunteers.

This was an open-label, randomized, single-dose, two-treatment, two-sequence, two-period, crossover study of the effect of food on the bioavailability and PK of a single 200 mg dose of pacritinib in healthy adult volunteers. Blood samples were collected for the measurement of plasma pacritinib concentration at multiple time points, from time t = 0 (predose) to t = 144 hours postdose.

PK parameters were comparable, whether pacritinib was taken after fasting or after a high-calorie, high-fat meal. Thus, the results of this study indicated that pacritinib can be taken without regard to meal content or timing.

Single-dose administration of pacritinib at an oral dose of 200 mg was well tolerated in healthy adult volunteers under both fed and fasted conditions.

Summary of Phase 1 and 2 Experience with Pacritinib in Myelofibrosis

- A total of 56 patients with MF were treated with escalating pacritinib doses in the phase 1 portions of studies SB1518-2007-001 and -2008-003. An additional 65 patients were treated in the phase 2 portions of these studies in Australia and the United States (US).
- A total of 36 patients with starting platelet counts ≤ 150,000/μL were treated, with an apparent response rate similar to those with higher platelet counts and no consistent treatment-related platelet suppression. Pacritinib did not appear to increase anemia or RBC transfusion requirements.
- Pacritinib has a favorable safety profile; it was generally well tolerated in patients with MF, including those with PMF, PPV-MF, and PET-MF. Side effects were predominantly GI and readily managed with symptomatic treatment and/or study drug interruption or dose reduction. Reported AEs associated with myelosuppression were uncommon, and pacritinib was well tolerated and active in patients with cytopenias, particularly thrombocytopenia.
- Median baseline platelet counts in patients treated with pacritinib in phase 2 studies were 126,000/μL and 119,000/μL—approximately half those in the ruxolitinib studies (262,000/μL and 244,000/μL).
- There was an overall 17% incidence of all grades of thrombocytopenia reported as AEs in efficacy and safety trials with pacritinib. Twelve percent (12%) of patients experienced a ≥ 2-grade shift in platelet counts from baseline to worst platelet count, and 5% experienced a ≥ 2-grade shift in platelet counts from baseline to end of study.

Ongoing Phase 3 Pacritinib Studies

Study PAC325 (PERSIST-1)

Study Title: A Randomized Controlled Phase 3 Study of Oral Pacritinib versus Best Available Therapy in Patients with Primary Myelofibrosis, Post-Polycythemia Vera Myelofibrosis, or Post-Essential Thrombocythemia Myelofibrosis

This is an ongoing multicenter, randomized, controlled, phase 3 study. It will compare the safety and efficacy of pacritinib with that of best available therapy (BAT) in patients with PMF, PPV-MF, or PET-MF. Eligible patients are centrally randomized in a 2:1 allocation to pacritinib (400 mg, taken orally QD) or BAT. BAT includes any physician-selected treatment for PMF, PPV-MF, or PET-MF with the exclusion of JAK inhibitors (inhibitors of Janus kinases), and may include any treatment received before study entry.

This is an ongoing study; thus, data from PERSIST-1 are preliminary.

The study's primary objective is to compare the efficacy of pacritinib with that of best available therapy (BAT) in patients with primary myelofibrosis (PMF), post-polycythemia vera myelofibrosis (PPV-MF), or postessential thrombocythemia myelofibrosis (PET-MF); the efficacy measure for this analysis is the proportion of patients achieving a ≥ 35% reduction in spleen volume from baseline to Week 24, as measured by magnetic resonance imaging (MRI) or computed tomography (CT) scan. The key secondary objective is the proportion of patients with ≥ 50% reduction in total symptom score (TSS) from baseline to Week 24 on the Myeloproliferative Neoplasm Symptom Assessment Form (MPN-SAF TSS 2.0). Other secondary objectives are to compare pacritinib with BAT with respect to:

- Proportion of patients with baseline platelet count < 100,000/μL achieving ≥ 35% reduction in spleen volume from baseline to Week 24 as measured by MRI or CT [computed tomography] scan
- Proportion of patients with baseline platelet count < 100,000/μL achieving ≥ 50% reduction in TSS from baseline to Week 24
- Proportion of patients with baseline platelet count < 50,000/μL achieving ≥ 35% reduction in spleen volume from baseline to Week 24 as measured by MRI or CT scan
- Proportion of patients with baseline platelet count < 50,000/μL achieving ≥ 50% reduction in TSS from baseline to Week 24

The pharmacokinetic (PK) and pharmacodynamic (PD) objectives are to assess exposure and exposure-response relationships pertaining to the safety and efficacy of pacritinib.

Patients on BAT can cross over to pacritinib treatment at the time of splenic progression, at any time after splenic progression, or after completing 24 weeks of treatment, with or without progression.

The statistical analysis will be performed when the last patient reaches the Week 24 evaluation. An Independent Data Monitoring Committee (IDMC) will evaluate the safety of pacritinib. No interim efficacy analysis is planned.

Study PAC326 (PERSIST-2)

Study Title: A Randomized Controlled Phase 3 Study of Oral Pacritinib versus Best Available Therapy in Patients with Thrombocytopenia and Primary Myelofibrosis, Post-Polycythemia Vera Myelofibrosis, or Post-Essential Thrombocythemia Myelofibrosis

This is an ongoing multicenter, randomized, controlled, phase 3 study. It compares the efficacy and safety of two dose schedules of pacritinib in pooled and individual group analyses versus BAT in patients with thrombocytopenia and PMF, PPV-MF, or PET-MF. Eligible patients are randomized in a 1:1:1 allocation to pacritinib 400 mg, dosed QD; pacritinib 200 mg, dosed BID; or BAT. BAT includes any physician-selected treatment for PMF, PPV-MF, or PET-MF, such as approved JAK2 inhibitors (administered according to package insert for patients with thrombocytopenia), and may include any treatment received before study entry. For example, BAT may include ruxolitinib, other approved JAK2 inhibitors, hydroxyurea, glucocorticoids, erythropoietic agents, immunomodulatory agents, mercaptopurine, danazol, interferons, cytarabine, melphalan, or other agents. BAT also includes no treatment and symptom-directed treatment without MF-specific treatment.

This is an ongoing study; thus, data from PERSIST-2 are preliminary.

The primary objective of this study is to compare the efficacy of two dose-schedule arms(s) of pacritinib (pooled once daily [QD] and BID dosing arms) with that of best available therapy (BAT) in patients with thrombocytopenia and primary myelofibrosis (PMF), post-polycythemia vera myelofibrosis (PPV-MF), or post-essential thrombocythemia myelofibrosis (PET-MF). The efficacy co-endpoints for this analysis are the proportion of patients achieving a ≥ 35% reduction in spleen volume from baseline to Week 24, as measured by magnetic resonance imaging (MRI) or computed tomography (CT) scan and the proportion of patients achieving a ≥ 50% reduction in total symptom score (TSS) from baseline to Week 24 as measured by the Myeloproliferative Neoplasm Symptom Assessment Form 2.0 (MPN-SAF TSS 2.0). Secondary objectives are:

1 To compare the efficacy of QD pacritinib with that of BAT, as assessed by the proportion of patients achieving a ≥ 35% reduction in spleen volume from baseline to Week 24 by MRI or CT and the proportion of patients achieving a ≥ 50% reduction in the TSS from baseline to Week 24 on the MPN-SAF TSS 2.0.
2 To compare the efficacy of BID pacritinib with that of BAT, as assessed by the proportion of patients achieving a ≥ 35% reduction in spleen volume from baseline to Week 24 by MRI or CT and the proportion of patients achieving a ≥ 50% reduction in the TSS from baseline to Week 24 on the MPN-SAF TSS 2.0.

The pharmacokinetic (PK) and pharmacodynamic (PD) objectives are to assess exposure and exposure-response relationships on the safety and efficacy of pacritinib.

Each patient is to receive pacritinib or BAT until progression of disease, the occurrence of unacceptable toxicity, or the patient no longer derives benefit from treatment. Patients on BAT may cross over to pacritinib at the time of splenic progression, at any time after splenic progression (if leukemic transformation, splenectomy, and splenic irradiation have not occurred), or after completing

24 weeks of treatment, with or without progression.

An Independent Data Monitoring Committee (IDMC) will evaluate the safety of pacritinib. No interim efficacy analysis is planned.

Description

Chemical name:	((2*E*,16*E*)-11-[2-(pyrrolidine-1-yl)ethoxy]-14,19-dioxa-5,7,27-trizatetracyclo[19.3.1.1^{2,6}.1^{8,12}]heptacosa-1(25),2,4,6,8,10,12(26),16,21,23-decaene
Laboratory name(s):	SB1518
Pharmacologic class:	JAK2-FLT3 inhibitor

Formulation

For use in clinical studies as an oral agent, pacritinib is supplied as size #0 hard gelatin capsules with gray bodies and red caps. Capsules contain 100 mg pacritinib (free base) and the following inactive ingredients: microcrystalline cellulose NF, polyethylene glycol 8000 (PEG 8000) NF, and magnesium stearate NF. The capsule gelatin is bovinederived.

Pharmacies at investigational sites will receive 120 capsules packaged in 200 mL HDPE bottles which have child-resistant closures.

Storage Conditions

Drug product should be stored in the pharmacy, hospital, clinic, or warehouse at controlled room temperature, 20° to 25°C (68° to 77°F), with excursions allowed between 15° to 30°C (59° to 86°F). Patients should be instructed that storage temperatures in the home should be below 30°C (86°F).

Disposal of Study Drug

Any unused or expired study drug will be disposed of per MD Anderson Cancer Center policy.

Nonclinical Studies

Non Clinical Pharmacology

Pacritinib was designed as an inhibitor of JAK2 kinase. The spectrum of kinases inhibited by pacritinib was evaluated by in vitro assays against a total of 46 kinases and 2 mutants that constitute a representative cross section of the kinase groups in the human kinome. Initial screens were performed using a single concentration of pacritinib (0.1 μM), and selected kinases were then evaluated further in dose response titration.

The antiproliferative activity of pacritinib was evaluated in vitro against a variety of cancer cell lines, including lines with and without *JAK2* and *FLT3* mutations. To demonstrate that pacritinib exerts its antiproliferative effects through its putative targets, effects of pacritinib on substrates downstream of JAK2 signaling were assessed, including phos-STAT3 expression in Karpas 1106P and 32D cells and phos-STAT5 expression in BaF3 cells. Cell cycle analysis was performed to demonstrate pacritinib-induced cell cycle arrest and induction of apoptosis in MV4-11 and BaF3 cells.

The in vivo antitumor activity of pacritinib was assessed in the MV4-11 AML model driven by a *FLT3* mutation, in the BaF3-JAK2V617F model of myeloproliferative disease driven by a *JAK2* mutation, and in two models independent of *FLT3* or *JAK2* mutations: the HL60 AML model and HCT116 colorectal cancer model.

For in vitro safety pharmacology evaluation, pacritinib was screened at at 10 $_{u}$M against a panel of 56 pharmacologically important receptors and enzymes. In these experiments, pacritinib caused > 50% inhibition against 4 enzymes and 12 receptors. Based on these findings, cardiovascular safety pharmacology studies were conducted.

Primary Pharmacodynamics

JAK2 and FLT3 as Drug Targets

The Janus kinases (JAK) are a family of cytoplasmic tyrosine kinases consisting of JAK1, JAK2, JAK3 and TYK2. They play a pivotal role in the signaling pathways of numerous cytokines, hormones and growth factors[(1)]. Their intracellular substrates include the family of proteins called Signal Transducer and Activator of Transcription (STAT). The JAK-STAT pathways, through the proper actions of the ligands, regulate important physiological processes such as immune response to viruses, hematopoiesis, lactation, lipid homeostasis, etc. However, dysfunctional signaling caused by a myriad of factors results in pathological conditions such as allergies, asthma, rheumatoid arthritis, severe combined immune deficiency, hematological malignancies, etc. In particular, mutations in *JAK2* have been associated with myeloproliferative disorders (including PV, ET, and idiopathic MF) and a wide range of leukemias and lymphomas[(2)]. Importantly, the myeloproliferative disorders constitute an area of unmet medical need in which treatment modalities have not been updated over the past few decades[(3)].

JAK2 mutations have been recently established as the most common genetic lesions in MPNs, and the putative role of *JAK2* in promoting lymphoid malignancies has been reported. The constitutively activated fusion protein Tel-JAK2 has been reported in rare cases of lymphoid leukemia[(4-6)]. Ectopic expression of various Tel-JAK2 constructs was shown to induce lymphoproliferative diseases in mouse models[(7-9)]. Recent reports of JAK2 overexpression due to *SOCS-1* inactivation in human lymphoma cell lines and primary samples further substantiate a role for JAK2 in lymphoproliferative diseases[(10-12)]. Together, these observations argue for the clinical evaluation of JAK2 inhibition as a novel therapeutic approach in lymphoid malignancies with *JAK2* activation and in myeloproliferative disorders.

As an inhibitor of FLT3, pacritinib has potential application for the treatment of leukemia. A family of Class III receptor tyrosine kinases (RTK), including c-fms, c-Kit, fms-like receptor tyrosine kinase 3 (FLT3), and platelet-derived growth factor receptors (PDGFR α and β) are important in the maintenance, growth, and development of both hematopoietic and non-hematopoietic cells[13]. Overexpression and activating mutations of these RTKs are involved in the pathophysiology of diverse human cancers from both solid and hematologic origins[14]. FLT3 mutations were first reported as internal tandem duplications (ITD) of the juxtamembrane domain-containing sequence[15]; subsequently, point mutations, deletions, and insertions surrounding the D835 coding sequence also were found[15]. FLT3 mutations are the most frequent genetic mutations reported in AML and are involved in the signaling pathway of autonomous proliferation and differentiation block in leukemia cells[16]. Several clinical studies have confirmed that *FLT3/ITD* is strongly associated with a poor prognosis[16]. Because high-dose chemotherapy and stem cell transplantation (SCT) cannot overcome the adverse effects of *FLT3* mutations[16], the development of FLT3 inhibitors is a promising therapeutic strategy.

Kinase Spectrum of Pacritinib

The spectrum of kinases inhibited by pacritinib was evaluated by in vitro assays against a total of 46 kinases and 2 mutants, which constitute a representative cross section of the kinase groups in the human kinome. Initial screens were performed using a single concentration of 0.1 μM pacritinib; selected kinases were then evaluated further in dose response titration.

Pacritinib is a potent inhibitor of both wild-type JAK2 (IC_{50} = 23 nM) and the mutant form of JAK2V617F that is present in patients with MPNs (IC_{50} = 19 nM). Importantly, pacritinib demonstrated an approximately 61-fold selectivity for JAK2 relative to JAK1 and 25-fold selectivity for JAK2 relative to JAK3. As JAK3 deficiency has been shown to lead to severe immune disorders in humans[17], it would appear desirable to avoid inhibiting this JAK isoform when developing a JAK2 inhibitor for MPNs. This specificity for JAK2 inhibition compared with JAK1 inhibition may contribute to differences in clinical efficacy and/or safety, given their differential involvement in cytokine signal transduction[18].

Pacritinib is also a potent inhibitor of FLT3 (IC_{50} = 22 nM) and its drug-resistant D835Y mutant (IC_{50} = 6 nM). However, pacritinib differs from other FLT3 compounds that have been studied in clinical trials, as it has no activity against most of the other class III RTKs[16]. Because of its potency and specificity, pacritinib may offer a unique therapeutic application in AML.

In Vitro Cellular Activity

The antiproliferative activity of pacritinib was evaluated in vitro against cancer cell lines obtained from either ATCC (Manassas, Virginia, US), ECACC (Salisbury, Wiltshire, UK), or, for murine BaF3 cells expressing the JAK2V617F mutant kinase, from Dr. Martin Sattler at the Dana Farber Cancer Institute (Boston, Massachusetts, US). Cells were cultivated according to instructions provided by the suppliers. Cells were seeded in 96-well plates at their exponential growth phase; plates were incubated at 37°C, 5% CO_2, for 24 hours. Cells were then treated with compounds at various concentrations for 96 hours; each compound was tested in triplicate.

Tumor cell growth was monitored using the CellTiter 96® Aqueous One Solution cell proliferation assay (Promega). Dose-response curves were plotted to determine IC_{50} values for the compounds using XL-fit (IDBS Ltd).

Results of testing, summarized in Table--1, showed that pacritinib potently inhibited the proliferation of only a few tumor cell lines at submicromolar concentrations, consistent with its target selectivity.

Table--1
In Vitro Antiproliferative Activity of Pacritinib in Tumor Cell Lines

Cancer Type	Cell Line	Cellular IC_{50} (nM)[a]
AML	MV4-11	32
Megakaryoblastic (ET)	SET2	213
Myeloid (Murine)	32D (+IL-3)	160
B-cell Lymphoma	Karpas 1106P	240
AML	HL60	410
Breast	MCF7	430
Prostate	PC3	540
Ovary	A2780	690
Burkitt's Lymphoma	Ramos	920
Multiple Myeloma	U266	1100
AML	HEL	1200
Colon	Colo205	1200
Colon	HCT116	1300
Brain (Glioblastoma)	U87MG	2100
Lung	H460	2800

[a] IC_{50} values are averages of at least 2 independent experiments. CV was generally within ± 30%.

Abbreviations:

AML = acute myeloid leukemia CV = coefficient of variation ET = essential thrombocythemia
IC_{50} = 50% inhibitory concentration nM = nanomolar.

The most sensitive cell lines were either JAK2 dependent, including murine 32D (IC_{50} = 160 nM) and human Karpas 1106P (IC_{50} = 240 nM), or contained a mutant FLT3 protein (MV4-11, IC_{50} = 32 nM). The *JAK2* genotype of the 4 BaF3-JAK2V617F clones was verified by sequencing the plasmids to make sure they contain sequences for *JAK2V617F*. The ectopic expression of JAK2 in these cells was also verified by western blot. When tested with pacritinib in cell proliferation assays, these 4 clones gave IC_{50} ranging from 147 to 461 nM (Table--3).

Cell Line[a]	Cellular IC_{50} (nM)[b]
BaF3-JAK2V617F (SD1)	147
BaF3-JAK2V617F (SD5)	278
BaF3-JAK2V617F (SD6)	461
BaF3-JAK2V617F (SD7)	161

[a] In all assays, the cancer type was Pro B (murine).

[b] The cellular IC_{50} values are averages of 3 independent experiments. The CV was generally within ± 30%.

Abbreviations:

CV = coefficient of variation IC_{50} = 50% inhibitory concentration nM = nanomolar.

The IC_{50} values are apparently related to the level of basal phos-JAK2; the higher the basal phos-JAK2, the more resistant the clone was to pacritinib. For example, cell line SD1 showed the lowest phos-JAK2 level and lowest IC_{50} of 147 nM, while cell line SD6 showed the highest level of phos-JAK2 and highest IC_{50} of 461 nM.

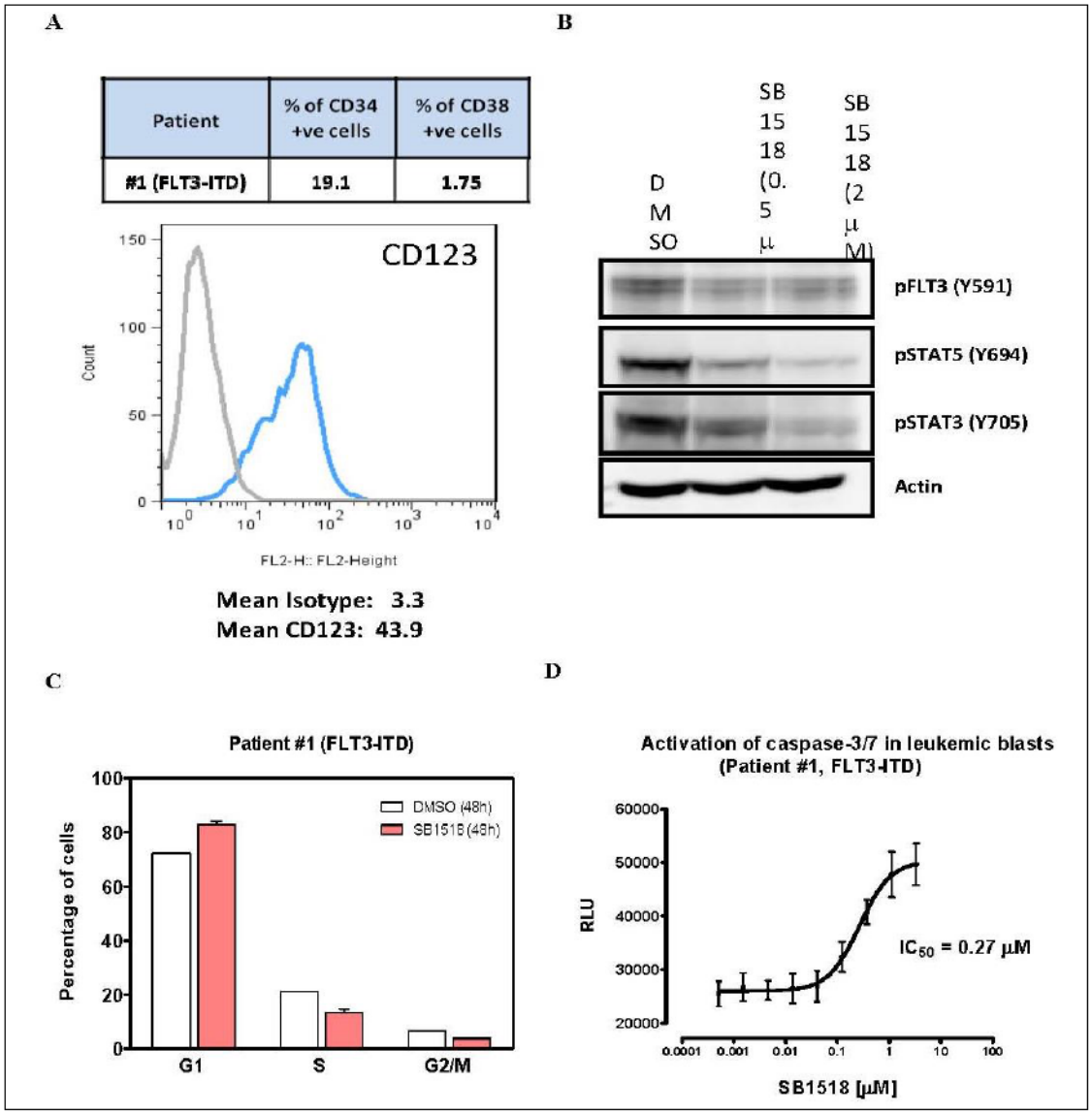
The effects of pacritinib were also examined on ex vivo expanded erythroid progenitors (EPs) from healthy volunteers and patients with PV; PV is an MPN in which the erythroblastic lineage acquires a hyperproliferative potential, but retains normal maturation, leading to elevated hematocrit and its attendant risks[19]. The results showed that pacritinib inhibited phos-STAT5 levels in a dose-dependent manner (IC_{50} < 200 nM) and reduced the viability of expanded EPs from both normal volunteers with *JAK2wt* (mean IC_{50} = 260 nM) and PV patients with *JAK2V617F* (mean IC_{50} = 230 nM), with no significant differences observed between groups. Moreover, pacritinib treatment had no effect on the *JAK2V617F* allele frequency in EPs from PV patients, indicating similar drug sensitivity for EPs from the same patient, regardless of the presence of *JAK2* mutation. This suggests that the constitutively activated

JAK2 mutant and the EPO-activated wild-type JAK2 exert equivalent quantitative impact on proliferative pathways in EPs. Similar effects were observed with other JAK inhibitors of different chemical classes and kinase selectivity profiles.

Since genetic and protein aberrations of FLT3 are most frequently encountered in AML, the effects of pacritinib were studied in primary blast cells expanded from PBMCs of thirteen patients with AML. Pacritinib inhibited the viability of blast cells with a mean IC_{50} of 0.47 $\pm$ 0.32 μM. Flow cytometric analyses verified that the expanded cells were predominantly leukemic stem cells, specifically CD38– /CD123+ (
Figure--1, 1A). Pacritinib caused blockade of FLT3 signaling pathway, as shown by the dose-dependent reduction of FLT3 receptor phosphorylation as well as downstream markers pSTAT5 and pSTAT3 (
Figure--1, 1B). The signaling inhibition resulted in cell cycle arrest at the G1 phase (
Figure--1, 1C) and subsequent apoptotic induction (
Figure--1, 1D). These collective data suggest that pacritinib holds potential for AML therapy.

Figure--1

Pacritinib (SB1518) Potently Blocked FLT3 Signaling and Induced Apoptosis in Expanded AML Blast



Peripheral blood mononuclear cells (PBMCs) or bone marrow mononuclear cells (BMMCs) were purchased from AllCells LLC (Emeryville, California, USA) and ProteoGenex Inc. (Culver City, California, USA). Cells were grown for 12 days in an AML blast expansion medium (StemSpan Serum-Free Expansion Medium containing 100 ng/mL of FLT3 ligand, 100 ng/mL

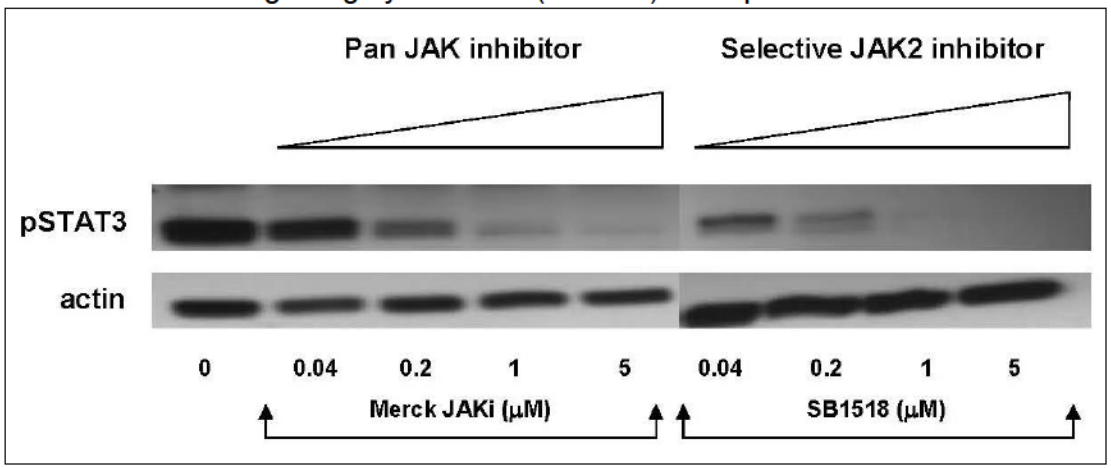
of stem cell factor, 20 ng/mL of interleukin-3 and 20 ng/mL of interleukin-6). (A) Flow cytometric analysis of CD34, CD38 and CD123. (B) AML blast cells were treated with pacritinib for 3 h, lysed and analyzed by western blot for FLT3, STAT3 and STAT5 phosphorylation. (C) AML blast cells were treated with 0.35 □M pacritinib for 48 h and the cell cycle distribution of the cell was analyzed using propidium iodide staining. (D) AML blast cells were treated with pacritinib for 16 h and Caspase-3/7 activity was measured using the Promega Caspase-Glo 3/7 assay.

Effects of Pacritinib on Intracellular JAK2 Signaling

To ascertain that compound exposure led to target inhibition in cells, we examined the effects of pacritinib on protein substrates downstream of JAK2 signaling. The STAT3 protein is a known substrate of JAK2. Constitutive activation of STAT3, resulting in high basal levels of phos-STAT3, has been found in multiple myeloma, leukemias, lymphomas and several types of solid tumors such as breast and prostate cancer[20]. In the case of Karpas 1106P cells, the genetic deletion of negative regulator gene *SOCS-1* results in enhanced stability and constitutive activation of JAK2, providing the basis for high basal levels of phos-STAT3. In the 32D murine myeloid cell line, treatment with interleukin-3 activates the JAK2 signaling pathway, leading to phosphorylation of STAT3.

As shown below, phos-STAT3 was reduced in a dose-dependent manner in Karpas 1106P (Figure--2) and murine 32D (Figure--3) cells. In the Karpas cells (Figure--2), the strong signal reduction at 0.2 □M was consistent with the observed cellular IC_{50} values.

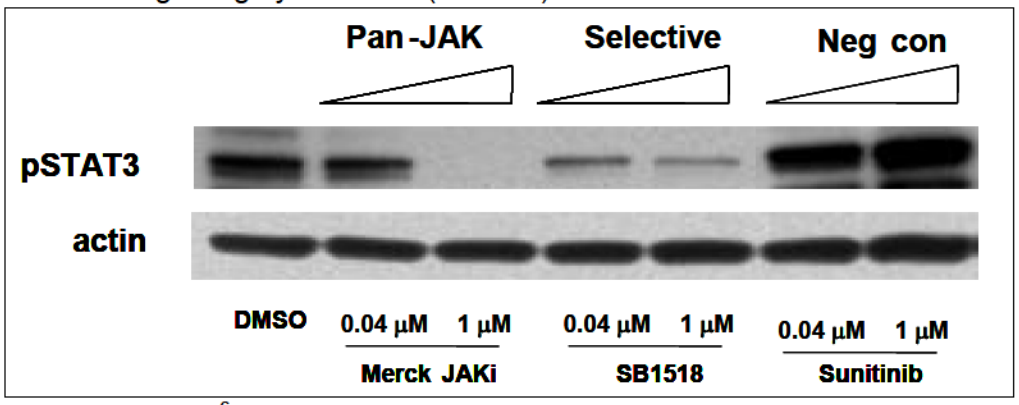
Figure--2
Inhibition of JAK2 Signaling by Pacritinib (SB1518) in Karpas 1106P Cells



Karpas cells (1 x 10^6) were treated with various concentrations of pacritinib or Merck JAKi for 4 h. 30 μg of cell lysates were resolved on SDS-PAGE, transferred onto PVDF membranes, and probed with the respective antibodies. Merck JAKi, a pan-JAK inhibitor, is

commercially available from Calbiochem catalog by the name of JAK Inhibitor I or Pyridone-6.

Figure--3
Inhibition of JAK2 Signaling by Pacritinib (SB1518) in Murine 32D Cells

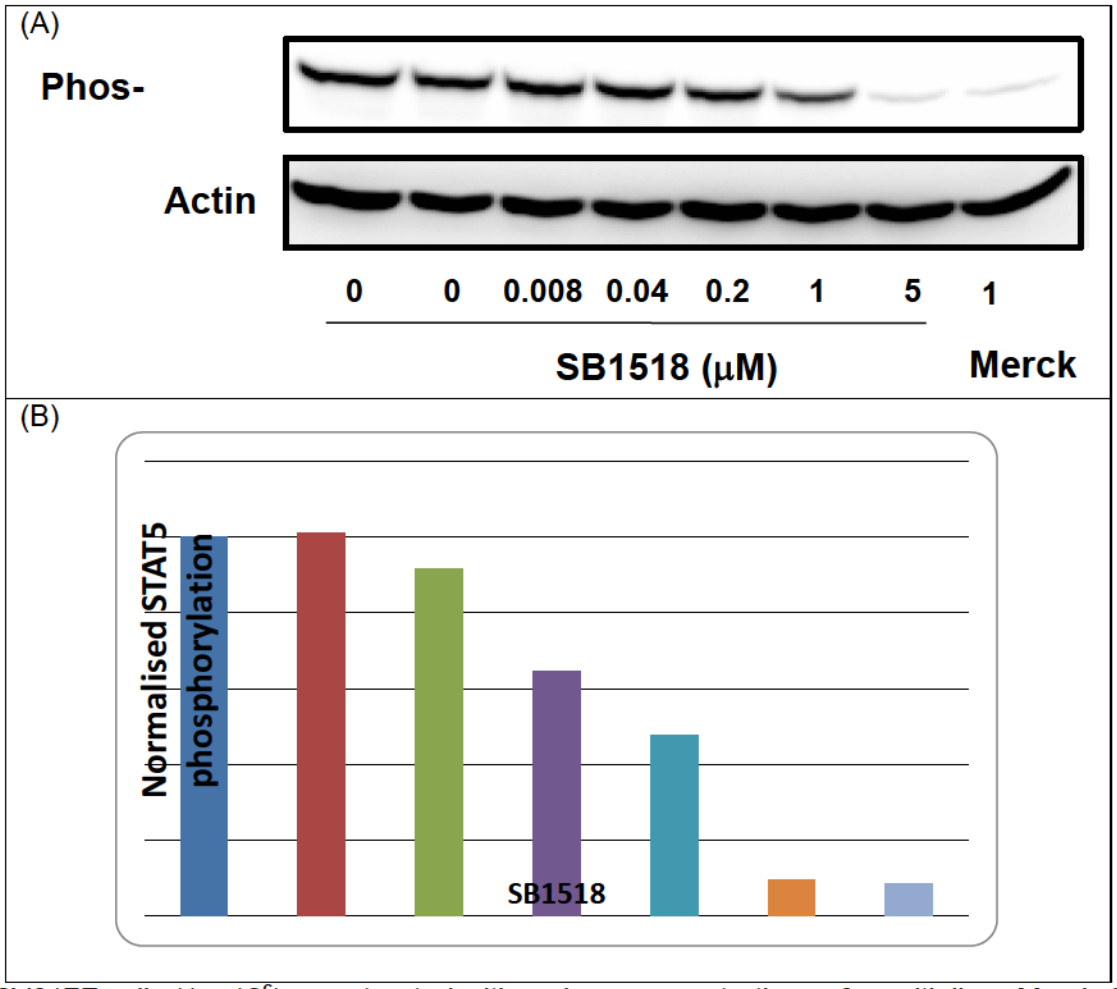


32D cells (1 x 10^6) were treated with various concentrations of the above compounds for 3 h. 30 µg of cell lysates were resolved on SDS-PAGE, transferred onto PVDF membranes, and probed with the respective antibodies. Sunitinib is an approved drug with the trade name of SUTENT and is a potent inhibitor of *FLT3*, but not *JAK2*.

In IL3-treated 32D cells (Figure--3), phos-STAT3 levels were reduced specifically by pacritinib (SB1518) at 0.4 and 1 µM, but not by sunitinib (Sutent®, an inhibitor of FLT3, but not JAK2, activity). This comparison led to the conclusion that the potent FLT3 activity in pacritinib is not responsible for blocking STAT3 phosphorylation.

With the BaF3-JAK2V617F clone SD6, the effect of pacritinib was also shown to inhibit the signaling pathway of JAK2 (Figure--4).

Figure--4
Inhibition of JAK2 Signaling by Pacritinib (SB1518) in BaF3- $JAK2^{V617F}$ Cells (Clone SD6)



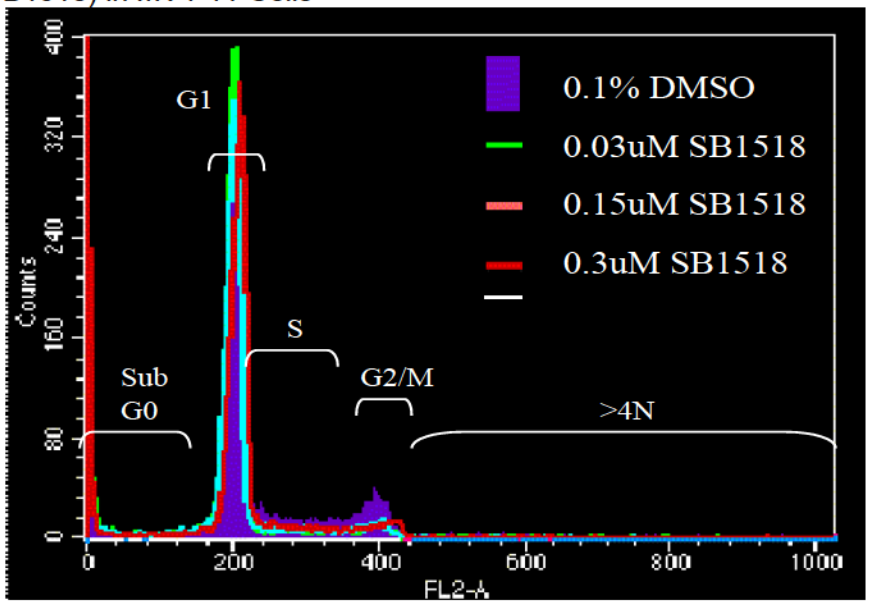
(A) BaF3-JAK2V617F cells (1 x 10^6) were treated with various concentrations of pacritinib or Merck JAKi for 4 h. 30 µg of cell lysates were resolved on SDS-PAGE, transferred onto PVDF membranes and probed with the respective antibodies. (B) Densitometric scans of Western blots in panel A were used to quantitate phosphorylated STAT5 and actin levels. Levels of phospho-STAT5 were normalized to actin.

Overall, these data have provided evidence that pacritinib can permeate tumor cell lines to modulate its kinase targets.

Pacritinib-Induced Cell Cycle Arrest and Apoptotic Killing in MV4-11 Cells

To investigate the effect of FLT3 inhibition on cell cycle progression, FACS analysis with propidium iodide staining was performed on MV4-11 cells treated with pacritinib or vehicle control. Results from representative experiments are depicted in Figure--5. Pacritinib arrested the cells at G_1/S phase and increased the sub-G_o apoptotic cell population.

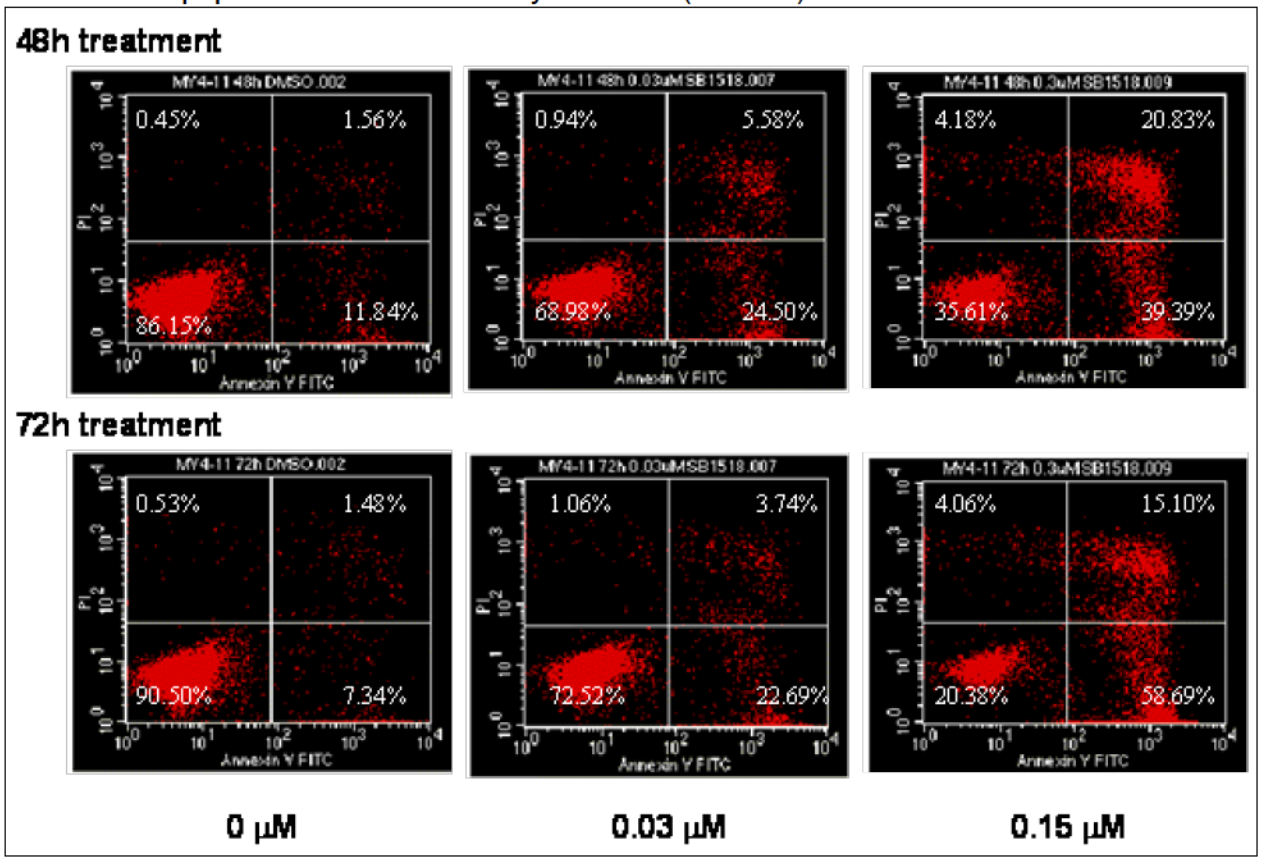
Figure--5
Cell Cycle Effects of Pacritinib (SB1518) in MV4-11 Cells



MV4-11 cells were treated with the indicated concentrations of pacritinib (approximately 1x, 5x, and 10x cellular IC_{50}) for 24 h. After treatment, cells were fixed, stained for DNA with propidium iodide, and analyzed on a FACS Calibur cell sorter. Data were quantified using CellQuest software (BD Bioscience).

The apoptotic effects were further confirmed by treating cells with pacritinib or vehicle for 48 or 72 hours, and then performing FACS analysis on annexin V-FITC and propidium-iodide co-stained cells (Figure--6). Increased, dose-dependent apoptosis (as demonstrated by increased annexin V staining) was seen in pacritinib-treated cells compared with control. Particularly seen at the higher pacritinib dose (0.15 μM), longer treatment time resulted in a greater percent of apoptotic cells with lower propidium iodide staining intensity, consistent with increased apoptosis in a sub-G_o cell population.

Figure--6
Induction of Apoptosis in MV4-11 Cells by Pacritinib (SB1518)



MV4-11 cells were treated with vehicle, 0.03 and 0.15 µM pacritinib (approximately 1x and 5x cellular IC_{50}) for 48 or 72 hours. After treatment, cells were fixed, stained with propidium iodide and annexin V-FITC, and analyzed on a BD Bioscience FACS Calibur cell sorter. Data were quantified using CellQuest software (BD Bioscience).

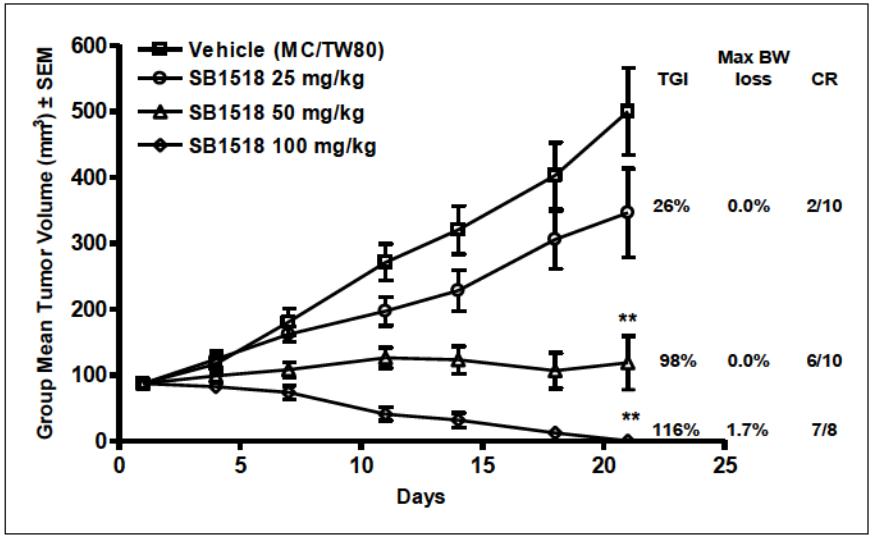
In Vivo Antitumor Activity

MV4-11 Model (FLT3 Mutant)

The antitumor activity of pacritinib was evaluated in a model of human AML with FLT3 mutation, MV4-11. MV4-11 cells (5×10^6 in Matrigel, BD Biosciences) were implanted SC in the flanks of female BALB/c athymic nude mice who were 10 to 12 weeks of age. When tumors reached a volume of 63 to 126 mm^3, treatment was initiated with daily PO doses of pacritinib (0, 25, 50, or 100 mg/kg) for 21 consecutive days. At the end of the 21-day treatment period, antitumor response was assessed by tumor growth inhibition (TGI, by one-way ANOVA followed by Dunnett's multiple comparison test).

Results showed a dose-dependent inhibition of tumor growth, with TGI of 26%, 98%, and 116% at 25, 50, and 100 mg/kg pacritinib, respectively (see Figure--7); tumor volumes were significantly different ($p < 0.01$) for the mid- and high-dose groups compared with vehicle controls.

Figure--7
Antitumor Activity of Pacritinib (SB1518) in a Murine Model of AML with FLT3 Mutation (MV4-11)



MV4-11 tumors were established in female BALB/c nude mice by s.c. implantation of 5×10^6 cells with Matrigel (BD Biosciences). All treatments were initiated on Day 1 when the group mean tumor volume reached 87 mm^3 and continued until Day 21. n = 8 for the 100 mg/kg group, n =10 for all the other groups. . * $p < 0.05$, ** $p < 0.01$ by one-way ANOVA followed by Dunnett's Multiple Comparison Test

Complete tumor regressions (CR, defined as tumor shrinkage to below measurable size 3 × 3 mm for 3 consecutive measurements) were observed for 2/10, 6/10, and 7/8 mice at 25, 50, and 100 mg/kg, respectively. All but 2 CRs were maintained for 28 days after the last dose. All the doses of pacritinib were well tolerated, with no significant body weight loss in any study group.

BaF3 Engraftment Model (JAK2 Mutant)

The antitumor activity of pacritinib also was evaluated in a human leukemia-like model with *JAK2* mutation. BaF3-JAK2V617F cells injected IV into nude mice led to leukocytosis, splenomegaly, hepatomegaly, and, in the late stage, hematopoietic crisis and death.

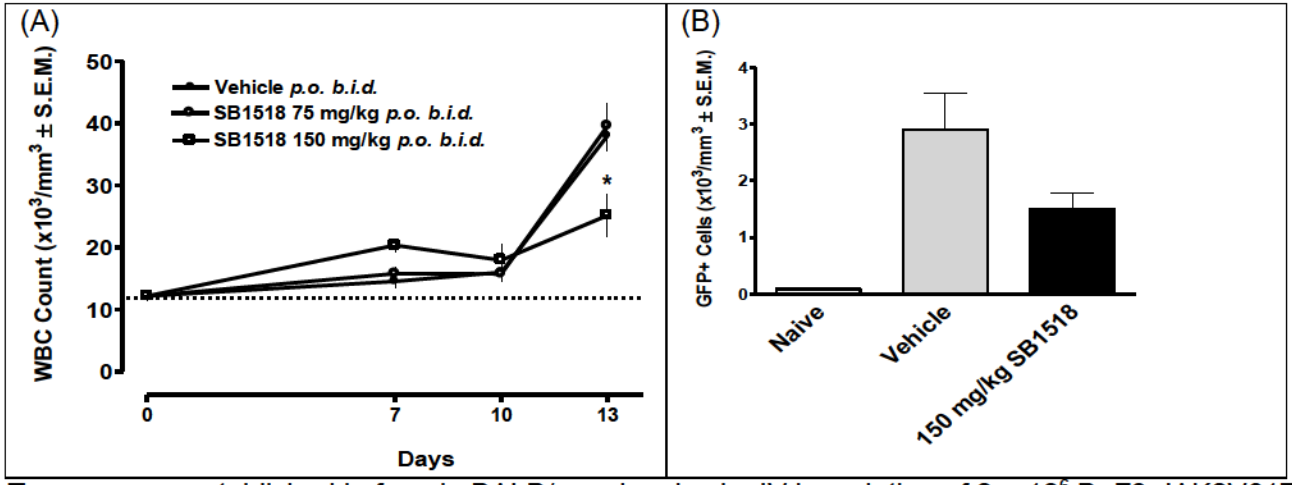
In this study, BaF3-JAK2V617F cells (SD6 in Table--3, 2×10^6 BaF3-JAK2V617F-GFP-Luc cells in serum-free RPMI1640 medium) were injected IV into the tail vein of female BALB/c nude mice. Three days after injecting the cells, treatment was initiated with daily PO doses of vehicle control, pacritinib at 75 mg/kg BID, or pacritinib at 150 mg/kg BID for 14 consecutive days. At the end of the treatment period, mice were sacrificed by elevating the carbon dioxide levels and blood was collected by cardiac puncture. Spleens and livers were removed and weighed. Two-way ANOVA followed by Dunnett's multiple comparison test was used to determine the statistical significance of various measurements between treated and control groups.

At the end of the treatment period, vehicle control mice showed spleen/liver hyperplasia and severe leukocytosis. Only the high-dose pacritinib treatment regimen caused a significant reduction in leukocytosis, paralleled by a decrease in tumor cell burden, as measured by GFP-labeled BaF3-JAK2V617F cells (see

Figure--8).
At the anatomical level, pacritinib treatment led to a dose-dependent reduction in both spleen and liver enlargement, as shown in

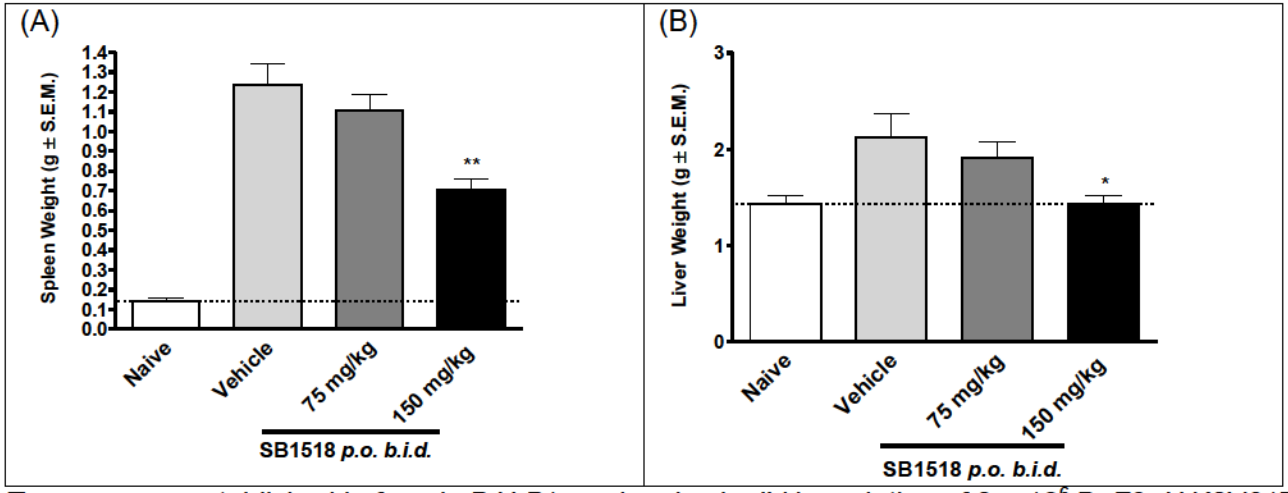
Figure--9. The higher dosage required to achieve comparable therapeutic effects in the BaF3 model relative to the MV4-11 model (300 mg/kg vs 100 mg/kg daily) is consistent with the relative potency for the anti-proliferative effects of pacritinib on these cells in vitro (IC_{50} = 461 nM and 32 nM, respectively). Together, these studies demonstrate the potential therapeutic benefits of pacritinib in human diseases that are driven by mutant *JAK2*.

Figure--8
Effect of Pacritinib (SB1518) on Leukocytosis and Tumor Cell Load in the BaF3 Model



Tumors were established in female BALB/c nude mice by IV inoculation of 2 × 10^6 BaF3-JAK2V617F cells at Day -4, followed by daily treatment of mice with vehicle or pacritinib (SB1518), as indicated. (A) Blood cell counts were taken by tail vein bleeding at Days 0, 7, 10, and 13. Blood samples were taken by cardiac puncture and analyzed using a Scilvet ABC animal blood counter. The total white blood cell (WBC) count is shown. * = $p < 0.05$ in ANOVA/Dunnett's multiple comparison test compared with vehicle. (B) At Day 13, whole blood cell counts were taken, and fixed-lysed whole blood samples were prepared. The percent of GFP-positive cells was quantified by FACS analysis, and the total numbers were calculated from WBC counts of corresponding animals. The 75 mg/kg pacritinib bone marrow samples were not analyzed for GFP cells.

Figure--9
Effect of Pacritinib (SB1518) on Spleen and Liver Enlargement in the BaF3 Engraftment Model



Tumors were established in female BALB/c nude mice by IV inoculation of 2 × 10^6 BaF3-JAK2V617F cells at Day -4, followed by daily treatment of mice with pacritinib (SB1518) or vehicle control, as indicated. One additional group of mice remained naïve (n = 3, no cells/no treatment). All groups of mice were sacrificed on Day 6. The **(A)** spleen and **(B)** liver weights. ** = $p<0.001$ and * = $p<0.01$ in ANOVA/Dunnett's multiple comparison test, compared to vehicle.

HL60 and HCT116 Models

Pacritinib was also tested in two xenograft models lacking *FLT3* or *JAK2* mutations, the HCT116 SC model for human colorectal carcinoma and the HL60 orthotopic model for human non-FLT3-dependent AML.

In the HCT116 model, 5 × 10^6 HCT116 cells were implanted SC in the flank of female BALB/c athymic nude mice (10 per treatment group) at 10 to 12 weeks of age. When the group mean tumor volume reached 133 to 135 mm^3, treatment was initiated with daily PO dosing of pacritinib (0, 25, 50, or 100 mg/kg) for 14 days. At the end of treatment, no tumor growth inhibition was observed (TGIs of 8%, 18%, and -8% for the 25, 50, and 100 mg/kg groups, respectively). No weight loss was observed for treated animals compared with controls.

In the HL60 model, 1 × 10^7 HL60 cells were inoculated IV into 6- to 7-week-old female NOD/SCID mice (9 per treatment group). Fourteen days later, treatment was initiated with daily PO doses of pacritinib (0, 50, 100, and 150 mg/kg) for 22 days. Vehicle control animals developed bilateral hind-leg paralysis, a hallmark for this animal model, starting 29 days after cell inoculation. Animals were followed for survival. There were no differences in median survivals for the vehicle control and treated groups (median survival of 42, 43, 43, and 42 days at 0, 50, 100 and 150 mg/kg, respectively). No weight loss was seen in treated animals compared with controls.

The lack of efficacy for pacritinib in models that have no mutations in *FLT3* or *JAK2* genes and its excellent tolerability in these models (no body weight loss), shows the specificity of pacritinib for JAK2 or FLT3-driven disease.

The antiproliferative activity of pacritinib was evaluated in vitro against cancer cell lines obtained from either ATCC (Manassas, Virginia, US), ECACC (Salisbury, Wiltshire, UK), or, for murine BaF3 cells expressing the JAK2V617F mutant kinase, from Dr. Martin Sattler at the Dana Farber Cancer Institute (Boston, Massachusetts, US). Cells were cultivated according to instructions provided by the suppliers. Cells were seeded in 96-well plates at their exponential growth phase; plates were incubated at 37°C, 5% CO_2, for 24 hours. Cells were then treated with compounds at various concentrations for 96 hours; each compound was tested in triplicate. Tumor cell growth was monitored using the CellTiter 96® Aqueous One Solution cell proliferation assay (Promega). Dose-response curves were plotted to determine IC_{50} values for the compounds using XL-fit (IDBS Ltd).

Results of testing, summarized in Table--1, showed that pacritinib potently inhibited the proliferation of only a few tumor cell lines at submicromolar concentrations, consistent with its target selectivity.

Table--2
In Vitro Antiproliferative Activity of Pacritinib in Tumor Cell Lines

Cancer Type	Cell Line	Cellular IC_{50} (nM)[a]
AML	MV4-11	32
Megakaryoblastic (ET)	SET2	213
Myeloid (Murine)	32D (+IL-3)	160
B-cell Lymphoma	Karpas 1106P	240
AML	HL60	410
Breast	MCF7	430
Prostate	PC3	540
Ovary	A2780	690
Burkitt's Lymphoma	Ramos	920
Multiple Myeloma	U266	1100
AML	HEL	1200
Colon	Colo205	1200
Colon	HCT116	1300

Table--2 In Vitro Antiproliferative Activity of Pacritinib in Tumor Cell Lines		
Cancer Type	**Cell Line**	**Cellular IC_{50} (nM)[a]**
Brain (Glioblastoma)	U87MG	2100
Lung	H460	2800

[a] IC_{50} values are averages of at least 2 independent experiments. CV was generally within ± 30%.

Abbreviations:

AML = acute myeloid leukemia CV = coefficient of variation ET = essential thrombocythemia

IC_{50} = 50% inhibitory concentration nM = nanomolar.

The most sensitive cell lines were either JAK2 dependent, including murine 32D (IC_{50} = 160 nM) and human Karpas 1106P (IC_{50} = 240 nM), or contained a mutant FLT3 protein (MV4-11, IC_{50} = 32 nM). The *JAK2* genotype of the 4 BaF3-JAK2V617F clones was verified by sequencing the plasmids to make sure they contain sequences for *JAK2V617F*. The ectopic expression of JAK2 in these cells was also verified by western blot. When tested with pacritinib in cell proliferation assays, these 4 clones gave IC_{50} ranging from 147 to 461 nM (Table--3).

Table--3 In Vitro Antiproliferative Activity of Pacritinib in Four BaF3-JAK2V617F-Positive Cell Clones	
Cell Line[a]	**Cellular IC_{50} (nM)[b]**
BaF3-JAK2V617F (SD1)	147
BaF3-JAK2V617F (SD5)	278
BaF3-JAK2V617F (SD6)	461
BaF3-JAK2V617F (SD7)	161

[a] In all assays, the cancer type was Pro B (murine).

[b] The cellular IC_{50} values are averages of 3 independent experiments. The CV was generally within ± 30%.

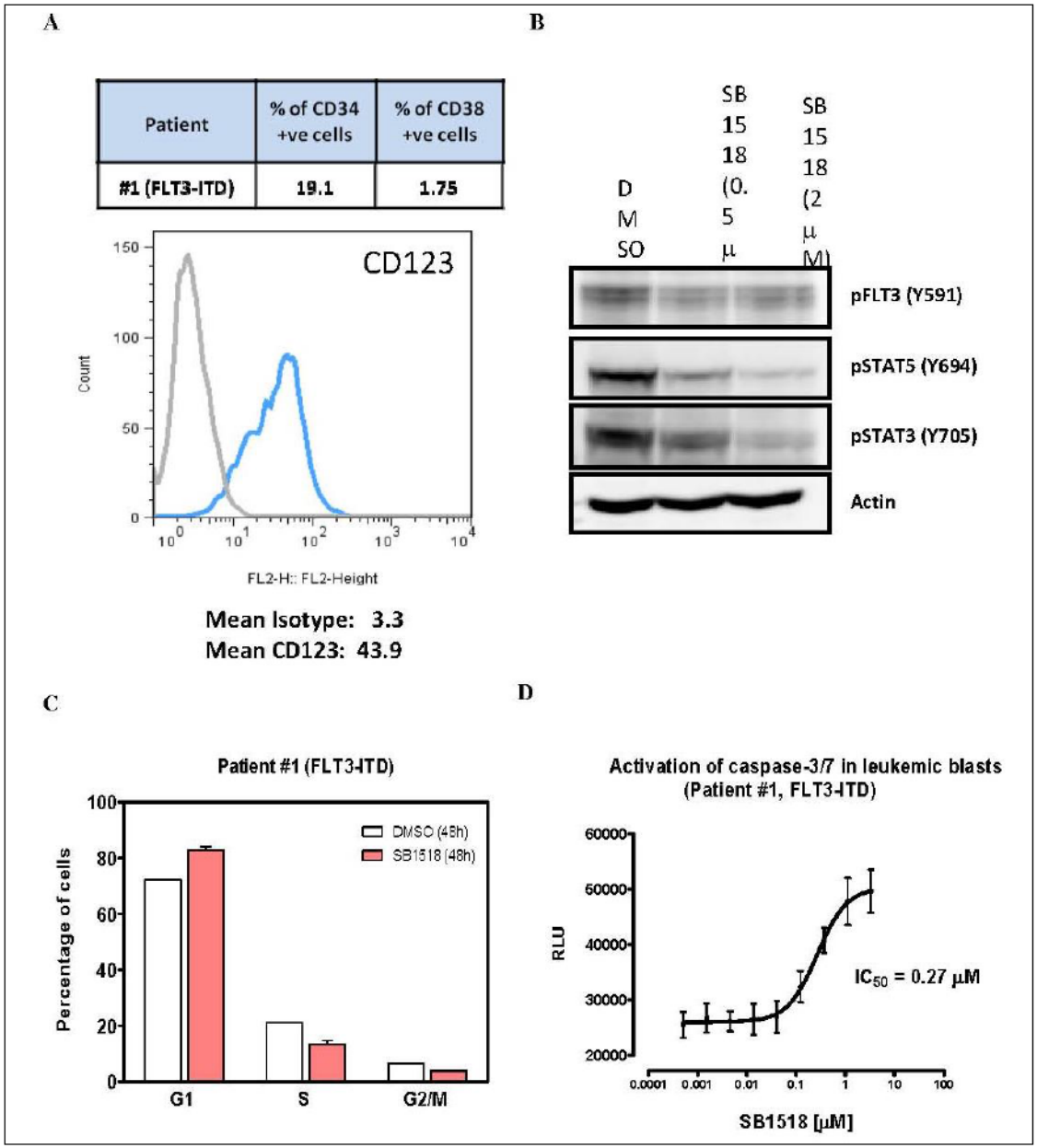
Abbreviations:

CV = coefficient of variation IC_{50} = 50% inhibitory concentration nM = nanomolar.

The IC_{50} values are apparently related to the level of basal phos-JAK2; the higher the basal phos-JAK2, the more resistant the clone was to pacritinib. For example, cell line SD1 showed the lowest phos-JAK2 level and lowest IC_{50} of 147 nM, while cell line SD6 showed the highest level of phos-JAK2 and highest IC_{50} of 461 nM.

The effects of pacritinib were also examined on ex vivo expanded erythroid progenitors (EPs) from healthy volunteers and patients with PV; PV is an MPN in which the erythroblastic lineage acquires a hyperproliferative potential, but retains normal maturation, leading to elevated hematocrit and its attendant risks[19]. The results showed that pacritinib inhibited phos-STAT5 levels in a dose-dependent manner (IC_{50} < 200 nM) and reduced the viability of expanded EPs from both normal volunteers with *JAK2wt* (mean IC_{50} = 260 nM) and PV patients with *JAK2V617F* (mean IC_{50} = 230 nM), with no significant differences observed between groups. Moreover, pacritinib treatment had no effect on the *JAK2V617F* allele frequency in EPs from PV patients, indicating similar drug sensitivity for EPs from the same patient, regardless of the presence of *JAK2* mutation. This suggests that the constitutively activated JAK2 mutant and the EPO-activated wild-type JAK2 exert equivalent quantitative impact on proliferative pathways in EPs. Similar effects were observed with other JAK inhibitors of different chemical classes and kinase selectivity profiles.

Since genetic and protein aberrations of FLT3 are most frequently encountered in AML, the effects of pacritinib were studied in primary blast cells expanded from PBMCs of thirteen patients with AML. Pacritinib inhibited the viability of blast cells with a mean IC_{50} of 0.47 $\pm$ 0.32 µM. Flow cytometric analyses verified that the expanded cells were predominantly leukemic stem cells, specifically CD38– /CD123+ (
Figure--1, 1A). Pacritinib caused blockade of FLT3 signaling pathway, as shown by the dose-dependent reduction of FLT3 receptor phosphorylation as well as downstream markers pSTAT5 and pSTAT3 (
Figure--1, 1B). The signaling inhibition resulted in cell cycle arrest at the G1 phase (
Figure--1, 1C) and subsequent apoptotic induction (
Figure--1, 1D). These collective data suggest that pacritinib holds potential for AML therapy.
Figure--10
Pacritinib (SB1518) Potently Blocked FLT3 Signaling and Induced Apoptosis in Expanded AML Blast


A
Patient
% of CD34 +ve cells
% of CD38 +ve cells
#1 (FLT3-ITD)
19.1
1.75
CD123
Count
FL2-H:: FL2-Height
Mean Isotype: 3.3
Mean CD123: 43.9
B
DMSO
SB1518 (0.5 µM)
SB1518 (2 µM)
pFLT3 (Y591)
pSTAT5 (Y694)
pSTAT3 (Y705)
Actin
C
Patient #1 (FLT3-ITD)
DMSO (48h)
SB1518 (48h)
Percentage of cells
G1
S
G2/M
D
Activation of caspase-3/7 in leukemic blasts (Patient #1, FLT3-ITD)
RLU
IC50 = 0.27 µM
SB1518 [µM]

Peripheral blood mononuclear cells (PBMCs) or bone marrow mononuclear cells (BMMCs) were purchased from AllCells LLC (Emeryville, California, USA) and ProteoGenex Inc. (Culver City, California, USA). Cells were grown for 12 days in an AML blast expansion medium (StemSpan Serum-Free Expansion Medium containing 100 ng/mL of FLT3 ligand, 100 ng/mL of stem cell factor, 20 ng/mL of interleukin-3 and 20 ng/mL of interleukin-6). (A) Flow cytometric analysis of CD34, CD38 and CD123. (B) AML blast cells were treated with pacritinib for 3 h, lysed and analyzed by western blot for FLT3, STAT3 and STAT5 phosphorylation. (C) AML blast cells were treated with 0.35

Safety Pharmacology

Receptor and Enzyme Profiling

To assess the potential for safety issues in preclinical animal and clinical studies, pacritinib was profiled against an in vitro panel of 56 pharmacologically important receptors. In these screens, 10 μM pacritinib caused > 50% inhibition against 4 enzymes and 12 receptors (Table--4). Based on these findings, cardiovascular safety pharmacology studies were conducted.

Table--4
In Vitro Receptor Profiling for Pacritinib

Receptor	IC_{50} (μM)	Receptor	IC_{50} (μM)
Adenosine A_{2A}	0.40	Sigma σ_1	4.4
Insulin Receptor	0.50	Sigma σ_2	5.5
Serotonin 5-HT_4	1.3	Dopamine D_1	5.6
Sodium Channel, Site 2	1.4	Adrenergic α_{1B}	6.4
Norepinephrine Transporter	2.2	Acetylcholinesterase	8.0
Calcium Channel, L-type	2.8	Adenosine A_1	8.1
Monoamine Oxidase MAO-A	3.5	Dopamine D_{2S}	8.6
Potassium Channel hERG	3.5	Adrenergic α_{2A}	18.8

Abbreviations:
μM = micromolar concentration hERG = human ether-à-go-go-related gene IC_{50} = 50% inhibitory
MAO = monoamine oxidase.

Cardiovascular Safety Pharmacology

Cardiovascular Effects in Conscious Dogs

A GLP safety pharmacology study was conducted to evaluate cardiovascular system effects following a single oral administration of gel capsules containing 30 mg/kg pacritinib (or empty capsules) to groups (2/sex/group) of conscious, fasted dogs. Electrocardiographic examination, determination of blood pressure, and recordings of respiratory and heart rates were performed one week prior to study, immediately prior to dosing, and then 1, 3, 6, and 24 hours after dosing. Body temperature was monitored and blood samples for serum chemistry (ALT, AST, LDH, CK, and glucose) were performed in the week prior to study, immediately before dosing, and then at 1, 2, 3, 6, and 24 hours after dosing. Clinical signs and symptoms were assessed three times during the day of dosing, then twice daily during the observation period.

Administration of pacritinib induced vomiting (up to 1 min duration) in 3 of 4 dogs (2M, 1F) within 30 to 120 minutes after dosing; no other clinical symptoms were observed. Despite the observed gastrointestinal effect of treatment, no treatment-related changes were observed in cardiac electrophysiology (P, R-R, PQ, QRS, QT, and QTc intervals), heart rate, respiratory rate, body temperature, blood pressure, or activity following a single oral dose of 30 mg/kg pacritinib.

hERG Binding

Binding of pacritinib to potassium channel hERG was evaluated using human recombinant HEK-293 cells, as part of the enzyme profiling described earlier. Pacritinib showed binding to hERG with an IC_{50} of 3.51 μM.

Nonclinical Pharmacokinetics

Absorption

PK following a single intravenous or oral administration of pacritinib was evaluated in mice, rats and dogs. Following oral administration, pacritinib showed rapid absorption in mice (T_{max} from 0.5 to 1.3 h) and moderately fast absorption in rats and dogs (T_{max} ~4 h). The oral terminal half-life was 2.2, 5.7, and 4.4 h in mice, rats, and dogs, respectively. Pacritinib showed a high volume of distribution in mice (V_{ss}= ~ 65 L/kg), rat (V_{ss}=13 L/kg) and dogs (V_{ss}=8.5 L/kg). The systemic clearance of pacritinib in terms of liver blood flow was high in mice (8 L/h/kg) and dogs (1.6 L/h/kg), and moderate in rats (1.6 L/h/kg). The terminal half-life for IV administration was 5.6, 6, and 4.6 h, in mice, rats, and dogs, respectively. The oral bioavailability of pacritinib was 39%, 10% and 24% in mice, rats, and dogs, respectively (Table--5).

Table--5
Intravenous and Oral Pharmacokinetic Parameters of Pacritinib in the Mouse, Rat, and Dog

PK Parameters	Species, Route of Administration, and Pacritinib Dose

	Mouse (n=2 Females)		Rat (n=3 Males)		Dog (n=1 Male)	
	IV 10 mg/kg	**PO** 50 mg/kg	**IV** 2 mg/kg	**PO** 10 mg/kg	**IV** 1 mg/kg	**PO** 5 mg/kg
C_{max} (ng/mL)	NA	1108	NA	114	NA	89
T_{max} (h)	NA	1.3	NA	4	NA	4
AUC $_{0\text{-}24h}$ (ng-h/mL)	1231	2438	1243	599	632	735
t½ (h)	5.6	2.2	6.0	5.7	4.6	4.4
Cl or Cl/F (L/h/kg)	8.0	17.6	1.6	15.6	1.6	6.6
V_d or V_d/F (L/kg)	64.6	57.0	13.2	13.0	8.5	42.2
F (%)	NA	39	NA	10	NA	24

Abbreviations:

AUC = area under the concentration vs time curve

Cl = clearance C_{max} = maximum concentration F = bioavailability h = hour(s)

IV = intravenous(ly) L/kg = liters per kilogram L/h/kg = liters per hour per kilogram

mg/kg = milligrams per kilogram NA = not applicable ng-h/mL = nanogram-hour per milliliter

ng/mL = nanograms per milliliter PK = pharmacokinetic PO = per os, oral(ly)

$t_{1/2}$ = half-life T_{max} = maximum time V_d = volume of distribution

V_d/F = volume of distribution at steady state.

Distribution

Plasma Protein Binding

Plasma protein binding of pacritinib was measured by adding pacritinib (1000 ng/mL) to mouse, dog, or human plasma and performing equilibrium dialysis against phosphate-buffered saline (PBS) at 37°C for 4 hours. Pacritinib was extracted from the dialysate and analyzed by LC-MS/MS. In all 3 species, > 98% of the pacritinib was bound to plasma proteins (Table--6).

Given the generally higher systemic exposure observed in humans, a plasma protein binding study was conducted in human plasma using pacritinib plasma concentrations of 10-50 µg/mL. The results of this study indicate that mean plasma protein binding of

pacritinib in humans is 98.8% at clinically relevant plasma concentrations (~10 µg/mL) (see Table--7).

Table--6 Pacritinib Binding to Human, Dog, and Mouse Plasma Proteins	
Species[a]	**% Bound ± SD (n = 3/Species)**
Mean Plasma Protein Binding (± SD)	99.88 ± 0.01

Table--7 Pacritinib Binding To Human Plasma Proteins	
Pacritinib Concentration (µg/mL)	**% Bound ± SD (N = 3/Concentration)**
10	98.8 ± 0.32
25	97.7 ± 0.10
50	97.1 ± 0.84
Abbreviations: µg/mL = microgram per milliliter N = number SD = standard deviation.	

Tissue Distribution

In a preliminary study, limited tissue distribution was assessed in female BALB/c mice following a single oral administration of 50 mg/kg pacritinib. Pacritinib levels were higher in lungs (tissue:plasma ratio of 19) than in brain (tissue:plasma ratio of 1.8). In a definitive quantitative whole-body autoradiography (QWBA) in male BALB/cAnNCrl mice, the highest presence of radioactivity was detected in the small intestine (469.675 µg- equiv/g at 1 h); the lowest levels were in brain tissue, eye lens and testes, suggesting that pacritinib does not distribute into the CNS. Concentrations of drug-derived radioactivity decreased rapidly and most tissues were

below levels of quantification by 24 hr post-dose, indicating no drug retention in any tissue compartment (see Section **Error! eference source not found.**).

Genotoxicity

In a preliminary study, no mutagenic activity was observed for pacritinib, as assessed by the induction of DNA base-pair substitution or frame-shift mutations in *Salmonella typhimurium* strains (Ames test). The results from the preliminary study were confirmed with a bacterial reverse mutation test under GLP conditions. Five histidine-dependent strains of *S typhimurium* (TA98, TA100, TA1535, TA1537 and TA102) were used to evaluate the mutagenic potential of pacritinib, both in the presence and absence of metabolic activation (± S9). The study was carried out using the plate incorporation method and the pre-incubation methods. The vehicle used was dimethylsulfoxide (DMSO), and the maximum solubility of pacritinib in DMSO was 20 mg/mL. When tested up to the maximum achievable dose level of 2000 µg/plate (limit of solubility in the vehicle = 20 mg/mL), using both the plate incorporation and pre-incubation methods, pacritinib induced no biologically significant increases in the number of revertant colonies in the five *S typhimurium* strains used (TA98, TA100, TA1535, TA1537 and TA102), with or without metabolic activation.

Phototoxicity

The phototoxic potential of pacritinib citrate salt was evaluated in female Crl:SKH1-hr hairless mice. Groups of six mice were administered PO (via gavage) (1) pacritinib citrate salt at a dose of 150 mg/kg salt, BID for 4 consecutive days, (2) vehicle (0.5% methylcellulose and 0.1% Tween-80) in the same manner as for pacritinib, and (3) a single 50 mg/kg dose of 8-methoxypsoralen (8-MOP), the positive control. After 8 doses of pacritinib or vehicle and a single dose of 8-MOP, at 60 ± 10 minutes following the last dose, mice were exposed to UVR from a xenon lamp (to simulate sunlight). An instrumental UVR exposure dose equivalent to 0.5 minimal erythema dose (MED, a UVR dosage adequate to elicit a barely perceptible response in skin) was delivered to each mouse over a period of 30 ± 5 minutes. No skin reactions indicative of phototoxicity were observed following administrations of the vehicle or pacritinib citrate salt at 150 mg/kg/dosage. A single administration of 8-MOP resulted in skin reactions consistent with phototoxicity (erythema, edema, scab[s]), validating the assay. Pacritinib citrate salt was present in the formulation at the protocol-required specifications.

Carcinogenicity

No carcinogenicity studies have been performed.

Reproductive and Development Toxicity

The potential toxic effect of pacritinib on male fertility in mice was evaluated following treatment of the males only. Three treatment groups of 30 male BALB/c mice/group were administered pacritinib at respective dose levels of 30, 100, or 300 mg/kg/day (15, 50, or 150 mg/kg/dose BID). One additional group of 30 males served as the control and received the vehicle, 0.5% methylcellulose and 0.1% Tween-80 in deionized water. The vehicle or pacritinib was administered BID to all groups by oral gavage with approximately 8 hours between doses. Dosing of the males began 29 days prior to the first pairing with untreated females, and continued through the 21-day pairing period of the second pairing and post mating periods to euthanasia. The study was initially designed to have a single pairing period to assess reproductive performance and fertility. However, due to a lower-than-expected pregnancy rate in all groups in the first pairing, dosing of males was extended to provide for a second pairing with a second shipment of untreated females. Overall, males were treated for a total of 106 to 108 consecutive days.

Observations of the males included survival, daily clinical observations, body weights and body weight change, and food consumption. Females were also observed for survival and weight during pairing, gestation (first pairing), and post-pairing until scheduled termination. In the first pairing, treated males were paired with untreated females for up to 21 days or until evidence of mating (vaginal copulatory plug) was observed. Mated females from the first pairing were euthanized on presumed gestation day (GD) 13 and subjected to a complete necropsy; the total number of corpora lutea, embryos, resorptions, and the number of total implantations was recorded. In the second pairing, males were co-housed with untreated females for 15 consecutive days. Females were examined daily for the presence of a vaginal copulatory plug, but remained with the males for the entire period. Females from the second pairing that appeared to be pregnant based on body weight (≥ 28 g) were euthanized; the total numbers of corpora lutea, embryos, resorptions, and implantations were recorded. Females that did not appear pregnant from weight gain were euthanized and similarly examined 2 weeks following completion of the pairing period. Complete necropsies were performed on all animals; organs and tissues from the males were collected, weighed, and preserved. Sperm analyses (motility, concentration, and morphology) were also conducted. Homogeneity of the dosing formulations at the low and high concentrations were evaluated and confirmed at Week 1. Periodic evaluations for concentrations of dosing formulations used on study were conducted and ranged from 86.3% to 104.3% of nominal (acceptance criteria for suspension was ± 15% of target).

No effect of pacritinib on mortality at the dose levels evaluated was evident among the males. Lower body weights and effects on weight change considered toxicologically significant were seen in male mice treated at 300 mg/kg/day. Additional toxicity seen at 300 mg/kg/day included clinical findings (decreased activity, salivation, hunched posture, and white ocular discharge) and lower food consumption. Effects on reproductive performance (lower mating and fertility indices) were seen at 300 mg/kg/day, but no effects at any of the dose levels evaluated were seen from uterine implantation data, macroscopic findings, reproductive organ weights, or sperm analyses. Thus, the NOAEL with pacritinib for male reproductive performance and fertility was 100 mg/kg/day.

Local Tolerance

No local tolerance studies have been performed.

Effects in Humans

Six clinical studies with pacritinib have been completed and are summarized in Table—16

Effects in Humans

Six (6) clinical studies with pacritinib have been completed and are summarized in Table--8.

Table--8
Completed Clinical Studies with Pacritinib (SB1518)

Study No.: Study Title	Phase; Total Enrolled Patients	Dose Regimen	Study Results[a]
SB1518-2007-001: Phase 1/2 Study of SB1518 for the Treatment of Advanced Myeloid Malignancies	Phase 1; 43	Escalating dose cohorts: doses @ 100, 150, 200, 300, 400, 500, 600 mg. PO administration; 25 days of dosing in cycle 1 and 28 days in subsequent cycles; cycles repeated every 28 days for minimum of 2 cycles; addi-tional cycles allowed if tolerating treatment and no PD.	Study is completed and CSR is pending; thus, data are preliminary. **Dose (mg/day) / No. Pts / Observed DLTs** 100 / 3 / None 150 / 6 / Prolonged QTc 200 / 9 / None 300 / 6 / Diarrhea 400 / 6 / None 500 / 7 / None 600 / 6 / GI toxicity, dizziness, blurred vision, un-steady gait, worsened performance status ***Efficacy:*** 44% of patients achieved maximum reduction in spleen volume ≥ 35% by Week 24.
	Phase 2; 31	PO administration @ 400 mg; 28 days of dosing; repeated every 28 days for minimum of 2 cycles; additional cycles allowed if tolerating treatment and no PD.	Study is completed and CSR is pending; thus, data are preliminary. ***Efficacy:*** 16% of patients achieved maximum reduction in spleen volume ≥ 35% by Week 24.
SB1518-2007-002: Phase 1 Study of SB1518 for the Treatment of	35	Escalating dose cohorts: dose levels @ 100, 200, 300, 400, and 600 mg.	Study is completed and CSR is pending; thus, data are preliminary. **Dose (mg/day) / No. Pts / Observed DLTs**

<table>
<tr><th colspan="4">Table--8
Completed Clinical Studies with Pacritinib (SB1518)</th></tr>
<tr><th>Study No.:
Study Title</th><th>Phase; Total Enrolled Patients</th><th>Dose Regimen</th><th>Study Results[a]</th></tr>
<tr><td>Advanced Lymphoid Malignancies</td><td></td><td>PO administration; 28 days of dosing; repeated every 28 days.</td><td>
<table>
<tr><td>100</td><td>3</td><td>None</td></tr>
<tr><td>200</td><td>7</td><td>None</td></tr>
<tr><td>300</td><td>6</td><td>Neutropenia</td></tr>
<tr><td>400</td><td>7</td><td>Thrombocytopenia</td></tr>
<tr><td>600</td><td>12</td><td>Sepsis</td></tr>
</table>
Efficacy: 4 PR, 17 SD</td></tr>
<tr><td rowspan="2">SB1518-2008-003:
Phase 1/2 Study of SB1518 in Subjects with Chronic Idiopathic Myelofibrosis</td><td>Phase 1; 20</td><td>Escalating dose cohorts: doses @ 100, 200, 400, 500, and 600 mg.
PO administration; 28 days of dosing; repeated every 28 days; treated for up to 1 year if tolerating treatment and no PD.</td><td>Study is completed and CSR is pending; thus, data are preliminary.
<table>
<tr><th>Dose (mg/day)</th><th>No. Pts</th><th>Observed DLTs</th></tr>
<tr><td>100</td><td>3</td><td>None</td></tr>
<tr><td>200</td><td>3</td><td>None</td></tr>
<tr><td>400</td><td>4</td><td>None</td></tr>
<tr><td>500</td><td>6</td><td>None</td></tr>
<tr><td>600</td><td>4</td><td>Diarrhea, nausea, fatigue, dehydration</td></tr>
</table>
Efficacy: 1 PR, 3 clinical improvement, 16 SD</td></tr>
<tr><td>Phase 2; 34</td><td>PO administration @ 400 mg; 28 days of dosing; repeated every 28 days for up 1 year if tolerating treatment and no PD.</td><td>Study is completed and CSR is pending; thus, data are preliminary.

Efficacy: 24% of patients achieved maximum reduction in spleen volume ≥ 35% by Week 24</td></tr>
<tr><td>SB1518-2010-004:
A Randomized, Single-Dose, Three-Treatment, Six-Sequence, Three-Period Crossover Study of the Pharmaco-</td><td>24</td><td>Single dose @ 100, 200, or 400 mg.</td><td>Study and CSR are completed.

Half-life was approximately 34 hr and independent of dose

Mean plasma concentrations increased in a dose-related manner, but increases in exposure were less than dose proportional.</td></tr>
</table>

Table--8 Completed Clinical Studies with Pacritinib (SB1518)			
Study No.: Study Title	**Phase; Total Enrolled Patients**	**Dose Regimen**	**Study Results[a]**
kinetics and Sources of Variability of SB1518 after Oral Administration to Healthy Volunteers Under Fasted Conditions.			
SB1518-2010-005: A Phase 2 Safety and Efficacy Study of SB1518 for the Treatment of Advanced Lymphoid Malignancies	28	PO administration @ 400 mg: 28 days of dosing, repeated every 28 days, for at least 2 cycles; may continue past Cycle 12 if tolerating treatment and no PD.	Study is completed and CSR is pending; thus, data are preliminary. ***Efficacy***: 1 CR, 1 PR, 17 SD
SB1518-2010-006: A Randomized, Single-Dose, Two-Treatment, Two-Sequence, Two-Period Crossover Study of the Effect of Food on the Bioavailability and Pharmacokinetics of SB1518 in Healthy Volunteers	18	Single dose @ 200 mg.	Study and CSR are completed. Food had no significant effect on pacritinib PK. Capsule could be taken without regard to timing of meals.
[a] See Section 1.2 for safety results from these studies. **Abbreviations:** CR = complete response CSR = clinical study report DLT = dose-limiting toxicity GI = gastrointestinal mg = milligram(s) No. = number PD = progressive disease PK = pharmacokinetic(s) PR = partial response Pts = patients PO = per os, oral(ly) PR = partial response			

Table--8 Completed Clinical Studies with Pacritinib (SB1518)			
Study No.: Study Title	**Phase; Total Enrolled Patients**	**Dose Regimen**	**Study Results[a]**
SD = stable disease.			

Study Title: Phase 1/2 Study of SB1518 for the Treatment of Advanced Myeloid Malignancies

The oral PK of pacritinib was assessed in the study when dosed as a single agent. During the study's phase 1, in Cycle 1, the starting dose was 100 mg daily for 25 consecutive days of a 28-day cycle, followed by 3 days of rest for PK sampling. From Cycle 2 onward, patients were to be treated daily for 28 consecutive days of each 28-day cycle. Patients in the study's phase 2 were to be treated daily for 28 consecutive days of a 28-day cycle.

This study has been completed. A CSR is pending for Study SB1518-2007-001; thus, data are preliminary.

In the study's phase 1, the primary objective of the study was to establish the maximum tolerated dose (MTD) of pacritinib as a single agent when administered orally daily in subjects with advanced myeloid malignancies. Secondary phase 1 objectives were to assess the safety and tolerability of pacritinib, administered orally once daily in subjects with advanced myeloid malignancies, assess the pharmacokinetic (PK) profile of pacritinib, and evaluate pharmacodynamic (PD) activity of pacritinib. In the study's phase 2, the objectives were to assess the spleen response rate in subjects with CIMF who are treated with pacritinib at the RD; assess the duration of spleen response at the RD; and assess the safety and tolerability of pacritinib, administered orally once daily at the RD in subjects with CIMF.

At the end of phase 1, there were 8 cohorts: Cohort 1 (100 mg); Cohort 2 (150 mg); Cohorts 3 (200 mg pacritinib HCl) and 6 (200 mg pacritinib citrate); Cohort 4 (300 mg); Cohort 5 (400 mg); Cohort 7 (600 mg); and Cohort 8 (500 mg). (The two salt forms of pacritinib, HCl and citrate, provided comparable systemic exposures, as discussed in section and are referred to interchangeably as *pacritinib*. Only the citrate form was and is being tested in pacritinib phase 2 and 3 studies.) Plasma samples for full PK assessments were taken just prior to dosing (Time 0) and at 0.5, 1, 2, 3, 4, 5, 6, 8, and 24 ± 2 hours after dosing on Days 1 (Dose 1) and 15 (± 3) of Cycle 1; and at 24, 48, and 72 hours after the end of the first 25 days of consecutive dosing for Cycle 1.

Pacritinib was quantified in human plasma with a validated LC-MS/MS method. Noncompartmental PK parameters C_{max}, T_{max}, and AUC_{0-24} were estimated using WinNonlin (Version 5.2, Pharsight). The preliminary results of PK analysis on Days 1 and 15 are summarized in Table--9. On Day 1, the mean C_{max} ranged from 2.8 to 8.3 µg/mL, the mean T_{max} from 3.5 to 12 h, the mean AUC_{0-24} from 51 to 141 µg-h/mL and the mean $t_{1/2}$ from 22 to 79 h. On Day 15, the mean C_{max} ranged from 6.2 to 9.3 µg/mL, the mean T_{max} ranged from 3 to 5 h, the mean AUC_{0-24} ranged from 119 to 193 µg·h/mL and the mean $t_{1/2}$ ranged from 38 to 108 h.

Table--9 Study SB1518-2007-001: Summary of Pharmacokinetic Parameters (Phase 1)

Parameter[a]	Cohort 1 100 mg (n = 3)	Cohort 2 150 mg (n = 5)	Cohorts 3,6 200 mg (n = 9)[b]	Cohort 4 300 mg (n = 6)	Cohort 5 400 mg (n = 3)	Cohort 8 500 mg (n = 7)	Cohort 7 600 mg (n = 6)
Day 1							
C_{max}(µg/mL)	3.7 ± 1.7	2.8 ± 1.2	5.0 ± 1.4	5.9 ± 1.5	3.6 ± 1.4	7.1 ± 3.9	8.3 ± 2.1
T_{max} (h)	4.3 ± 1.2	3.5 ± 0.6	4.6 ± 1.8	8.6 ± 8.8	12.3 ± 10	4.4 ± 1.6	4.2 ± 1.2
AUC_{0-24} (µg-h/mL)	66 ± 27	51 ± 19	93 ± 25	105 ± 28	72 ± 33	141 ± 86	134 ± 65
$t_{1/2}$ (h)	76 ± 67	74 ± 47	79 ± 82	43 ± 25	22[c]	49 ± 17	28 ± 13
Day 15							
C_{max}(µg/mL)	6.4 ± 1.6	6.8 ± 3.6	6.7 ± 1.9	6.4 ± 2.1	6.2 ± 4.3	8.7 ± 3.8	9.3 ± 2.3
T_{max} (h)	3.3 ± 0.6	3 ± 2	4.9 ± 2.5	3.8 ± 1.2	4 ± 1.7	3.9 ± 1.1	5 ± 2
AUC_{0-24} (µg-h/mL)	119 ± 31	142 ± 84	125 ± 44	132 ± 50	125 ± 89	178 ± 88	193 ± 56
$t_{1/2}$ (h)	50 ± 41	80 ± 59	73 ± 45	108 ± 154	49 ± 23	38 ± 10	38 ± 21

Source: Preliminary data; CSR in progress.

[a] Data are expressed as mean ± SD.

[b] Cohorts 3 (HCl) and 6 (citrate) data were pooled, because they were at the same dose; the two salt forms have been shown to be pharmaceutically equivalent (section **Error! Reference ource not found.**).

[c] Data from a single patient.

Abbreviations:

µg-h/mL = microgram-hour per milliliter µg/mL = micrograms per milliliter

AUC_{0-24} = area under the concentration vs time curve for time = 0 to 24h

C_{max} = maximum concentration h = hours mg = milligram(s)

n = number of patients in each cohort SD = standard deviation

$t_{1/2}$ = half-life

T_{max} = time to reach maximum concentration.

In the phase 2 portion of the study, 31 patients were treated with pacritinib at 400 mg once daily for 28 consecutive days per cycle. The results from a preliminary PK analysis are summarized in Table--10. The mean T_{max}, C_{max}, AUC, and $t_{1/2}$ were 5 h, 7.1 µg/mL, 134 µg-h/mL, 41 h and 4.6 h, 7.8 µg/mL, 163 µg-h/mL and 65 h on Days 1 and 15, respectively.

Table--10
Study SB1518-2007-001: Summary of Pharmacokinetic Parameters (Phase 2)

Parameter	Day 1[a]	Day 15[a]
T_{max} (h)	5 ± 1.5	4.6 ± 1.6
C_{max} (µg/mL)	7.1 ± 2.8	7.8 ± 2.9

AUC_{0-24} (µg-h/mL)	134 ± 59	163 ± 67
$t_{1/2}$ (h)	41 ± 19	65 ± 45

Source: Preliminary data; CSR in progress.
[a]Data are expressed as mean ± SD.
Abbreviations:
µg-h/mL = microgram-hours per milliliter µg/mL = micrograms per milliliter
AUC_{0-24} = area under the concentration vs time curve at time = 0 to 24 h
C_{max} = maximum concentration h = hour(s) SD = standard deviation
$t_{1/2}$ = half-life T_{max} = time to maximum concentration.

Study Title: Phase 1 Study of SB1518 for the Treatment of Advanced Lymphoid Malignancies

The oral PK of pacritinib was assessed in this study when dosed as a single agent in a 28-day cycle. Pacritinib capsules were administered orally once daily in the morning before breakfast for 28 consecutive days, in 5 dose cohorts: Cohort 1 (100 mg), Cohort 2 (200 mg), Cohort 3 (300 mg), Cohort 4 (400 mg), and Cohort 5 (600 mg). Plasma samples for full PK assessments were taken just prior to dosing (Time 0) and at 0.5, 1, 2, 3, 4, 5, 6, 8, and 24 ± 2 hours after dosing on Days 1 and 15 (± 3) of Cycle 1. Pacritinib was quantified in human plasma with a validated LC-MS/MS method. Noncompartmental PK parameters C_{max}, T_{max}, and AUC_{0-24h} were estimated using WinNonlin (Version 5.2, Pharsight).

This study has been completed. A CSR is pending for Study SB1518-2007-002; thus, data are preliminary.

The preliminary results of PK analysis on Days 1 and 15 are summarized in Table--11. On Day 1 the mean C_{max} ranged from 4.4 to 11.1 µg/mL, the mean T_{max} from 5 to 9 h, the mean AUC_{0-24} from 81 to 157 µg-h/mL, and the mean $t_{1/2}$ from 43 to 63 h. On Day 15, the mean C_{max} ranged from 6.9 to 12.5 µg/mL, the mean T_{max} from 2 to 12 h, the mean AUC_{0-24} from 133 to 71 µg·h/mL, and the mean $t_{1/2}$ from 29 to 108 h.

Table--11
Study SB1518-2007-002: Summary of Pharmacokinetic Parameters

Parameter[a]	Pacritinib Dose (mg)				
	100 mg (n = 3)[b]	**200 mg** (n = 6)	**300 mg** (n = 6)	**400 mg** (n = 7)	**600 mg** (n = 5)
Day 1					
C_{max} (µg/mL)	4.4	5.1 ± 1.8	5.4 ± 2	11.1 ± 4.5	10.0 ± 5.4
T_{max} (h)	5	9 ± 8	9 ± 7	5 ± 1	5.5 ± 1.5
AUC_{0-24h} (µg-h/mL)	81	101 ± 36	112 ± 50	147 ± 55	157 ± 130
$t_{1/2}$ (h)	51	49 ± 9	43 ± 27	61 ± 29	63[a]
Day 15					
C_{max} (µg/mL)	6.9	7 ± 2.5	7.9 ± 4.9	9.7 ± 5.8	12.5 ± 7.4

Table--11 Study SB1518-2007-002: Summary of Pharmacokinetic Parameters					
Parameter[a]	Pacritinib Dose (mg)				
	100 mg (n = 3)[b]	200 mg (n = 6)	300 mg (n = 6)	400 mg (n = 7)	600 mg (n = 5)
T_{max} (h)	2	10 ±7	12 ± 11	7 ± 2	4 ± 2
AUC_{0-24h} (µg-h/mL)	133	153 ± 53	177 ± 117	220 ± 136	271 ± 157
$t_{1/2}$ (h)	102	59 ± 14	29	108 ± 49	98[c]

Source: Preliminary data; CSR in progress.
[a]Data are expressed as mean ± SD.
[b]Mean values reported as PK parameters were estimated from 2 patients.
[c]Estimated in a single patient.

Abbreviations:

µg-h/mL = microgram-hour per milliliter µg/mL = micrograms per milliliter
AUC_{0-24h} = area under the concentration vs time curve at time = 0 to 24 hours
C_{max} = maximum concentration h = hour(s) n = number of patients per cohort
$t_{1/2}$ = half-life T_{max} = time to maximum concentration.

Study SB1518-2007-003
Study Title: Phase 1/2 Study of SB1518 in Patients with Chronic Idiopathic Myelofibrosis

The oral PK of pacritinib was assessed in the study when dosed as a single agent, once daily, in a 28-day cycle. There were 5 cohorts: Cohort 1 (100 mg), Cohort 2 (200 mg), Cohort 3 (400 mg), Cohort 4 (600 mg), and Cohort 5 (500 mg). Pacritinib capsules were administered PO once daily in the morning before breakfast for 28 consecutive days. Plasma samples for full PK assessments were taken just prior to dosing (Time 0) and at 0.5, 1, 2, 3, 4, 5, 6, 8, and 24 ± 2 hours after dosing on Days 1 (dose 1) and 15 (± 3) of Cycle 1, prior to dosing on Day 1 of Cycle 2 and Cycle 3 from patients participating in Phase 1. Pacritinib was quantified in human plasma with a validated LC-MS/MS method. Noncompartmental PK parameters C_{max}, T_{max} and AUC_{0-24h} were estimated using WinNonlin (Version 5.2, Pharsight).

This study has been completed. A CSR is pending for Study SB1518-2008-003; thus, data are preliminary.

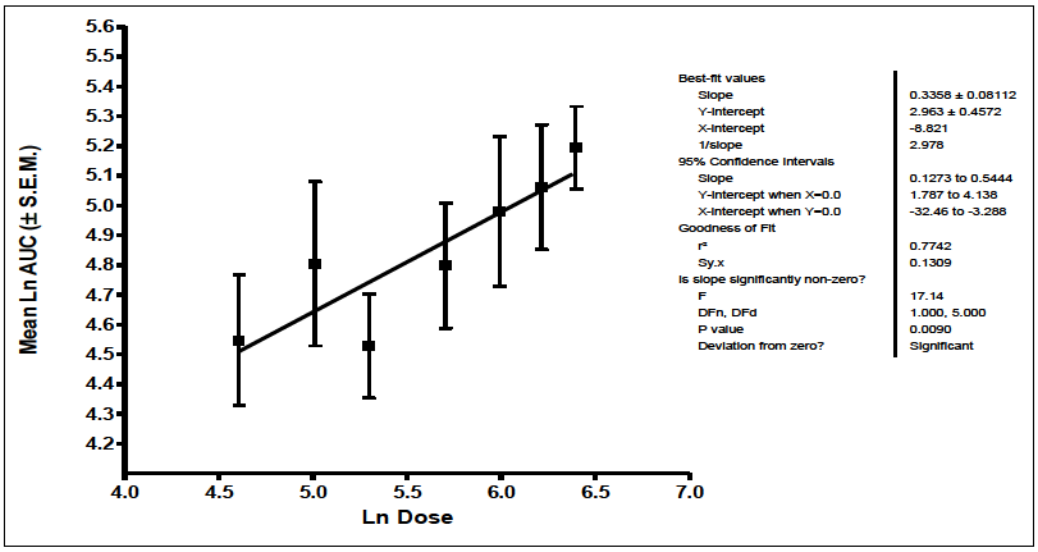
Preliminary results of PK analysis on Days 1 and 15 are summarized in Table--12. On Day 1 the mean C_{max} ranged from 2 to 9 µg/mL, the mean T_{max} from 4 and 6 h, the mean AUC_{0-24} from 41 and 149 µg-h/mL, and the mean $t_{1/2}$ from 24 to 55 h. On Day 15, the mean C_{max} ranged from 3 to 12 µg/mL, the mean T_{max} from 3 to 6 h, the mean AUC_{0-24} from 54 to 211µg-h/mL, and the mean $t_{1/2}$ from 26 to 48 h. A combined analysis of data from the three studies showed that pacritinib displayed dose-related but less than dose-proportional increase in exposure.

Table--12 Study SB1518-2008-003: Summary of Pharmacokinetic Parameters

Parameter[a]	Pacritinib Dose				
	100 mg (n = 3)	**200 mg** (n = 3)	**400 mg** (n = 4)	**500 mg** (n = 6)	**600 mg (n = 4)**
Day 1					
C_{max}(µg/mL)	3 ± 1	2 ± 1.6	9 ± 3	8 ± 4	6 ± 3
T_{max} (h)	5 ± 1.2	5 ± 0	4 ± 1.5	5 ± 1	6 ± 2
AUC_{0-24} (µg-h/mL)	44 ± 15	41 ± 32	149 ± 55	149 ± 72	122 ± 41
$t_{1/2}$ (h)	28 ± 6	27 ± 10	24 ± 7	25 ± 14	55[b]
Day 15					
C_{max}(µg/ml)	4[b]	3 ± 1	12 ± 2.5	8 ± 4	9 ± 4
T_{max} (h)	3[b]	6 ± 2	4 ± 1	6 ± 1	5 ± 3
AUC_{0-24} (µg-h/mL)	77[b]	54 ± 33	211 ± 64	170 ± 95	186 ± 82
$t_{1/2}$ (h)	36[b]	26[b]	33 ± 15	48[b]	45[c]

Source: Preliminary data; CSR in progress.
[a]Data are expressed as mean ± SD.
[b]Mean values from 2 patients.
[c]Estimate from a single patient.
Abbreviations:
µg-h/mL = microgram-hour per milliliter µg/mL = micrograms per milliliter C_{max} = maximum concentration
h = hour(s) SD = standard deviation $t_{1/2}$ = half-life
T_{max} = time to maximum concentration.

Figure--11
Study SB1518-2008-003: Pacritinib Dose-AUC Relationship for Day 1 Assessments[a]



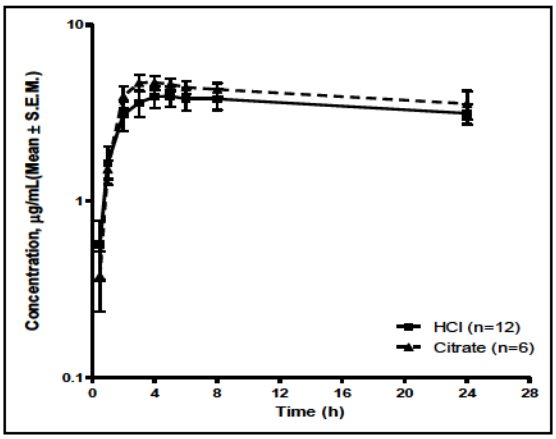
Source: Preliminary data; CSR in progress.

A Comparative Analysis of Exposures from the Pacritinib HCl and Citrate Salts

A comparative analysis of exposures from the HCl and citrate salts was performed comparing Day 1 PK data from the 200 mg cohorts (n = 12 patients dosed with the HCl salt) from studies SB1518-2007-001, SB1518-2007-002, and SB1518-2008-003 with Cohort 6 (n = 6 patients dosed with the citrate salt) from SB1518-2007-001. The mean PK profiles of pacritinib when dosed as citrate and HCl salts were comparable (Figure--12). The mean AUC_{0-24} of the citrate salt was not statistically significantly different from the mean AUC_{0-24} of HCl ($p > 0.05$, Student's t test [unpaired, 2- tailed]), indicating that the two salt forms of pacritinib are pharmaceutically equivalent.

These studies have been completed. Clinical study reports are pending for Studies SB1518-2007-001, -2007-002, and -2008-003; thus, data are preliminary.

Figure--12
Mean Concentration-Time Profiles of Pacritinib Dosed as HCl and Citrate Salts at 200 mg



Source: Preliminary data; CSR in progress. The HCl curve was obtained by pooling the concentrations from the 200 mg cohorts of SB1518-2007-001, SB1518-2007-002, and SB1518-2008-003. The citrate curve is the mean concentrations from 6 patients from cohort 6 (SB1518-2007-001).

1.1 Pharmacodynamics in Humans

Validation of the assays to be used for assessing the pharmacodynamic biomarkers of JAK2 inhibition in clinical trials was reported in ASH 2009[(1)].

Western blot analysis with densitometry was used successfully to demonstrate the following:

- Pacritinib induced blockade of STAT signaling in vitro in cell lines with activated JAK2 or FLT3 pathway signaling;
- Inhibition of pSTAT5 in tumor tissue from a murine xenograft model of FLT3 mutant AML after treatment with a single dose of pacritinib (100 mg/kg);
- Inhibition of pJAK2 and pSTAT5 after *ex vivo* treatment with pacritinib of PBMCs from healthy human volunteers.

Flow cytometric analysis was used successfully to demonstrate the following:

- Dose-dependent inhibition of pSTAT5 in whole blood samples from pacritinib–treated normal mice
- Inhibition of pSTAT5 after ex vivo treatment with pacritinib of whole blood from healthy volunteers

In preliminary analyses on patient blood samples from the pacritinib trials, both the Western blot and flow cytometric analyses demonstrated significant inhibition of the JAK2-STAT5 pathway within 2 h (Day 1) at the lowest dose of 100 mg per day.

These studies have been completed. CSRs are pending for Studies SB1518-2007-001, -2007-002, -2008-003, and -2010-005; thus, data are preliminary.

1.2 Safety

The safety data for pacritinib include all data from completed studies as of the data cutoff date

for this IB version (see cover page). The AEs were tabulated by system organ class (SOC) and preferred term using the *Medical Dictionary for Regulatory Activities Terminology* (MedDRA). The National Cancer Institute Common Toxicity Criteria for Adverse Events (NCI-CTC) version 3.0 was used to classify the severity of AEs.

1.2.1 Study SB1518-2007-001

Study Title: A Phase 1/2 Study of SB1518 for the Treatment of Advanced Myeloid Malignancies

A phase 1/2 study of pacritinib for the treatment of advanced myeloid malignancies was initiated (first patient enrolled) in 2008. The first part of the study was a phase 1 dose-escalation study to determine the MTD and DLT of pacritinib when given as a single agent orally once daily in patients with advanced myeloid malignancies. Cohorts of 3 to 4 patients were enrolled at each dose level with expansion to 6 patients if necessary to assess toxicity. Patients at each dose level were treated and observed through the end of the first cycle before treatment of patients at the next-higher dose level of pacritinib was initiated. If no DLT was observed, or 1 out of 6 patients experienced a DLT, then the dose was escalated to the next dose level. The increment in dose was determined by the investigators and sponsor.

This study has been completed. A CSR is pending for Study SB1518-2007-001; thus, data are preliminary.

Following the identification of 400 mg as the MTD, the phase 2 portion of the study evaluated the efficacy (based on spleen response rate) and safety profile of single agent pacritinib at 400 mg in patients with MF (including PMF, PPV-MF, or PET-MF). The spleen response rate was defined as the proportion of patients achieving a ≥ 35% reduction in spleen volume from baseline to Week 24, as measured by MRI. The recommended dose for phase 2 was 400 mg daily and was chosen based on exposure, safety, pharmacodynamic, and clinical benefit data from phase 1.

A total of 74 patients were treated in the study (43 in phase 1 and 31 in phase 2). Of the 43 patients enrolled in the phase 1 portion of the study, 27 were male and 16 female. Most patients were Caucasian (86%), with the remaining 14% being Asian, African American, or classified as other. The median age was 70 years (range 48 to 85 years). For phase 2, 31 patients (22 males and 9 females) were enrolled, with a median age of 67 (range 47 to 83) years. Thirty patients were Caucasian (97%) and 1 was African American (3%). Seven patients with AML and 36 with MF were enrolled in the phase 1 portion of the study. All subjects in the phase 2 portion of the study had MF.

Deaths and Withdrawals from Study Treatment Due To Adverse Events

Fourteen deaths were reported in the phase 1 portion: 4 due to disease progression, 7 due to an AE/SAE, and 3 due to unknown causes. The AE/SAEs leading to death included 1 subdural hematoma, 1 cardiorespiratory arrest, 1 anemia, 1 hemorrhage intracranial, 1 asthenia, 1 septic shock, and 1 AML. Six patients discontinued treatment due to an AE/SAE.

There were 9 deaths reported in the phase 2 portion of the study: 1 due to disease progression, 3 due to an AE/SAE, and 5 due to unknown causes. AE/SAEs leading to death were cachexia, bacterial sepsis, and hemorrhage following an arterial laceration during a procedure. Two patients discontinued treatment due to an AE; both events were diarrhea and neither was deemed serious. None of the deaths was considered related to study drug.

Serious Adverse Events

In phase 1, a total of 33 SAEs were reported for 19 patients. Of these, 5 related SAEs were reported for 5 patients including 2 pleural effusions (1 at 100 mg and 1 at 200 mg), tumor lysis syndrome (400 mg), congestive heart failure (400 mg), and *Clostridium difficile* diarrhea (600 mg). The other SAEs reported during phase 1 were anemia, neutropenia, cardiorespiratory arrest, right ventricular failure, abdominal pain, diarrhea, pyrexia, asthenia, chest pain, pneumonia, abscess, bacterial infection, bronchitis, sepsis, septic shock, subdural hematoma, arthralgia, acute myeloid leukemia, bladder transitional cell carcinoma, cerebrovascular accident, intracranial hemorrhage, syncope, and acute renal failure.

In phase 2, 38 SAEs were reported for 13 patients. Two related SAEs, diarrhea and dehydration, were each reported in 1 patient. The other SAEs reported during phase 2 were anemia, pancytopenia, splenic infarction, splenomegaly, cardiac failure congestive, acute myocardial infarction, atrial fibrillation, complete atrioventricular block, myocardial ischemia, pericarditis, hematemesis, intraabdominal hematoma, obstructive umbilical hernia, chest pain, peripheral edema, abdominal abscess, bacterial sepsis, lobar pneumonia, pneumonia, tracheobronchitis, urinary tract infection, subdural hematoma, cachexia, failure to thrive, back pain, pain in extremity, myelofibrosis, peripheral neuropathy, mental status changes, epistaxis, hemoptysis, pulmonary hypertension, and hemorrhage.

Adverse Events

Phase 1

DLTs included grade 3 QTc prolongation (150 mg), grade 3 diarrhea (300, 500, and 600 mg), nausea and vomiting (600 mg) and grade 2 dizziness, blurred vision, unsteadiness and worsened performance status (600 mg). The MTD was determined to be 500 mg daily.

The most commonly reported (> 10%) TEAEs were diarrhea, nausea, vomiting, fatigue, peripheral edema, pyrexia, constipation, anemia, dyspnea, asthenia, abdominal distension, thrombocytopenia, epistaxis, back pain, abdominal pain, hyperuricemia, dizziness, insomnia, night sweats, cardiac murmur, hyperbilirubinemia, pain in extremity, neutropenia, chills, anorexia, dehydration, cough, petechiae, arthralgia, and skin lesion. The most commonly reported AEs considered related to pacritinib administration were diarrhea (65%, n = 28), nausea (35%, n = 15), vomiting (23%, n = 10), and thrombocytopenia (12%, n = 5). These GI toxicities were dose dependent and were managed by dose interruption/reduction and/or antidiarrheal and antiemetic treatment. Increasing dose did not result in more frequent grade 3 or 4 AEs.

Phase 2

For phase 2, the most commonly reported (> 10%) TEAEs were diarrhea, fatigue, nausea, vomiting, peripheral edema, abdominal pain, insomnia, pruritus, night sweats, pain in extremity, anemia, bone pain, dyspnea, constipation, asthenia, pyrexia, cardiac murmur, weight decreased, hyperuricemia, and rash. The most commonly reported AEs considered related to pacritinib administration were diarrhea (90%, n = 28), nausea (48%, n = 15%), vomiting (29%, n = 9), and fatigue (13%, n = 4).

Summary

A total of 74 patients were treated with pacritinib in this study, and of these, 67 had MF. The recommended phase 2 dose of 400 mg daily was well tolerated. The most common TEAEs were mild to moderate GI toxicities that were managed with dose reduction and/or antinausea or antidiarrheal medications.

This study has been completed. A CSR is pending for Study SB1518-2007-001; thus, data are preliminary.

Study No. SB1518-2007-002

Study Title: A Phase 1 Study of SB1518 for the Treatment of Advanced Lymphoid Malignancies

This phase 1 open-label dose-escalation study assessed the MTD and DLT of pacritinib when given as a single agent orally once daily in patients with advanced lymphoid malignancies. The objective of the study was to determine the MTD and the DLT of pacritinib when given as a single agent orally once daily. Patients were enrolled at doses of 100 to 600 mg. DLTs observed were neutropenia (300 mg), thrombocytopenia (400 mg) and sepsis (600 mg). The MTD was not identified. The recommended phase 2 dose was 400 mg daily.

This study has been completed. A CSR is pending for Study SB1518-2007-002; thus, data are preliminary.

Thirty-five patients were enrolled and 34 patients were treated in this study (24 males and 11 females) with a median age of 49 years (range of 22 to 80 years). Most (91%) patients were Caucasian; the remaining 9% were African American or classified as other. Of the 35 patients enrolled, 15 had HL, 10 had FL, 4 had DLBCL, 5 had MCL, and 1 had SLL.

Deaths and Withdrawal from Study Treatment Due To Adverse Events

Three deaths were reported in this study. One was due to progressive disease, 1 was attributed to an AE/SAE of acute renal failure that was not related to study drug, and 1 was due to an unknown cause. Five patients discontinued treatment due to an AE/SAE, including 1 patient with cerebrovascular ischemia (200 mg), 1 with pneumonia (300 mg), 1 with neutropenia (400 mg), 1 with sepsis (600 mg), and 1 with fatigue (600 mg).

1.2.1.1 **Serious Adverse Events**

Ten patients reported a total of 17 SAEs. Of these, cerebrovascular ischemia (200 mg dose level) was considered possibly related to pacritinib administration, and sepsis (600 mg dose level) and pulmonary embolism (600 mg dose level) were considered probably related to pacritinib administration. The other reported SAEs were abdominal pain, retroperitoneal hemorrhage, pyrexia, hyperbilirubinemia, pneumonia, bacteremia, pyelonephritis, skin infection, musculoskeletal pain, acute renal failure, and pleural effusion.

Adverse Events

The most commonly reported (> 10%) TEAEs were diarrhea, constipation, nausea, pyrexia, fatigue, anemia, thrombocytopenia, cough, abdominal pain, neutropenia, decreased appetite, dyspnea, vomiting, chills, pneumonia, upper respiratory tract infection, peripheral edema,

hypokalemia, and hypomagnesemia. The most commonly reported related treatment-emergent AEs (> 10%) were diarrhea (50%, n = 17), nausea (38%, n = 13), constipation (29%, n = 10), fatigue (24%, n = 8), thrombocytopenia (24%, n = 8), pyrexia (18%, n = 6), anemia (18%, n = 6), neutropenia (15%, n = 5), vomiting (15%, n = 5), abdominal pain (12%, n = 4), and decreased appetite (12%, n = 4). The majority of AEs were mild to moderate in severity. Increasing dose did not result in more frequent grade 3 or 4 AEs.

Summary

The safety profile in this study population showed that pacritinib was well tolerated, with the majority of AEs being mild to moderate in severity. Dose-dependent AEs included nausea and diarrhea that were readily managed with the use of antinausea or antidiarrheal medications. The MTD was not identified; however, based on the exposure and safety data from all 3 phase 1 patient studies, 400 mg was deemed the recommended dose for the phase 2 trial in this indication.

This study has been completed. A CSR is pending for Study SB1518-2007-002; thus, data are preliminary.

Study SB1518-2008-003

Study Title: A Phase 1/2 Study of oral SB1518 in Subjects with Chronic Idiopathic Myelofibrosis

This phase 1/2 study of oral pacritinib in subjects with MF was initiated in 2008. The first part of the study was a phase 1 dose-escalation portion to determine the MTD and DLT of pacritinib when given as a single agent orally once daily in patients with MF. The phase 2 portion of the study evaluated the efficacy (based on spleen response rate) and safety profile of single agent pacritinib at the recommended dose in patients with MF. The spleen response rate was defined as the proportion of patients achieving a ≥ 35% reduction in spleen volume from baseline to Week 24, as measured by MRI.

This study has been completed. A CSR is pending for Study SB1518-2008-003; thus, data are preliminary.

In the phase 1 portion, a total of 20 patients were enrolled (12 males, 8 females) with a median age of 58.5 years (range of 47 to 74 years). Most (80%) patients were Caucasian, and the remaining were Asian (15%) or classified as other (5%). The recommended phase 2 dose was 400 mg daily. For the phase 2 portion, 34 patients with MF were enrolled (25 males and 9 females). Most (97%, n = 33) patients were Caucasian; one (3%) patient was Asian. The median age was 69 years (range of 44 to 84 years).

1.2.1.2 Deaths and Withdrawal from Study Treatment Due To Adverse Events

Phase 1
Three deaths were reported in the phase 1 portion. All were due to an AE/SAE, but no deaths were related to study drug (AML, pneumonia and acute renal failure). Seven patients discontinued treatment due to an AE/SAE.

Phase 2- In phase 2, 5 deaths were reported, 2 due to disease progression and 3 due to an AE/SAE. One death was considered possibly related to pacritinib (subdural hematoma); the

other 2 were considered not related (septic shock, sepsis). Nine patients discontinued treatment due to an AE/SAE.

Serious Adverse Events

Phase 1- 22 SAEs were reported in 8 patients. Of these, 3 related SAEs were reported for 2 patients, including anemia (100 mg), dehydration (600 mg), and soft tissue infection (600 mg). The other SAEs reported during phase 1 were splenic infarction, abdominal pain, gastrointestinal hemorrhage, esophageal varices hemorrhage, anal abscess, cellulitis, orchitis, pneumonia, electrolyte imbalance, fluid imbalance, acute myeloid leukemia, acute renal failure, urinary retention, skin ulcer, hematoma, and peripheral vascular disorder.

Phase 2- 33 SAEs were reported for 14 patients. Of these, 11 SAEs in 7 patients were considered probably related to pacritinib, including one each of the following: hyperbilirubinemia, dehydration, thrombocytopenia, subdural hematoma, anticoagulation drug level above therapeutic, myocardial infarction, hyponatremia, hyperuricemia, septic shock, hypersensitivity, and febrile neutropenia. The other SAEs reported during phase 2 were anemia, atrial fibrillation, atrial flutter, cardiac failure congestive, gastrointestinal hemorrhage, abdominal pain, pyrexia, cellulitis, pneumonia, sepsis, spinal compression fracture, tibia fracture, traumatic brain injury, renal cell carcinoma, pulmonary embolism, pulmonary hypertension, pulmonary infarction, and deep vein thrombosis.

Adverse Events

Phase 1- The observed DLTs included intermittent diarrhea, nausea, fatigue, and dehydration (all occurred at 600 mg). The MTD was determined to be 500 mg daily. The recommended phase 2 dose was 400 mg.

Of the 20 patients in phase 1, the most commonly reported (>10%) TEAEs were diarrhea, nausea, vomiting, fatigue, constipation, abdominal pain, dizziness, ALT increased, anorexia, cough, anemia, abdominal distension, bone pain, pain in extremity, headache, rash, peripheral edema, arthralgia, back pain, lymphadenopathy, thrombocytopenia, cellulitis, urinary tract infection, gastroenteritis, dysgeusia, epistaxis, and pruritus. The most commonly reported (> 10%) related treatment-emergent AEs were diarrhea (90%, n = 18), nausea (45%, n = 9), vomiting (35%, n = 7), fatigue (25%, n = 5), ALT increased (25%, n = 5), abdominal pain (20%, n = 4) rash (20%, n = 4), peripheral edema (15%, n = 3), and dysgeusia (15%, n = 3). Increasing dose did not result in more frequent grade 3 or 4 AEs. The low frequency of severe cytopenias and lack of an apparent dose relationship in this patient population suggests that pacritinib has limited potential, if any, for myelosuppression.

Phase 2- The most common (> 10%) TEAEs observed in the phase 2 portion of this study were diarrhea, nausea, anemia, fatigue, vomiting, abdominal pain, thrombocytopenia, pruritus, flatulence, dehydration, dizziness, aspartate aminotransferase increased, hyperuricemia, anorexia, constipation, asthenia, pyrexia, hypomagnesemia, musculoskeletal pain, dysgeusia, headache, dyspnea, insomnia, and alopecia. The most commonly reported (> 10%) related treatment-emergent AEs were diarrhea (75%, n = 25), nausea (41%, n = 14), vomiting (24%, n = 8), thrombocytopenia (21%, n = 7), fatigue (21%, n = 7), anemia (21%, n = 7), flatulence (18%, n = 6), aspartate aminotransferase increased (15%, n = 5), abdominal pain (12%, n = 4), alopecia (12%, n = 4), and anorexia (12%, n = 4).

Summary
In this study, 54 patients with MF were treated with pacritinib: 20 patients in the phase 1 portion and 34 patients in the phase 2 portion. The recommended phase 2 dose of 400 mg daily was well tolerated. The most common TEAEs were mild-to-moderate GI toxicities that were managed with dose reduction and/or with antinausea or antidiarrheal medications. To date, hematologic AEs have been uncommon.

This study has been completed. A CSR is pending for Study SB1518-2008-003; thus, data are preliminary.

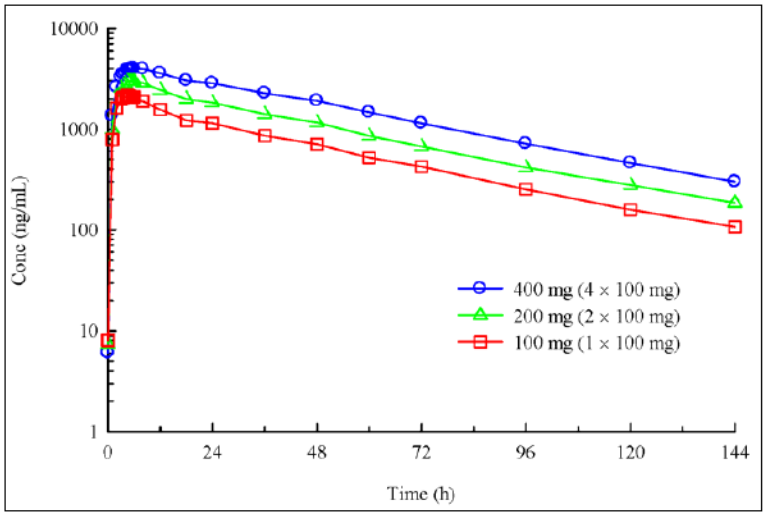
Study SB1518-2010-004
Study Title: A Randomized, Single Dose, Three-Treatment, Six-Sequence, Three-Period Crossover Study of the Pharmacokinetics and Sources of Variability of SB1518 after Oral Administration to Healthy Volunteers under Fasted Conditions

The primary objectives of this three-treatment, six-sequence, three-period, open-label, crossover PK study were to assess the inter- and intra-individual variability of pacritinib PK and assess the linearity and dose proportionality of pacritinib after administration of single oral doses in healthy volunteers. Twenty-four subjects were randomly assigned into 6 sequence groups, with 4 subjects per group. Each subject received all treatments in the study in a crossover manner. Following an overnight fast (≥ 10 hours), subjects received either a single 100 mg, 200 mg, or 400 mg oral dose of pacritinib, according to the randomization schedule. Subjects fasted for 4 hours after each study drug administration, with water consumption restricted from 1 hour prior to dosing until 1 hour post-dose (with the exception of 240 mL water taken with the study drug). A washout period of 14 days was given between treatments. Blood samples for measuring pacritinib plasma concentrations were collected immediately prior to dosing and at 1, 2, 3, 3.5, 4, 4.5, 5, 5.5, 6, 8, 12, 18, 24, 36, 48, 60, 72, 96, 120, and 144 hours post-dose during each treatment period. Pacritinib plasma concentrations were quantified using a validated LC-MS/MS method. All PK parameters were calculated using noncompartmental analysis.

This study is complete, and a CSR is available.

The mean pacritinib plasma concentrations increased in a dose-related, but less than dose-proportional, manner after administration of single 100, 200, and 400 mg doses (Figure--13).

Figure--13
Study SB1518-2010-004: Mean Plasma Concentrations of Pacritinib After Single 100, 200, and 400 mg Oral Doses to Healthy Subjects Under Fasted Conditions (Pharmacokinetic Analysis Population)



Source: CSR, Study SB1518-2010-004, Figure 11-1, lower panel.

The median T_{max} ranged from 4.5 to 5.5 h, and there was no apparent change with dose. The mean $t_{½}$ was 33.5 to 34.5 h and was not dose dependent. The increases in systemic exposures were linear and appeared to be less than dose proportional (the exponent of the power model was < 1, and the 95% CIs did not include 1) over the tested dose range. The between-subject CV for natural log-transformed C_{max}, AUC_{0-t}, and AUC_{0-inf} ranged from 28.28% to 45.01%, indicating relatively high variability among subjects; however, the within-subject CVs for all 3 parameters were relatively low, ranging from 13.38% to 15.33%. **Error! Reference source not ound.** presents a summary of the mean PK parameters.

Table--13
Study SB1518-2010-004: Summary of Pacritinib Pharmacokinetic Parameters (Pharmacokinetic Analysis Population)

PK Parameter	**100 mg** (1 × 100 mg) N = 24	**200 mg** (2 × 100 mg) N = 24	**400 mg** (4 × 100 mg) N = 24
C_{max} (ng/mL)	2224 (616)	3106 (862)	4153 (1116)
T_{max} (h)[a]	4.50 (2.0, 6.0)	5.00 (3.0, 8.0)	5.50 (3.0, 12.0)
AUC_{0-t} (ng•h/mL)	86759 (29898)	137351 (62218)	214712 (86642)
AUC_{0-inf} (ng•h/mL)	92722 (34409)	147888 (72914)	232108 (100988)
λ_z (L/h)	0.0217 (0.0052)	0.0218 (0.0048)	0.0214 (0.0056)
$t_{1/2}$ (h)	33.5 (7.38)	33.5 (7.83)	34.5 (8.74)
CL/F (L/h)	1.25 (0.54)	1.64 (0.69)	2.04 (0.84)
V_d (L)	56.7 (16.8)	73.5 (21.3)	94.4 (28.6)

Source: CSR SB1518-2010-004, Table 11-2.
[a] T_{max} was reported as median (min, max).
Abbreviations:
AUC_{0-inf} = area under the concentration versus time curve at time = 0 to infinity
AUC_{0-t} = area under the concentration versus time curve at time = 0 to time t
CL/F = oral clearance C_{max} = maximum concentration h = hour(s)
L/h = liters per hour
mg = milligram(s) ng•h/mL = nanograms $t_{1/2}$ = half-life T_{max} = time to maximum

Table--13 Study SB1518-2010-004: Summary of Pacritinib Pharmacokinetic Parameters (Pharmacokinetic Analysis Population)			
PK Parameter	**100 mg** (1 × 100 mg) N = 24	**200 mg** (2 × 100 mg) N = 24	**400 mg** (4 × 100 mg) N = 24
concentrationV_d = volume of distribution.			

Patients were also monitored for safety with serial serum chemistries, CBCs, urinalyses, ECGs, vital signs, physical examinations, and AE monitoring. All 24 subjects in the study received 3 doses of pacritinib.

Pacritinib was well tolerated at the lower (100 and 200 mg) doses. The number of subjects experiencing AEs following administration of 400 mg pacritinib (n = 11; 46%) was greater than after administration of the 100 (n = 3; 13%) or 200 mg (n = 2; 8%) doses. The AEs that occurred at the 400 mg dose were primarily GI and nervous system disorders, including diarrhea (n = 7), nausea (n = 3), vomiting (n = 2), and headache (n = 2). In contrast, no AE was experienced by > 1 subject at the 100 or 200 mg dose. The investigator considered all AEs to be mild in intensity, and most (18 of 20) AEs were considered to have a possible causal relationship to study drug.

There were no serious AEs reported during the study, and no subject experienced an AE that resulted in treatment discontinuation or withdrawal from the study. No clinically important changes in laboratory values, vital signs, ECGs, or physical examinations were reported.

In summary, single-dose PO administration of pacritinib at doses of 100 to 400 mg was well tolerated in healthy adult volunteers. While the 400 mg pacritinib dose was associated with mild AEs, the frequency of AEs was greater for the 400 mg pacritinib dose compared with the lower doses.

Study SB1518-2010-005
Study Title: A Phase 2 Safety and Efficacy Study of SB1518 for the Treatment of Advanced Lymphoid Malignancies

The objectives of this open-label phase 2 study were to assess responses, survival, and safety and tolerability of pacritinib in patients with advanced lymphoid malignancies. Twenty-eight patients (19 males and 9 females) were enrolled, with a median age 61.5 years (range 20 to 91 years). Most (89%) patients were Caucasian and the rest were black or African American. The dose of pacritinib was 400 mg daily. Twenty patients received at least two 28-day cycles of pacritinib. Patients in this study received up to 10 cycles.

This study has been completed. A CSR is pending for Study SB1518-2010-005; thus, data are preliminary.

Deaths and Withdrawal from Study Treatment Due to Adverse Events
Five deaths were reported: 1 due to progressive disease and 4 due to AE/SAEs. None of these events (acute respiratory failure, multiorgan failure, cardiac arrest, and asthenia) were considered related to study drug. Five patients discontinued treatment due to an AE/SAE.

Serious Adverse Events
There were 14 SAEs reported in 8 patients. Respiratory failure and stroke were each reported in 2 patients. Two SAEs in 2 patients were considered possibly related to pacritinib: cerebrovascular accident and pneumonia. The other SAEs were febrile neutropenia, pancytopenia, cardiac arrest, pericardial effusion, asthenia, multiorgan failure, sepsis, convulsion, acute respiratory failure, respiratory failure, acute myeloid leukemia, and cerebrospinal fluid reservoir placement.

Adverse Events- The most commonly reported (>10%) TEAEs were diarrhea, nausea, fatigue, thrombocytopenia, constipation, anemia, headache, cough, dizziness, abdominal pain, anorexia, peripheral edema, and pneumonia. The most commonly reported (> 10%) related treatment-emergent AEs were diarrhea (61%, n = 17), nausea (43%, n = 12), vomiting (21%, n = 6), fatigue (18%, n = 5), constipation (14%, n = 4), thrombocytopenia (14%, n = 4), anemia (11%, n = 3), and dizziness (11%, n = 3).

This study has been completed. A CSR is pending for Study SB1518-2010-005; thus, data are preliminary.

Study SB1518-2010-006
Study Title: A Randomized, Single Dose, Two-Treatment, Two-Sequence, Two-Period Crossover Study of the Effect of Food on the Bioavailability and Pharmacokinetics of SB1518 in Healthy Volunteers

The objective of this two-treatment, two-sequence, two-period crossover study was to characterize the effect of a high-calorie, high-fat meal on the bioavailability and PK of pacritinib in healthy volunteers. Eighteen subjects were randomly assigned into 2 sequence groups (9 subjects per group) to receive a single 200–mg dose of SB1518 twice—once under fed conditions and once under fasted conditions. Samples from all subjects completing both periods of the study were analyzed. On Day 1, following completion of the overnight fast, subjects randomized to receive pacritinib in a fasted state received a single 200-mg dose of pacritinib with 240 mL of water. Those subjects randomized to receive pacritinib under fed conditions received a high-fat/high-calorie meal (containing ~ 150 protein calories, 250 carbohydrate calories, and 500 to 600 fat calories) prior to receiving a single 200 mg dose of pacritinib; the meal was to be ingested starting approximately 30 minutes prior to dosing and completed within the 5 minutes preceding the scheduled dosing time. Each dose was separated by a washout period of at least 14 days. Blood samples for the measurement of pacritinib plasma concentrations were collected immediately prior to dosing and at 1, 2, 3, 3.5, 4, 4.5, 5, 5.5, 6, 8, 12, 18, 24, 36, 48, 60, 72, 96, 120, and 144 hours postdose during each treatment period. Concentrations in plasma were quantified using a validated LC-MS/MS method. All PK parameters were calculated using noncompartmental analysis.

This study is complete, and a CSR is available.

The mean pacritinib plasma concentrations after administration of a single 200 mg dose under fed conditions were slightly higher than after administration under fasted conditions. Mean C_{max}, AUC_{0-t}, and AUC_{0-inf} values were higher after administration under fed conditions. The fed-to-fasted geometric mean ratios (GMRs) were 111.18% to 117.36%, and the associated 90% CIs for all three parameters were contained within 80% and 125%, demonstrating the lack of a food

effect with respect to these exposure parameters. Based on these PK analyses, 100-mg pacritinib capsules can be taken without regard to meal content or timing.

Table--14
Study SB1518-2010-006: Statistical Comparison of Pacritinib Pharmacokinetic Data in Healthy Human Volunteers When Administered Under Fed and Fasted Conditions

Parameter (Units)[a]	Geometric Means		Geometric Mean Ratio (90% CI)
	Fed (Test)	Fasted (Reference)	
C_{max} (ng/mL)	3433	3087	111.18 (104.99, 117.73)
AUC_{0-t} (ng-hr/mL)	169475	144408	117.36 (111.16, 123.90)
AUC_{inf} (ng-hr/mL)	181537	155507	116.74 (110.42, 123.42)

Source: CSR SB1518-2010-006, Table 11-3, Table 15.2.6.
[a] Analysis was performed using an analysis of variance statistical model with sequence, period, treatment, and subject within sequence as the classification variables; data were natural log-transformed before analysis.
Abbreviations:
AUC_{0-t} = area under the concentration vs time curve from time = 0 to time = t
AUC_{inf} = area under the concentration vs time curve from time = 0 to time = infinity
CI = confidence interval C_{max} = maximum concentration ng-hr/mL = nanogram-hour per milliliter.

No SAEs were reported during the study, and no subject experienced an AE that resulted in treatment discontinuation or withdrawal from the study. No clinically important changes in laboratory values, vital signs, ECGs or physical examinations were reported. Three subjects had 5 AEs. Two subjects had nausea: 1 subject in the fasted group and 1 in the fed group. All 5 reported AEs were considered by the investigator to have a possible causal relationship with pacritinib, and most (4 of the 5 AEs) were considered mild in intensity; 1 AE of headache was considered moderate in intensity. The safety profile of single-dose pacritinib did not appear to be altered by the consumption of a high-fat, high-calorie meal.

Summary of Safety Data
A total of 191 patients with hematologic malignancies have been exposed to pacritinib and 187 (98%) patients reported AEs.

These studies have been completed. Clinical study reports are pending for Studies 1518-2007-001, -2007-002, -2008-003, and -2010-005; thus, data are preliminary.

Safety data were consistent across all studies; TEAEs are summarized for all studies in and are also shown by indication, MF;**Error! Reference source not found.** lymphoma). Side effects ere predominantly GI and mild to moderate. Thirty-three (33) patients (17%) reported Grade 3/4 GI AEs. Treatment-emergent diarrhea was reported by 139 (73%) patients, with 15 (8%) reporting Grade 3/4 diarrhea.

Clinical AEs associated with myelosuppression (eg, hemorrhage, febrile neutropenia) were rare

and did not appear to be dose dependent. Since patients with advanced myeloid or lymphoid malignancies have compromised hematopoiesis, pacritinib appears to have limited potential for myelosuppression.

Among patients with MF and advanced myeloid malignancies, side effects were predominantly GI. The most common AEs were diarrhea (81%), nausea (50%), fatigue (41%), vomiting (36%), anemia (26%), abdominal pain (23%), peripheral edema (24%), and constipation (20%). Grade 3/4 AEs were reported by 77% of patients. The most frequently occurring Grade 3/4 events in patients with MF were anemia (19%), thrombocytopenia (15%), fatigue (12%), and diarrhea (11%).

QTc Toxicities

CTI performed a post-hoc review of the phase 1-2 S*BIO studies, SB1518-2007-001 and SB1518-2008-003, for potential cardiac toxicity, based upon a single case report of a QTc prolongation reported as a dose-limiting toxicity in the SB1518-2007-001 study. In these studies, ECGs were performed at baseline, periodically during the first 30 days of treatment, at termination, and as clinically indicated.

These studies have been completed. Clinical study reports are pending for Studies SB1518-2007-001 and -2008-003; thus, data are preliminary.

- Based on the identified DLT, CTI performed a full review of the patient profiles for all patients enrolled in these two studies. Review of these data identified the following observations:
- Nine patients across the studies had at least 1 reported QTc exceeding 500 ms, including the patient with the DLT. Of these 9 patients, 1 patient was receiving pacritinib at 150 mg per day, 1 at 450 mg per day, and the remaining 7 were receiving 400 mg per day. Baseline pre-pacritinib QTc intervals ranged from 441 to 491 ms. A majority (5) of the QTc prolongations were identified at the Day 29 time point, with the others identified on Days 1, 10, and 18.
  - At the time of the QTc events, 8 patients were on concomitant medications associated with QTc prolongation, including ondansetron (5 patients), levofloxacin (2 patients), venlafaxine (2 patients), and amiodarone, solifenacin, fluconazole, voriconazole, citalopram, cilostazol, and prochlorperazine in 1 patient each.
  - Pre-study cardiac history was also noted in 7 of the patients, including atrial fibrillation (3 patients), coronary artery disease (3 patients), myocardial infarction (3 patients), congestive heart failure (1 patient), heart murmur (2 patients), and syncope (2 patients).

- In addition to the DLT event, there were two QTc prolongations reported as non-serious AEs.

Pacritinib was discontinued soon after the QTc observations in the patient with the DLT (3 days after the DLT) and in the patients with the reported non-serious AEs (11 and 47 days after the reported AE, respectively). In the remaining patients, treatment continued for extended periods of time [27 days (Patient 004-0909), 474 days (Patient 004-0913), 574 days (Patient 004-0919), 562 days (Patient 004-0920), 560 days (Patient 004-0921), and 277 days (Patient 010-0404)] after the QTc event, which was deemed not clinically significant by the investigator.

One patient (Patient 007-0505) had Grade 3 congestive heart failure requiring hospitalization 35

days after QTc prolongation was first reported that was deemed possibly related to pacritinib. No other post-QTc cardiac events attributable to pacritinib were observed.

As a result, investigators are advised to exclude from the study patients with a baseline QTc > 450 ms.

Pacritinib in Thrombocytopenic Patients

A post hoc analysis of the pooled phase 2 myelofibrosis patients from the SB1518-2007-001 and -2008-003 studies was performed to evaluate the efficacy and safety of pacritinib in patients with baseline platelet counts of ≤ 100,000 and in patients with baseline platelet counts of > 100,000. These two subpopulations were analyzed in parallel for the efficacy measures of spleen volume changes by MRI and changes in patient-reported symptoms. They were also analyzed for safety outcomes, including investigator-reported adverse events, serious adverse events, thrombocytopenia-related adverse events, and laboratory-based toxicity evaluations of platelet counts and hemoglobin levels.

These studies have been completed. Clinical study reports are pending for Studies SB1518-2007-001 and -2008-003; thus, data are preliminary.

Baseline demographics, including age, gender, and frequency of JAK2 positivity were similar between the two groups. The ≤ 100k cohort had more patients with primary MF and fewer with PPV-MF relative to the > 100k cohort (75% versus 51% and 18% versus 30%, respectively). In addition, a higher proportion of patients in the ≤ 100k cohort were in the Dynamic International Prognostic Scoring System (DIPSS) Intermediate-2 risk group (39% versus 24%), while the > 100k cohort had similar proportions of patients in the Intermediate-1, Intermediate-2, and High-risk DIPSS risk categories. All of these patients were initially prescribed oral pacritinib at 400 mg once daily and were observed to have similar (≤ 100k, > 100k cohorts) mean delivered doses (359 mg/day, 349 mg/day), dose intensity (90%, 87%), and duration of therapy (46.6 weeks, 43 weeks). Similar efficacy was achieved in the two cohorts, with 44% and 31% (respectively) achieving ≥ 35% reduction in spleen volume as assessed by MRI during the study and 46% and 50% respectively achieving ≥ 50% reduction in patient-reported symptom scores using the CIMF Quality of Life and Symptom Assessment Tool[(2)].

Safety was also similar in the 2 cohorts. When normalized to baseline values (100%), no significant differences were observed in treatment-emergent changes in platelet counts or in hemoglobin values. No significant trends could be discerned in the analysis of the most frequently occurring AEs of all grades, of Grade 3/4 events, nor of SAEs. Qualitatively, there were more AEs involving bleeding or bruising among the patients with baseline platelets of ≤ 100k, but there were no unexpected safety observations.

The overall incidence of all grades of thrombocytopenia reported as AEs in efficacy and safety trials with pacritinib was 17%. Twelve percent (12%) of patients experienced a 2-grade or greater shift in platelet counts from baseline to worst platelet count, and 5% experienced a 2-grade or greater shift in platelet counts from baseline to end of study (Table--15).

In summary, pacritinib is well tolerated in patients with various hematologic malignancies including patients with significantly impaired bone marrow function associated with cytopenias, including thrombocytopenia.

Table--15 Shift from Baseline Platelet Count by CTC Grade in Phase 2 of Studies SB1518-2007-001 and -2008-003					
CTC Grade	**Baseline Platelet Count CTC Grade (N = 65)**				
	Grade 0	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**
Nadir Platelet Count CTC Grade (N = 64)					
0	16 (25.0%)	0	0	0	0
1	11 (17.2%)	3 (4.7%)	0	0	0
2	0	4 (6.3%)	5 (7.8%)	0	0
3	1 (1.6%)	5 (7.8%)	6 (9.4%)	4 (6.3%)	0
4	1 (1.6%)	0	1 (1.6%)	5 (7.8%)	2 (3.1%)
End-of-Study Platelet Count CTC Grade (N = 64)					
0	20 (31.3%)	4 (6.3%)	0	0	0
1	7 (10.9%)	2 (3.1%)	2 (3.1%)	0	0
2	0	5 (7.8%)	6 (9.4%)	1 (1.6%)	0
3	1 (1.6%)	1 (1.6%)	4 (6.3%)	6 (9.4%)	1 (1.6%)
4	1 (1.6%)	0	0	2 (3.1%)	1 (1.6%)
Source: Derived from clinical database as of 20 August 2012. **Abbreviations:** CTC = Common Toxicity Criteria N = number.					

Reference Safety Information

The expectedness of ADRs from completed studies will be determined based on the indication-specific ADR information from completed studies presented in the Appendix in **Error! eference source not found.**, **Error! Reference source not found.**, and **Error! Reference source not found.**. Table--16 summarizes the content of these tables, including the studies included in each appendix table.

Table--16 Reference Safety Information	
Type of Study	**Table Number**
All studies[a]	**Error! Reference source not found.**
Myelofibrosis studies[b]	**Error! Reference source not found.**
Lymphoma studies[c]	**Error! Reference source not found.**
[a] Includes Studies SB1518-2007-001P1, -2007-001P2, -2007-002, -2008-003P1, -2008-003P2, and -2010-005. [b] Includes Studies SB1518-2007-001P1, -2007-001P2, -2008-003P1, and -2008-003P2. [c] Includes Studies SB1518-2007-002 and -2010-005.	

Myelofibrosis Efficacy:
In the SB1518-2007-001 and SB1518-2008-003 phase 1 studies, response was determined according to the IWG criteria for MF.[11]

These studies have been completed. Clinical study reports are pending for Studies SB1518-2007-001 and -2008-003; thus, data are preliminary.

Among 36 patients with MF in the SB1518-2007-001 phase 1 study, 12 (33%) achieved clinical improvement (CI) and 22 (61%) had stable disease (SD). In 2 patients with spleens between 5 and 10 cm below the left costal margin at baseline, the spleen became non-palpable. Eight patients with spleens >10 cm below the left costal margin at baseline had a greater than 50% reduction in palpable splenomegaly. In 9 patients with baseline leukocytosis (6 of whom were dosed with ≥400 mg), white blood cell (WBC) counts normalized during treatment.

Among 20 patients with MF in the SB1518-2008-003 phase 1 study, 3 (15%) achieved CI, 1 (5%) had PR, and 16 (80%) had SD. In 3 patients with spleens ≥5 cm below the left costal margin at baseline, the spleen became non-palpable. In 6 patients with baseline leukocytosis (all of whom were dosed with ≥400 mg), WBC counts normalized during treatment.

In the phase 2 portions of the SB1518-2007-001 and SB1518-2008-003 studies, both MRI and physical examination were used to measure treatment effect on splenomegaly. Treatment with pacritinib resulted in reduction of splenomegaly by both MRI and physical examination. Results through Week 24 showed that 29 of 49 (59%) evaluable patients had a 25% or greater reduction in spleen volume and 13 (27%) had a 35% or greater reduction in spleen volume by MRI.

Spleen response was even more favorable among patients with platelet counts <100,000/μL. The proportion of patients with ≥25% reduction in spleen volume was 61%, and the proportion with ≥35% reduction in spleen volume was 30%. Thus, pacritinib may provide a therapeutic option for thrombocytopenic patients with MF in Table 25.

Spleen response	All Patients N = 65[a,b]	Patients with Baseline Platelet Count ≤ 100,000/μL N = 28[a,b]
≥ 35% reduction in spleen volume by MRI	13/49 (27%)	7/23 (30%)
≥ 50% reduction in spleen length by PE (IWG-MRT criteria for Clinical Improvement)	26/62 (42%)	12/28 (43%)

Source: Verstovsek et al 2013[(4)].
[a] Sixty-five (65) patients (28 with platelet counts ≤ 100,000/μL) were available for assessment. Only patients with both baseline and at least one post-baseline assessment were included in the evaluable population and used in the calculation of response rate.
[b] Includes Studies SB1518-2007-001 and -2008-003.

In addition to evaluating treatment effect on splenomegaly, a patient-reported outcome tool (Myelofibrosis Symptom Assessment Form, or MF-SAF) was used to measure the treatment effect on MF-related symptoms. Symptom severity by the MF-SAF varied at baseline both between symptoms for individual patients and among patients. Symptom improvements in Studies SB1518-2007-001 and -2008-003 were analyzed. Patients with both baseline and follow-up assessments were included in these analyses. At Week 12, symptom relief was observed for all symptoms except worse fatigue (Table--17).

Table--17 Change from Baseline to Week 12 in MF-SAF Symptom Severity Scores (Combined Phase 2 Studies in Patients with MF: N = 65)				
Symptom	**N[a]**	**Baseline Score Mean (SD)**	**Week 12 Score Mean (SD)**	**Change in Score (Mean)**
Abdominal Pain	46	2.9 (2.8)	2.5 (2.8)	-0.4
Bone Pain	48	2.5 (3.1)	2.3 (3.1)	-0.3
Early Satiety	48	4.7 (2.8)	3.3 (2.5)	-1.5
Fatigue - Worst	48	5.4 (2.8)	5.5 (2.6)	0.0
Inactivity	47	3.9 (2.6)	3.3 (2.6)	-0.5
Night Sweats	48	2.4 (3.0)	2.0 (3.1)	-0.4
Pruritus	47	2.5 (3.0)	1.4 (2.1)	-1.1
Source: Derived from clinical database as of 07 September 2012. [a] Includes Studies SB1518-2007-001 and -2008-003.				

At Week 24, even greater score improvements were seen; scores improved for all symptoms (Table--18).

Table--18 Change from Baseline to Week 24 in MF-SAF Symptom Severity Scores (Combined Phase 2 Studies in Patients with MF: N = 65)				
Symptom[a]	**N**	**Baseline Score Mean (SD)**	**Week 24 Score Mean (SD)**	**Change in Score (Mean)**
Abdominal Pain	41	3.0 (2.7)	1.8 (2.1)	-1.2
Bone Pain	42	2.3 (3.0)	1.6 (2.6)	-0.7
Early Satiety	42	4.6 (2.7)	3.0 (2.0)	-1.6
Fatigue - Worst	42	5.5 (2.7)	4.8 (2.6)	-0.8
Inactivity	42	3.7 (2.3)	2.6 (1.8)	-1.1
Night Sweats	42	2.2 (3.0)	1.1 (1.7)	-1.1
Pruritus	42	2.5 (3.0)	0.8 (1.3)	-1.7
Source: Derived from clinical database as of 07 September 2012. [a] Includes Studies SB1518-2007-001 and -2008-003.				

Similar improvements were observed in patients with baseline platelet counts of ≤ 100,000/μL at Weeks 12 (Table--19) and 24 (Table--20).

Table--19 Change from Baseline to Week 12 in MF-SAF Symptom Severity Scores for Patients With Baseline Platelet Count ≤ 100,000/μL (Combined Phase 2 Studies in Patients With MF: N = 28)

Symptom	N[a]	Baseline Score Mean (SD)	Week 12 Score Mean (SD)	Change in Score (Mean)
Abdominal Pain	22	2.7 (2.9)	2.0 (2.0)	-0.7
Bone Pain	23	2.4 (3.0)	2.1 (2.8)	-0.3
Early Satiety	23	4.6 (2.9)	3.3 (2.5)	-1.3
Fatigue - Worst	23	5.3 (3.0)	5.7 (2.7)	0.4
Inactivity	22	4.0 (2.8)	2.7 (2.1)	-1.3
Night Sweats	23	2.4 (3.1)	1.2 (1.8)	-1.2
Pruritus	22	1.9 (2.8)	0.7 (1.1)	-1.1
Source: Derived from clinical database as of 06 September 2012. [a] Includes Studies SB1518-2007-001 and -2008-003.				

Table--20
Change from Baseline to Week 24 in MF-SAF Symptom Severity Scores for Patients With Baseline Platelet Count ≤ 100,000/μL (Combined Phase 2 Studies in Patients With MF: N = 28)

Symptom	N[a]	Baseline Score Mean (SD)	Week 24 Score Mean (SD)	Change in Score (Mean)
Abdominal Pain	18	3.4 (2.9)	1.9 (2.3)	-1.5
Bone Pain	18	2.7 (3.2)	2.6 (3.3)	-0.1
Early Satiety	18	4.8 (2.9)	3.6 (1.9)	-1.2
Fatigue - Worst	18	5.8 (2.9)	5.0 (2.8)	-0.8
Inactivity	18	4.3 (2.6)	2.4 (1.8)	-1.8
Night Sweats	18	2.5 (3.2)	0.8 (1.2)	-1.7
Pruritus	18	2.1 (3.0)	0.6 (1.0)	-1.4
Source: Derived from clinical database as of 06 September 2012. [a] Includes Studies SB1518-2007-001 and -2008-003.				

Lymphoma

Responses for lymphoma patients in Study SB1518-2007-002 were assessed by the Revised Response Criteria for Malignant Lymphoma[(5)]. Of 31 patients with at least one follow-up tumor assessment, 4 had partial responses (2 patients with MCL at 300 and 400 mg/d, and 2 with FL at 400 and 600 mg/d). Seventeen (17) had stable disease (7 with FL, 7 with HL, 2 with MCL, and 1 with SLL). Thirteen (13) of 17 patients with stable disease had tumor mass reductions of 4% to 45%.

In Study SB1518-2010-005, of 24 patients with at least 1 follow-up tumor assessment, 1 patient with FL had PR and 1 with FL had CR. Seventeen patients had SD (5 patients with HL, 4 with MCL, and 8 with FL). Four (4) of 16 patients with SD had tumor mass reductions > 50% from baseline.

Pacritinib has encouraging activity in patients with relapsed lymphomas; clinical benefit was observed in several lymphoma subtypes at doses ≥ 300 mg/d and was well tolerated at doses up to 600 mg/d. The recommended dose for phase 2 study is 400 mg/d.

These studies have been completed. Clinical study reports are pending for Studies SB1518-2007-002 and -2010-005; thus, data are preliminary.

Marketing Experience
There is no marketing experience with Pacritinib.

Summary
Pacritinib is a novel small molecule inhibitor of JAK2 and FLT3, with anti-tumor activity in murine models of acute myeloid leukemia and JAK2-dependent leukemia and activity in human clinical studies of myelofibrosis and lymphoma.

3.0 PATIENT SELECTION

Inclusion criteria

1. Signed informed consent indicating that patients are aware of the investigational nature of this study, in keeping with the policies of MDACC, must be obtained prior to any study specific procedures.
2. Patients with a histologically confirmed diagnosis of MDS by World Health Organization (WHO) classification and lower-risk MDS as defined by the IPSS classification[2] (Low or Int-1 disease) or R-IPSS classification[12] (Very Low or Low) are eligible. Patients with MDS/MPD overlap syndromes including CMML are also eligible if they have Low or Int-1 disease per IPSS. Patients may have received MDS-directed therapy (i.e. lenalidomide), although patients with prior exposure to hypomethylating agents (e.g. 5-azacitidine or decitabine) are not eligible.
3. The interval from prior treatment to time of study drug administration is at least 1 week (except for hydroxyurea or steroid therapy) with recovery from all prior therapy-related toxicities
4. Age $\geq$ 18 years old.
5. Eastern Cooperative Oncology Group (ECOG) performance status of 0 to 2.
6. Adequate liver function, as evidence by serum bilirubin $\leq$ 2x the laboratory normal range (except for patients with Gilbert's Disease), or an aspartate aminotransferase (AST) or alanine aminotransferase (ALT) of $\leq$ 2.5x the upper limit of normal (ULN) or $\leq$ 5x ULN if hepatic disease involvement is present as determined by the investigator.
7. Serum creatinine (Cr) $\leq$ 2x ULN or 24-hour creatinine clearance $\geq$50 ml/min
8. Subjects of reproductive potential must agree to the use of acceptable contraceptive methods for the duration of the time on study and a further 6 months after completion of treatment. Women of childbearing potential must have a negative blood or urine pregnancy test within 72 hours of start of treatment.

Exclusion criteria

1. Subjects with any prior exposure to the hypomethylating agents (5-azacitadine or decitabine) are excluded.
2. Subjects with any prior exposure to JAK2 inhibitor therapy (i.e. ruxolitinib or prior pacritinib therapy) are excluded.

3. Any prior or coexisting medical condition that in the investigator's judgment will substantially increase the risk associated with the subject's participation in the study.
4. Psychiatric disorders or altered mental status precluding understanding of the informed consent process and/or completion of the necessary study procedures.
5. Active uncontrolled serious infection or sepsis at study enrollment. Patients receiving antibiotics for infections that are under control may be included in the study.
6. Gastrointestinal disorders that may significantly interfere with absorption of study drug.
7. Subjects have received potent CYP3A inhibitors within 7 days prior to the initiation of study treatment.
8. History of myocardial infarction, severe/unstable angina, or symptomatic congestive heart failure (New York Heart Failure (NYHA) Class III or IV congestive heart failure) within 6 months prior to study enrollment or LVEF <50%
9. Impaired cardiac function including ongoing cardiac dysrhythmias of Grade $\geq$ 2, ejection fraction < 50%, atrial fibrillation of any grade, or QTc prolongation > 450 ms, or other factors that increase the risk of QT prolongation (i.e. family history of long QT interval syndrome, hypokalemia defined as serum potassium < 3.0 mEq/L)
10. Diagnosis of other malignancies within the last 3 years other than curatively treated non-melanoma skin cancer, carcinoma in situ of the cervix, organ-confined or treated non-metastatic prostate cancer, in situ breast carcinoma after complete surgical resection, or superficial transitional cell bladder carcinoma.
11. Known active Hepatitis A, B or C.
12. Known HIV seropositivity.
13. Women who are pregnant or lactating.

4.0 TREATMENT PLAN

Study Design
This is a Phase II study to evaluate the safety and efficacy of Pacritinib for the treatment of lower-risk MDS patients. The primary efficacy endpoint is the overall response rate (ORR) based on the IWG-2006 criteria, which includes complete remission (CR), partial remission (PR), and major hematologic improvement (HI). Patient's overall response will be assessed after 1 cycle of treatment but no more than after 4 cycles of treatment; cycle length is 28 days. If no response is attained after 4 cycles of treatment or if progression occurs during therapy at any time, the patient is eligible to continue on Pacritinib therapy with the addition of hypomethylating agent therapy (HMA) [Part 2]. A maximum total of 40 evaluable patients (n=40) will be enrolled.

Agent Administration
All patients will be registered through Protocol Data Management System (PDMS) or CORe.

One initial schedule of Pacritinib will be studied: 200 mg orally twice daily (every 12 hours) without regards to per oral (p.o.) intake. Study cycles will be administered every 28 days $\pm$ 5 days or upon resolution of any clinically significant study drug related adverse event (AE) to grade 0-1, whichever occurs first.

Patients will receive the first course of therapy without interruption regardless of degree of myelosuppression unless patients with an un-transfused pre-treatment platelet count $\geq$50 x

10^9/L drops to less than 10 x 10^9/L. In that case, drug will be held until platelets return $\geq$ 10 x 10^9/L and resumed at the next lower dose if appropriate. If prolonged myelosuppression is observed with evidence of a hypocellular marrow (marrow cellularity less than 5% without evidence of recurrent MDS) drug will be stopped. Subsequent courses of Pacritinib will be given at the next lower dose, once counts recover. Count recovery will be considered if the ANC is $\geq$ 1 x10^9/L and platelets $\geq$ 50 x10^9/L. For other clinical situations, subsequent courses of therapy will be administered at the discretion of the treating physician. Patients with residual disease may start the next cycle with neutrophil or platelet counts lower than these, if judged to be in the best interest of the patient to initiate the next cycle. The decision to treat should be documented in the patient's medical record.

Patients will be assessed for disease response at the completion of 4 cycles of treatment. If there has been no response to therapy (i.e. stable disease per IWG-2006), or if disease progression is identified, patients will be eligible to continue on the study with the addition of 5-azacitadine or decitabine, with continuation of Pacritinib, in **Part 2** (Pacritinib + HMA). Patients responding to therapy will continue on Pacritinib monotherapy. Patients with disease progression prior to 4 cycles of Pacritinib are also eligible to initiate the Pacritinib + HMA study portion prior to the completion of 4 cycles. In these patients, disease assessment including bone marrow aspiration should be performed prior to the initiation of Part 2. Patients receiving Pacritinib + HMA will be re-assessed for disease response after 4 cycles of *combination* treatment on Part 2. Patients on pacritinib monotherapy in Part 1 who manifest disease progression at any time, including before or after completion of 4 cycles of treatment, can continue on study in Part 2 with the addition of a hypomethylating agent.

The addition of HMA should be initiated on Day 1 of the next Pacritinib treatment cycle whenever possible. HMA dosing and dosing adjustments should be performed according to the approved package insert, or at a dose of decitabine 20mg/m2 IV over 1 hour for 5 days every 4 weeks and according to standard of care (see Section 5.0 for rules of dose reduction or delay).

Duration of Therapy
All patients must complete a screening visit, day 1 visits for each new cycle (+/- 5 days), end of treatment visit and whenever possible, an end of study safety visit 28 days after the last dose of Pacritinib (+/- 7 days).

Patients may remain on study for up to 12 months if the patient demonstrates clinical benefit and no excessive toxicity (i.e. no clinically significant study-drug related grade $\geq$ 3 toxicity. Patients who are experiencing clinical benefit beyond 12 months and have not experienced excessive toxicity may be eligible to continue therapy after discussion with the PI and the discussion documented in the patient's medical record. After the end of therapy and/or 30 days after your last dose of study drug, the study staff will follow your health status by phone call every 2 months (+/- 2 months) until you receive another cancer treatment.

Treatment is expected to continue until one of the following criteria applies:

1. Possibility of undergoing allogeneic bone marrow transplantation
2. Unacceptable adverse event(s)
3. Discontinuation of study drug for more than 6 weeks
4. Clinically significant intercurrent illness that prevents further administration of treatment
5. Patient request
6. General or specific changes in the patient's condition that render the patient unacceptable for further treatment in the judgment of the investigator or treating physician.
7. Note that patients with progressive disease or lack of objective response after 4 cycles of Pacritinib monotherapy are eligible to continue on study with the addition of a hypomethylating agent (azacitadine or decitabine) to Pacritinib. In patients with clinically significant progressive disease and/or lack of objective response after 4 cycles of *combination* therapy, treatment will be discontinued.

After the first course of therapy, interval between cycles of therapy can be spaced out at the discretion of the treating physician, although a schedule as close as possible to the proposed one should be attempted.

5.0 DOSING DELAYS/DOSE MODIFICATIONS

Dose Modification of Pacritinib within Patients

The dose of Pacritinib will be adjusted according to the guidelines shown in the following tables for study drug-related and **clinically significant** toxicity. If toxicity is not covered in the table, doses may be reduced or held at the discretion of the investigator for the patient's safety and documented in the patient's medical record.

Patients with study-drug related toxicities that are manageable with supportive therapy may not require dose reductions.

Table 4. Pacritinib dose de-escalation

Clinically significant non-hematologic treatment related toxicity and dose management	
Toxicity Grade	*Management/Action*
1 or 2	No change required. If the toxicity persists at Grade 1 or 2, then reduction to a lower dose level is allowed according to Investigator judgment and the likelihood that the toxicity is related to Pacritinib.
3	Hold treatment. Treatment may resume if toxicity is resolved to Grade ≤ 1 or returned to baseline. If the toxicity resolves within 7 days, Pacritinib may be resumed at the same level or at the next lower dose level at the discretion of the Investigator. Toxicity lasting more than 7 days will require dose reduction to the next lower dose level.
4	Hold treatment. Treatment may resume if toxicity is resolved to Grade ≤ 1 or returned to baseline at the discretion of the Investigator. Treatment will be reduced by two dose levels from the level at which the toxicity was observed. If Grade 4 toxicity occurs at 200 mg/d, the patient should be discontinued from the study.

Table 5. Dose reduction levels

Starting dose level	400 mg/d
Dose level -1	300 mg/d
Dose level -2	200 mg/d

Patients that require reduction of more than 2 dose levels will be removed from the study.

Part 2 Dose Schedule

The pacritinib dose will be decreased to 200 mg in AM and 100 mg in the evening for the first cycle of combined therapy. The dose may be reduced further to 100 mg in AM and 100 mg in PM if grade 2 non-hematopoietic toxicity is observed. Pacritinib will be held for up to 14 days if grade 3 non-hematopoietic toxicity is observed and then restarted at 100 mg BID (every 12 hours). If no toxicity is observed in the first cycle of combined therapy, the pacritinib dose may be increased to 200 mg BID (every 12 hours) on subsequent cycles of combined therapy.

Patients with drug-related toxicities that are manageable with supportive therapy may not require dose reductions. Otherwise, the following dose adjustment rules apply:

Table 6:

Clinically significant treatment related toxicity and dose management

NCI CTCAE Grade	*Management/Action*
0-2 non-hematologic toxicity	No dose reduction. Grade 2 toxicities that are persistent and intolerable (i.e. stomatitis) can result in dose delays or dose reductions to the next lower dose level.
3-4 clinically significant non-hematologic toxicity	Hold until recovery to NCI CTCAE grade 0-1. If recovery occurs within 2 weeks after treatment has been held, dose should be reduced to -1 dose level, if appropriate.

Table 7: Suggested dose reductions for azacitadine and decitabine*

Dose Level	Decitabine (in mg/m2 for 5 days)	Azacitadine (in mg/m2 for 7 days)
0	20	75
-1	15	50
-2	10	25
-3	10 for 4 days	25 for 5 days

*Dose reductions different than those described above are acceptable after discussion with the investigator, and require documentation of the rationale for such action in the patient's medical record.

Study cycles should be administered every 28 days $\pm$ 5 days or upon resolution of any clinically significant study drug related AE to grade 0-1, whichever occurs first. Subsequent cycles will begin on the first day of Decitabine or Azacitidine therapy, and in the absence of toxicity, Pacritinib administration will not be interrupted. If a subsequent cycle is delayed due to adverse events related to HMA-agent, or if it is considered in the best interest of the patient to delay administration of HMA, administration of the Pacritinib may continue as planned.

Concomitant and Excluded Therapies

Patients may not receive other investigational agents, chemotherapy, or immunotherapy for the treatment of hematologic malignancy during the study apart from azacitadine or decitabine in Part 2 as described in Section 4.0, Treatment Plan.

Patients must discontinue treatment with any potent CYP3A4 inhibitors for 1 week prior to administration of study drug. Every effort should be made to avoid CYP3A4 inducers or inhibitors throughout the course of the study. If concomitant therapy with CYP3A4 inducers or inhibitors is medically necessary and there are no available or suitable substitutes, this should be documented in the patient's medical record and extreme caution taken with their continued use during the study duration.

Patients should not receive treatment with medications with a possible or conditional risk of causing Torsades de Pointes, or those associated with prolongation of the QT interval" unless medically necessary. If medically necessary, close follow up and monitoring is recommended.

All medications (other than study drug) and significant non-drug therapies administered after the patient starts treatment with study drug, with any changes in dosing must be recorded in the electronic medical record."

Erythropoietin, granulocytic or other hematopoietic colony stimulating factors for treatment of cytopenias are discouraged. If administered, the subject will not be eligible for a hematologic improvement (HI) response assessment within 2 weeks of administration.

6.0 AGENT FORMULATION AND PROCUREMENT

Pacritinib study drug description and storage

Pacritinib for oral administration is supplied in capsules containing 100 mg as free base SB1518 in dark green or red cap/gray body size "zero" opaque hard gelatin capsules. The inactive ingredients are microcrystalline cellulose, magnesium stearate, and polyethylene glycol 8000.

Pacritinib capsules should not be opened or crushed. Direct contact of the powder in Pacritinib capsules with the skin or mucous membranes should be avoided. If such contact occurs, affected areas should be washed thoroughly with water.

Pacritinib capsules should be stored at controlled room temperature, excursions allowed from 15 to 30ºC or 59 to 86ºF. All study supplies must be kept in a restricted-access area.

Observed Risks

Based on the adverse effects observed in toxicity studies, as well as data from phase 1 and 2 clinical studies in patients with advanced MF, AML, and lymphoma, the investigator is advised to observe the following precautions:

- **Hematologic**: Reversible myelosuppression; leukopenia, neutropenia, thrombocytopenia, and anemia have been observed; these events are common in hematologic malignancies and may not be related to pacritinib administration. Thrombocytopenia, anemia, neutropenia, and leukopenia ADRs were more common in lymphoid than myeloid patients. Patients participating in clinical trials of pacritinib will be monitored frequently for myelosuppression. Provisions for interruption of treatment and dose reduction in the event of myelosuppression are outlined in clinical trial protocols.
- **Gastrointestinal**: Treatment with pacritinib is associated with dose-related diarrhea and nausea. Vomiting, abdominal pain, abdominal distension, anorexia, and constipation have also been reported to be related to pacritinib administration. Diarrhea and vomiting ADRs were more common in myeloid studies than lymphoid studies, but the reverse was true for constipation. Nausea was the same in both populations. Antiemetic and antidiarrheal medications should be prescribed prophylactically to control symptoms. Fluid and electrolytes should be replaced as needed to prevent dehydration. Pre-existing nausea, vomiting, and diarrhea should be adequately controlled before beginning therapy. Patients with significant GI symptoms despite optimal supportive care may have study drug interrupted or have the dose of study drug reduced per protocol.

- **Cardiac**: QTc prolongation has been reported in association with pacritinib therapy. A post-hoc review of the patients in the phase 1-2 Studies SB1518-2007-001 and SB1518-2008-003 identified 9 patients with at least one reported QTc exceeding 500 ms; all but one were treated at a pacritinib dose of ≥ 400 mg QD. Therapy was continued in most of these patients for extended periods of time after the observations of QTc prolongation. In addition, pre-study cardiac history was noted in 7 patients, and 8 patients were taking concomitant medications associated with QTc prolongation. Other cardiac TEAEs and ADRs were rare in all pacritinib-treated populations. Based on these observations, investigators are advised to exclude any patient with a baseline QTc > 450 ms and to perform routine ECG monitoring during treatment. For patients with identified changes in treatment-emergent ECG abnormalities or other cardiac events, study drug may be interrupted or the dose may be reduced per protocol.
- **Hepatic**: Animal studies suggest that pacritinib treatment may cause dose-related hepatotoxicity. There has been little evidence of hepatotoxicity in clinical trials to date. Significant increases in aminotransferases have been observed infrequently and have not been accompanied by concomitant increases in bilirubin. Most reported hepatic toxicities were seen in the myeloid population. No instances of hepatic dysfunction meeting Hy's Law criteria have been observed. Hepatic function (AST/SGPT, ALT/SGOT, alkaline phosphatase, and total bilirubin) will continue to be assessed at frequent intervals during treatment with pacritinib.
- **Renal**: Animal studies suggest that treatment may cause dose-related renal toxicity. To date, little impact on renal function has been observed in human clinical trials. Assessment of renal function (creatinine, BUN, sodium, potassium) will be undertaken frequently in patients participating in clinical trials of pacritinib.
- **General**: Additional adverse effects commonly reported during pacritinib administration have included infections, fatigue, asthenia, peripheral edema, pyrexia, and rash. Infectious ADRs were more common in lymphoid patients, while the other adverse effects were more common in myeloid patients. Bleeding and bruising complications have been reported, most of which were low grade and generally more common in the myeloid population than in the lymphoid population. Patients participating in clinical trials of pacritinib will be monitored closely for these adverse effects.

No reproductive or developmental toxicity studies have been performed; therefore, the effects of pacritinib on female and male fertility are unknown. If fertile, both males and females must agree to use effective birth control. Women of childbearing potential must use highly effective methods of birth control for the duration of study treatment and for 12 months after last dose of study drug. The contraceptive methods considered highly effective are intrauterine devices and hormonal contraceptives (contraceptive pills, implants, transdermal patches, hormonal vaginal devices, or injections with prolonged release).

No drug-drug interaction studies have been conducted with pacritinib. In vitro studies indicate that pacritinib has no significant potential to inhibit or induce CYP450 isozymes and has no significant involvement with p-glycoprotein mediated transport. Pacritinib is believed to be metabolized by CYP3A4. Therefore, caution is advised when considering the coadministration of potent inhibitors of CYP3A4 in conjunction with pacritinib. Specific exclusions are outlined in the protocols for human clinical trials of pacritinib.

Gastrointestinal Toxicity Management

Treatment with pacritinib is associated with dose-related diarrhea and nausea. Vomiting, abdominal pain, abdominal distension, anorexia, and constipation have also been reported

to be related to pacritinib administration. Diarrhea and vomiting ADRs were more common in myeloid studies than lymphoid studies, but the reverse was true for constipation. Nausea was the same in both populations. Antiemetic and antidiarrheal medications should be prescribed prophylactically to control symptoms. Fluid and electrolytes should be replaced as needed to prevent dehydration. Pre-existing nausea, vomiting, and diarrhea should be adequately controlled before beginning therapy. Patients with significant GI symptoms despite optimal supportive care may have study drug interrupted or have the dose of study drug reduced per protocol.

Study Drug Dose, Route, and Mode of Administration
Patients will be supplied with 100 mg pacritinib capsules. Two capsules should be taken twice a day at approximately the same time of day without regard of food. Dose reductions will be allowed if the Investigator determines an event is related to study treatment as per Section 5.0.

Pacritinib will be administered primarily on an outpatient basis, but may be administered as an inpatient. Vomited doses will not be made up on the same day. Unused study drug returned by the patient will be destroyed in accordance with institutional policy.

Decitabine and azacitadine may be administered by local doctor or at MD Anderson. Cycle 1 of Part 2 will be administered at MDACC. Commercial supplies of decitabine and azacitadine will be used.

Outside physician participation during treatment is acceptable. Documentation of MDACC physician communication with the outside physician will be required. Protocol required evaluations outside MDACC will be documented by telephone, fax or email. Fax and/or email will be dated and signed by the MDACC physician. Changes in HMA drug dose and/or schedule must be discussed with and approved by the MDACC PI or Investigator, or their representative prior to initiation, and will be documented in the patient record.

7.0 CORRELATIVE/SPECIAL STUDIES

Samples will be collected for potential studies and will be stored in the laboratory of Dr. Guillermo Garcia-Manero in the Department of Leukemia at MD Anderson Cancer Center. Samples will be collected as follows:

Bone marrow aspirate: pre-therapy (within 28 days of study start), Day 28 of first cycle, and Day 28 of fourth cycle (or earlier if disease progression), and Day 28 of the fourth cycle of Part 2 (pacritinib + HMA).

Peripheral blood (including serum): on Days 0, 7, 14, 21 and 28 of the first cycle.

All samples can be obtained +/- 5 days.

Samples could be used, but not limited, to analysis of cytokine profile, genetic and epigenetic lesions and molecular characterization, and STAT3 phosphorylation.

Mutational profiling by the Molecular Diagnostics Lab (MDL) will be performed on pre-therapy and sequential bone marrow aspirates.

8.0 PATIENT EVALUATION

Pre-treatment
Results from tests performed as standard of care up to 28 days prior to study entry can be used for determining eligibility and establishing baseline measurements. Any clinically significant abnormal results (except for bone marrow aspiration) should be repeated within 48 hours of beginning therapy to establish a baseline.

- A complete history and physical exam including height, weight, and vital signs (blood pressure, heart rate, breathing rate, and temperature), disease documentation, and performance status (PS) should be performed 72 hours before initiation of study, and subsequently as indicated.

- Document significant baseline abnormalities and concomitant medications within 28 days of initiation of study.

- CBC, platelet count, creatinine, total bilirubin, AST and/or ALT, potassium and blood or urine pregnancy test (when indicated) should be performed 72 hours before initiation of study. Pregnancy testing will be conducted for any female that is of childbearing age that has not been surgically sterilized or without a menses for 12 consecutive months.

- Bone marrow aspirate and/or biopsy during the last 28 days preceding study initiation

- FLT3 and JAK2 mutational status (including FLT3 allelic ratio if mutation present). This can be performed on the pre-study bone marrow aspirate or from peripheral blood

- Cytogenetics will be obtained within 28 days prior to therapy, and should be repeated with all bone marrow assessments performed on study.

- EKG with documentation of QTc interval

- Echocardiogram or MUGA for evaluation of left ventricular ejection fraction

- Pretreatment correlative studies if patient signed for them during consent process.

During Treatment
- CBC, platelet count, creatinine, AST and/or ALT and total bilirubin weekly during first course of therapy, and prior to each course of therapy.

- Document adverse event assessment and concomitant medications at each clinic visit.

- EKG will be performed 4 hours post dosing on cycle 1 day 1 (+/- 1 hour) , on cycle 1 day 28 (+/- 5 days), cycle 4 day 28 (+/- 5 days), and as clinically indicated.

- Bone marrow aspiration on day 28 (+/- 5 days) of Cycle 1 and Cycle 4 of Part 1, and after

Cycle 4 of Part 2 (Pacritinib + HMA). Additional interim aspirations are allowed as indicated by the treating physician. If patients require the addition of a HMA prior to the end of cycle 4, a bone marrow aspiration prior to HMA start should be performed. Cytogenetic analysis should be performed for all bone marrow assessments.

- Peripheral blood for correlative studies as described above (+/- 5 days) and as shown in Table 8.

End-of-Treatment
Whenever possible, twenty-eight days following the last dose of study drug (+/- 7 days):

- CBC with chemistry
- Bone marrow aspirate, if clinically indicated.
- Document adverse event assessment and concomitant medications.

9.0 STUDY CALENDAR DURING (all pretreatment tests within 28 days unless specified otherwise)

Table 8. Study Calendar

		Course 1				End of Treatment
	PreTx	**W1**	**W2**	**W3**	**W4**	
Complete history, physical exam including height, weight, vital signs, and PS	X	**X**				
Concomitant medication and significant baseline abnormality assessment	X	X	X	X	X	X
CBC[1,3]	X	X	X	X	X	X
Chemistries[3]	X	X	X	X	X	X
Pregnancy test[3]	X					
EKG and EF assessment by ECHO or MUGA	X	X			D28	
Bone marrow aspirate	X				D28	X[4]

and/or biopsy with cytogenetics[2]						
JAK2 and FLT3 mutational status[5]	X					
Correlative Studies[6]						

Footnotes:
PreTx, pretreatment; W, week; D day.
1 CBC includes complete differential and platelets;
2 Bone marrow aspirate and/or biopsy with cytogenetics should be performed within 28 days at pretreatment, on D28 (+/- 5 days) of Cycle 1 and Cycle 4, and D28 (+/- 5 days) of Cycle 4 after initiation of HMA agent + Pacritinib on Part 2. If HMA agent is added prior to end of Cycle 4, bone marrow aspirate and/or biopsy should be performed before prior to Part 2 start.
3 within 72 hours at pretreatment
4 if clinically indicated
5 mutational status can be assessed by peripheral blood or bone marrow as appropriate
6 Correlatives obtained during - Bone marrow aspirate sample: pre-therapy (within 28 days of study
start), Day 28 (+/- 5 days) of first cycle, and
Day 28 (+/- 5 days) of fourth cycle (or earlier if disease progression), and Day 28 (+/- 5 days) of the fourth cycle of Part 2
(pacritinib + HMA) Peripheral blood (including serum): on Days 0, 7, 14, 21 and 28 of the first cycle (+/- 5 days).
7 A single 12-lead EKG will be performed at screening, at 4 hours post dosing on cycle 1 day
1,on cycle 1 day 28 (+/- 5 days), cycle 4 day 28 (+/- 5 days), and as clinically indicated".

10.0 CRITERIA FOR RESPONSE

The primary endpoint is the overall response rate (ORR) per IWG-2006 criteria[13]. This is summarized in table 9 and 10. Response will include complete remission, partial remission, marrow complete remission and any hematological improvement achieved any time during the duration of the therapy.

Table 9. Proposed modified International Working Group response criteria for altering natural history of MDS[7]

Category	Response criteria (responses must last at least 4 wk)
Complete remission	Bone marrow: ≤5% myeloblasts with normal maturation of all cell lines*

	Persistent dysplasia will be noted*†
	Peripheral blood‡ • Hgb ≥11 g/dL • Platelets ≥100 x 10^9/L • Neutrophils ≥1.0 x 10^9/L† • Blasts 0%
Partial remission	All CR criteria if abnormal before treatment except: • Bone marrow blasts decreased by ≥50% over pretreatment but still > 5% • Cellularity and morphology not relevant
Marrow CR†	Bone marrow: ≤5% myeloblasts and decrease by ≥50% over pretreatment† Peripheral blood: if HI responses, they will be noted in addition to marrow CR†
Stable disease	Failure to achieve at least PR, but no evidence of progression for > 8 wks
Failure	Death during treatment or disease progression characterized by worsening of cytopenias, increase in percentage of bone marrow blasts, or progression to a more advanced MDS FAB subtype than pretreatment
Relapse after CR or PR	At least 1 of the following: • Return to pretreatment bone marrow blast percentage • Decrement of ≥50% from maximum remission/response levels in granulocytes or platelets • Reduction in Hgb concentration by ≥1.5 g/dL or transfusion dependence
Cytogenetic response	Complete • Disappearance of the chromosomal abnormality without appearance of new ones Partial • At least 50% reduction of the chromosomal abnormality
Disease progression	For patients with: • Less than 5% blasts: ≥50% increase in blasts to > 5% blasts • 5%-10% blasts: ≥50% increase to > 10% blasts • 10%-20% blasts: ≥50% increase to > 20% blasts • 20%-30% blasts: ≥50% increase to > 30% blasts Any of the following: • At least 50% decrement from maximum remission/response in granulocytes or platelets • Reduction in Hgb by ≥2 g/dL • Transfusion dependence
Survival	Endpoints: • Overall: death from any cause • Event free: failure or death from any cause • PFS: disease progression or death from MDS • DFS: time to relapse

- Cause-specific death: death related to MDS

Table 10. Proposed modified International Working Group response criteria for hematologic improvement[7]

Hematologic improvement*	Response criteria (responses must last at least 8 wk)†
Erythroid response (pretreatment, < 11 g/dL)	Hgb increase by ≥1.5 g/dL Relevant reduction of units of RBC transfusions by an absolute number of at least 4 RBC transfusions/8 wk compared with the pretreatment transfusion number in the previous 8 wk. Only RBC transfusions given for a Hgb of ≤9.0 g/dL pretreatment will count in the RBC transfusion response evaluation†
Platelet response (pretreatment, < 100 x 10^9/L)	Absolute increase of ≥30 x 10^9/L for patients starting with > 20 x 10^9/L platelets Increase from < 20 x 10^9/L to > 20 x 10^9/L and by at least 100%†
Neutrophil response (pretreatment, < 1.0 x 10^9/L)	At least 100% increase and an absolute increase > 0.5 x 10^9/L†
Progression or relapse after HI‡	At least 1 of the following: At least 50% decrement from maximum response levels in granulocytes or platelets Reduction in Hgb by ≥1.5 g/dL Transfusion dependence

Secondary endpoints include:

1. To evaluate efficacy of Pacritinib based on the presence of JAK2 and FLT3 mutations at diagnosis.
2. To evaluate efficacy based on IPSS, R-IPSS and LR-MDS risk classification systems at diagnosis.

11.0 CRITERIA FOR REMOVAL FROM THE STUDY

This will include:

1. Possibility of undergoing allogeneic bone marrow transplantation
2. Unacceptable adverse event(s)
3. Discontinuation of study drug for more than 6 weeks
4. Clinically significant intercurrent illness that prevents further administration of treatment
5. Patient request
6. General or specific changes in the patient's condition that render the patient unacceptable for further treatment in the judgment of the investigator or treating physician.
7. Note that patients with progressive disease or lack of objective response after 4 cycles of Pacritinib monotherapy are eligible to continue on study with the addition of a hypomethylating agent (azacitidine or decitabine) to Pacritinib. In patients with clinically significant progressive disease and/or lack of objective response after 4 cycles of *combination* therapy, treatment will be discontinued.

12.0 STATISTICAL CONSIDERATIONS

General Description

This is a Phase II open-label study designed to evaluate the efficacy and toxicity of Pacritinib as single agent (Part 1) and Pacritinib + HMA (Part 2) in subjects with lower-risk myelodysplastic syndrome. The primary efficacy outcome of both parts is the overall response rate (ORR) based mainly on hematologic improvement defined by IWG-2006 criteria, and which also includes complete remission (CR), partial remission (PR) and marrow complete remission. In Part 1, patient's overall response will be assessed after 1 cycle of treatment but no more than after 4 cycles of treatment; cycle length is 28 days. If no response is attained after 4 cycles of treatment or if progression occurs during therapy at any time, the patient is eligible to continue on Pacritinib + HMA (Part 2). Part 2 will not continue if Part 1 is stopped early. A maximum of 40 total evaluable patients (n=40) will be enrolled.

Study design: Part 1

The standard practice is to put these patients on observation, an ORR rate of about 15% is clinically relevant. The target ORR with the experimental treatment is 15%. The regimen of the Pacritinib will be considered worthy of further investigation if it elicits an increase in ORR to 15% with acceptable toxicity. A >15% therapy related non-hematological grade 3/4 toxicity rate is considered unacceptable. Thus, interim monitoring rules, assuming the prior distributions below, were constructed that meet the following two conditions,

1) Stop if Prob{ p(ORR,E)< 0.15 | data} > 0.975, or
2) Stop if Prob{ p(TOX,E)>0.15| data}>0.975,

where P(ORR, E) and P(TOX, E) are the true ORR and toxicity rates for the Pacritinib, and p(ORR, H) is the true ORR rate of the standard treatment. The first rule provides for stopping the study if the data suggest that it is unlikely (i.e., probability < 2.5%) that ORR rate of the Pacritinib treatment is greater than 15.0%. The second condition will stop the study early if excessive therapy-related non-hematological grade 3/4 toxicity (>15%) is highly probable (i.e., probability >97.5%) for the Pacritinib. Monitoring for toxicity and futility will not begin until 5 patients have been evaluated, and cohort size for future evaluations is 5.

The monitoring rule for the toxicity rate, based on these assumptions and monitoring conditions above is found in Table 11. For example, accrual will cease if 4 or more patients experience toxicities among the first 5 patients.

Table 11. Stop accrual if the number of drug-related non-hematological grade 3/4 toxicities is greater than or equal to indicated (i.e., # patients with toxicities) among the number of patients evaluated

# patients evaluated	5	10	15	20	25	30	35	40
# patients with toxicities	4-5	5-10	6-15	7-20	8-25	10-30	11-35	Always stop with this many patients

Monitoring the ORR rate, based on the above assumptions and monitoring conditions is found in Table 12. For example, accrual will cease if 0 patients experience an overall response within 4 cycles in the first 15 patients treated.

Table 12. Stop accrual if the number with overall response is less than or equal to indicated (i.e., # patients with overall response) among the number of patients evaluated

# patients evaluated	5-10	15-25	30-35	40
# patients with overall response	Never Stop with this many patients	0	0-1	Always stop with this many patients

Multc Lean Desktop (version 2.1.0) was used to generate the toxicity and futility stopping boundaries and the OC table (Table 13). In order to utilize the software for the design, a response constant rate of 0.15 and beta (0.3, 1.7) priors were assumed for the standard treatment response distribution and experimental treatment response prior distribution, respectively. In addition, a 15% toxicity constant rate and beta (0.3, 1.7) priors were assumed for the standard treatment toxicity constant rate and experimental treatment toxicity prior distribution, respectively.

The probability of stopping the study early if the true ORR of the Pacritinib was 15% and the true toxicity rate was 15% was 21.9%. Probabilities of stopping early for high true toxicity rates (i.e., 40%) were 95.3% when the true ORR was 15%, and 94.4% when true ORR rate was 30%.

Table 13. Operating characteristics for simultaneous monitoring response and toxicity rates for patients treated with Pacritinib treatment		
True Toxicity Rate	True ORR	Prob(stop the trial early)
0.05	0.05	0.6908
	0.10	0.3528
	0.15	0.1654
	0.20	0.0754
	0.25	0.0338
	0.30	0.0150
0.10	0.05	0.6944
	0.10	0.3603
	0.15	0.1750
	0.20	0.0861
	0.25	0.0450
	0.30	0.0264
0.15	0.05	0.7109
	0.10	0.3947
	0.15	**0.2194**
	0.20	0.1352
	0.25	0.0964
	0.30	0.0787
0.20	0.05	0.7533
	0.10	0.4836
	0.15	0.3341
	0.20	0.2623
	0.25	0.2292
	0.30	0.2141
0.30	0.05	0.8956
	0.10	0.7815
	0.15	0.7182
	0.20	0.6879
	0.25	0.6378
	0.30	0.6675
0.40	0.05	0.9826
	0.10	0.9635
	0.15	**0.9529**
	0.20	0.9478
	0.25	0.9455
	0.30	**0.9444**
0.50	0.05	0.9990
	0.10	0.9979
	0.15	0.9973
	0.20	0.9970
	0.25	0.9969
	0.30	0.9968

Study design: Part 2

The standard practice is to put these patients on observation, an ORR rate of about 30% is

clinically relevant. The target ORR with the experimental treatment is 30%. The regimen of the Pacritinib + HMA will be considered worthy of further investigation if it elicits an increase in ORR to 30% with acceptable toxicity. A >30% therapy related non-hematological grade 3/4 toxicity rate is considered unacceptable. Thus, interim monitoring rules, assuming the prior distributions below, were constructed that meet the following two conditions,

1) Stop if Prob{ p(ORR,E)< 0.30 | data} > 0.975, or
2) Stop if Prob{ p(TOX,E)>0.30| data}>0.95,

where P(ORR, E) and P(TOX, E) are the true ORR and toxicity rates for the Pacritinib + HMA, and p(ORR, H) is the true ORR rate of the standard treatment. The first rule provides for stopping the study if the data suggest that it is unlikely (i.e., probability < 2.5%) that ORR rate of the Pacritinib + HMA treatment is greater than 30.0%. The second condition will stop the study early if excessive therapy-related non-hematological grade 3/4 toxicity (>30.0%) is highly probable (i.e., probability >95.0%) for the Pacritinib + HMA. Monitoring for toxicity and futility will not begin until 5 patients have been evaluated, and cohort size for future evaluations is 5. The monitoring rule for the toxicity rate, based on these assumptions and monitoring conditions above is found in Table 14. For example, accrual will cease if 4 or more patients experience toxicities among the first 5 patients.

Table 14. Stop accrual if the number of drug-related non-hematological grade 3/4 toxicities is greater than or equal to indicated (i.e., # patients with toxicities) among the number of patients evaluated

# patients evaluated	5	10	15	20	25	30	35	40
# patients with toxicities	4-5	6-10	8-15	10-20	12-25	14-30	16-35	Always stop with this many patients

Monitoring the ORR rate, based on the above assumptions and monitoring conditions is found in Table 15. For example, accrual will cease if 0 patients experience an overall response within 4 cycles in the first 10 patients treated.

Table 15. Stop accrual if the number with overall response is less than or equal to indicated (i.e., # patients with overall response) among the number of patients evaluated

# patients evaluated	5	10	15	20	25	30	35	40
# patients with	Never stop	0	0-1	0-2	0-3	0-4	0-5	Always stop

overall response	with this many patients							with this many patients

Multc Lean Desktop (version 2.1.0) was used to generate the toxicity and futility stopping boundaries and the OC table (Table 16). In order to utilize the software for the design, a response constant rate of 0.30 and beta (0.6, 1.4) priors were assumed for the standard treatment response distribution and experimental treatment response prior distribution, respectively. In addition, a 30% toxicity constant rate and beta (0.6, 1.4) priors were assumed for the standard treatment toxicity constant rate and experimental treatment toxicity prior distribution, respectively.

The probability of stopping the study early if the true ORR of the Pacritinib + HMA was 30% and the true toxicity rate was 30% was 14.3%. Probabilities of stopping early for high true toxicity rates (i.e., 50%) were 84.4% when the true ORR was 30%, and 83.6% when true ORR rate was 40%.

Table 16 Operating characteristics for simultaneous monitoring response and toxicity rates for patients treated with Pacritinib treatment

True Toxicity Rate	True ORR	Prob(stop the trial early)
0.10	0.10	0.9090
	0.20	0.3994
	0.30	0.0848
	0.40	0.0127
	0.50	0.0020
0.20	0.10	0.9103
	0.20	0.4079
	0.30	0.0979
	0.40	0.0268
	0.50	0.0163
0.30	0.10	0.9202
	0.20	0.4730
	0.30	**0.1970**
	0.40	0.1337
	0.50	0.1243
0.40	0.10	0.9505
	0.20	0.6734
	0.30	0.5023
	0.40	0.4631

	0.50	0.4573
0.50	0.10	0.9849
	0.20	0.9005
	0.30	0.8484
	0.40	0.8365
	0.50	0.8347

Statistical Analysis Plan

All patients who received any dose of the study agent will be included in the analysis for efficacy and safety. Demographic/clinical characteristics (i.e. including duration of response) and safety data of the patients will be summarized using descriptive statistics such as mean, standard deviation, median and range. For the primary efficacy analysis, we will estimate the ORR for the Pacritinib and Pacritinib + HMA, along with the 95% confidence interval. Patients who drop out of the study before completing all the cycles will be treated as "failures" for the primary analysis. Overall response rate (ORR) during the study period will also be presented with the 95% confidence interval. The association between ORR and patient's clinical characteristics will be examined by Wilcoxon's rank sum test or Fisher's exact test, as appropriate. Toxicity type, severity and attribution will be summarized for each patient using frequency tables. The distribution of time-to-event endpoints (EFS and OS) including overall survival and event free survival will be estimated using the method of Kaplan and Meier. Comparisons of time-to-event endpoints by important subgroups will be made using the log-rank tests. The additional treatment in part 2 will be included in time-to-event analysis as time-dependent covariates.

12.0 PROTOCOL ADMINISTRATION

Protocol specific data will be entered into PDMS/CORe.

Protocol amendments
Changes to the protocol will be made only when protocol amendments have been signed by the principal investigator and approved by the ethical committee of MD Anderson Cancer Center.

Archival of data

All patient data (including source data) generated in connection with this study will be kept in the archives of the MDACC in accordance with 21 CFR 312.62. All data will be available for inspection by company representatives of the Medical Department and by regulatory authorities.

13.0 REPORTING REQUIREMENTS

The Investigator or physician designee is responsible for verifying and providing source documentation for all adverse events and assigning the attribution for each event for all subjects enrolled on the trial.The PI or designee will be responsible for determining attribution for all events.

Grade 3 or 4 related and/or unexpected adverse events and protocol specific data will be entered into PDMS/CORE. PDMS/CORE will be used as the electronic case report form for this protocol.

Reporting Requirements
The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 4.0 will be utilized for adverse event reporting. (http://ctep.cancer.gov/reporting/ctc.html).

Adverse event reporting will be as per the NCI criteria and the MDACC Leukemia Specific Adverse Event Recording and Reporting Guidelines.

Adverse event is any untoward medical occurrence that may present during treatment with a pharmaceutical product but which does not necessarily have a causal relationship with this treatment.

Adverse drug reaction is a response to a drug which is noxious and unintended and which occurs at doses normally used in man for prophylaxis, diagnosis, or therapy of disease or for the modification of physiologic function.

Assessing causal connections between agents and disease is fundamental to the understanding of adverse drug reactions. In general, a drug may be considered a contributory cause of an adverse event if, had the drug not been administered, 1) the event would not have happened at all, 2) the event would have occurred later than it actually did, or 3) the event would have been less severe.

Adverse Events (AEs) will be evaluated according to the most current version of the Pacritnib Investigator Brochure for expectedness. The current CTC version will be used to assess and grade AE severity. Only unexpected AEs will be recorded in the Case Report Form (CRF). Expected events during leukemia therapy are:

1. *Myelosuppression related events (due to disease or leukemia therapy)*
    a. *febrile or infection episodes not requiring management in the intensive care unit*
    b. *epistaxis or bleeding except for catastrophic CNS or pulmonary hemorrhage*
    c. *anemia, neutropenia, lymphopenia, thrombocytopenia, leukopenia, leukocytosis*
2. *Disease related events*
    a. *symptoms associated with anemia*
        i. *fatigue*
        ii. *weakness*

iii. shortness of breath

b. electrolyte abnormalities (sodium, potassium, bicarbonate, CO2, magnesium)
c. chemistry abnormalities (LDH, phosphorus, calcium, BUN, protein, albumin, uric acid, alkaline phosphatase, glucose)
d. coagulation abnormalities
e. disease specific therapy (induction, maintenance, salvage, or stem cell therapy)
f. alopecia
g. bone, joint, or muscle pain
h. liver function test abnormalities associated with infection or disease progression
i. disease progression

3. General therapy related events
a. catheter related events
b. renal failure related to tumor lysis syndrome or antibiotic/antifungal therapy
c. rash related to antibiotic use

4. Hospitalization for the management of any of the above expected events

Abnormal hematologic values will not be recorded on the CRF. For abnormal chemical values grade 3 or 4, the apogee will be reported per course in the CRF.

Serious Adverse Event Reporting (SAE)

An adverse event or suspected adverse reaction is considered “serious” if, in the view of either the investigator or the sponsor, it results in any of the following outcomes:

- Death
- A life-threatening adverse drug experience – any adverse experience that places the patient, in the view of the initial reporter, at immediate risk of death from the adverse experience as it occurred. It does not include an adverse experience that, had it occurred in a more severe form, might have caused death.
- Inpatient hospitalization or prolongation of existing hospitalization
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions.
- A congenital anomaly/birth defect.

Important medical events that may not result in death, be life threatening, or require hospitalization may be considered a serious adverse drug experience when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. 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 (21 CFR 312.32).

- Important medical events as defined above, may also be considered serious adverse events. Any important medical event can and should be reported as an SAE if deemed appropriate by the Principal Investigator or the IND Sponsor, IND Office.

- All events occurring during the conduct of a protocol and meeting the definition of a SAE must be reported to the IRB in accordance with the timeframes and procedures outlined in ôThe University of Texas M. D. Anderson Cancer Center Institutional Review Board Policy

for Investigators on Reporting Unanticipated Adverse Events for Drugs and Devicesö. Unless stated otherwise in the protocol, all SAEs, expected or unexpected, must be reported to the IND Office, regardless of attribution (within 5 working days of knowledge of the event).

- All life-threatening or fatal events, that are unexpected, and related to the study drug, must have a written report submitted within 24 hours (next working day) of knowledge of the event to the Safety Project Manager in the IND Office.

- Unless otherwise noted, the electronic SAE application (eSAE) will be utilized for safety reporting to the IND Office and MDACC IRB.

- Serious adverse events will be captured from the time of the first protocol-specific intervention, until 30 days after the last study treatment/intervention, unless the participant withdraws consent. Serious adverse events must be followed until clinical recovery is complete and laboratory tests have returned to baseline, progression of the event has stabilized, or there has been acceptable resolution of the event.

- Additionally, any serious adverse events that occur after the 30 day time period that are related to the study treatment must be reported to the IND Office. This may include the development of a secondary malignancy.

  - Serious adverse events will be forwarded to FDA by the IND Sponsor (Safety Project Manager IND Office) according to 21 CFR 312.32.

    It is the responsibility of the PI and the research team to ensure serious adverse events are reported according to the Code of Federal Regulations, Good Clinical Pactices, the protocol guidelines, the sponsor's guidelines, and Institutional Review Board policy.

Reporting of external SAEs

- The MDACC institutional policy for reporting of external SAEs will be followed.

It is the responsibility of the PI and the research team to ensure serious adverse events are reported according to the Code of Federal Regulations, Good Clinical Practices, the protocol guidelines, the sponsor's guidelines, and Institutional Review Board policy.

14.0 REFERENCES

1. Vardiman JW, Thiele J, Arber DA, et al: The 2008 revision of the WHO classification of myeloid neoplasms and acute leukemia: rationale and important changes. Blood 114:937-951, 2009
2.Greenberg P, Cox C, LeBeau MM, et al: International scoring system for evaluating prognosis in myelodysplastic syndromes. Blood 89:2079-88, 1997
3. Garcia-Manero G, Shan J, Faderl S, et al: A prognostic score for patients with lower risk myelodysplastic syndrome. Leukemia 22:538-43, 2008
4. Silverman LR, Demakos EP, Peterson BL, et al: Randomized controlled trial of azacitidine in patients with the myelodysplastic syndrome: a study of the cancer and leukemia group B. J Clin Oncol 20:2429-40, 2002
5. List A, Kurtin S, Roe DJ, et al: Efficacy of lenalidomide in myelodysplastic syndromes. N Engl J Med 352:549-57, 2005
6. Kantarjian H, Issa JP, Rosenfeld CS, et al: Decitabine improves patient outcomes in myelodysplastic syndromes: results of a phase III randomized study. Cancer 106:1794-803, 2006
7. Fenaux P, Mufti GJ, Hellstrom-Lindberg E, et al: Azacitidine prolongs overall survival compared with conventional care regimens in elderly patients with low bone marrow blast count acute myeloid leukemia. J Clin Oncol 28:562-9, 2010
8. Kantarjian H, Oki Y, Garcia-Manero G, et al: Results of a randomized study of three schedules of low-dose decitabine in higher risk myelodysplastic syndrome and chronic myelomonocytic leukemia. Blood, 2006
9. Cutler CS, Lee SJ, Greenberg P, et al: A decision analysis of allogeneic bone marrow transplantation for the myelodysplastic syndromes: delayed transplantation for low-risk myelodysplasia is associated with improved outcome. Blood 104:579-85, 2004
10. Tefferi A, Vaidya R, Caramazza D, et al: Circulating Interleukin (IL)-8, IL-2R, IL-12, and IL-15 Levels Are Independently Prognostic in Primary Myelofibrosis: A Comprehensive Cytokine Profiling Study. J Clin Oncol 29:1356-63
11. Tefferi A, Vardiman JW: Classification and diagnosis of myeloproliferative neoplasms: the 2008 World Health Organization criteria and point-of-care diagnostic algorithms. Leukemia 22:14-22, 2008
12. Greenberg PL, Tuechler H, Schanz J, et al: Revised international prognostic scoring system for myelodysplastic syndromes. Blood 120:2454-65, 2012
13. Cheson BD, Greenberg PL, Bennett JM, et al: Clinical application and proposal for modification of the International Working Group (IWG) response criteria in myelodysplasia. Blood 108:419-25, 2006