Identification of a First-In-Class PRMT5 Inhibitor with Potent \textit{In Vitro} and \textit{In Vivo} Activity in Preclinical Models of Mantle Cell Lymphoma


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- Stockholder in and Employee of: Epizyme, Inc.
Protein methyltransferase (PMTs) are part of a regulatory system that controls gene expression, called epigenetics.

PMTs regulate gene expression by placing methyl marks on nuclear and cytoplasmic substrates.

Genetic alterations can alter PMT activity making them oncogenic due to misregulated gene expression.

96-member target class, 20 prioritized based on oncogenic mechanism.
PMTs – Equally Divided Between KMTs and RMTs

Arginine Methyl Transferases (RMTs)  Lysine Methyl Transferases (KMTs)

Copeland 2013 Clinical Cancer Research
PMTs as Drivers of Cancer

Arginine Methyl Transferases (RMTs)

PRMT1: AML, Glioblastoma
PRMT7: Breast
PRMT5: Lymphoma
CARM1: Breast, Prostate
NSUN2: Breast

DOT1L: MLL-r
AML, ALL

PRDM14: Breast

Lysine Methyl Transferases (KMTs)

SMYD3: Breast, Liver, Colon, Gastric
SMYD2: Esophageal Squamous

EZH2: NHL, INI1, Breast, Prostate, Colon, Gastric, Bladder, Liver, Melanoma

SUV39H1: Colon
EHM2: Lung, Prostate, HCC
SETDB1: Melanoma

NSD1: AML
WHSC1L1: Breast
WHSC1: Multiple Myeloma

MLL4: Pancreatic, Glioblastoma
MLL: Leukemia

Copeland 2013 Clinical Cancer Research
PRMT5 is a Type II Arginine Methyltransferase

- The mammalian family of Arginine Methyltransferases (RMTs) contains 11 members
- PRMT5 is the pre-dominant Type II RMT that is responsible for the symmetric dimethylation of arginine residues
- PRMT5 has been shown to methylate numerous nuclear and cytoplasmic substrates; some of which are postulated to drive tumorigenesis
- PRMT5 has been shown to be upregulated in several human malignancies including lymphomas
PRMT5 Overexpression in Mantle Cell Lymphoma (MCL)

- PRMT5 Overexpression identified in Mantle Cell Lymphoma (MCL)

- Anti-proliferative effects observed upon PRMT5 KD in Jeko-1, a MCL cell line

- MCL is one of the rarest forms of non-Hodgkin’s lymphomas (NHLs) representing ~6% of NHL cases or ~4000 new cases per year in the United States

- MCL is defined by the t(11;14) translocation resulting in overexpression of cyclin D1

Pal et al. 2007 *EMBO*  
Chung et al. 2013 *JBC*
EPZ015666 – First-in-class PRMT5 Inhibitor

- **Potent** inhibition of PRMT5:MEP50 complex
  - SAM uncompetitive, peptide competitive inhibition

- Highly **selective** vs. other PMTs
  - Biochemical – >20,000-fold by $K_i$
  - Biochemical $K_i$ : 5 nM
  - Cell Biochemical (In-Cell-Western) IC50 : 8 nM

- **Orally bioavailable**

- Potent **methyl mark inhibition** with excellent correlation to killing of cells *in vitro*

- Potent **in vivo efficacy** in animal models of MCL following inhibition of target methyl mark
EPZ015666 Inhibits Symmetric Arginine Di-methylation in a Dose-Dependent Manner

- On target inhibition of EPZ015666 demonstrated by strong correlations between biochemical, cell biochemical, and phenotypic IC50s

Symmetric Di-Methyl Arginine (SDMA) is a pan-dimethyl arginine antibody (motif Ab)
MCL Cell Lines are Sensitive to EPZ015666 Treatment

**Z-138**

**Methylation Day 4 IC$_{50}$ = 44 nM**

<table>
<thead>
<tr>
<th>MCL Cell Line</th>
<th>Day 12 Proliferation IC$_{50}$ (nM)</th>
<th>SDMA Western Blot IC$_{50}$ (nM)</th>
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<tbody>
<tr>
<td>Z-138</td>
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<td>Mino</td>
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<td>Jeko-1</td>
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</table>

Penebre et al. submitted
Z-138 Xenografts Are Highly Sensitive to Orally Dosed EPZ015666

**21-day Efficacy Study**

- Vehicle, 0.5%MC BID
- 25mg/kg BID
- 50mg/kg BID
- 100mg/kg BID
- 200mg/kg BID

Tumor Volumes (mm³)

Days of Treatment

**Target Inhibition in Day 21 Tumors (SDMA ELISA)**

- Z-138

Maver-1

**22-day Efficacy Study**

- Vehicle (0.5% MC) BID
- 25mg/kg BID
- 50mg/kg BID
- 100mg/kg BID
- 200mg/kg BID

Tumor Volumes (mm³)

Days of Treatment

**Target Inhibition in Day 22 Tumors (SDMA ELISA)**

- vehicle
- 25mg/kg BID
- 50mg/kg BID
- 100mg/kg BID
- 200mg/kg BID

- No significant body weight loss observed during the studies
EPZ015666: First RMT Inhibitor Showing *In Vitro* and *In Vivo* Activity in Pre-clinical Models of MCL

- EPZ015666 is a potent, selective and orally bioavailable inhibitor of PRMT5
- EPZ015666 demonstrated potent cellular activity as measured by its ability to block symmetric dimethylation of SmD3 and inhibit proliferation of MCL cell lines
- EPZ015666 displays robust anti-tumor activity as a single agent in MCL xenograft animal models
- Pre-clinical studies of the effects of PRMT5 inhibition in other cancer indications is currently being studied
We would like to thank the principal investigators and their institutions, the employees of Epizyme and GSK.