Nature Reviews Molecular Cell Biology | AOP, published online 19 September 2007; doi:10.1038/nrm2269

DOI:
10.1038/nrm2269

POST-TRANSLATIONAL MODIFICATION

Mimicking the unknown

Di- and trimethylation of Lys9 on histone H3 (H3K9me2/me3) creates a binding site for chromodomain-containing proteins that belong to the heterochromatin protein-1 (HP1) family. This methylation-dependent recruitment of HP1 is thought to induce changes in chromatin structure that result in gene repression. HP1 recruitment can be antagonized by phosphorylation of Ser10 on H3, which overrides the recruitment mediated by H3K9 methylation. This example demonstrates the intricacies of combinatorial post-translational modifications (PTMs) of histones that regulate protein recruitment, chromatin structure and gene expression. Srihari Sampath, Alexander Tarakhovsky and colleagues report evidence indicating that this 'histone code', which is not fully understood, might also be applicable to non-histone proteins.

The authors showed that G9A, an H3K9 methyltransferase, contains a conserved core motif that resembles the sequence surrounding Lys9 of H3 that is recognized by HP1. Consistently, they showed that Lys165 of G9A is di- and trimethylated *in vitro* and *in vivo* by automethylation. Previously, it has been shown that the HP1γ isoform interacts with G9A, and the authors investigated whether this interaction depended on G9A-K165 automethylation. G9A-K165me2 peptides that encompassed the conserved core motif interacted with endogenous HP1γ from 293T cells, unlike unmethylated and G9A-K165A peptides. Similarly, expression of G9A-K165A in 293T cells abolished the interaction with HP1γ, indicating that methylation of Lys165 is necessary and sufficient to mediate the interaction of HP1 with G9A.

Using nuclear magnetic resonance (NMR) analysis, the authors investigated the HP1γ–G9A interaction further and showed that the chromodomain of HP1γ directly interacts with di- and trimethylated Lys165 of G9A. Furthermore, they showed that the NMR profiles resulting from this interaction were similar to those exhibited by H3K9me3 peptides with HP1γ and that the affinity of the HP1γ chromodomain for methylated Lys on either protein was comparable. To further examine the similarities between the effects of H3K9 methylation with those of G9A-K165 methylation, the authors analysed the consequences of G9A-T166 phosphorylation. Residue Thr166 is part of the conserved core motif of G9A that interacts with the chromodomain of HP1γ. The authors showed that expression of a phosphomimetic G9A-T166E mutant abolished the methylation-dependent interaction with HP1γ and that this effect was independent of, and therefore dominant over, K165 methylation, analogous to the effect of Ser10 phosphorylation of H3 on HP1 recruitment.

So, what is the biological significance of this PTM-dependent interaction between non-histone proteins? The expression of a G9A mutant that has no methyltransferase activity in mouse embryonic fibroblasts caused global gene expression changes owing to the abrogated recruitment of HP1 by H3K9 methylation. However, expression of the G9A-K165A mutant did not reproduce this effect, possibly because of the combinatorial aspect of such PTMs or functional redundancy with the related protein GLP (G9A-like protein), leaving this question unanswered. Nevertheless, accumulating evidence suggests that histone-code PTMs could be relevant to non-histone proteins that contain similar PTM-dependent binding motifs to those found in histones — this phenomenon is termed histone mimicry. In addition, studies of these PTM-dependent protein–protein interactions in non-histone proteins could shed light on the elusive histone code.

Gemma K. Alderton

ORIGINAL RESEARCH PAPER Sampath, S. C. *et al.* Methylation of a histone mimic within the histone methyltransferase G9a regulates protein complex assembly. *Mol. Cell* **27**, 596–608 (2007)