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Thyrosin kinase inhibitors: nilotinib

Nilotinib is an aminopyrimidine derivative that inhibits the tyrosine kinase activity of the chimeric protein BCR-ABL. The unregulated activity of the ABL tyrosine kinase in the BCR-ABL protein causes CML, and inhibition of tyrosine kinase activity is key to the treatment of the disorder. Nilotinib acts via competitive inhibition at the binding site of the BCR-ABL protein in a similar manner to imatinib, although nilotinib has a higher binding affinity and selectivity for the ABL kinase. Once bound to the ATP-binding site, nilotinib inhibits tyrosine phosphorylation of proteins involved in BCR-ABL-mediated intracellular signal transduction.

The inhibitory activity of nilotinib in CML cell is markedly higher than that of imatinib. In sensitive CML cell lines, the inhibitory ctivity of nilotinib is 20-50 times that of imatinib. For example, mean 50% inhibitory concentrations (IC_{50}) of cellular BCR-ABL autophosphorylation and proliferation of Ba/F3 BCR-ABL cells were 21 and 25 nmol/L, respectively, for nilotinib compared with 220 and 649 nmol/L for imatinib. The inhibitory activity of nilotinib is 3- to 7-fold that of imatinib in imatinib-resistant. Nilotinib demonstrated inhibitory activity in 32 of 33 imatinib-resistant CML cell lines The only BCR-ABL mutant that was not inhibited by nilotinib was T315I.

Pharmacokinetic profile

The extent of nilotinib absorption following oral administration was estimated to be approximately 30%. The bioavailability of nilotinib was increased when given with a meal. Compared to the fasted state, the systemic exposure (AUC) in the fed state increased by 15% (drug administered 2 hours after a light meal), 29% (30 minutes after a light meal), or 82% (30 minutes after a high fat meal), and the C_{max} increased by 33% (2 hours after a light meal), 55% (30 minutes after a light meal), or 112% (30 minutes after a high fat meal). In the large phase I study, with oral nilotinib at dosages of 50-1200 mg once daily, as well as 400 or 600 mg twice daily, trough serum concentrations exceeded the IC_{50} for cellular phosphorylation of BCR-ABL. With once daily dosing at steady-state, C_{max} and AUC increased with increasing dose from 50 mg to 400 mg in a generally dose-proportional manner, but appeared to plateau at dose levels starting at 400 mg, remaining relatively constant over the dose range from 400 mg to 1200 mg. Dividing the daily dose in a

twice daily schedule overcame the dose-limiting exposure to some extent with daily steady-state serum exposure to nilotinib with 400 mg twice daily dose being approximately 35% greater than with 800 mg once daily dose. However, there was no further relevant increase in exposure to nilotinib when given 600 mg dose with the twice daily schedule (1200 mg/day). With multiple oral doses of nilotinib, steady-state conditions were achieved by day 8 after initiating nilotinib treatment. There was a 2-fold or 3.8-fold accumulation with once daily dosing or twice daily dosing, respectively, in serum concentrations between the first-dose and steady-state. The median time to reach $_{ma}$ of nilotinib (t_{max}) was 3 hours. Drug elimination half-lives calculated during the dose interval averaged 17 hours for once daily dosing, consistent with the observed accumulation in serum concentrations. Elimination half-lives of the biphasic plasma profile were 1.5 h (83% of AUC) and 116 h for the rat. The binding of nilotinib to plasma proteins was high and there were no species differences. When radiolabeled nilotinib was administered intravenously to rats, drug-related radioactivity was widely distributed to most tissues, consistent with a large volume of distribution. There was a minimal passage for drug-related radioactivity across the blood:brain and blood:testis barriers. Nilotinib was eliminated in the rats mainly via oxidative metabolism. Based on its inhibition (IC_{50} ≤7.5 μM) for CYP enzymes, 2C8, 2C9, 2C19, 2D6, and 3A4/5, nilotinib may inhibit the metabolic clearance of comedications metabolized by these CYP enzymes, if sufficiently high concentrations of nilotinib are achieved *in vivo*. Exposure to nilotinib in female patients was approximately 20% greater than in male patients. There was no difference in nilotinib apparent clearance between Caucasians and non- Caucasians. The population pharmacokinetic analysis showed that nilotinib pharmacokinetics is not affected by age. Pharmacokinetics of nilotinib has not been investigated in subjects with impaired hepatic function. Impaired renal function is not expected to influence nilotinib pharmacokinetics.

Efficacy

Phase I study

A Phase I study was conducted in 119 patients with imatinib resistant CML in BP (n=33), AP (n=56), CP (n=17) and Ph pos ALL (n=13).[39] Patients in this study were treated at once daily dose levels ranging from 50 mg QD to 1200 mg QD, and at twice daily dose levels of 400 mg BID and 600 mg BID. Intrapatient dose escalations were permitted. The 400 mg BID dose level was chosen for further development in Phase II trials, with dose escalation to 600 mg BID permitted for lack of response.

Complete hematologic and cytogenetic response by CML disease classification (n=106) is shown in Table 1. The response in Ph pos ALL patients (n=13) is not shown.

Table 1. Response in Phase I study by disease classification

	CML-CP	*CML-AP*	*CML-AP (clonal evolution)*[1]	*CML-BP (myeloid and lymphoid)*
	N=17 (%)	N=46 (%)	N=10[2] (%)	N=33 (%)
Complete Hematologic Response	11 (92)	28 (61)	5 (100)	2 (8)
Complete Cytogenetic Response	6 (35)	9 (20)	5 (50)	2 (6)

[1]CML-AP (clonal evolution) includes patients whose only AP criterion was clonal evolution; [2]CML-AP clonal evolution group had a total n of 10 patients, but only 5 were evaluable for hematologic response.

Phase II study in late chronic phase

A phase II open-label study (NCT0010 9707) was designed to evaluate the safety and efficacy as defined by hematologic/cytogenetic response rates (HR/CgR) of nilotinib administered at a daily dose of 400 mg BID to imatinib resistant or intolerant CML-CP patients. Daily doses of nilotinib could be escalated to 600 mg BID for patients who did not adequately respond to treatment, and in the absence of safety concerns.

The majority of patients were imatinib resistant (91 patients, 69%) and 41 (31%) were imatinib intolerant. Nearly half (49%) had the disease for ≥5 years. Patients have been treated with nilotinib for a median duration of 226 days (range 3-379 days). Complete hematologic response (CHR) in patients without CHR at baseline was achieved in 69% of patients (60 of 87 patients). Major cytogenetic response was achieved in 42% (55 of 132 patients). A complete cytogenetic response was achieved in 33 (25%) and 22 (17%) were partial cytogenetic responses in this imatinib resistant/intolerant population. The median time to CHR was 1.4 months and MCyR was 2.6 months. A total of 50 (38%) patients have discontinued treatment (26 for adverse events, 15 for disease progression, 7 for other/administrative problems, and 2 for patient deaths (one myocardial infarction in a patient with previous infarctions noted and one with progressive disease). Overall, most frequent Grade 3 or 4 adverse events included thrombocytopenia in 34 patients (26%), neutropenia in 24 (18%), elevated serum lipase in 10 (8%) and anemia in 9 (7%).

Phase II studies in accelerated phase and in blast phase

119 patients with IM-resistant or intolerant Ph pos CML in AP were treated with NIL 400 mg twice daily.[44] A confirmed CHR was obtained in 26% of patients, and a PHR in 21% of patients. The CgR was complete in 16% of pts, partial in 14% of patients, and minor in 13% of pts, for an overall CgR rate of 43%. After 6 months, 40% of pts were still on treatment, while 60% had discontinued, for disease progression (29%), AEs (13%), or other causes (18%).

132 patients with IM-resistant or intolerant Ph pos CML in BP were treated with NIL 400 mg twice daily.[45] A HR was achieved in 39% of patients, and was complete in 25%.

Nilotinib front-line in early CP CML, phase II and III studies

A phase 2 exploratory study of NIL front-line, 400 mg twice daily, is currently running at the MD Anderson Hospital, Houston. Twenty-two patients were treated for at least 3 months and 95% of them achieved a CCgR. Eleven patients were treated for 12 months, and were all in CCgR. Grade 3/4 neutropenia and thrombocytopenia were observed in 7% and 3% of patients. Other grade 3/4 AEs included transitory elevation of lipase (9%), bilirubine (6%), amylase (3%), and infection (3%).

A phase 2 exploratory study of NIL front-line in Ph pos CML has been promoted and sponsored by the GIMEMA CML WP (EUDRACT No. 2007 000597 22). The study has enrolled 73 patients from July 2007 to February 2008. First available results have been presented at 2008 EHA meeting: the complete cytogenetic response and the major molecular response were 84% and 62% at 3 months and 97% and 74% at 6 months, respectively. A phase III study of NIL vs IM in the treatment front-line of Ph pos CML has been promoted and sponsored by Novartis Pharma, and has already enrolled more than 200 patients (Study CAMN107A2303, EUDRACT 2007-000208-34).

Tolerability

Nilotinib non-hematologic adverse events

The most common adverse events reported in the phase I study with nilotinib in patients with ima tinib-resistant CML or Ph+ ALL were mild-to-moderaterashes, transient and clinically insignificant elevations of indirect bilirubin levels, and myelosuppression, which was an important dose-limit- ing adverse event. Grade 3 or 4 neutropenia occurred in 22% of patients treated with nilotinib 600 mg twice daily compared with 9% of those who received 400 mg twice daily. The maximum tolerated dose of nilotinib was 600 mg twice daily and the recommended dose for phase II studies was 400 mg twicedaily, with dose escalation to 600 mg twice daily for patients with an inadequate response. Tolerability data from 316 imatinib-resistant or intolerant patients with chronic-phase CML who received nilotinib 400 mg twice daily, increasing to 600 mg twice daily if necessary. The most frequently reported adverse events (regardless of causality) were rash, nausea, headache, pruritus and fatigue, which were generally mild to moderate severity. The median number of days of exposure to nilotinib was 247 and the medan dose intensity was 797 mg/day. Thrombocytopenia was reported in 58% of patients, including 29% with grade 3 or 4 severity, and led to dosage reduction or interruption in 24% of patients and discontinuation of therapy in 6% of patients. Neutropenia was reported in 50% of of patients, including 28% with grade 3 or 4 severity, and led to dosage modification in 10% of patients and discontinuation of therapy in 6% of patients. Anaemia occurred in half of the 316 patients, including 9% with grade 3 or 4 anaemia. Other frequently reported laboratory abnormalities (incidence >40% for all grades) included abnormal elevations of AST or ALT levels, which led to dosage modification in 2% of patients and discontin uation of therapy in 3%, hyperbilirubinaemia, which led to dosage modification in 3% and discontinuation of therapy in 2% of patients, and lipase elevation or pancreatitis, which led to dosage modification in 4% and discontinuation of therapy in <1% of study patients. An exploratory analysis of ECG data in the phase I trial identified a prolongation of the corrected QT interval as the only abnormality associated with nilotinib; therefore, ECGs were also evaluated in patients with imatinib-resistant or intolerant CML. Less than 1% of patients had a corrected QT interval that was increased from baseline by >500 msec, and 2% of patients had a corrected QT interval that was prolonged by >60 msec; there were no episodes of Torsade de pointes. Fluid retention, oedema and weight gain, which can occur commonly with imatinib, were infrequently reported among nilotinib recipients. In the phase I trial in 119 patients treated with nilotinib, there were no reports of fluid retention, oedema, weight gain or pleural effusion. In the phase II trial in 316 patients, the incidence of peripheral oedema (all grades) was 11% but there were no grade 3 or 4 cases. Pleural effusion, pericardial effusion and pulmonary oedema of any grade were each reported in <1% of patients in the large phase II trial. In general, the tolerability profile of nilotinib, including biochemistry and myelosuppression abnormalities, was similar in the phase II trial in 64 patients with imatinib-resistant or intolerant CML in accelerated phase to that in the large phase II trial. Rash (34% of patients), constipation (28%) and 2% of pruritus (27%) were the most frequently reported adverse events regardless of causality, and grade 3 or 4 adverse events were uncommon. Grade 3 or 4 neutropenia and thrombocytopenia occurred in 45% and 40% of patients, respectively. On the basis of results of the large, dose-escalating phase I trial with nilotinib primarily in patients phase (AP), or blast crisis (BC) chronic

myeloid leukemia with imatinib-resistant CML (section 3), the recommended dose of nilotinib is 400 mg orally twice daily, which can be increased to 600 mg twice daily tinib mesylate resistance in chronic myeloid leukemia (CML) in patients with an inadequate response who do not have any significant tolerability issues.

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