

Degron-induced inactivation of H2A.Z isoforms in mammalian cells

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The H2A.Z histone variant participates in gene expression control and genetic stability. In mammals, it is encoded by two genes which produce two highly related isoforms (called H2A.Z.1 and H2A.Z.2) differing at three positions. The respective roles of these two isoforms in genome transactions are still unclear.

By genome editing, we generated cell lines derived from U2OS osteosarcoma cells in which these two isoforms are tagged individually or in combination with the FKBP degron. Addition of dTag molecule leads to the rapid and efficient degradation of FKBP-tagged H2A.Z isoforms. We found that the inactivation of either isoform alone does not change cell cycle distribution neither cell viability. However, clonogenic assays show that depletion of both isoforms together results in a strong decrease in the number of clones, indicating that H2A.Z expression is required for cell viability and that the two isoforms are redundant in this respect.

In order to identify in an unbiased fashion the cellular processes in which H2A.Z isoforms are involved, we performed screenings with a human whole genome CRISPR/Cas9 library to identify genes synthetic lethal with H2A.Z.1 or H2A.Z.2 depletion. Gene ontology analyses of hits obtained with both proteins uncover an enrichment in genes linked to RNA processing, including premRNA processing.

In conclusion, we have constructed valuable tools facilitating the analysis of H2A.Z isoform functions. Our first results uncover a genetic link between H2A.Z and premRNA processing. At the meeting I will also present ongoing analyses of genome expression data following the rapid depletion of H2A.Z isoforms alone or in combination.