Epigenetic therapy reprograms hereditary disease

José M. Bautista  COMPLUTENSE UNIVERSITY OF MADRID

In this issue of Blood, Makarona et al demonstrate that histone deacetylase (HDAC) inhibitors (HDACis) in glucose-6-phosphate dehydrogenase (G6PD)-deficient cells reinstates enzyme activity by boosting gene transcription.1 This therapeutic approach opens new avenues for preclinical and clinical studies to treat not only chronic nonspherocytic hemolytic anemia caused by severe G6PD variants,2 but also other genetic diseases.

Epigenetic mechanisms control gene expression in different cell types without changing the genome sequence. These mechanisms include histone modification, DNA methylation, and small regulatory RNAs. Specific targeting of these elements transforms the packaging of nuclear DNA and its expression. Chemical groups including acetyl, methyl, and phosphate can modify histones at particular amino acids, which modify protein–protein and protein–DNA interactions, and consequently the cell phenotype. Two types of nuclear enzymes with several families, classes and members, histone acetyltransferases (HATs) and HDACs, play key roles in mastering the acetylation pattern of the local chromatin structure through the next cell generations. In this way, acetylation patterns are transmitted and interconnected with protein interactions in the chromatin to define specific cell types and functions. In this context, the abundance of elements controlling gene transcription makes the research on epigenetics appealing because it is likely to contribute with new insights and targets for treatment. Rather than targeting the altered cell or its outcome, epigenetic therapy endeavors to reprogram the behavior of the dysfunctional cell to normality by turning genes on or off.

Erythrocyte G6PD deficiency is a common X-linked hereditary disorder mostly caused by missense mutations at the housekeeping G6PD gene. Overall, more than 160 different mutations have been described. Hemizygous (males) and homozygous individuals with deficient alleles are susceptible to hemolytic anemia. The spatial position of the mutation in the enzyme determines the severity of the disease, from mild/acute forms—triggered by drugs, fava beans, or infections—to severe/chronic forms requiring transfusions.3 Most G6PD mutations cause dysfunction by decreasing protein stability and never entirely abolishing enzyme activity, because its complete loss is lethal.3 Sporadic de novo mutations are rare but very often cause severe chronic nonspherocytic hemolytic anemia. On the other hand, a high prevalence of G6PD deficiency (>1%) is found in populations where malaria is or was endemic because of the selective advantage of these mild G6PD-deficient alleles against severe malaria. Nevertheless, in malaria-endemic countries, the large percentage of G6PD-deficient individuals impedes the radical use of some antimalarials, such as primaquine,3 and requires additional medical interventions to manage the adverse hemolytic toxicity induced by drug eradication strategies.6

Makarona and colleagues provide new insights into the selectiveness and mechanism underlying transcriptional upregulation of G6PD in response to HDACi. Within a background of 3% up- or downregulated genes by HDACi (in more than 1000 genes), from the 16 genes analyzed in the glycolysis and pentose phosphate shunt that are commonly involved in inherited erythroenzymopathies, only G6PD messenger RNA levels increased upon HDACi treatment. The researchers, using neat in vitro experiments with human B cells and erythroid precursor cells, expand the present knowledge to describe the chain of consecutive events that enhance specific transcription of G6PD (see figure). HDACi induces histone hyperacetylation that displaces the HAT/HDAC equilibrium at the G6PD promoter by increasing the enrollment of different acetylation/deacetylation enzymes from several families. This results in 1 particular transcription factor (Sp1) binding to specific G6PD promoter motifs and mediating transcription, boosting the core promoter through the RNA polymerase II machinery. Promoter-associated RNA polymerase II regulates transcription through collective positioning of participating components at the core promoter,7 including phosphorylation.8 Thus, variable DNA elements within the core promoter of different genes direct the efficiency of their transcription initiation; consequently, not all genes are expected to behave equally to changes in promoter accessibility by epigenetic therapy. In fact, the authors show that G6PD selectively upregulated by HDACi is in contrast with most other genes that remain unaltered. This is explained as a consequence of the equilibrium dynamics of both HATs and HDACs bound on the chromatin of active genes with acetylated histones8 to further be able to hyperacetylate them by the direct effect of HDACi on HDAC. Besides, the treatment by HDACi does not influence the transcription of silent genes because they are not bound by HDACs. Thus, the authors discovered a prospective general use of HDACi on active housekeeping genes. Additionally, in their gene expression profiling, the authors identify 65 other genes responsible for inherited diseases, upregulated by HDACi sodium butyrate.

Taking into account the various elements revealed by Makarona et al in the upregulation of G6PD by HDACi, it can be anticipated that a variety of strategies for specific epigenetic reprogramming based on combinatorial chromatin changes in the methyl, acetyl, and phosphate groups as well as on the abundance of the various binders in the core promoter. In addition, strategies to fine-tune gene expression by class isoform–specific HDAC inhibitors that could function on protein–protein interactions within the multiprotein transcriptional complexes could also expand the therapeutic benefit of epigenetic intervention.

The investigators went further to test HDACi treatments on nucleated cells carrying different G6PD alleles, including severe and mild variants, to demonstrate that HDACis correct deficient phenotypes in primary
erythroid precursor cells. HDACi-upregulation of mutated G6PD-messenger RNAs favors the assembly of an increased number of G6PD monomers into more stable dimers and tetramers to overall increase the enzymatic activity just above normal levels. Apart from the direct potential therapeutics of G6PD deficiency, these last achievements are important for 3 other reasons. First, the findings consolidate previous data in other hematological hereditary diseases that reprogramming gene expression by chromatin modifiers rectifies the associated pathology.9,10 Second, these studies can provide a new strategy for boosting G6PD activity in red cells of those carriers of polymorphic G6PD variants that cannot be administered with effective antimalarials, such as primaquine, for the elimination of *Plasmodium vivax* and *Plasmodium ovale* hypnozoites causing relapse.6 Finally, these drug-reprogramming approaches of damaged cells carrying disease alleles can pave the way to other therapeutic scenarios for hereditary diseases different than those based on engineering gene transfers.

Conflict-of-interest disclosure: The author declares no competing financial interests.

REFERENCES


Epigenetic therapy reprograms hereditary disease

José M. Bautista