Characterization of Lysine Methyltransferase Inhibitors

Thomas Clayton, Wei Fang, Jon Cordia, Deborah Mortensen, Philip Chamberlain.
Celgene

Post-translation modification of histones via phosphorylation, acetylation, and methylation play a key role in chromatin remodeling. Taken individually or in concert, these changes can have significant effects on such processes as transcription, replication, DNA repair, and apoptosis (Trievel RC). Histone methyltransferases (HMTs) catalyze the mono, di, or trimethylation of arginine or lysine residues of H3 or H4 tails through their SET domains. Increased cellular levels of certain methyltransferases and the global changes in the particular histone methyl marks associated with those methyltransferase result in a reprogramming to an oncogenic epigenetic landscape. Here we present efforts to develop and characterize SET domain binding inhibitory molecules capable of reversing the oncogenic epigenetic reprogramming. Initial hits were identified using an Alpha screen, and were confirmed with a radioisotope assay. When crystallization trials with Compound A proved fruitless, additional thermal shift assay and dynamic light scattering experiments were performed to characterize the Compound A-SET domain interaction. The observed dose dependent loss of fluorescence in the thermal shift assay and the drastic increase in the hydrodynamic radius of the protein in the presence of compound established aggregation as the mechanism of action for Compound A series compounds. When Compound A series compounds were found to aggregate a spectrum of unrelated proteins, this series was abandoned in favor of alternate approaches.