DNMT1 Stability Is Regulated by Proteins Coordinating Deubiquitination and Acetylation-Driven Ubiquitination

Zhanwen Du1,2, Jing Song3*, Yong Wang1,2, Yiqing Zhao1,2, Kishore Guda2,4,5, Shuming Yang6, Hung-Ying Kao7, Yan Xu6,8, Joseph Willis2,9, Sanford D. Markowitz2,4,5, David Sedwick2,4, Robert M. Ewing1,3, and Zhenghe Wang1,2,10†

1 Department of Genetics, Case Western Reserve University, Cleveland, OH 44106, USA.
2 Case Comprehensive Cancer Center, Case Western Reserve University, Cleveland, OH 44106, USA.
3 Case Center for Proteomics and Bioinformatics, Case Western Reserve University, Cleveland, OH 44106, USA.
4 Department of Medicine, Case Western Reserve University, Cleveland, OH 44106, USA.
5 Howard Hughes Medical Institute, Cleveland, OH 44106, USA.
6 Cancer Pharmacology Core Facility, Case Western Reserve University, Cleveland, OH 44106, USA.
7 Department of Biochemistry, Case Western Reserve University, Cleveland, OH 44106, USA.
8 Department of Chemistry, Cleveland State University, Cleveland, OH 44115, USA.
9 Department of Pathology, Case Western Reserve University, Cleveland, OH 44106, USA.
10 Genomic Medicine Institute, Cleveland Clinic Foundation, Cleveland, OH 44195, USA.

* These authors contributed equally to this work.

Abstract: DNA methyltransferase 1 (DNMT1) is the primary enzyme that maintains DNA methylation. We describe a previously unknown mode of regulation of DNMT1 protein stability through the coordinated action of an array of DNMT1-associated proteins. DNMT1 was destabilized by acetylation by the acetyltransferase Tip60, which triggered ubiquitination by the E3 ligase UHRF1, thereby targeting DNMT1 for proteasomal degradation. In contrast, DNMT1 was stabilized by histone deacetylase 1 (HDAC1) and the deubiquitinase HAUSP (herpes virus–associated ubiquitin-specific protease). Analysis of the abundance of DNMT1 and Tip60, as well as the association between HAUSP and DNMT1, suggested that during the cell cycle the initiation of DNMT1 degradation was coordinated with the end of DNA replication and the need for DNMT activity. In human colon cancers, the abundance of DNMT1 correlated with that of HAUSP. HAUSP knockdown rendered colon cancer cells more sensitive to killing by HDAC inhibitors both in tissue culture and in tumor xenograft models. Thus, these studies provide a mechanism-based rationale for the development of HDAC and HAUSP inhibitors for combined use in cancer therapy.

† To whom correspondence should be addressed. E-mail: zhenghe.wang@case.edu