Roles of H3K36-specific histone methyltransferases in transcription: antagonizing silencing and safeguarding transcription fidelity

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Abstract Histone H3K36 methylation is well-known for its role in active transcription. In Saccharomyces cerevisiae, H3K36 methylation is mediated solely by SET2 during transcription elongation. In metazoans, multiple H3K36-specific methyltransferases exist and contribute to distinct biochemical activities and subsequent functions. In this review, we focus on the H3K36-specific histone methyltransferases in metazoans, and discuss their enzymatic activity regulation and their roles in antagonizing Polycomb silencing and safeguarding transcription fidelity.

Keywords H3K36 methylation, Histone methyltransferase, SETD2, Ash1L, NSD

INTRODUCTION

Chromatin features, including DNA modifications, histone modifications, histone variants, nucleosome occupation, and chromatin organization, regulate the regional accessibility of chromatin and thus modulate various chromatin-based biological processes, including replication, transcription, and repair. Histone acetylation and methylation at lysine residues are two of the most studied histone modifications, and they have interesting differences. Histone acetylation generally promotes active transcription by altering the positive charge at the lysine residues and the interactions between DNA and histone tails. Therefore, with few exceptions, the majority of histone acetyltransferases and deacetylases display broad substrate specificity and function for multiple lysine residues on various histones (Kouzarides 2007b). In contrast, histone lysine methylation does not change the charge at the histone tails and these methylated lysine moieties function by recruiting downstream reader proteins that are involved in gene activation or repression. The reader proteins generally display site-specificity due to the recognition of neighboring residues. This is probably the reason that histone methyltransferases and demethylases have co-evolved to have specific lysine site preferences (Kouzarides 2007a).

Histone H3K36 methylation is a hallmark of active transcription. Pioneer studies on budding yeast SET2, the first H3K36 methyltransferase (Strahl et al. 2002), have established a paradigm for the recruitment of SET2 and the function of H3K36 methylation: SET2 is recruited by Ser-2-phosphorylated Pol II during elongation to deposit H3K36me3, which functions as a docking site for the Rpd3S histone deacetylase complex to suppress cryptic transcription initiation (Venkatesh and Workman 2013). In metazoans, the characterization of multiple H3K36-specific methyltransferases has expanded the function of H3K36 methylation from transcription elongation to developmental gene regulation (Wagner and Carpenter 2012). In metazoans, SETD2 (also known as HYPB) is the sole enzyme responsible for H3K36me3 (Edmunds et al. 2008; Yuan...
enzymeres the main contributors of H3K36me2, and enzyme responsible for H3K36me3, NSD family adopted the common belief that SETD2 is the sole specifically dimethylases, displayed quite a number of dis-
tion. Notably, H3K36 methylation also participates in various developmental diseases (Bennett et al. 2011; Li 2013; McDaniel and Strahl 2006; Popovic et al. 2012; Wagner and Carpenter 2012). Moreover, the recent discoveries that ‘‘oncohistones’’ with H3K36M/I mutations can drive chondroblastoma tumorigenesis by inhibiting H3K36 methyltransferases and reprogramming the H3K36 methylation landscape further underscore the functional significance of H3K36 methylation (Fang et al. 2016; Lu et al. 2016). In vitro, H3K36 methylation directly inhibits PRC2, which catalyzes repressive H3K27 methylation (Schmitges et al. 2011; Yuan et al. 2011). In vivo, the genomic landscape of H3K36 and H3K27 methylation are anti-correlated (Gaydos et al. 2012; Lu et al. 2016; Papp and Muller 2006; Popovic et al. 2014; Yuan et al. 2011). These findings clearly underscore the role of H3K36 methylation in antagonizing Polycomb silencing. On the other hand, the distinct regulation of each H3K36-specific methyltransferase remains unclear. Here, we summarize the recent progress regarding the regulation of H3K36 methyltransferase activity and their roles in transcription. Notably, H3K36 methylation also participates in other aspects of chromatin events such as DNA repair and mRNA splicing, discussion of which is beyond the scope of this review, but has been reviewed by others (Fahey and Davis 2017; Li 2013; McDaniel and Strahl 2017; Wagner and Carpenter 2012).

AUTO-INHIBITION IS A CONSERVED REGULATORY MECHANISM OF H3K36 METHYLTRANSFERASES

Studies reporting the characterization of the substrate specificity of H3K36-specific methyltransferases, especially dimethylases, displayed quite a number of disputes. After years of study, chromatin researchers have adopted the common belief that SETD2 is the sole enzyme responsible for H3K36me3, NSD family enzymes are the main contributors of H3K36me2, and Ash1/Ash1L is an enzyme governing H3K36me2 at specific regions (An et al. 2011; Dorighi and Tamkun 2013; Edmunds et al. 2008; Huang et al. 2017; Kuo et al. 2011; Li et al. 2009b; Miyazaki et al. 2013; Qiao et al. 2011; Streubel et al. 2018; Tanaka et al. 2007; Yuan et al. 2009, 2011). One likely explanation for the initial conflicting observations is that most H3K36-specific methyltransferases are nucleosome-specific enzymes; these methyltransferases are highly specific for H3K36 methylation at nucleosome substrates, but they display weak non-specific activities for non-nucleosomal histones (An et al. 2011; Byrd and Shearn 2003; Gregory et al. 2007; Li et al. 2009b; Tanaka et al. 2007; Yuan et al. 2009). The structural basis for the nucleosome-specific activities of H3K36-specific methyltransferases has not yet been resolved, and the exact molecular mechanisms of how nucleosomes confine the specificity and stimulate the catalytic activity of H3K36-specific methyltransferases remain unclear. Nevertheless, the structures of the catalytic domains of all three sub-types of H3K36 methyltransferases have been resolved, and interestingly, all of them share a conserved auto-inhibitory mechanism (Fig. 1) (An et al.
A loop at the post-SET region occupies the binding channel for the histone H3 tail, thus blocking the access of lysine 36 to the catalytic center. Obviously, this loop must undergo a conformational change to remove this steric hindrance upon activation. Indeed, a half-opened conformation of the inhibitory loop was observed in SETD2, indicating the dynamic nature of this loop (Yang et al. 2016). Furthermore, when engaged with a K36M-mutated H3 peptide, which mimics the methylated product but cannot be released from the catalytic center, a fully opened state was observed (Fig. 1D) (Yang et al. 2016). In addition, biochemical studies suggest that nucleosomal DNA may act as an allosteric effector for NSD proteins (Li et al. 2009b), and computational docking and simulation suggest that the inhibitory loop of NSD1 may come into contact with the DNA and lead to lysine binding channel widening (Qiao et al. 2011). Therefore, it is reasonable to speculate that the engagement of nucleosome substrates with H3K36-specific methyltransferases may alter the auto-inhibitory loop to a conformation that favors H3K36 methylation catalysis.

Auto-inhibition is also an intrinsic characteristic of E(z), the catalytic subunit of the H3K27 methyltransferase PRC2. While E(z) alone is inactive (Antonysamy et al. 2013; Wu et al. 2013), engagement with two other core subunits of PRC2—EED and SUZ12—alters the configuration of its catalytic center and transforms it into an active conformation (Brooun et al. 2016; Jiao and Liu 2015; Justin et al. 2016). Therefore, another interesting speculation is that certain interaction partner(s) may induce a conformational change and activate H3K36-specific methyltransferases. SETD2 and Ash1/Ash1L have stable interaction partners (Huang et al. 2017; Schmahling et al. 2018; Yuan et al. 2009). In human cells, SETD2 interacts stably with HnRNP-L, which facilitates H3K36me3 deposition in vivo. However, HnRNP-L does not stimulate the enzymatic activity of SETD2 in vitro, which suggests that HnRNP-L does not function as a catalytic activator of SETD2 (Yuan et al. 2009). Recently, we and others demonstrated that Drosophila Ash1 and human Ash1L form stable complexes with Mrg15 (or human Morf4L1/2) and Nurf55 (or human RbAp46/48) (Huang et al. 2017; Schmahling et al. 2018). Interestingly, MRG domain-containing proteins, including Mrg15 and human Morf4L1/2, stimulate the catalytic activity of Ash1/Ash1L significantly. It will be interesting to determine whether Mrg15 activates Ash1 by inducing a conformational change that eliminates the blockage of the catalytic center by the auto-inhibitory loop of Ash1. Although the exact mechanism of induction remains to be determined, this is the first case of allosteric activation directed by an interaction partner among H3K36-specific methyltransferases.

In addition to the positive regulation of catalytic activities, all three sub-types of mammalian H3K36-specific methyltransferases are inhibited directly by H2A ubiquitination (Yuan et al. 2013). Whether such negative regulation involves the stabilization of the auto-inhibitory loop is an interesting question for future exploration.

The physiological roles of this auto-inhibition remain unclear, but this process has been proposed to protect enzymes from hyperactivation (Wang et al. 2015). An intriguing hypothesis is that auto-inhibition and its bypass are ideal interfaces for additional regulation, such as signaling events. Thus, the catalytic process of H3K36-specific methyltransferases is subject to intricate regulations directed by both intrinsic and external mechanisms.

**Diversified Chromatin Recruitment of H3K36-Specific Methyltransferases Assigns Distinct Physiological Function to H3K36 Methylation**

In Saccharomyces cerevisiae, SET2 generates all forms of H3K36 methylation, with trimethylation being the primary effective mark; however, H3K36me2 can work as efficiently as H3K36me3 in recruiting Rpd3S and suppressing cryptic initiation (Li et al. 2009a), suggesting indiscriminate roles of di- and trimethylation under this context. In metazoans, ChIP-sequencing showed that H3K36me2 demarcates chromatin differently from H3K36me3: within genic regions, H3K36me2 is preferentially enriched proximal to the transcription start sites and gradually decays downstream into the H3K36me3-enriched 3’ region; in addition, the large amount of H3K36me2 spread across intergenic regions implies a role at distal regulatory elements (Kuo et al. 2011). Although SETD2 preserves the capacity to generate all three states of methylation, SETD2 depletion affects only H3K36me3, not H3K36me1/2, at the bulk level (Edmunds et al. 2008; Yuan et al. 2009), indicating the specific assignment of trimethylation for transcription elongation. Moreover, the evolution of several methyltransferases specific for H3K36me2 and the fact that mutations and deregulation of these enzymes cause varied developmental diseases further suggest that H3K36me2 may have a function distinct from that of H3K36me3. In convergence, these di-methyltransferases are recruited to chromatin differently from SETD2 (see discussion below), which expands the H3K36 methylation territory and diversifies the function of H3K36.
methylation. H3K36 also exists in mono-methylated state. However, as an intermediate product of methylation reactions catalyzed by the aforesaid di- and tri-methyltransferases, the chromatin distribution and functional role of H3K36me1 remain poorly characterized.

**SET2/SETD2 and H3K36me3: recruited by elongating Pol II to safeguard transcription fidelity**

As mentioned, in *Saccharomyces cerevisiae*, SET2 is recruited by the Ser-2-phosphorylated C-terminal domain (CTD) of elongating RNA polymerase II, and it deposits H3K36 methylation at the gene bodies of active genes (Krogan et al. 2003; Li et al. 2002, 2003; Xiao et al. 2003). Successive transcription may cause histone hyperacetylation in gene bodies, which allow cryptic transcription initiation. To prevent such deleterious events, transcription elongation-coupled H3K36 methylation serves as a docking site for the histone deacetylase complex Rpd3S, which restores the repressive chromatin environment following Pol II passage to prevent cryptic transcription initiation (Carrozza et al. 2005; Joshi and Struhl 2005; Keogh et al. 2005; Li et al. 2007).

Transcription elongation-coupled SETD2 recruitment and H3K36me3 deposition are conserved in mammals, as well as the role of H3K36me3 to prevent aberrant transcription initiation; however, whether the repressive environment promoted by H3K36me3 depends on histone deacetylases remains unexplored. Intriguingly, in mammals, the PWWP domain containing de novo DNA methyltransferases DNMT3A/B is recruited by H3K36me3 to methylate intragenic DNA, which may, in turn, recruit methylated DNA-binding proteins and histone deacetylases (Jones 2012; Neri et al. 2017), to safeguard transcription initiation. Indeed, mouse ES cells lacking DNA methylation exhibited intragenic transcription initiation (Neri et al. 2017).

Overall, SETD2 deposits H3K36me3 in gene bodies to recruit downstream machineries to restore the non-permissive chromatin state following Pol II passage and to maintain transcription fidelity at the genome level.

**H3K36me2 methyltransferases and H3K36me2: demarcating active chromatin by antagonizing silencing**

Loss-of-function studies in multiple species, including *C. elegans*, *Drosophila*, and mammalian cells, indicate that NSD family proteins are responsible for bulk chromatin H3K36me2 levels (Bell et al. 2007; Bender et al. 2006; Kuo et al. 2011), implying the widespread distribution of these enzymes.

In *C. elegans*, MES-4, a homolog of mammalian NSD proteins, is vital for germ cell viability (Bender et al. 2006) and is highly abundant in H3K36me2-enriched autosomes, but not in the H3K27me3-enriched X chromosome in germline cells (Bender et al. 2006; Fong et al. 2002). Moreover, an MES-4 ChIP-chip analysis of early embryos revealed that MES-4 is distributed around the gene bodies of approximately 20% of genes, among which germline-specific genes are highly enriched (Rechtsteiner et al. 2010). MES-4 signals arise near TSS regions, peak proximally, and gradually decrease towards the 3’ end of gene bodies, correlating well with the pattern of H3K36me2 at genomic regions (Rechtsteiner et al. 2010).

In *Drosophila*, MES-4 is also enriched at the 5’ end of target genes (Bell et al. 2007). Importantly, dMES-4 is recruited by an insulator-binding protein to promote the transcription of flanking genes by antagonizing the spread of H3K27 methylation from nearby regions (Lhoumaud et al. 2014); this finding revealed the functional role of the MES-4/NSD family of enzymes at the intergenic cis-regulatory regions.

In mammals, there are three NSD family proteins, and their chromatin localization has not been thoroughly analyzed. Knocking down NSD1 in ESC cells reduces H3K36me2 levels throughout the genome—at gene promoters, gene bodies, and intergenic regions (Streubel et al. 2018). NSD2 localizes to active transcripts, with a greater preference for elongating regions and distal regulatory regions (Ram et al. 2011). Consistently, in multiple myeloma, t(4;14) chromosomal translocation resulting in NSD2 overexpression led to the aberrant accumulation of H3K36me2 at both the intragenic and intergenic regions, supporting the widespread targeting of NSD2 (Kuo et al. 2011; Popovic et al. 2014). The distribution of full-length NSD3 has not been reported, but a short isoform of NSD3 possessing the PWWP domain localizes preferentially to enhancers and promoters (Shen et al. 2015). Importantly, in chondroblastomas, recurrent H3K36M mutations reprogrammed the transcriptome through inhibiting and sequestering H3K36 methyltransferases, resulting in a global reduction in H3K36 methylation; in this process, intragenic and intergenic H3K36me2 were mediated largely by NSD proteins (Fang et al. 2016; Lu et al. 2016). These findings further underscore the physiologic significance of the broad targeting of NSD proteins.

NSD proteins can interact with nuclear receptors, suggesting their recruitment by transcription factors (Huang et al. 1998); however, the general targeting mechanism remains unknown, especially for the intergenic regions. Notably, NSD proteins contain multiple
chromatin reader modules, including the PWWP and PHD domains, which may contribute to the spread of NSD proteins (He et al. 2013; Sankaran et al. 2016). Overall, the NSD family of proteins targets numerous genes involved in many development pathways and extensive intergenic regions.

Different from the MES-4/NSD family of enzymes, the other H3K36me2-specific methyltransferase Ash1 functions as a trithorax protein in Drosophila to maintain the expression of a small collection of developmental genes, the HOX genes. ChIP analysis showed that Ash1 is distributed throughout its target genes (Huang et al. 2017; Schwartz et al. 2010). A subset of cis-regulatory elements in the Drosophila genome that can recruit Trithorax/Polycomb group proteins was identified and defined as Trithorax/Polycomb response elements (TRE/PRE) (Ringrose and Paro 2007). Transgenic TRE/PREs can recruit Trithorax/Polycomb proteins ectopically to maintain the active/repressive states of reporter genes, underscoring their significance in Trithorax/Polycomb recruitment. Importantly, the sequences of TRE/PREs are the same in different cell types, but Trithorax and Polycomb proteins, including Ash1, have different locations in each cell type, suggesting additional regulators beyond DNA sequences. Moreover, Ash1 cooperates with other Trithorax group members to maintain Hox gene expression, among which Trithorax and Kismet may directly promote the chromatin recruitment of Ash1: Ash1 and the N-terminus of Trithorax display an interdependency on chromatin localization (Schwartz et al. 2010); knocking down kismet, a CHD family chromatin remodeler, greatly reduces the chromatin retention of Ash1 (Srinivasan et al. 2008), suggesting that chromatin accessibility directed by a chromatin remodeler also affects Ash1 recruitment. Given that the Ash1 protein also contains multiple chromatin binding domains, including Bromo, BAH, and PHD, and that its partner protein Mrg15 also contains a Chromo domain that recognizes H3K36 methylation, it is natural to expect that the recognition of a combination of histone modifications will contribute another regulatory layer to Ash1 recruitment. Taken together, DNA elements, transcription factors, and the chromatin environment may function coordinate to shape the binding profile of Ash1. The recruitment of Ash1L in mammalian systems is not well studied, and PREs/TREs are not defined in mammals. Despite this, Ash1L also regulates the HOX genes in mammals, indicating the conservation of the recruitment and function of Ash1L (Miyazaki et al. 2013).

Overall, NSD proteins and Ash1L are associated with active transcription, the malfunction of which leads to gene inactivation. Mechanistically, H3K36me2/3 inhibits the catalytic activity of PRC2 (Schmitges et al. 2011; Yuan et al. 2011), and the mutually exclusive distribution of H3K36 methylation and H3K27 methylation along chromatin has been observed in many biological systems (Gaydos et al. 2012; Lu et al. 2016; Papp and Muller 2006; Popovic et al. 2014; Yuan et al. 2011). While the anti-silencing mechanisms of NSD proteins and Ash1L are similar, it would be of great interest to uncover their distinct recruitment mechanisms, which will help in the understanding of the biological impact of these distinct enzymes.

**SUMMARY AND PERSPECTIVES**

In higher eukaryotes, in addition to its conservative role in transcription elongation, H3K36 methylation has an additional function: anti-silencing. In addition to targeting certain developmental genes, PRC2-mediated transcription silencing through H3K27 methylation seems to establish and maintain a default repressive state for most of the inert genome. PRC2 may be recruited initially by cis-elements and then propagate along chromatin through a reinforced spreading mechanism (Wang et al. 2018). On the other hand, active genes and regulatory regions have adapted multiple mechanisms, including installation of H3K4 and H3K36 methylation, as well as an open chromatin status, which are all repulsive substrates for PRC2 catalysis, to overcome the silencing effect (Schmitges et al. 2011; Yuan et al. 2011, 2012). Both H3K36me2 and H3K36me3 can inhibit PRC2 activity efficiently *in vitro* (Schmitges et al. 2011; Yuan et al. 2011). However, they may antagonize PRC2 in different ways *in vivo*: PRC2 generally targets promoters and enhancers, but not gene bodies, for the initiation of silencing, so protecting these cis-elements with H3K36me2 via NSD/Ash1L may abolish the initial recruitment of PRC2; in addition, the general H3K36me2 enrichment and the gene body-enriched H3K36me3 at active genes may inhibit the spread of H3K27 methylation from adjacent regions (Fig. 2). Taken together, we propose that NSD/Ash1L-mediated H3K36me2 may act as the primary effector to actively repel PRC2 silencing at developmental genes, thus maintaining their expression.

Although we attempt to dissect the specific roles of each H3K36 methyltransferase and the different forms of H3K36 methylation to clarify their intrinsic biological functions, they often work in concert and affect each other in many cases. For instance, the transcription of genes maintained by the NSD family proteins/Ash1L will certainly upregulate SED2 and H3K36me3, and...
the loss of the NSD family proteins/Ash1L, resulting in transcription inactivation, will surely cause the loss of SETD2 and H3K36me3.

Now two decades old, our knowledge of H3K36-specific methyltransferases and H3K36 methylation is still expanding. The links between the deregulation of H3K36-specific methyltransferases and various biological outcomes in diseases have not been fully established. Hopefully, a thorough understanding of the mechanism and function of H3K36-specific methyltransferases will help to pave the way for designing specific and rational targeting strategies for these diseases in the future.

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**Compliance with Ethical Standards**

**Conflict of interest** Chang Huang and Bing Zhu declare that they have no conflict of interest.

**Human and animal rights and informed consent** This article does not contain any studies with human or animal subjects performed by any of the authors.

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**References**

An S, Yeo KJ, Jeon YH, Song JJ (2011) Crystal structure of the human histone methyltransferase ASH1L catalytic domain and its implications for the regulatory mechanism. J Biol Chem 286:8369–8374

Antonysamy S, Condon B, Druzin A, Bonanno JB, Gheyi T, Zhang F, MacEwan I, Zhang A, Ashok S, Rodgers L, Russell M, Gately Luz J (2013) Structural context of disease-associated mutations and putative mechanism of autoinhibition revealed by X-ray crystallographic analysis of the EZH2-SET domain. PLoS ONE 8:e84147

Bell O, Wirbelauer C, Hild M, Scharf AN, Schweiger M, MacAlpine DM, Zilbermann F, van Leeuwen E, Bell SP, Imhof A, Garza D, Peters AH, Schübel D (2007) Localized H3K36 methylation states define histone H4K16 acetylation during transcriptional elongation in *Drosophila*. EMBO J 26:4974–4984

Bender LB, Suh J, Carroll CR, Fong Y, Fingerman IM, Briggs SD, Cao R, Zhang Y, Reinke V, Strome S (2006) MES-4: an autosomal-associated histone methyltransferase that participates in silencing the X chromosomes in the C. elegans germ line. Development 133:3907–3917

Bennett RL, Swaroop A, Troche C, Licht JD (2017) The role of nuclear receptor-binding SET domain family histone lysine methyltransferases in cancer. Cold Spring Harb Perspect Med https://doi.org/10.1101/cshperspect.a026708

Brooun A, Gajiwala KS, Deng YL, Liu W, Bolanos B, Bingham P, He YA, Diehl W, Grable N, Kung PP, Sutton S, Maegley KA, Yu X, Stewart AE (2016) Polycomb repressive complex 2 structure with inhibitor reveals a mechanism of activation and drug resistance. Nat Commun 7:11384

Byrd KN, Shearn A (2003) ASH1, a *Drosophila* trithorax group protein, is required for methylation of lysine 4 residues on histone H3. Proc Natl Acad Sci USA 100:11535–11540

Carrozza MJ, Li B, Florens I, Suganuma T, Swanson SK, Lee KK, Shia WJ, Anderson S, Yates J, Washburn MP, Workman JL (2005) Histone H3 methylation by Set2 directs deacetylation of coding regions by Rpd3S to suppress spurious intragenic transcription. Cell 123:581–592
Dorighi KM, Tamkun JW (2013) The trithorax group proteins Kismet and ASH1 promote H3K36 dimethylation to counteract Polycomb group repression in Drosophila. Development 140:4102–4119

Edmunds JW, Mahadevan LC, Clayton AL (2008) Dynamic histone H3 methylation during gene induction: HY/P/Setd2 mediates all H3K36 trimethylation. EMBO J 27:406–420

Fahey CC, Davis IJ (2017) SETting the stage for cancer development: SETD2 and the consequences of lost methylation. Cold Spring Harb Perspect Med. https://doi.org/10.1101/cshperspect.a026468

Fang D, Gan H, Lee JH, Han J, Wang Z, Riester SM, Jin L, Chen J, Zhou H, Wang J, Zhang H, Yang N, Bradley EW, Ho TH, Rubin BP, Bridge JA, Thibodeau SN, Ordog T, Chen Y, van Wijnen AJ, Oliveira AM, Xu RM, Westendorf JJ, Zhang Z (2016) The histone H3K36M mutation reprograms the epigenome of chondroblastosomas. Science 352:1344–1348

Fong Y, Bender L, Wang W, Strome S (2002) Regulation of the different chromatin states of autosomes and X chromosomes in the germ line of C. elegans. Science 296:2235–2238

Gaydos LJ, Rechtsteiner A, Egelhofer TA, Carroll CR, Strome S (2012) Antagonism between MES-4 and Polycomb repressive complex 2 promotes appropriate gene expression in C. elegans germ cells. Cell Rep 2:1169–1177

Gregory GD, Vakoc CR, Rozovskaia T, Zheng X, Patel S, Nakamura T, Canaani E, Blobel GA (2007) Mammalian ASH1L is a histone methyltransferase that occupies the transcribed region of active genes. Mol Cell Biol 27:9466–9479

He C, Li F, Zhang J, Wu J, Shi Y (2013) The methyltransferase NSD3 has chromatin-binding motifs, PHD5-C5HCH, that are distinct from other NSD (nuclear receptor SET domain) family members in their histone H3 recognition. J Biol Chem 288:4692–4703

Huang N, vom Baur E, Garnier JM, Lerouge T, Vonesch JL, Li F, Zhang J, Wu J, Shi Y, Garcia BA, Li W, Gozani O (2011) NSD2 links dimethylation of histone H3 at lysine 36 to oncogenic programming. Mol Cell 44:609–620

Lhoumaud P, Hennon M, Gamot A, Cudapah S, Quelle J, Liang J, Micas G, Morillon P, Urbach S, Bouchez D, Severac D, Embery E, Zhao K, Cuvier O (2014) Insulators recruit histone methyltransferase dMes4 to regulate chromatin of flanking genes. EMBO J 33:1599–1613

Li GM (2013) Decoding the histone code: role of H3K36me3 in mismatch repair and implications for cancer susceptibility and therapy. Cancer Res 73:6379–6383

Li J, Moazed D, Gygi SP (2002) Association of the histone methyltransferase Set2 with RNA polymerase II plays a role in transcription elongation. J Biol Chem 277:49383–49388

Li B, Howe L, Anderson S, Yates JR 3rd, Workman JL (2003) The Set2 histone methyltransferase functions through the phosphorylated carboxyl-terminal domain of RNA polymerase II. J Biol Chem 278:8897–8903

Li B, Gogol M, Carey M, Lee D, Seidel C, Workman JL (2007) Combined action of PHD and chromo domains directs the Rpd3S HDAC to transcribed chromatin. Science 316:1050–1054

Li B, Jackson J, Simon MD, Fleharty B, Gogol M, Seidel C, Workman JL, Shilatifard A (2009a) Histone H3 lysine 36 dimethylation (H3K36me2) is sufficient to recruit the Rpd3S histone deacetylase complex and to repress spurious transcription. J Biol Chem 284:7970–7976

Li Y, Trojer P, Xu CF, Cheung P, Kuo A, Drury WJ 3rd, Qiao Q, Neubert TA, Xu RM, Gozani O, Reinberg D (2009b) The target of the NSD family of histone lysine methyltransferases depends on the nature of the substrate. J Biol Chem 284:34283–34295

Liu C, Jain SÜ, Hoelper D, Bechet D, Molden RC, Ran L, Murphy D, Venneti S, Hameed M, Pawel BR, Wunder JS, Dickson BC, Lundgren SM, Jani KS, De Jay N, Papillon-Cavanagh S, Andrilus E, Sawyer SL, Greenspan D, Turcotte RE, Nadaf J, Fahmiminyah S, Muir TW, Majewski J, Thompson CB, Chi P, Garcia BA, Allis CD, Jabado N, Lewis PW (2016) Histone H3K36 mutations promote sarcomagenesis through altered histone methylation landscape. Science 352:844–849

McDaniel SL, Strahl BD (2017) Shaping the cellular landscape with Set2/SETD2 methylation. Cell Mol Life Sci 74:3317–3334

Miyazaki H, Higashimoto K, Yada Y, Endo TA, Komori T, Miyazaki H, Hirose S, Nishioka K (2013) Ash1 l methylates histone H3K36M mutation reprograms the epigenome of chondroblastosomas. Science 352:844–849

Nerri F, Rapelli S, Kreplelova A, Incarnato D, Parlato C, Basile G, Maldotti M, Anselmi F, Oliviero S (2017) Intragenic DNA methylation prevents spurious transcription initiation. Nature 543:72–77

Papp B, Muller J (2006) Histone trimethylation and the maintenance of transcriptional ON and OFF states by trxG and PcG proteins. Genes Dev 20:2041–2054

Popovic R, Martinez-Garcia E, Giannopoulou EG, Zhang Q, Zhang Q, Espondra T, Shah MY, Zheng Y, Will CM, Early EC, Hua Y, Bulic M, Jiang Y, Carrara M, Calogero RA, Kelleher N, Wang JP, Elemento O, Licht JD (2014) Histone
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methyltransferase MMSET/NSD2 alters EZH2 binding and reprograms the myeloma epigenome through global and focal changes in H3K36 and H3K27 methylation. PLoS Genet 10:e1004566

Qiao Q, Li Y, Chen Z, Wang M, Reinberg D, Xu RM (2011) The structure of NSD1 reveals an autoregulatory mechanism underlying histone H3K36 methylation. J Biol Chem 286:8361–8368

Ram O, Goren A, Amit I, Shores N, Yosef N, Ernst J, Kellis M, Gymrek M, Issner R, Coyne M, Durham T, Zhang X, Donaghey J, Epstein CB, Regev A, Bernstein BE (2011) Combinatorial patterning of chromatin regulators uncovered by genome-wide location analysis in human cells. Cell 147:1628–1639

Rechtstetter A, Ercan S, Takasaki T, Phippen TM, Egelhofer TA, Wang W, Kimura H, Lieb JD, Strome S (2010) The histone H3K36 methyltransferase MES-4 acts epigenetically to transmit the memory of germline gene expression to progeny. PLoS Genet 6:e1001091

Ringrose L, Paro R (2007) Polycomb/Trithorax response elements and epigenetic memory of cell identity. Development 134:223–232

Rogawski DS, Grembecka J, Cierpicki T (2016) H3K36 methyltransferases as cancer drug targets: rationale and perspectives for inhibitor development. Fut Med Chem 8:1589–1607

Sankaran SM, Wilkinson AW, Elias JE, Gozani O (2016) A PWWP domain of histone-lysine N-methyltransferase NSD2 binds to dimethylated Lys-36 of histone H3 and regulates NSD2 function at chromatin. J Biol Chem 291:8465–8474

Schmahling S, Meiler A, Lee Y, Mohammed A, Finkl K, Tauscher K, Israel L, Wirth M, Philippou-Massier J, Blum H, Habermann B, Imhof A, Song JJ, Müller J (2018) Regulation and function of H3K36 di-methylation by the trithorax-group protein complex AMC. Development 145

Schmitges FW, Prüstemann D, Tempst P, Chen S, Zhu B, Reinberg D (2009) Heterogeneous nuclear ribonucleoprotein L Is a subunit of human KMT3a/SETD2 complex required for H3 Lys-36 trimethylation activity in vivo. J Biol Chem 284:15701–15707

Yuan, W, Xu, M, B, Huang, C, Liu, N, Chen, S, Zhu, B (2011) H3K36 methylation antagonizes PRC2-mediated H3K27 methylation. J Biol Chem 286:7983–7989

Yuan, W, Wu, T, Fu, H, Dai, C, Wu, H, Liu, N, Li, X, Xu, M, Zhang, Z, Niu, T, Han, Z, Chai, J, Zhou, XJ, Gao, S, Zhu, B (2012) Dense chromatin activates Polycomb repressive complex 2 to regulate H3 lysine 27 methylation. Science 337:971–975

Yuan, G, Ma, B, Yuan, W, Zhang, Z, Chen, P, Ding, X, Feng, L, Shen, X, Chen, S, Li, G, Zhu, B (2013) Histone H2A ubiquitination inhibits the enzymatic activity of H3 lysine 36 methyltransferases. J Biol Chem 288:30832–30842

Zheng W, Ibanez G, Wu, H, Blum, G, Zeng, H, Dong, A, Li, F, Hajian, T, Allahi-Hassani A, Amaya MF, Siarheyeva A, Yu W, Brown PJ, Schapira M, Vedadi M, Min J, Luo M (2012) Sinelfin derivatives as inhibitors and structure probes of protein lysine methyltransferase SETD2. J Am Chem Soc 134:18004–18014

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Streubel G, Watson A, Jamulla SG, Scelfo A, Fitzpatrick DJ, Oliviero G, McCoie R, Conway E, Glancy E, Negri GL, Dillon E, Wynne K, Pasini D, Krogan NJ, Bracken AP, Cagney G (2018) The H3K36me2 methyltransferase Nsd1 demarcates PRC2-mediated H3K27me2 and H3K27me3 domains in embryonic stem cells. Mol Cell 70:371–379

Tanaka Y, Katagiri Z, Kawahashi K, Kioussis D, Kitajima S (2007) Trithorax-group protein ASH1 methylates histone H3 lysine 36. Gene 397:161–168

Venkatesh S, Workman JL (2013) Set2 mediated H3 lysine 36 methylation: regulation of transcription elongation and implications in organismal development. Wiley Interdiscip Rev Dev Biol 2:685–700

Wagner EJ, Carpenter PB (2012) Understanding the language of Lys36 methylation at histone H3. Nat Rev Mol Cell Biol 13:115–126

Wang Y, Niu Y, Li B (2015) Balancing acts of SRI and an auto-inhibitory domain specify Set2 function at transcribed chromatin. Nucleic Acids Res 43:4881–4892

Wang, C, Zhu, B, Xiong J (2018) Recruitment and reinforcement: maintaining epigenetic silencing. Sci China Life Sci 61:515–522

Wu H, Zeng H, Dong A, Li F, He H, Senisterra G, Seitova A, Duan S, Brown PJ, Vedadi M, Arrowsmith CH, Schapira M (2013) Structure of the catalytic domain of EZH2 reveals conformational plasticity in cofactor and substrate binding sites and explains oncogenic mutations. PLoS ONE 8:e83737

Xiao T, Hall H, Kizer KO, Shibata Y, Hall MC, Borchers CH, Strahl BD (2003) Phosphorylation of RNA polymerase II CTD regulates H3 methylation in yeast. Genes Dev 17:654–663

Yang S, Zheng X, Lu C, Li GM, Allis CD, Li H (2016) Molecular basis for oncohistone H3 recognition by SETD2 methyltransferase. Genes Dev 30:1611–1616

Yuan W, Xie J, Long C, Erdjument-Bromage H, Ding, X, Zheng, Y, Tempst, P, Chen, S, Zhu, B, Reinberg D (2009) Heterogeneous nuclear ribonucleoprotein L Is a subunit of human KMT3a/SETD2 complex required for H3 Lys-36 trimethylation activity in vivo. J Biol Chem 284:15701–15707

Yuan W, Xu, M, Huang, C, Liu, N, Chen, S, Zhu, B (2011) H3K36 methylation antagonizes PRC2-mediated H3K27 methylation. J Biol Chem 286:7983–7989