Mechanism of Action of SNS-032, a Novel Cyclin Dependent Kinase Inhibitor, in Chronic Lymphocytic Leukemia: Comparison with Flavopiridol

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Introduction

- Cyclin-dependent kinases (Cdks) not only drive cell cycle progression, but also control transcription. For example, Cdk7 and Cdk9 phosphorylate the Ser2 and Ser5 sites, respectively, on the C-terminal domain (CTD) of RNA polymerase II (pol II) to promote transcription initiation and elongation.
- Our previous experience with flavopiridol, a pan-Cdk inhibitor, in primary chronic lymphocytic leukemia (CLL) cells demonstrated inhibition of RNA pol II-driven mRNA synthesis, reduction of the expression of short lived anti-apoptotic proteins, and induction of apoptosis in CLL cells in vitro (Blood 106:2313-19, 2005).
- Here we studied a novel Cdk inhibitor SNS-032, a 2-aminothiazole derivative with potent and selective inhibitory activity against Cdk2, Cdk7, and Cdk9.

SNS-032

- SNS-032, formerly known as RMS-037032, is currently in a Phase 1 trial in patients with chronic lymphocytic leukemia and multiple myeloma. SNS-032 is administered using a pharmacologically-defined dose regimen, designed to sustain levels above 0.3 μM for over 6 hours, the EC_{50} concentration and time required for in vitro cytostasis.

Hypothesis

Because CLL cells are not actively cycling, and their viability is dependent upon the continuous expression of anti-apoptotic proteins, we hypothesized that SNS-032 would induce apoptosis in CLL cells through transcriptional inhibition of anti-apoptotic proteins.

Results

SNS-032 inhibits the phosphorylation of RNA pol II in CLL cells

- Time course of SNS-032 induced apoptosis
- Inhibition of RNA Pol II by Cdk7 & Cdk9

- SNS-032 potently induces apoptosis in CLL cells
- Recovery of CLL cells after washing into fresh medium without SNS-032

- SNS-032 reduces transcripts of anti-apoptotic proteins in CLL cells

- SNS-032 inhibits RNA synthesis

- SNS-032 induced cell death is not dependent on a functional p53 or ATM

Summary

- SNS-032 inhibits the phosphorylation of Cdk7/Cdk9-substrates Ser5/Ser2 of CTD of RNA Pol II, and reduces RNA synthesis.
- SNS-032 reduces the mRNA and protein levels of short-lived anti-apoptotic proteins Mcl-1 and XIAP.
- SNS-032 induces apoptosis in CLL cells.
- Killing of CLL cells by SNS-032 is maximal between 10 to 12 hr exposure.
- Induction of apoptosis appears independent of p53 or ATM.
- SNS-032 is about 10-50-fold more potent than flavopiridol.