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Insights into newly discovered marks and readers of epigenetic information

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Abstract

The field of chromatin biology has been advancing at an accelerated pace. Recent discoveries of previously uncharacterized sites and types of post-translational modifications (PTMs) and the identification of new sets of proteins responsible for the deposition, removal, and reading of these marks continue raising the complexity of an already exceedingly complicated biological phenomenon. In this Perspective article we examine the biological importance of new types and sites of histone PTMs and summarize the molecular mechanisms of chromatin engagement by newly discovered epigenetic readers. We also highlight the imperative role of structural insights in understanding PTM–reader interactions and discuss future directions to enhance the knowledge of PTM readout.

PTMs of histones and other components of the epigenetic machinery, including a network of chromatin-modifying enzymes, covalent modifications of DNA, histone variants, and noncoding RNAs regulate chromatin structure and function[1–3]. PTMs have been found in both flexible tails and globular domains of the core and linker histones and are often referred to as ‘epigenetic’ marks, because they elicit changes in genome function that are not mediated through a change in the DNA sequence itself. Epigenetic marks directly influence histone–DNA and histone–histone interactions and serve as docking sites for reader domains (Fig. 1a). In large part, the binding of a reader to its cognate histone PTM defines the place and timing of recruitment of the host protein within the genome. Many reader-containing proteins constitute multisubunit enzymatic complexes, in which several readers, often with specificities for different PTMs, are in close proximity (Fig. 1b). Combinatorial readout of the multiple marks by distinct sets of readers provides a lock-and-key mechanism for targeting a particular genomic site that, in turn, is essential for imparting specific biological response. The recruited enzymatic complexes (for example, histone methyltransferases, demethylases, acetyltransferases, deacetylases, and ATPases) further change the epigenetic state of chromatin by adding or removing PTMs or by altering its dynamics and structure. This chain of fine-tuned events is vital to the control of most nuclear processes, including

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DNA transcription, replication, recombination, and repair. Consistent with this fundamental role, impaired epigenetic mechanisms are at the root of many human diseases, most notably cancer[4–6].

Over the past decade a considerable effort has been put forth to elucidate epigenetic mechanisms and identify histone readers and their matching PTMs. Characterization of specific PTM–reader combinations has been an especially fast-moving field. The first reader of the acetyllysine PTM, a bromodomain (BD), was discovered in 1999 (ref. 7); this finding was followed by discovery that a chromodomain (CD)[8–10] and a plant homeodomain (PHD) finger[11–14] recognize methyllysine. Dozens of epigenetic readers have since been identified and examined[15–18] (Table 1). The biological consequences of 'reading' the histone PTMs are highly context dependent and vary for individual readers. Epigenetic studies continue shedding light on the dynamic nature of the chromatin landscape amended through spatial and temporal fluctuations of PTMs and a diverse repertoire of readers.

In this Perspective we highlight the most recent developments in the field of epigenetics biology, focusing on the molecular mechanisms for the recognition of epigenetic marks by novel readers. In addition, we discuss the effect of combinatorial readout involving multiple modifications and paired readers and summarize the biological importance of newly identified PTMs.

Newly identified PTMs and orphan marks

Recent breakthroughs in mass spectrometry (MS) proteomics have enabled the detection of a large number of new types and sites of histone PTMs[19–22] (~550 currently known histone marks are listed in ref. 23) (Fig. 2). In addition to well-characterized canonical PTMs such as methylation of lysine (Kme) in histone H3 (K4, K9, K27, K36, and K79) and H4 (K20), acetylation of lysine (Kac), most notably in H3 (K9, K14, K18, K23, K27, K36, and K56) and H4 (K5, K8, K12, K16, and K20), and phosphorylation of serine and threonine in histones H2A and H3, an extensive set of new PTMs has emerged.

Although the precise functional contributions of the newly discovered sites of histone PTMs have yet to be elucidated, a number of recent reports demonstrate significant progress in this direction. For example, the finding that K4 of histone H3 (H3K4) undergoes not only methylation but also acetylation suggests the presence of a 'methyl–acetyl' switch to activate transcription[24]. It is likely that such regulatory switches represent a common epigenetic mechanism, as most lysine residues targeted for acetylation are also found methylated. Another new PTM linked to active chromatin is citrullination of H3R26, which promotes estrogen receptor-α target gene activation[25]. Spontaneous conversion of an aspartate into an isoaspartate followed by methylation of the latter is associated with protein aging, and this PTM was found in histone H4 (H4D24iso/me)[22].

Some novel PTMs identified within the core domains of histones have been implicated in chromatin organization and transcriptional regulation. Acetylation of H3K64 (H3K64ac) facilitates nucleosome eviction and is associated with active chromatin, whereas its trimethylated counterpart, H3K64me3 has a repressive function (refs. 26,27). Dimethylation

of H3R42 (H3R42me2) stimulates gene expression from chromatinized templates, probably through a disruption of DNA rewrapping onto the nucleosome core[28], and H3T45ph or H3S57ph precludes HP1-mediated repression of a set of stress-response genes[29].

Although some insights into the biological importance of the novel modification sites have been obtained, much less is known about the newly discovered modification types, including lysine crotonylation (Kcr) and lysine 2-hydroxyisobutyrylation (Khib). Similarly to acetylation and other acylation modifications, crotonylation neutralizes the positive charge and increases hydrophobicity of the lysine side chain. Unlike other acyl groups, the crotonyl group is an unsaturated moiety that contains a C–C double bond conjugated with the carbonyl group. The presence of the π-system offers additional opportunities for π–π or cation–π interactions not available to saturated acyllysine PTMs. Interestingly, histone Kcr levels are substantially elevated at promoters and show, unlike H3K4me3, symmetrical distribution around transcription start sites[20]. A breakthrough in the understanding of Kcr functioning has come from recent studies establishing a direct role for H3K18cr in the stimulation of gene transcription[30]. Because Kcr is widely distributed, further investigation is needed to determine whether distinct crotonylation sites impart other functions and whether metabolic programs contribute to the regulation of unique transcription through controlling distinct acyl-CoA forms (propionylation and butyrylation, like crotonylation, are derived from CoA precursors whose levels are dynamically controlled via metabolic flux).

One of the most abundant new histone modifications, Khib, contains a functional hydroxyl group that enables it to form additional hydrogen bonds. H4K8hib mark correlates with high transcriptional activity in meiotic and post-meiotic male germ cells and can be distinguished from the analogous Kac and Kcr PTMs at different spermatogenic stages[21]. Although its mechanism of action in meiosis is poorly understood, we envision that Khib might either directly mediate the recruitment of yet-to-be-identified chromatin regulators to promote transcription or act as a dynamic switch that interferes with activities of acetyllysine regulators. Given that biological data have been obtained for only a few modifications discussed above, it will be important to functionally characterize other newly discovered PTMs and determine whether these marks can be read by specific readers.

Novel readers and mechanisms of chromatin engagement

A number of epigenetic readers identified and studied in the past decade have been reviewed and therefore are not discussed here[15–18]. Below we provide an overview of recently discovered readers and summarize mechanistic details and biological implications of their interactions with PTMs.

YEATS domain recognizes H3Kcr

The family of acyllysine readers, comprising BD, double bromodomain (DBD), a double PHD finger (DPF), and a double pleckstrin homology (DPH) domain (Table 1), has been expanded with the discovery that the Yaf9–ENL–AF9–Taf14–Sas5 (YEATS) domains of yeast Taf14 and human AF9 and YEATS2 recognize crotonyllysine marks[31–33] and, to a lesser degree, other acyllysines, including acetyllysine[34,35]. Binding of the AF9 YEATS domain to H3K18cr or H3K9ac stimulates transcription, and the latter interaction links the

H3K79 methyltransferase DOT1L to H3K9ac-enriched chromatin[32,34]. Likewise, binding of the Taf14 YEATS domain to H3K9ac is required for gene activation and DNA damage repair in yeast[35,36].

The unique mechanism for recognition of Kcr has been elucidated from structures of the YEATS domains of Taf14, AF9 and YEATS2 bound to crotonylated histone peptides[31–33]. The structure of the Taf14 YEATS–H3K9cr complex shows a novel π–π–π stacking binding mode that had not been observed previously in protein interactions[31]. In the complex, the H3K9cr peptide adopts an extended conformation that is stabilized through an extensive set of direct and water-mediated hydrogen bonds with Taf14 (Fig. 3a). The crotonyllysine side chain transverses the narrow tunnel, crossing the β-sandwich structure of the protein in a corkscrew-like manner. The planar crotonyl group inserts between the aromatic rings of W81 and F62 that lay parallel to each other and at equal distance from the crotonyl group, creating a unique aromatic-amide–aliphatic-aromatic π–π–π stacking system[31] (Fig. 3a,b). The side chain of W81 adopts two conformations, each providing maximum π stacking with either the crotonyl alkene group or the crotonyl amide. The apparent dual conformation of W81 is probably due to the conjugated nature of the double bond and carbonyl π orbitals of the crotonyl group.

The side chain amide of K9cr is additionally constrained through a set of polar interactions. The K9cr amide nitrogen and carbonyl oxygen form hydrogen bonds with T61 and W81 and a water-mediated hydrogen bond with G82 (ref. 31) (Fig. 3b). Whereas a similar pattern of stabilizing hydrogen bonds is observed upon interaction with the H3K9ac peptide in the Taf14 YEATS–H3K9ac complex, W81 adopts only one conformation, being involved in π stacking with the acetyl amide[35]. The π stacking binding mode and the hydrogen bond network are conserved and mediate binding of the AF9 YEATS domain to H3K9cr and H3K9ac[32,34] (Fig. 3c), though the aromatic ring of Y78 is tilted and thus is likely to contribute to the π–π interaction to a lesser degree than a tryptophan (W81 in Taf14).

Both AF9 and Taf14 YEATS domains show preference for the RKacyl motif present in the H3R8K9, H3R17K18, and H3R26K27 sequences but not in H3G13K14 (refs. 31,32,34,35). The guanidino group of H3R8 forms a salt bridge with an aspartate, which is conserved in AF9 and Taf14 (Fig. 3a,c), and substitution of R8 with an alanine decreases binding of AF9 ~200-fold[34]. However, the YEATS domain of YEATS2 lacks this aspartate. The structure of the YEATS2 YEATS–H3K27cr complex reveals that R26 is entirely solvent exposed and the H3K27cr peptide is bound in an orientation opposite to that of the H3K9cr peptide in the Taf14 and AF9 complexes[33] (Fig. 3d). Whereas the histone residues N terminal to the acyllysine modification mediate the majority of contacts with the Taf14 and AF9 YEATS domains, the YEATS2 YEATS domain interacts with the histone residues that are C terminal to K27cr (Fig. 3d). Importantly, despite the overall different binding mode of YEATS2 compared to that of Taf14 and AF9, the mechanism for coordination of K27cr is conserved. The acyllysine recognition mechanism of the YEATS domains differs substantially from the mechanism for methyllysine recognition by Kme readers, where the methylammonium group of lysine is engaged in cation–π interactions with the aromatic side chains typically positioned perpendicularly to each other (discussed below).

BD selects for acetyllysine and propionyllysine

A comprehensive screen of bromodomains reveals that the majority (but not all) BDs bind acetyllysine and propionyllysine marks almost equally well, and a few BDs (BRD9 and CECR2) associate with butyryllysine[31,32,37,38]. These observations suggest that the varied preferences for acyllysines may mediate unique functions of these proteins. The structures of BD of BRD4 in complex with propionylated H3K23 (H3K23pr), butyrylated H3K14 (H3K14bu), and H3K14ac peptides and of BDs of BRD9 and TAF1 in complex with H4K5buK8bu or H4K5crK8cr provide insight into the selectivity of bromodomains for short acyllysine modifications, such as acetyllysine and propionyllysine, and their inability to bind longer acyllysine modifications, such as butyryllysine and crotonyllysine[37,38]. Much like in the canonical BD–Kac complex, the long-chain acyllysines are anchored through direct and water-mediated hydrogen bonds formed between the acyllysine carbonyl oxygen and an asparagine and a tyrosine (N140 and Y97 in BD1 of BRD4) (Fig. 3e,f). However, the invariable signature water shell, which lines the acyllysine-binding pocket of BDs restricting its size, allows only the short acyl chain to be accommodated and excludes the long one. Whereas propionyl and butyryl moieties can adopt bended conformations to avoid disturbing the water shell[38], the rigid planar crotonyl group displaces two water molecules in the water shell of the binding pocket of TAF1 BD, which probably accounts for the much weaker association (K_d = 100 μM) with this PTM[37]. Interestingly, BRD9 and CECR2 are members of the ATP-dependent remodeling complexes that may have evolved the capacity to read a range of short acyl modifications as part of a mechanism to differentially affect chromatin organization. It will be of interest to test whether crotonyllysine acts as a negative switch for these complexes.

Spindlin reads H3K4me3R8me2a

Spindlin is a novel reader capable of recognizing two consecutive PTMs, H3K4me3 and asymmetrically dimethylated H3R8 (H3K4me3R8me2a)[39]. This domain consists of three Spin–Ssty repeats and is found in proteins involved in transcriptional and cell cycle regulation, such as Spindlin1 (refs. 39–41). Spindlin appears to be the most robust epigenetic reader found to date, with a binding affinity of 45 nM[39]. The mechanism for the combinatorial methyllysine–methylarginine readout was deciphered from the structure of the H3K4me3R8me2a-bound Spindlin[39]. The Spin–Ssty repeats fold into Tudor-like β-barrels that are packed against each other to form a triangular assembly (Fig. 4a). The H3K4me3R8me2a peptide lays across the repeats, with K4me3 occupying an aromatic cage in the second repeat and R8me2a occupying an aromatic cage of the first repeat. Notably, the extended conformation of the bound H3K4me3R8me2a peptide allows to precisely match the distance between the two PTMs to the distance between the two aromatic cages (Fig. 4a,b). The K4me3 group is enclosed in a characteristic methyllysine-recognizing binding site consisting of four aromatic residues. The aromatic side chains are positioned almost orthogonally to each other and are involved in cation–π and hydrophobic interactions with the trimethylammonium group of lysine. In contrast, the methylarginine-recognizing aromatic cage is larger—it contains five aromatic residues and a glutamate (E64) that together with Y98 restrains the guanidino group of R8me2a via two hydrogen bonds (Fig. 4b). The methyllysine–methylarginine binding activity of Spindlin1 has been found to have a role in activation of the Wnt signaling pathway[39].

SAWADEE favors H3K9me over unmethylated H3K9

The SAWADEE domain of the plant protein SHH1 associates with H3K9me, and this interaction is required for the maintenance of small interfering RNA (siRNA) and methylated DNA levels and for the plant RNA polymerase Pol-IV to occupy target genes[42]. SAWADEE does not differentiate among methylation states *in vitro* and binds mono-, di-, and trimethylated H3K9 with similar affinity of 2 μM, yet methylation of H3K9 is essential, because binding to unmodified H3 is 17-fold weaker[42]. The structures of SAWADEE in complex with H3K9me1, H3K9me2, and H3K9me3 peptides provide a mechanistic explanation for this selectivity. The side chain of K9me inserts into the aromatic cage of Tudor1 in a tandem Tudor-like structure of SAWADEE (Fig. 4c). Whereas the hydrophobic character and the size of the aromatic cage preferentially select for tri-and dimethylated H3K9 species, the monomethylated species forms additional energetically favorable hydrogen bond with the protein.

PZP is a promiscuous reader of unmodified H3

Structural studies of the PHD–zinc-knuckle–PHD (PZP) domain of AF10 and BRPF1 reveal that this reader comprises three integrated zinc-binding modules—two PHD fingers linked by a single zinc finger[43,44]. Remarkably, although the two PZP domains have very similar structures, their functions differ (Fig. 5). The PZP domain of AF10 recognizes the middle portion (residues 22–27) of the histone H3 tail and is highly sensitive to methylation of H3K27, which abrogates this interaction even when K27 is monomethylated[43]. The PZP domain of BRPF1 instead associates with the N terminus of H3 and DNA[44]. The results of comprehensive biochemical and *in vivo* analyses underscore the significance of these interactions for proper functioning of AF10 and BRPF1. AF10 is a cofactor of the H3K79 methyltransferase DOT1L, and binding of its PZP domain to H3 is required for H3K79 dimethylation[43]. Histone- and DNA-binding activities of the BRPF1 PZP domain are essential for the recruitment of the MOZ/MORF acetyltransferase complex to chromatin and histone acetylation[44]. The bivalent interaction of BRPF1 PZP with H3 and DNA also affects nucleosome dynamics, shifting the DNA unwrapping–rewrapping equilibrium toward the unwrapped state and increasing DNA accessibility.

The H3 peptide is bound in a deep channel within the AF10 PZP domain[43] (Fig. 5a). Numerous intermolecular contacts restrain the histone residues T22–K27. Particularly, the side chain amino group of K27 is locked by hydrogen bonds formed with three backbone carbonyls of the protein, the loss of which due to H3K27 methylation would be energetically unfavorable. In the BRPF1 PZP domain, the first PHD finger retains its histone-binding function, also observed in an isolated PHD1 finger construct of BRPF1/2, which interacts with residues 1–7 of histone H3 (refs. 44,45). The H3(1–7) peptide forms the third antiparallel β-strand and pairs with the double-stranded β-sheet of BRPF2 PHD1 (ref. 45). Notably, the H3(1–7)-binding site of PZP is located on the side opposite to the H3(22–27)-binding site. The zinc-knuckle–PHD2 part of the BRPF1 PZP domain is enriched in positively charged residues and is implicated in DNA binding[44]. The PZP domain represents a fascinating example of functional plasticity of some readers and adds another layer of complexity in efforts to establish and generalize epigenetic mechanisms.

Crosstalk of paired readers

A wide variety of nuclear proteins contain multiple reader domains that show specificity for distinct PTMs and act in a combinatorial manner, mediating recruitment of the proteins to specific genomic regions. Concurrent binding of paired readers to modifications on the same histone tail and on separate histone tails is referred to as *cis*- and *trans*-readout, respectively.

Bromo–ZnF–PWWP of ZMYND11 recognizes H3.3S31K36me3

A combinatorial *cis*-readout regulates function of ZMYND11, a candidate tumor suppressor and transcriptional regulator. ZMYND11 contains contiguous PHD finger, BD, zinc finger (ZnF), and PWWP domains. The BD–ZnF–PWWP region recognizes K36me3 in the histone variant H3.3 and also binds DNA[46]. Although histone H3.3 differs from its canonical H3.1 form by only five amino acids, one residue in particular (S31 in H3.3 and A31 in H3.1) is essential for the ZMYND11 interaction. Structural comparison of BD–ZnF–PWWP complexes with H3.3K36me3 and H3.1K36me3 peptides reveals that K36me3 occupies the same aromatic cage of the PWWP domain in both complexes; however, several hydrogen bonds involving S31 and other residues in the A29–G33 region of H3.3 are lost in the corresponding H3.1 complex[46] (Fig. 6a). ZMYND11 represents the first example of a paired reader that is selective for a particular histone variant, and it will be of interest to explore whether other variant-specific readers exist.

BAH–PHD of ORC1 binds unmodified H3

A new *cis*-readout mechanism has been reported for the bromo-adjacent homology (BAH)–PHD cassette of the plant protein ORC1b. The ORC1b PHD finger sequence is embedded in the sequence of BAH, and this unique assembly of readers recognizes unmodified histone H3 tail (residues 1–8)[47]. In the BAH–PHD–H3 complex, the peptide adopts an extended conformation and occupies a long groove at the interface of the readers (Fig. 6b). Both readers create the binding site and surround the same region of the peptide. The extensive set of hydrogen bonds formed between BAH–PHD and the side chains of the histone R2, T3, and K4 residues suggests that modification of these residues would prevent binding, and this has been confirmed experimentally[47].

A *trans*-readout of H3K9me2 by BAH and CD of ZMET2

Biochemical and structural characterization of the maize protein ZMET2 shows that CD and BAH, which are linked through the DNA methyltransferase domain, each recognize H3K9me2 through caging dimethyllysine in their individual aromatic pockets[48]. A triangular topology of the BAH–CD–methyltransferase region suggests a *trans*-readout mechanism in which CD and BAH can read the H3K9me2 marks on two histone tails (Fig. 6c). As further evidence for the dual-recognition mode, isothermal titration calorimetry (ITC) experiments yield a 1.8 stoichiometry for binding of the H3K9me2 peptide to ZMET2. A dual-recognition mode may be necessary to ensure a high fidelity of DNA methylation and may promote spreading of the methyltransferase activity[48].

Concluding remarks

The past few years have seen rapid advances in the field of epigenetics, especially in the identification and characterization of histone PTMs and the protein domains capable of reading these modifications. In addition to the well-established mechanisms that mediate recognition of methyllysine or methylarginine through cation–π interactions within the aromatic cage of the reader or dictate reader specificity toward a mono- or dimethyllysine species due to the presence of a negatively charged residue in the aromatic pocket, a diverse set of new binding modes has emerged. These include π– π–π stacking interactions involving the C–C double bond of crotonyllysine and a pair of aromatic residues of the reader; a sandwich-like insertion of acetyllysine between the aromatic residues; a water-shell control allowing differentiation of the acyllysine side chains; a hydrogen bond–mediated selectivity toward methylated states of lysine; and PTM rulers that govern binding of paired readers. In-depth characterization of these mechanisms is crucial not only for understanding a wide array of chromatin-related processes—most of all gene expression regulation—but also for the development of unique epigenetic-driven therapies. Aberrant epigenetic states have been linked to cancer, premature aging, immunodeficiency, and other human diseases and, as a result, histone readers have become attractive therapeutic targets[49,50].

Despite remarkable advances in the field, many more questions remain. For example, the list of histone readers has grown substantially, but binding partners for a number of currently known PTMs have yet to be elucidated. To facilitate the discovery of readers, new technologies and adaptions of current ones, including high-throughput microarrays capable of testing interactions with intact PTM-containing nucleosomes *in vitro* and in cells and animal models, need to be developed. A large number of PTMs have been discovered in the core domains of histone proteins, and it will be important to explore whether unique core PTM readers exist. Other pressing objectives are to establish the synergistic and antagonistic effects of PTMs at the genome-wide level; define the role of the regulatory methyl-, acyl-, and phospho-PTM switches[51–53]; and characterize bookmarking PTMs that may contribute to reactivation of transcriptional and chromatin-remodeling programs during cell division[54,55].

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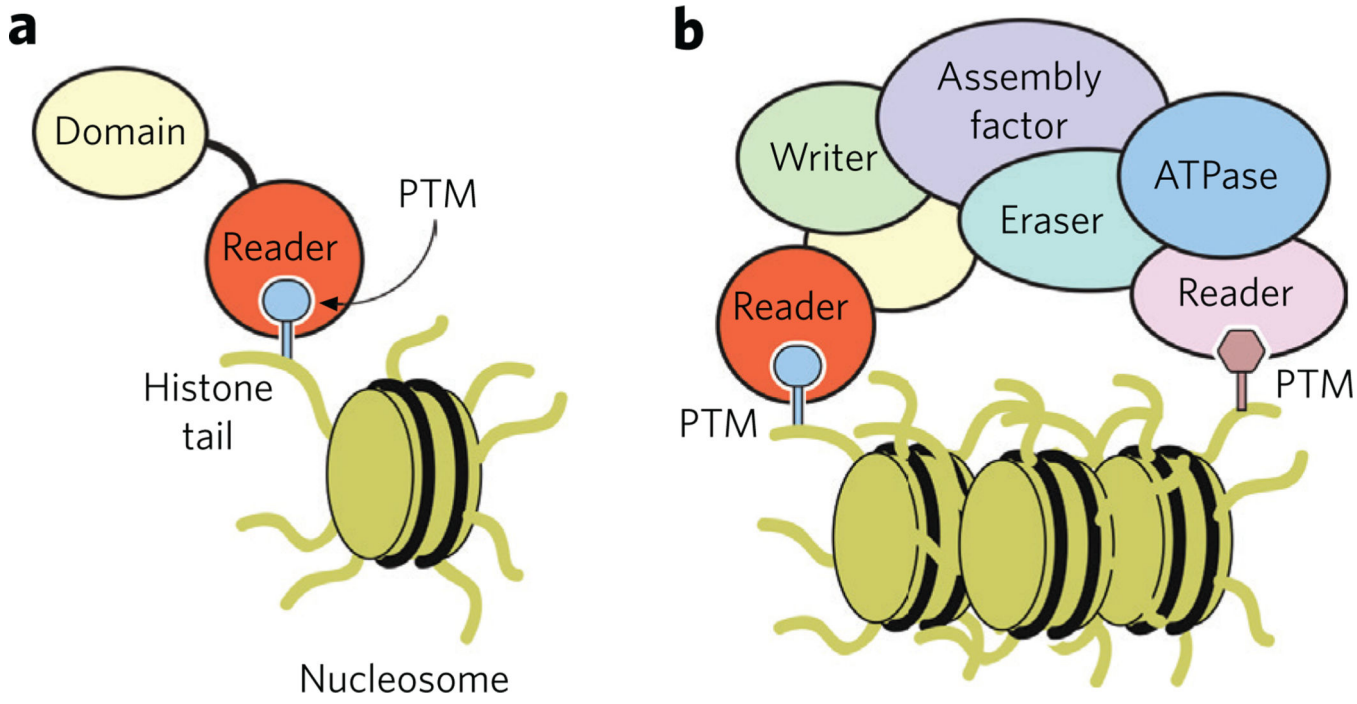

Figure 1. Recognition of epigenetic marks by histone readers

(**a**) A reader domain (orange circle) binds to its target PTM (blue circle) in the histone tail, tethering the host protein to chromatin. (**b**) Multivalent engagement with chromatin through interactions of multiple readers, in the same protein or in different proteins (assembled in the complex), to enhance or regulate overall binding affinity and specificity. Complexes often contain proteins or subunits with catalytic domains (writers, erasers, or ATPase remodelers) and scaffolding domains necessary for the complex assembly.

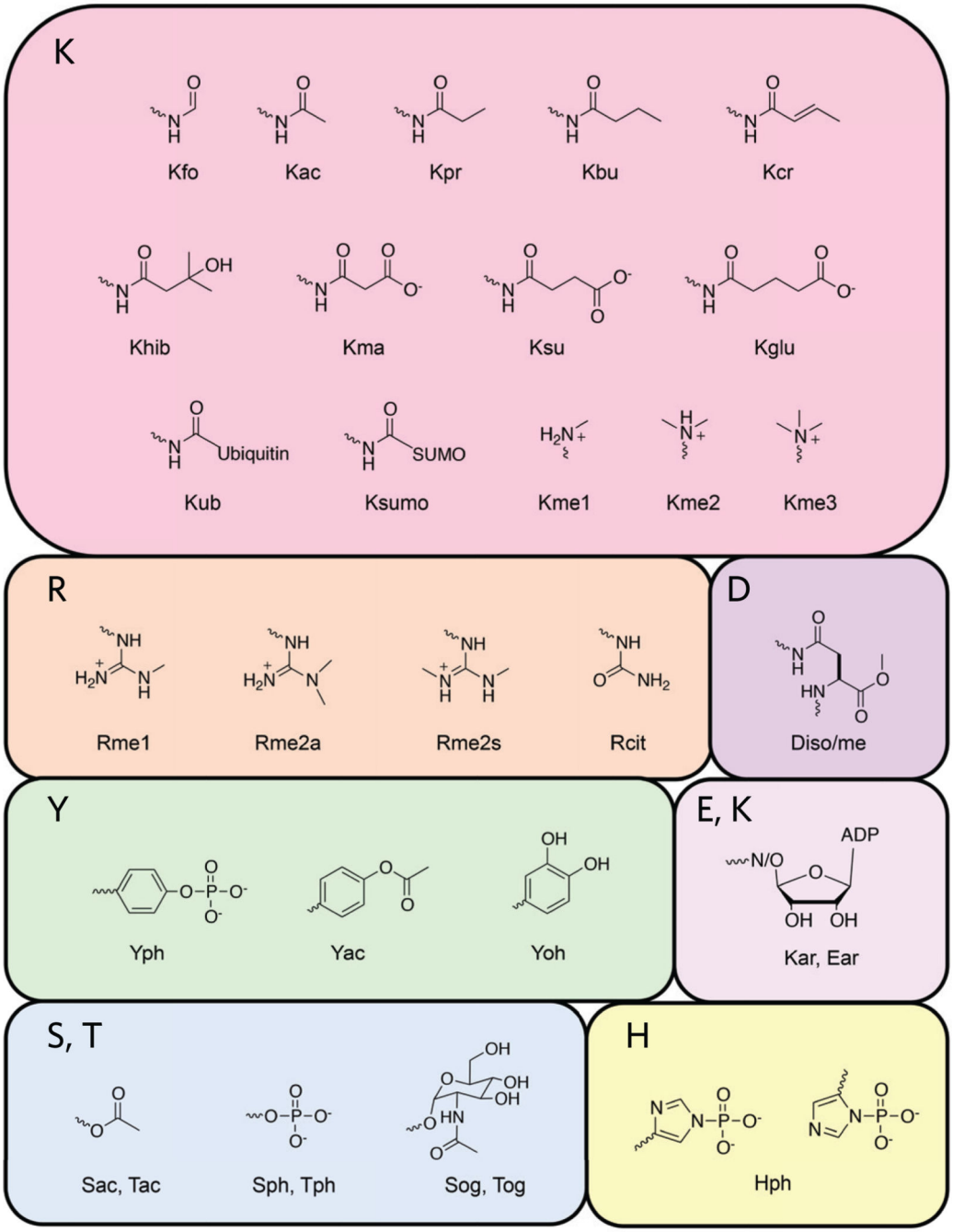

Figure 2. Modifications identified in histone proteins

fo, formylation; ma, malonylation; su, succinylation; glu, glutarylation; ub, ubiquitination; cit, citrullination; oh, hydroxylation; ar, ADP ribosylation; og, *O*-GlcNAcylation.

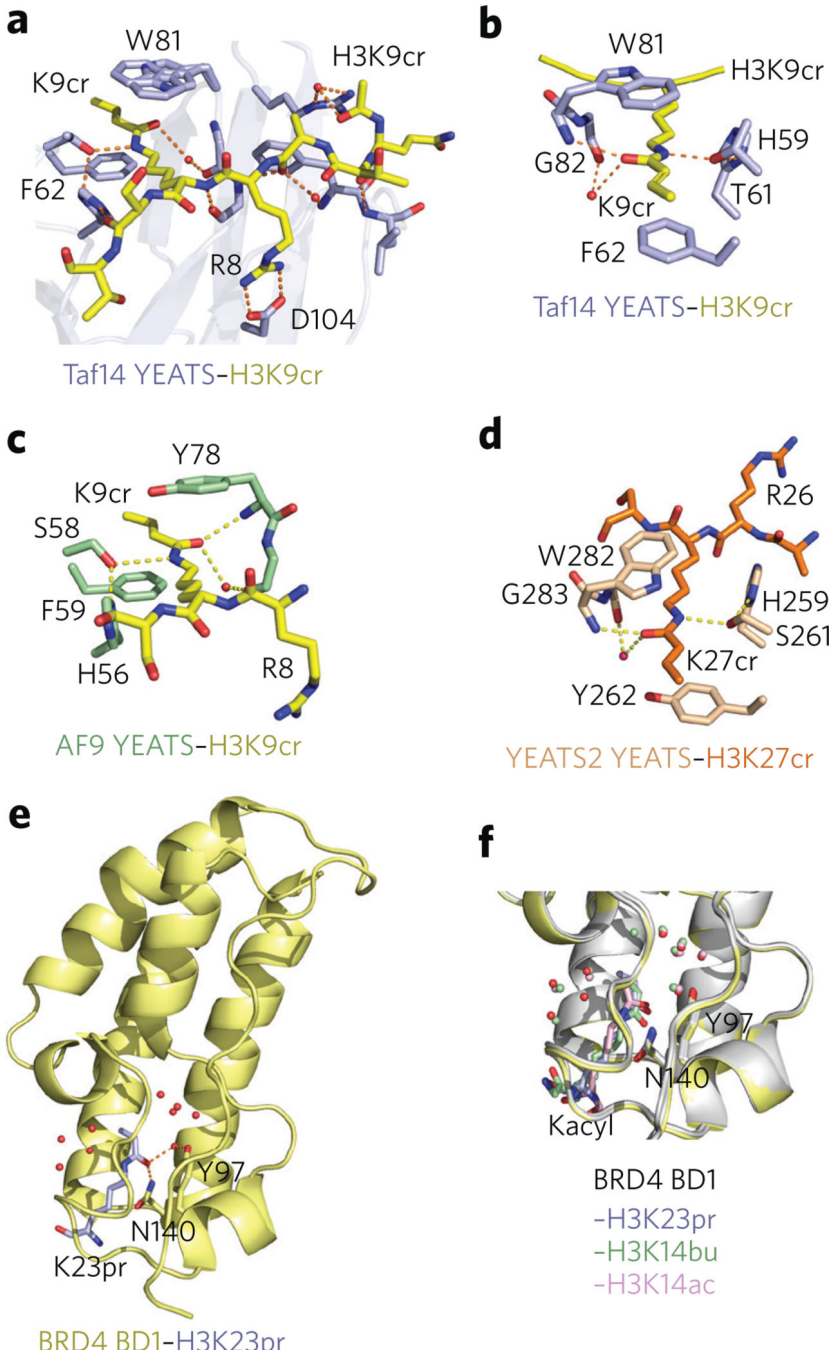

Figure 3. Novel acyllysine readers and their binding mechanisms

(**a**) Structure of the Taf14 YEATS domain (blue) in complex with H3K9cr peptide (yellow stick). Red dashed lines represent hydrogen bonds; spheres represent water molecules. (**b**) The **π**–**π**–**π** stacking mechanism involving the alkene moiety of Kcr in Taf14 YEATS–H3K9cr (PDB 5IOK). (**c**) AF9 YEATS domain (green) in complex with H3K9cr (yellow; PDB 5HJB). (**d**) The H3K27cr-binding site of the YEATS2 YEATS domain (beige; PDB 5IQL). H3K27cr is shown in orange. (**e**) The structure of BD of BRD4 (yellow) in complex with the H3K23pr (light blue; PDB 3MUK). (**f**) Overlay of the structures of the BRD4 BD1

in complex with H3K23pr (light blue), H3K14bu (light green; PDB 3MUL), and H3K14ac (pink; PDB 3JVK), with water shells shown as red, light green, and pink spheres, respectively.

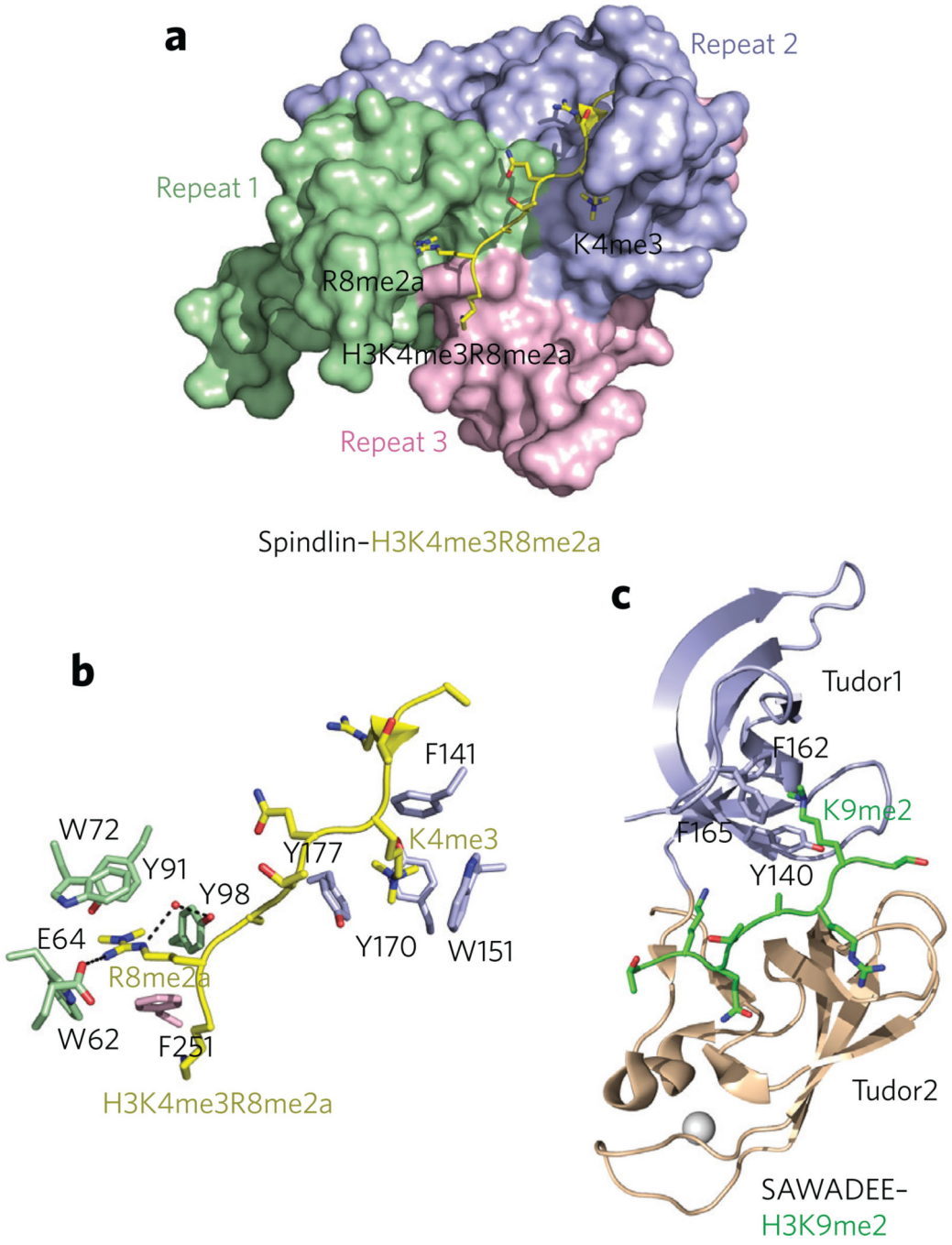

Figure 4. New methyllysine readers

(**a**) The structure of Spindlin of the protein Spindlin1 in complex with H3K4me3R8me2a peptide (PDB 4MZF). (**b**) Close-up view of the two aromatic cages of Spindlin. (**c**) The structure of the SHH1 SAWADEE–H3K9me2 complex (PDB 4IUT).

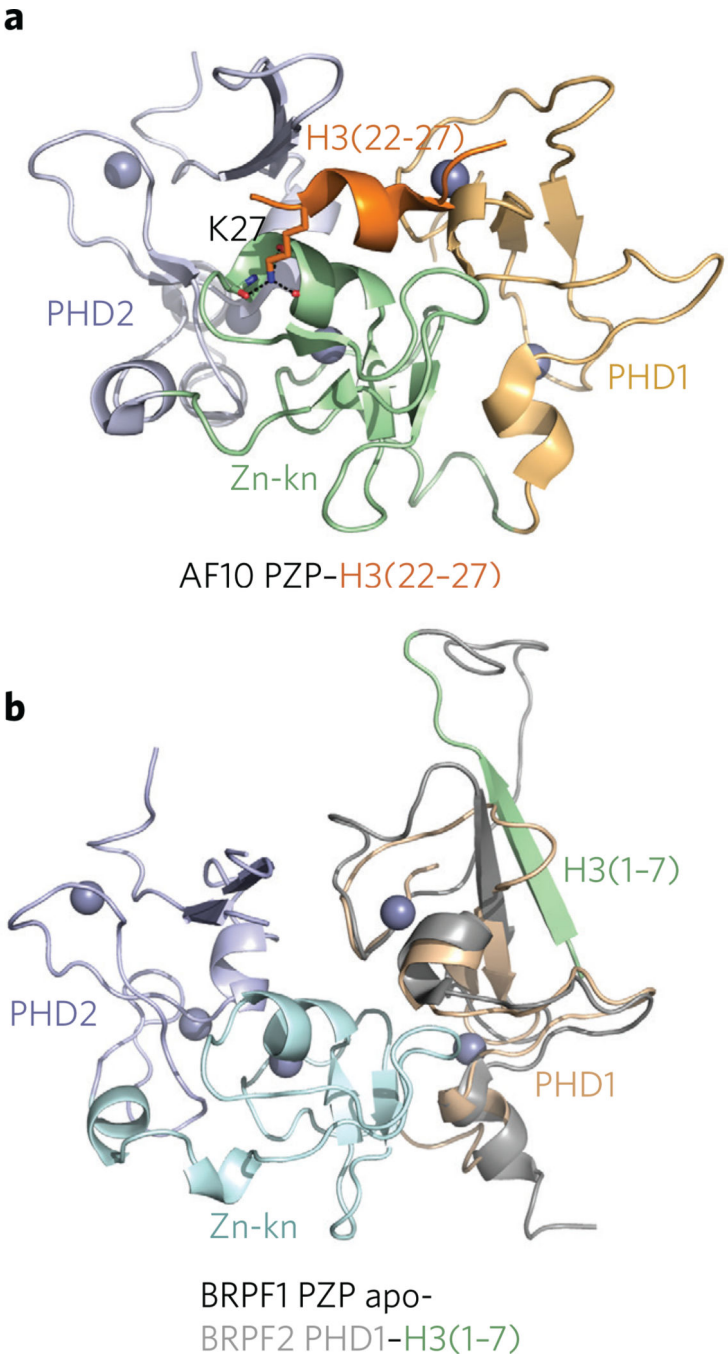

Figure 5. Novel readers of unmodified histone H3

(**a**) The structure of the AF10 PZP–H3(22–27) complex (PDB 5DAH). Zn-kn, zinc knuckle. (**b**) Superimposed structures of the BRPF1 PZP domain (brown, cyan, and blue) and the PHD1 finger (gray) of orthologous BRPF2 fused to a sequence corresponding to histone H3(1–7) tail (light green) (PDB 5ERC and 2L43). Spheres represent zinc ions.

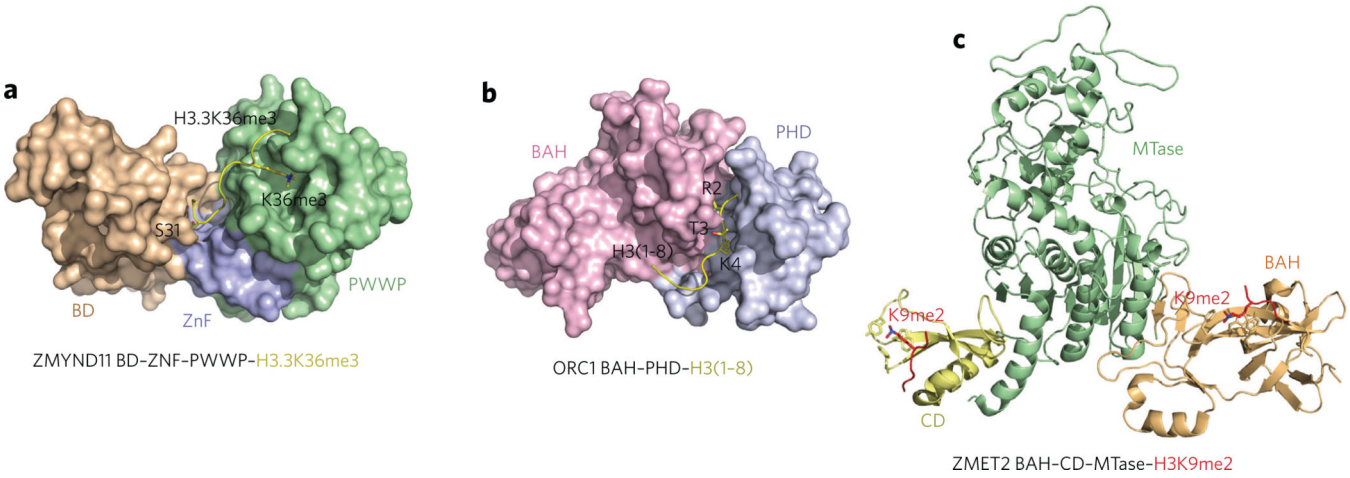

Figure 6. Crosstalk of PTMs and paired readers

(**a**) The structure of the ZMYND11 BD–ZnF–PWWP region in complex with H3.3K36me3 peptide (yellow) (PDB 4N4I). (**b**) The structure of ORC1 BAH–PHD in complex with H3(1–8) (yellow) (PDB 5HH7). (**c**) Superimposed structures of two complexes of the ZMET2 BAH–CD–methyltransferase region bound to different H3K9me2 peptides (red) (PDB 4FT2 and 4FT4). MTase, methyltransferase.

Table 1

Readers of histone PTMs

Epigenetic mark	Reader	Histone PTM
Kac	BD	H3Kac, H4Kac, H2AKac, H2BKac
	DBD	H3KacKac, H4KacKac
	DPF	H3K14ac
	DPH	H3K56ac
Kpr	BD	H3Kpr
Kcr	YEATS	H3Kcr
Kme	ADD	H3K9me3
	Ankyrin	H3K9me2, H3K9me1
	BAH	H4K20me2, H3K27me3
	Chromobarrel	H3K36me3, H3K36me2, H4K20me1, H3K4me1
	CD	H3K9me3, H3K9me2, H3K27me3, H3K27me2
	DCD	H3K4me3, H3K4me2, H3K4me1
	MBT	H3Kme1, H3Kme2, H4Kme1, H4Kme2
	PHD	H3K4me3, H3K4me2, H3K9me3
	PWWP	H3K36me3, H4K20me1, H4K20me3, H3K79me3
	SAWADEE	H3K9me1, H3K9me2, H3K9me3
	Spindlin	H3K4me3R8me2a
	TTD	H3K4me3, H3K9me3, H4K20me2
	Tudor	H3K36me3
	WD40	H3K27me3, H3K9me3
	CW	H3K4me3
Rme	ADD	H4R3me2s
	Tudor	H3Rme2, H4Rme2
	WD40	H3R2me2
Sph, Tph	14-3-3	H3S10ph, H3S28ph
	BIR	H3T3ph
	Tandem BRCT	H2AXS139ph
H3 unmodified	ADD	H3
	BAH–PHD	H3
	PHD	H3
	PZP	H3
	WD40	H3