

ABSTRACT

JHDMS (JmjC-domain-containing histone demethylases) are the largest class of demethylase enzymes, contain a Jumonji C (JmjC) domain and catalyze lysine demethylation of histones through an oxidative reaction that requires Fe(II) ion and α -ketoglutarate (2OG) as cofactors. The misregulation of these enzymes, in particular JMJD2 subfamily, has been significantly implicated in cancer initiation and progression. Potent and specific inhibitors of these enzymes have not been identified yet, most of them inhibiting many other Fe(II)/2OG dependent oxygenases or being affected by undesirable characteristics.

Here, we describe the discovery by high throughput screening (HTS) of a bunch of novel hit compounds active against KDM4s and the subsequent hit validation stage to select the most interesting ones for further derivatization. The use of a multiple combined approach of different in vitro techniques led us to select the hit **EML586** as starting point for the development of novel optimized derivatives.

The substitution of quinoxaline ring with more aliphatic portions gave derivatives such as **EML678** and **EML684**, which demonstrate a better activity against *h*KDM4A compared to the starting hit compound. Furthermore, they induced a marked reduction in methylation of lysines H3K9 and H3K27 in a cell-based assay together with a marked arrest in the S phase of cell cycle.