

Development of KDM5 covalent inhibitors as chemical probes

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Histone lysine demethylase (KDMs) are involved in the dynamic regulation of gene expression by reversible regulation of the methylation levels on lysine residues in histone tails. Among the KDMs, the jumonji (JmjC)-domain-containing KDMs (KDM2-7) are Fe(II), 2-OG (α -ketoglutarate) and molecular oxygen-dependent enzymes that employ an oxygenase mechanism to demethylate specific methylation states at various histone sites. KDMs play a critical role in several biological processes such as cell differentiation, inflammation, cancer progression and resistance. Achieving selectivity over the different families of KDMs has been a major challenge. Here we report potent and selective KDM5 covalent inhibitors designed to target a cysteine residue only present in the KDM5 sub-family. *In vitro* assays show that compounds are selective for the KDM5 sub-family, showing potencies in the low nanomolar range, with higher affinity for KDM5A/B. The covalent binding to the targeted proteins was proved by MS. A kinetic approach was studied in order to describe the components of overall inhibitor potency (reversible binding and chemical reactivity), showing a time-dependent decrease of IC_{50} values for irreversible inhibition. Additional 2-OG competition assays show that compounds were non 2-OG competitive and target engagement assays showed that the compounds inhibited the KDM5 members in cells. Full description of the results will be presented in the meeting.