EPIGENETICS

Disrupting histone lysine methylation

A small molecule targeting the protein-protein interaction between a chromatin binding protein and an oncogenic transcription factor shows therapeutic potential in a subtype of acute myeloid leukemia

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Histone lysine methylation states are regulated by methyltransferases and demethylases. Histone methyltransferase enzymes often function as integral components of multisubunit protein complexes and require the presence of these complex partners to exhibit catalytic activity. WD repeat domain 5 (WDR5) is a component of a number of methyltransferase complexes that specifically methylate histone H3 at lysine 4, a histone modification that is associated with transcriptional activation. WDR5 directly binds SETD1A, SETD1B or one of four MLL homologous methyltransferases[1,2], and this interaction is required for the enzymes to be catalytically competent. Targeting aberrant histone lysine methylation signaling has recently emerged as a promising cancer therapeutic strategy, and rapid progress is being made toward the development of small-molecule inhibitors of these enzymes[3]. In this issue, Grebien *et al.*[4] report on the discovery, characterization and application of OICR-9429, a small molecule that directly binds the chromatin-associated protein WDR5 and disrupts its interaction with partner proteins.

OICR-9429 is an improved, cell-active representative of a chemotype that was previously identified by the same group[5,6]. OICR-9429 competes with the WDR5-interacting (WIN) region of the methyltransferases. Disruption of the protein-protein interaction results in inactivation of the enzyme and perhaps destabilizes the methyltransferase complex. OICR-9429 is highly selective for WDR5, as demonstrated by enzymatic and unbiased chemoproteomic studies. The high-resolution co-crystal structure reveals residues that are critical for compound binding. OICR-9429 binding results in a conformational rearrangement of the WIN-binding pocket of WDR5. In a cellular context, OICR-9429 abrogates the physical interaction between WDR5 and MLL at submicromolar concentrations, making it a suitable probe compound to investigate disease biology.

Disruption of the MLL1-WDR5 interaction has been proposed previously as a strategy to target MLL1 methyltransferase activity and to inhibit the growth of acute myeloid leukemias (AML) that harbor MLL1 translocations[7]. Grebien *et al.* suggest that this strategy may extend to cases of AML that are driven by the oncogenic p30 isoform of the transcription factor CCAAT-enhancer binding protein alpha (C/EBPα), a myeloid master regulator[4]. The *CEBPA* gene, encoding C/EBPα, is recurrently mutated in AML. These mutations frequently lead to the expression of a short translational isoform that is termed p30. To gain insight into the mechanism underlying the p30 pro-oncogenic function in AML, the authors used an elegant proteomics approach. They identified WDR5 among a number of proteins that preferentially interacted with p30 compared to the longer, non-oncogenic p42 isoform of C/EBPα. This finding led to the hypothesis that truncated C/EBPα utilizes the chromatin-associated protein WDR5 and associated histone methyltransferases to modulate H3K4 methylation and to drive oncogenic transcriptional programs. Indeed, depletion of Wdr5, Mll1 or Mll3 by RNAi or pharmacological suppression of Wdr5-protein interactions by OICR-9429 lead to mouse leukemia cell differentiation (**Fig. 1**). Moreover, Wdr5 depletion or OICR-9429 treatment reduced p30-dependent malignant transformation of mouse hematopoietic progenitor cells. Finally, OICR-9429 preferentially caused viability defects in cells derived from patients with AML carrying N-terminal *CEBPA* mutations as compared to AML cells with other oncogenic driver mutations.

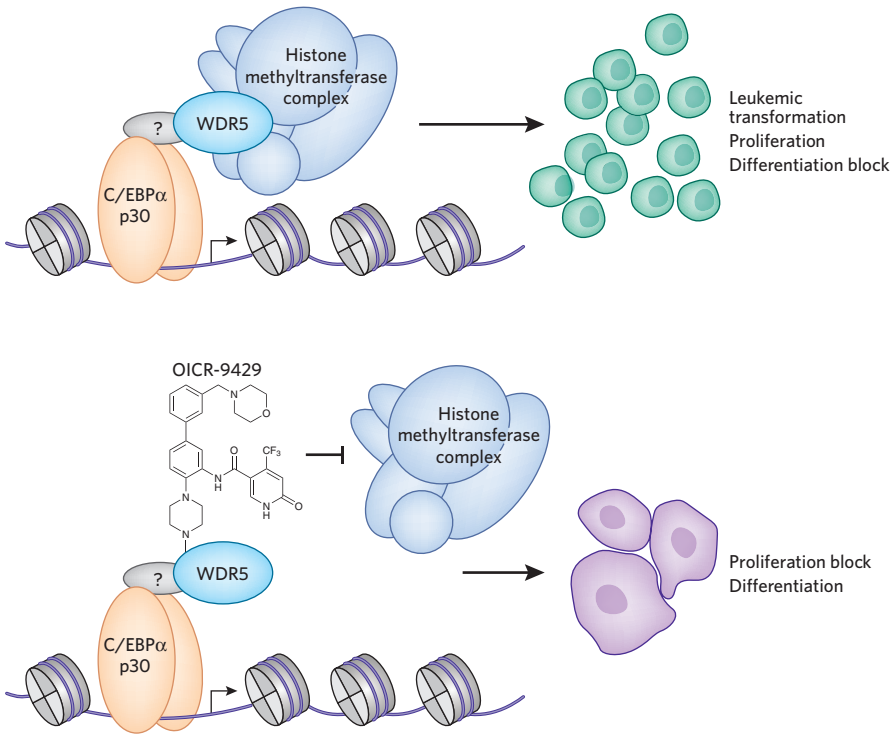

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Figure 1 | The small molecule OICR-9429 disrupts the interaction between the chromatin-binding protein WDR5 and a histone methyltransferase complex that is required for the oncogenic p30 isoform of the transcription factor C/EBPα to suppress differentiation and promote proliferation of leukemic cells. WDR5 indirectly interacts with C/EBPα and the identity of the bridging factors is currently unknown (indicated by '?').

From a drug-discovery perspective, these data are encouraging because OICR-9429 offers an distinct approach to modulating histone lysine methylation, in contrast to small-molecule inhibitors that target the catalytic sites of methyltransferases and demethylases. However, disrupting high-affinity protein-protein interactions is a nontrivial undertaking, and it remains to be seen whether therapeutic effects can be achieved with such a compound *in vivo* at pharmacologically relevant doses. Also, questions regarding safety and therapeutic index remain to be addressed once the chemotype is sufficiently optimized. From an oncology perspective, the data make a compelling case that C/EBPα p30–driven leukemias depend on WDR5. The study opens the door for future investigations aimed at further understanding the molecular basis of WDR5 dependency and determining the scope of WDR5 dependencies in other hematologic malignancies. The interaction between C/EBPα p30 and WDR5 is likely to be indirect (**Fig. 1**), and one may speculate that, depending on context, the interaction may be modulated by these currently unknown bridging factors. Interestingly, OICR-9429 not only off-competes the enzyme but also seems to displace the RBBP5 methyltransferase complex subunit, even though its binding site is located on the opposite side of the compound-binding site. WDR5 also participates in the formation of various mutually exclusive complexes[8] and interacts with other oncogenic transcription factors such as MYC[9] via binding sites that are identical with those recognizing methyltransferase complex components. OICR-9429 potentially ablates these WDR5 protein interactions as well, which complicates interpretation of observed phenotypic effects.

One can now begin to compare this approach with other chromatin-targeted therapies in leukemia, such as inhibitors targeting the histone methyltransferase DOT1L, the MLL-associated protein MENIN and BET bromodomain-containing proteins. All of these have shown efficacy in the context of MLL-translocated leukemias. It will be exciting to explore, for available tool compounds such as OICR-9429, whether they offer any advantage over other modalities or can be successfully combined with any other modalities to increase their efficacy.

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Competing financial interests

The author declares no competing financial interests.