PRECLINICAL EVALUATION OF EZH2 INHIBITORS IN MODELS OF HUMAN SYNOVIAL SARCOMA

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Abstract

Objective: Posttranslational modification of histone proteins and ATP-dependent chromatin remodeling are crucial processes for the regulation of the fidelity of DNA replication and SWI/SNF complexes are often genetically altered in cancer. Importantly, these two complexes normally compete with each other in binding to and affecting chromatin; genetic alterations can create an imbalance in this antagonism. For EZH2 methyltransferase, and rhabdoid tumor models are sensitive to EZH2 small molecule inhibitors. In synovial sarcoma, another INI1-negative tumor type, a recurrent chromosomal translocation fuses the SS18 gene (a subunit of the SWI/SNF chromatin remodeling complex) on chromosome 18 to one of three related genes on the X chromosome, SSX1, SSX2 and rarely SSX4. This results in the expression of an oncogenic SS18-SSX fusion protein that binds to the SWI/SNF complex evicting both the wild-type SS18 and the tumor suppressor INI1, which are subsequently degraded. This results in aberrant gene expression and ultimately the development of cancer.

Methods: Synovial sarcoma cell lines and patient-derived xenograft (PDX) models were evaluated for their sensitivity to EZH2 inhibition in vitro and in vivo. In addition, histone methylation, changes in gene expression and histology endpoints were assessed.

Results: Here we show that EPZ-6438 (E7438), an early clinical-stage, selective and orally bioavailable small-molecule inhibitor of EZH2 enzymatic activity induces anti-proliferative activity in preclinical models of synovial sarcoma both as a single agent and in combination with chemotherapy. The compound induces dose-dependent cell growth inhibition and cell death specifically in SS18-SSX fusion-positive cells in vitro. Treatment of mice bearing either a cell line xenograft or two PDX models leads to dose-dependent tumor growth inhibition with correlative inhibition of tri-methylation levels of the EZH2-specific substrate, lysine 27 on histone H3, except for one SS18-SSX fusion-positive cell line.

Conclusion: These data demonstrate a dependency of SS18-SSX-positive synovial sarcomas on EZH2 enzymatic activity in xenograft models which warrants further investigations in the clinical setting.

Background

| Suggested Functions of PRC2-EZH3 in Synovial Sarcoma and an Antagonistic Interaction Between PRC2 and SWI/SNF |

- Generation of an aberrant SWI/SNF complex (SWI/SNF activity is mandatory for PRC2 activity)
- Antagonism of Polycomb (H3K27me3) target repression
- Generation of an aberrant SWI/SNF complex (innu_3/11 RNAi)
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Conclusions

- EPZ-6438 inhibits cell proliferation in SS18-SSX fusion positive synovial sarcoma cell lines in vitro.
- EPZ-6438 increases expression of genes that are aberrantly suppressed in synovial sarcoma.
- Antitumor activity was observed in 3 out of 4 synovial sarcoma in vivo models, including cell line and patient derived xenografts grown in mice.
- In the Fuji xenograft model EPZ-6438/Doxorubicin combination shows a superior antitumor effect when compared to each monotherapy; the combination dosage amounts need further optimization to improve tolerability.
- Although sensitive to EPZ-6438 in 2D cell culture, HS-SY-II cell line xenografts exhibit dose-related tumor volume increase in a mouse xenograft model, which is being further investigated.
- EPZ-6438 combined with Doxorubicin does not diminish the antitumor effect of Doxorubicin in the HS-SY-II model.
- E7438 (EPZ-6438) has transitioned into clinical development for evaluation in genetically defined cancers.

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