Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Structural Biology

Nucleosome adaptability conferred by sequence and structural variations in histone H2A–H2B dimers

Alexey K Shaytan, David Landsman and Anna R Panchenko

Nucleosome variability is essential for their functions in compacting the chromatin structure and regulation of transcription, replication and cell reprogramming. The DNA molecule in nucleosomes is wrapped around an octamer composed of four types of core histones (H3, H4, H2A, H2B). Nucleosomes represent dynamic entities and may change their conformation, stability and binding properties by employing different sets of histone variants or by becoming post-translationally modified. There are many variants of histones H2A and H2B. Specific H2A and H2B variants may preferentially associate with each other resulting in different combinations of variants and leading to the increased combinatorial complexity of nucleosomes. In addition, the H2A–H2B dimer can be recognized and substituted by chaperones/remodelers as a distinct unit, can assemble independently and is stable during nucleosome unwinding. In this review we discuss how sequence and structural variations in H2A–H2B dimers may provide necessary complexity and confer the nucleosome functional variability.

Address
National Center for Biotechnology Information, National Library of Medicine, National Institutes of Health, Bethesda, MD 20894, USA

Corresponding author: Panchenko, Anna R (panch@ncbi.nlm.nih.gov)

Current Opinion in Structural Biology 2015, **32**:48–57

This review comes from a themed issue on **Sequences and topology**

Edited by **M Madan Babu** and **Anna R Panchenko**

For a complete overview see the Issue and the Editorial

Available online 27th February 2015

http://dx.doi.org/10.1016/j.sbi.2015.02.004

Introduction

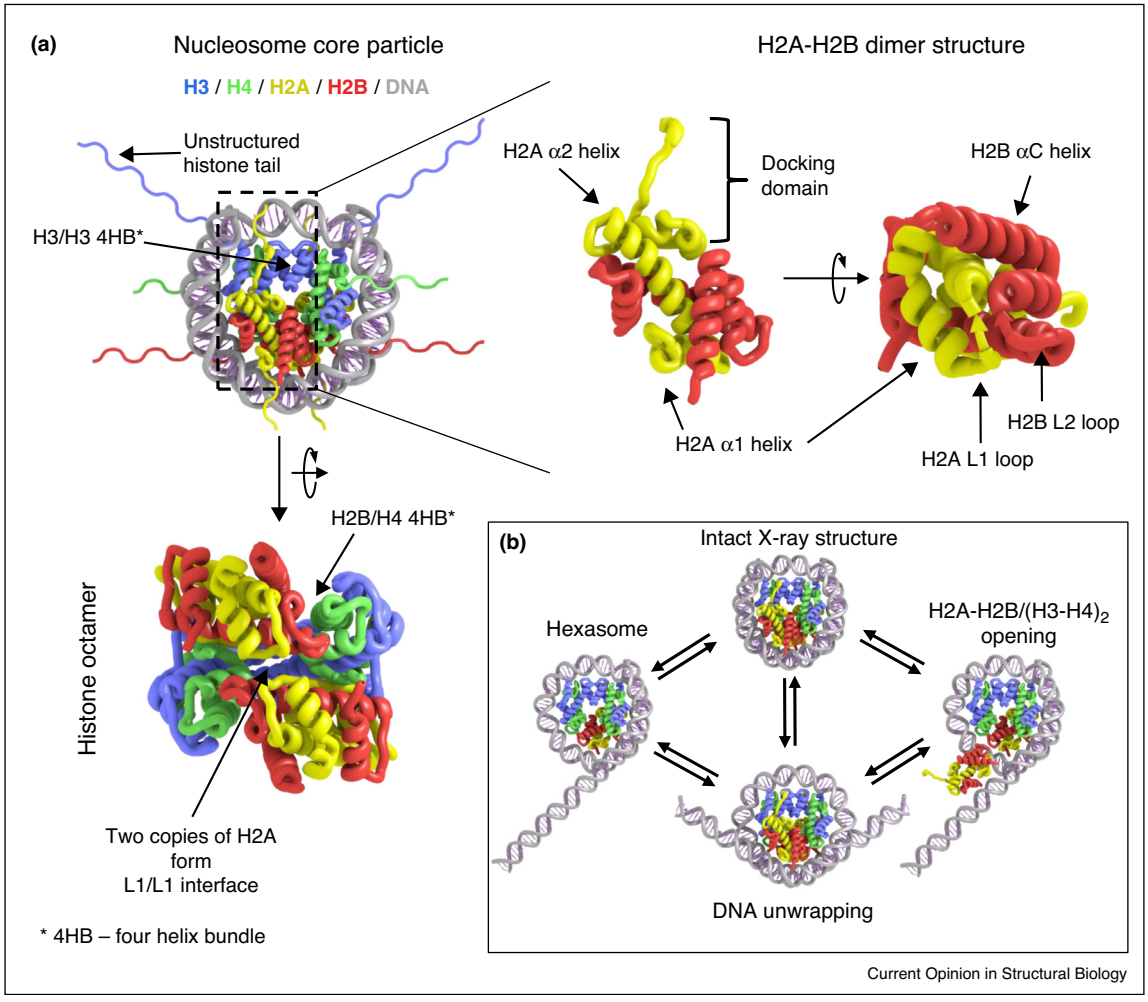
Chromatin packaging is tightly coupled to genome function and gene expression regulation. The basic unit of chromatin packing, the nucleosome, wraps ~145–147 bp of DNA in a ~1.7 left-handed super helical turns around an octamer composed of four types of core histones (H3, H4, H2A, H2B — two copies of each) [1]. The histone octamer is known to form a tripartite modular protein assembly where the $(H3–H4)_2$ tetramer is composed of two (H3–H4) heterodimers and organizes the inner turn of DNA, while two (H2A–H2B) heterodimers dock on both sides of tetramer in order to further wrap remaining ~40 bp of DNA on each end (Figure 1a) [2]. Nucleosomes are located at certain distances from each other along the DNA molecule and nucleosome spacing is shown to be species and tissue dependent [3,4]. Once thought to play merely structural role, nucleosomes now unravel their dual nature as key players in epigenetic regulation of transcription, replication and reprogramming. It is becoming more recognized that nucleosomes represent dynamic entities [5,6], namely, they may change their conformation, histone content, employ a set of different histone variants (see definition below), and become post-translationally modified upon certain conditions. Nucleosome variability is essential to adapt chromatin structure and function in order to respond to environmental stimuli and fulfill cellular function [7]. Some of the recent striking examples of nucleosome adaptability include: role of H2A.Z histone acetylation and deposition in memory formation [8,9], modulation of olfactory neurons life span by histone variant H2B.E [10•], H2A.Z participation in embryonic stem cell differentiation [11] and its role in acclimatization of common carp [12]. The histone H2A and H2B families contain many sequence variants which can confer the nucleosome structural and functional variability. Moreover, the H2A–H2B dimer can be recognized and substituted by chaperones and remodelers as a distinct unit, it may assemble independently before incorporation into nucleosomes and is stable during nucleosome unwinding (Figure 1b).

While many histone variants are known to be lineage specific, in this review we focus on a representative set of universal and mammalian specific H2A and H2B histone variants. We highlight recent advances in understanding of the sequence and structural variability of H2A–H2B dimers resulting in dynamical and functional changes in nucleosomes.

H2A–H2B dimer as a semi-independent unit in nucleosome dynamics

Nucleosomal dynamics is tightly coupled to genome markup and transcription. The unwrapping of DNA from the octamer surface is thought to be crucial for transcription factor (TF) binding in most cases [13]. Moreover, RNA Polymerase II (Pol II) pauses upon encountering the nucleosome and the DNA–octamer interactions may account for the nucleosomal barrier to Pol II and regulate the rate of transcription [14]. Moderately expressed genes, transcription by Pol II is accompanied by the displacement of one H2A–H2B dimer while $(H3–H4)_2$ tetramer remains intact, as a result a hexasome is formed (see Figure 1b). Hexasome survival is facilitated by electrostatic interactions between polymerase and histones [15,16]. Recent

Figure 1



Nucleosome structure and dynamics with focus on H2A–H2B dimer. **(a)** Nucleosome core particle, histone octamer and H2A–H2B dimer structures are shown in various orientations (PDB: 1AOI [2]) the unstructured histone tails are schematically shown as extensions protruding away from the core. Individual histones form two types of topologically similar heterodimers H3–H4 and H2A–H2B via a stable 'hand-shake' motif interaction between interdigitating helices of histone-folds (α1, α2 and α3) connected by two loops (L1, L2) (see also Figure 2). The dimers can then interact with each other via four-helical bundle motifs (4HB). Two H3–H4 dimers associate via a strong H3–H3 4HB interaction forming a tetramer. Two H2A–H2B dimers can further associate with a tetramer on each side upon DNA binding via a weaker H4–H2B 4HB interaction, supplemented by the interactions of the H2A docking domain with corresponding H3–H4 interface. Main histone-DNA binding sites are formed by the L1–L2 loop regions and α1 helical regions and usually have characteristic arginine side chains protruding into the minor groove of kinked DNA [70]. **(b)** Alternative conformational states of nucleosome may occur due to thermal fluctuations, active biochemical processes (such as transcription), histone variant substitution or post-translational modifications. These alternative conformational states include DNA unwrapping (the octamer is intact); hexasome formation when a loss of one H2A–H2B dimer leads to the DNA unwrapping from one end; opening of H2A–H2B/$(H3–H4)_2$ interface accompanied by changes in DNA conformations while the H2A–H2B dimer still remains bound to DNA.

characterization of the hexasome structure by small angle X-ray scattering revealed that the removal of one H2A–H2B dimer did not cause large structural changes in the remaining part of the nucleosome core [17].

Substantial evidence points to the fact that simple unwrapping of DNA ends from the octamer surface is not the only event in nucleosome unwinding/disassembly (Figure 1b). Nucleosome unwinding experiments using optical tweezers recently revealed the existence of multiple unwound states suggesting the symmetrical split of the octamer through H3–H3 interface under tension at specific conditions [18]. At the same time, equilibrium FRET experiments suggest an open intermediate state of the nucleosome, where the interface between $(H3–H4)_2$ tetramer and H2A–H2B dimer may be reversibly opened under physiological conditions [19]. The balance between these alternative nucleosome conformations should depend on sequence and structural variations within the H2A–H2B dimer and might have

profound functional implications. The dynamical opening of nucleosome structure can expose interfaces, which are otherwise inaccessible and not observed in static X-ray structures.

H2A–H2B dimers can be recognized, actively exchanged and deposited by different histone chaperones and nucleosome remodelers, however the details of these processes still remain elusive and not well studied. Recent advancements in this field include the structural characterization of ATP-dependent SWR1-complex, which is able to recognize nucleosomes with canonical H2A–H2B dimers and substitute it with an H2A.Z–H2B dimer [20]. In addition, recent insights into structure and function of histone chaperon FACT showed that its Spt16M domain binds H2A–H2B dimer via the H2B α1-helix region [21•], suggesting that FACT can block the interactions of H2B with DNA in nucleosome favoring unwound DNA conformation.

H2A and H2B histone variants and their functions

The four main types of core histones (H3, H4, H2A, H2B) are all structurally very similar within the histone-fold region, while sharing less than 25% sequence identity [22]. Every histone type is usually encoded by several different genes giving rise to histone variants (which may also arise due to the alternative splicing). Histone variants may be either universal to eukaryotes or species specific. The difference between variants and canonical histones can range from several amino acids up to the level marking the variance between different types of canonical histones (see Tables 1 and S1). Histones are usually subdivided into canonical replication-dependent (their expression coincides with the S-phase of cell cycle) and replication-independent histone variants, constitutively expressed during cell cycle [23,24] (referred to as 'variants' thereafter). In metazoans canonical genes are typically located within multigene clusters and use specific type of regulation at the RNA level with a stem loop structure instead of polyA tail [25]. Genes encoding the histone variants (sometimes called 'orphan genes') are typically located outside of these clusters and are regulated similar to normal genes [25]. A few known exceptions in mammals include testis-specific histones TS H2A.1 and TS H2B.1 [26••,27] and H2B.E in mice [10•], which are all located in gene clusters but can be expressed outside of the DNA replication phase. Interestingly, the number of histone variants tends to increase with the complexity of organism providing structural and functional diversification needed for genome functioning. The nomenclature for growing family of histone variants was recently suggested in [28•], and we follow this nomenclature in the current review.

Histone H2A has the highest number of known variants, while H2B is thought to be less variable. Tables 1 and S1 show the list of variants and summary of their known functions and localizations. The variants for H2A include widely studied universal variants H2A.Z and H2A.X, vertebrate specific mH2A, mammal specific H2A.B, as well as less studied testis-specific variants in mammals TS H2A.1, H2A.L. The H2B variants in mammals include testis-specific TS H2B.1, H2B.W, subH2B, and newly characterized variant H2B.E, shown to regulate olfactory neuron function in mice [10•]. Despite considerable progress in understanding the functions of histone variants, the complete picture remains elusive. Variants are associated with various functions such as up and down regulation of gene expression (H2A.Z, mH2A, H2A.B), DNA damage response (H2A.X, H2A.Z), epigenetic reprogramming (TS H2A.1, TS H2B.1), splicing (H2A.B), pericentric and telomere chromatin organization (H2B.W, H2A.Z, H2A.X), among others. All this suggests that the role of histone variants is often multifunctional and context dependent. With time, novel histones variants or new splice isoforms may be discovered. For example, an alternatively spliced isoform of H2A.Z.2 (H2A.Z.2.s2) has been recently identified in human brain tissues [29•]. Despite high sequence similarity, the splice isoforms have been shown to function differently during DNA damage repair [30].

Sequence variation and evolution of H2A and H2B histones

H2A variants H2A.Z, H2A.B and mH2A are known to have monophyletic origins with H2A.Z originating early in eukaryotic evolution [23], whereas H2A.X variants have diverged repeatedly [24]. While canonical histones (and ancient variants, like H2A.Z) are among the most conserved proteins across different species [31], certain variants do not follow this trend. For example, H2A.B and some testis-specific histones are considered quickly evolving hypervariable mammalian histones [32].

Histone variants may differ only by several amino acids. For example, only four or five amino acids are changed between H2B and H2B.E variants [10•] while two subvariants of H2A.Z (H2A.Z.1 and H2A.Z.2) in vertebrates vary by only three amino acids. The same is true of H2A.X, which mainly differs from the canonical H2A by a functionally important C-terminal phosphorylation motif Ser-Gln-(Glu/Asp)-Φ, where Φ represents a hydrophobic residue. Variant specific phosphorylation of serine in this motif can occur upon the formation of DNA double-strand breaks [33] and, may be important in engaging and retention of various chromatin remodeling factors in order to promote the double-strand break repair. On the other hand, major variants H2A.Z, mH2A, H2A.B show much lower sequence identity to canonical histones (about 40–60%) whereas members of H2A.L family (which is rather diverse and still awaits further investigation), like mouse H2A.L.3, show even lower sequence identity of 24% with canonical H2A.

Table 1

Representative list of known H2A and H2B histone variants and their characteristics. Letters in parenthesis indicate taxonomic span: M-mammals, m–mouse, V-vertebrates, H-human, D-drosophila.

Variant name[a] Alt. names	Known subtypes or splice isoforms[b]	Taxonomic distribution[c]	Proposed functions	Preferential localization/tissue specificity	Sequence features	Known human genes[d]	Sequence identity[e]	Ref
H2A variants								
H2A.X[f]		Eukaryotes except nematode	DNA damage response, chromatin remodeling	DNA double strand breaks, collapsed replication forks, heterochromatin	SQE/DΦ[g] motif at C-terminus, S is phosphorylated in DNA double strand breaks	H2AFX	78%	[24,57,58]
H2A.Z H2A.V(D)	H2A.V(D) H2A.Z.1(V) H2A.Z.2(V) H2A.Z.2.s2 (H)	Eukaryotes	Pol II recruitment, transcription regulation, DNA repair, suppression of antisense RNA, among others	Transcription start site (+1 and −1 nucleosomes), enhancers	Larger acidic patch, deletion and insertion in the core region	H2AFZ H2AFV	60%	[24,57,59,60]
mH2A macroH2A	mH2A.1.s1(M) mH2A.1.s2(M) mH2A.2(M)	Vertebrates	X-inactivation, positive or negative transcriptional regulation	Inactivated X chromosome (M), gene promoters, heterochromatin	Contains a ~30 kDa additional globular macrodomain attached to C-terminus	H2AFY H2AFY2	N/A	[61]
H2A.B H2A.Bbd, H2A.Lap1 (m)[h]		Mammals	Gene upregulation, gene splicing	Active genes, transcription start site in testis(m)	Short C-terminal tail, truncated acidic patch, arginine rich	H2AFB1 H2AFB2 H2AFB3	60%	[57,62]
H2A.J		Human, mouse			Very similar to canonical	H2AFJ	97%	[63]
TS H2A.1 TH2A		Mammals	Genome reprograming(m)	Testis, oocytes, fertilized eggs	Similar to canonical	HIST1H2AA	88%	[26••]
H2A.L[i] H2AL1/2(m) H2A.Lap2(m) H2A.Lap3(m) H2A.Lap4(m)	H2A.L.1(m)[j] H2A.L2(m) H2A.L3(m)	Certain mammals[k]	Pericentric chromatin organization in spermatids	Pericentric regions/ spermiogenic cells	Short C-terminal tail	None	41%/ 24%	[45,51,64]
H2B variants								
TS H2B.1 TH2B		Mammals	Reprograming, chromatin-to-nucleoprotamine transition	Testis, oocytes, fertilized eggs, spermatogenic cells	Similar to canonical	HIST1H2BA	86%	[26••,54,65]
H2B.W H2BFWT		Mammals	Spermiogenesis, telomere associated functions in sperm	Spermatogenic cells	Extended N-tail	H2BFWT[l]	33%	[66,67]
subH2B subH2Bv H2BL1(m)		Mammals	Spermiogenesis	Subacrosome of spermatozoa[m]	N-terminus has a bipartite nuclear localization signal	None	39%	[51,53,68]

Table 1 (*Continued*)

Variant name[a] Alt. names	Known subtypes or splice isoforms[b]	Taxonomic distribution[c]	Proposed functions	Preferential localization/tissue specificity	Sequence features	Known human genes[d]	Sequence identity[e]	Ref
H2B.E H2BE(m) H2B.1		Mouse, potentially mammals	Modulates olfactory neuron population dynamics (m)	Neurons	Very similar to canonical	HIST2H2BE	97%	[10•,69]

[a] According to new nomenclature suggested in Ref. [28•].
[b] Splice isoforms are denoted with a suffix 's', according to recommended convention [28•].
[c] The known exclusion is dinoflagellates, their chromosomes lack histones completely [55].
[d] According to HUGO nomenclature http://www.genenames.org/genefamilies/histones.
[e] Percent identity between variant and canonical histone (see Table S1 for details).
[f] In yeast H2A.X function is fulfilled by conventional H2A, while in *Drosophila melanogaster* by H2A.Z.
[g] Φ-represents a hydrophobic residue, usually Tyr in mammals, which is another phosphorylation site involved in DNA damage response [56].
[h] It is possible that mouse homolog H2A.Lap1 bears modified function with respect to human H2A.B. H2A.Lap1 has additional negative residue in acidic patch, which is thought to increase its propensity to compact nucleosomal arrays relative to human H2A.B [50].
[i] The variant family is rather variable, not all members are known to perform the same function.
[j] H2A.L1/2 is involved in forming a non-nucleosomal nucleoprotein structure in pericentric chromatin of mouse round spermatids, but might be incorporated in nucleosomes [51].
[k] Characterized in mice, so far not found in humans [51], but other mammals likely have similar proteins [28•].
[l] Another gene H2BFM encodes a somewhat similar protein at the same location on X chromosome, but has not yet been characterized.
[m] This variant is present outside of nucleus in acrosomal space of spermatozoa, extent of its involvement in nucleosome formation is unclear.

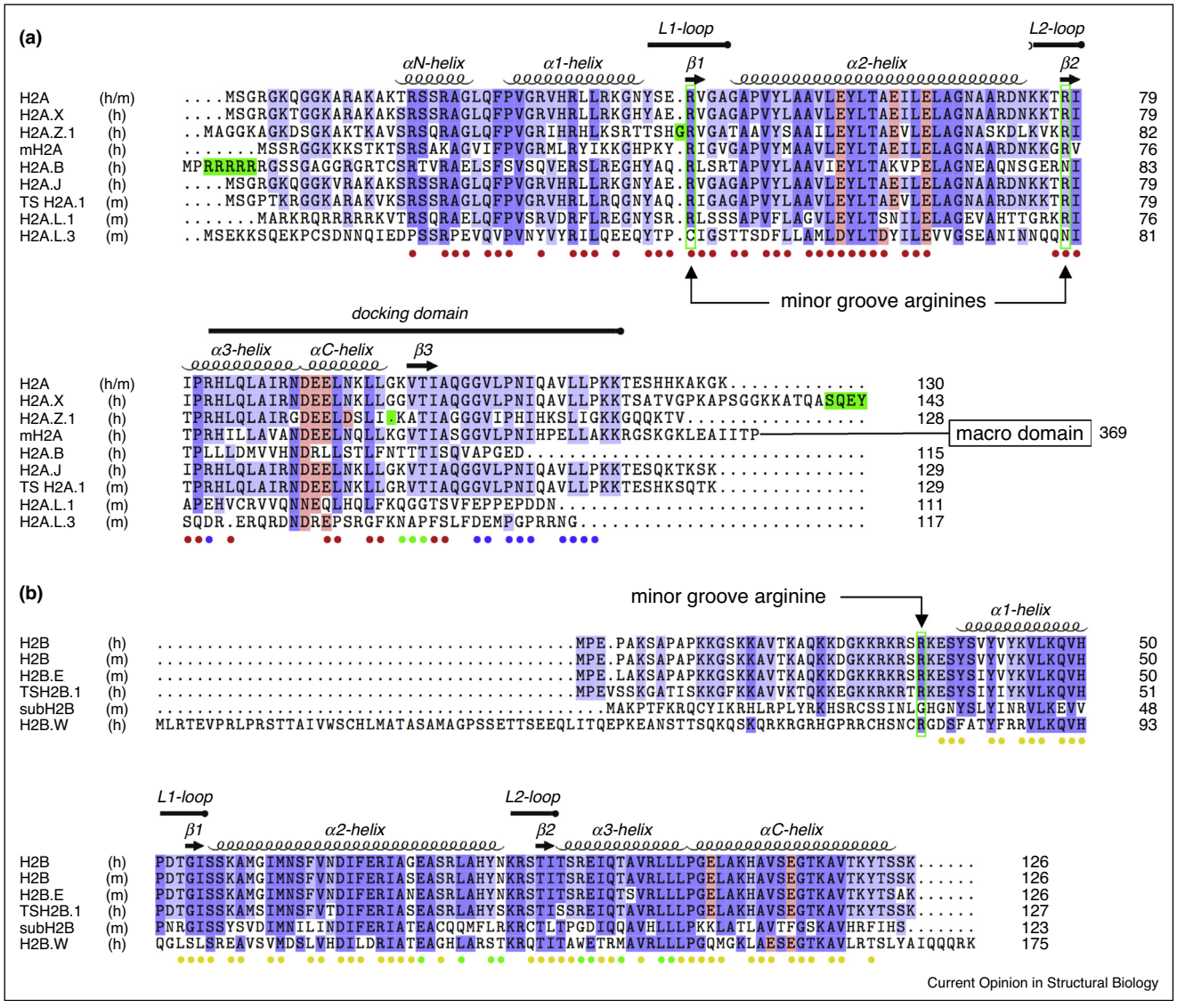
The histone fold regions (Figure 2) are well aligned based on sequence and structural comparisons and conserved between variants, with the exception of L1 and L2-loop regions of H2A and histone tails which are more divergent in terms of their sequences and lengths (Figures 2 and 3). A notable feature of H2A.Z with respect to H2A alignment is an amino acid insertion in α1-helix and one deletion in the docking domain (Figure 2). Previous studies of evolution of protein complexes showed that such insertions and deletions can mediate specific and preclude undesired interactions [34]. Moreover, certain variants differ in their amino acid composition, especially in their lysine to arginine ratio: there is only one lysine in human H2A.B compared to 14 lysines in a canonical H2A [32]. At the same time, the N-terminus of H2A histone has systematically acquired arginine amino acids as genomes expanded [35•]. In the next section we discuss how sequence differences in histone dimers are coupled with their structural variation.

Structure and stability of H2A and H2B variant nucleosomes

The atomic-resolution X-ray structures of variant nucleosomes are available for H2A.Z [36,37] and mH2A (histone domain) [38,39] variants, their structural superposition with canonical histones shows very similar conformations with the exception of the L1-loop regions of H2A (Figure 3a). In fact, this is the only region where the two H2A–H2B dimers interact. These structural differences can be explained by sequence variations as the evolutionary plasticity (the degree of structural change per unit of sequence change) is usually greater for loop regions compared to the protein core [40]. Indeed, the L1-loop region exhibits considerable sequence variation among different H2A variants, and is likely involved in conferring stability and functional specificity of variant nucleosomes. For example, a four amino acid difference between mH2A and H2A in the L1-loop was shown to be responsible for the increased salt-dependent stability of the variant histone octamer [39]. As to other variant subtypes, H2A.Z.1 and H2A.Z.2 histones differ by only S38T substitution within the histone fold. This substitution is located at the end of α1-helix, which precedes the L1-loop. X-ray structures revealed polymorphisms within L1-loop conformations between these subtypes, while *in vivo* mutagenesis experiments showed that S38T substitution might alter the mobility of different H2A.Z variants in cells [37].

Stability of nucleosomes depends on many variables and factors, such as histone sequences and structures, salt concentration, post-translational modifications and DNA sequence. There is an apparent controversy regarding the stability of some variant nucleosomes and its relation to their function. For certain variants, such as H2A.Z, no clear conclusion about their stability can be drawn due to the discrepancies between *in vivo* and *in vitro* studies [41]. However, certain sequence and structural features of

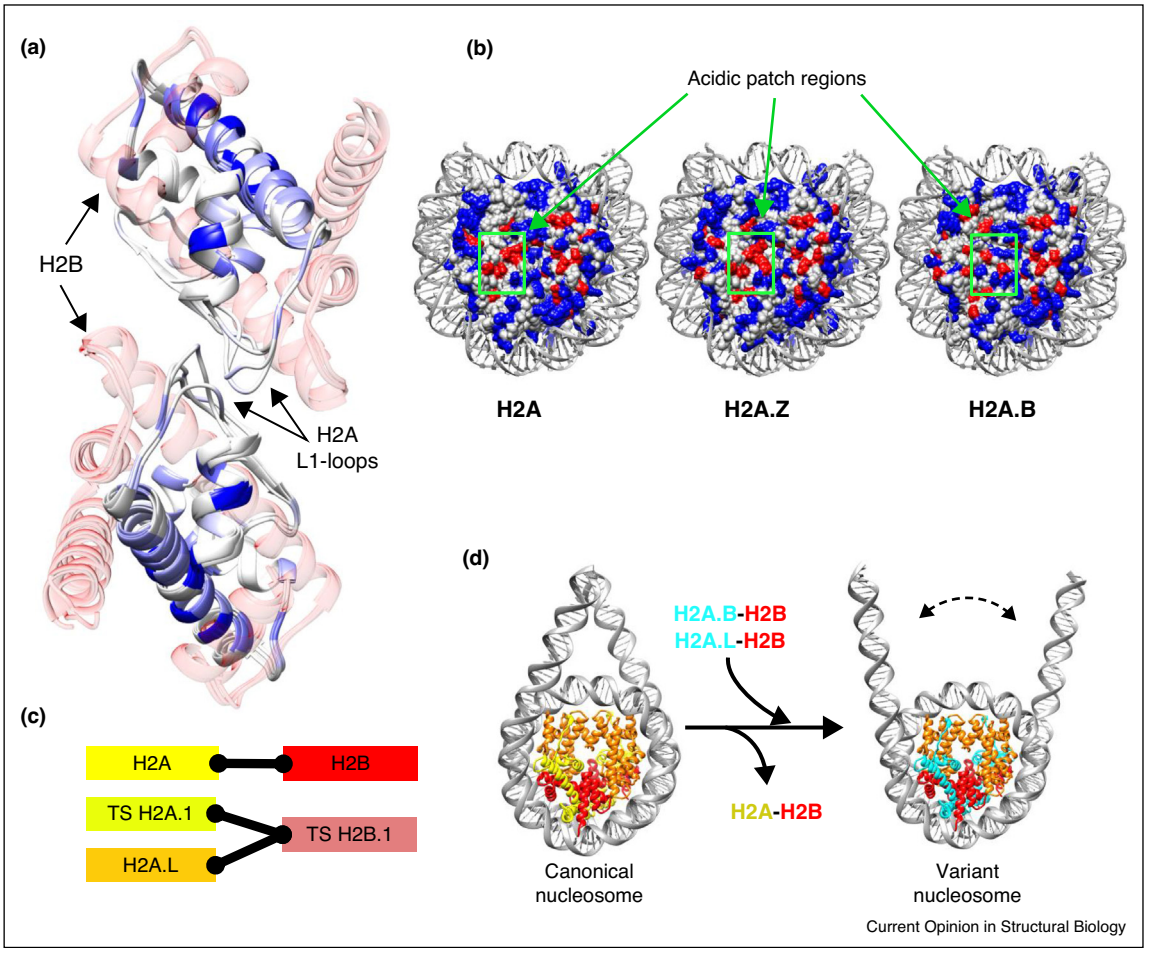
Figure 2



Multiple alignments of representative sequences of H2A **(a)** and H2B **(b)** histone variants from human and mouse (organism is specified next to the variant name as (h)-human, (m)-mouse). Variability along the sequence is highlighted in different shades of blue with dark blue corresponding to more conserved sites. The structural elements of each histone are annotated on top of the alignments. Dots beneath the alignment mark residues, which interact with residues in other histone chains, as reported by IBIS server [71] for PDB: 1AOI. The color of the dot specifies the interacting histone H3 — blue, H4 — green, H2A — yellow, H2B — red. Acidic patch residues are shaded in pink. The arginines penetrating into minor groove of DNA are shown with green frames. Variant specific features are highlighted in green and include: insertions and deletions in H2A.Z, arginine rich N-tail of H2A.B and characteristic phosphorylation motif in H2A.X. Multiple sequence alignments were visualized via TEX shade package [72].

histone variants (including the charge of the histone core [42]) might be responsible for a changed stability and have been confirmed by different experimental studies. For example, the conformation of H2A.B-variant nucleosome was recently characterized by small angle neutron scattering, which revealed that the DNA ends were detached from the histone core surface and flexibly expanded toward the solvent. At the same time, the histone tails seem to be more compact in this variant compared to tails in canonical nucleosomes [18]. H2A.B-containing nucleosomes are destabilized relative to canonical nucleosomes in a way similar to that seen in hyperacetylated histones [32], and associate with only 118–130 bp of DNA [43,44] (Figure 3d). Similarly it was shown that when the H2A.L variant is incorporated, only ~130 base pairs of DNA are wrapped around the nucleosome with subsequent nucleosome destabilization [45]. Such partial wrapping and destabilization of H2A.B and H2A.L containing

Figure 3



Structural features of variant nucleosomes. **(a)** Structural and sequence variation of H2A–H2B dimers in nucleosomes. Interacting H2A–H2B dimers from several variant nucleosome structures mH2A, H2A.Z.1 and H2A.Z.2 and canonical H2A (PDB IDs: 1KX5, 1U35, 3WA9, 3WAA), were structurally superimposed using UCSF Chimera [73]. The H2B histones are depicted in red, the H2A histones are colored in shades of blue according to site conservation between variants (blue — highly conserved, white — non-conserved). The L1 region of H2A histones shows considerable structural and sequence variation. **(b)** Differences in the surface charge and acidic patch configurations of histone octamers in variant nucleosomes. The molecular surface of the histone octamer is colored according to amino acid types (negatively charged — red, positively charged — blue, others — light blue). The acidic patch regions are highlighted with green frames. H2A and H2A.Z nucleosomes are taken from PDB 1AOI and 1F66; for H2A.B nucleosome a homology model was built using Modeller [74]. **(c)** Diagram of known preferential binding partners between various H2A and H2B variants. **(d)** Illustration of DNA opening in certain variant nucleosomes.

nucleosomes can be the result of a shorter C-terminal docking domain. In addition, H2A.Z.2.2 splice variant may form severely destabilized nucleosomes due to its truncated C-terminal tail [29•].

Nucleosomes may interact with the neighboring nucleosomes or other nuclear proteins through the acidic patch, a region on the nucleosome surface formed mainly by the acidic residues of H2A [2,46] (Figure 3b), and characterized by an increased counter ion density in its vicinity [47]. These interactions are largely responsible for chromatin compaction. Different variants show the variability in the acidic patches conferring various degrees of chromatin compaction and, consequently, may cause changes in regulation of transcription and replication. For instance, H2A.Z variants usually have an additional negatively charged residue (DEELD vs DEELN motif in the docking domain) that causes nucleosome arrays to be more compact [48], while the H2A.B variant lacks residues involved in the acidic patch resulting in a decreased tendency of chromatin fiber folding [49]. On the other hand, the mouse homolog of H2A.B (named H2A.Lap1) is known to have one additional negative residue in the acidic patch compared to human H2A.B, which increases its propensity to compact nucleosomal arrays [50].

Importantly, certain H2A–H2B variants have been shown to preferentially associate with each other and it might well be that a combinatorial complexity based on H2A–H2B variant combinations might exist and be functionally relevant in nucleosomes. For instance, some H2A.L histones display a strong preference for dimerization with TS H2B.1 rather than with the canonical H2B (Figure 3c). Furthermore, canonical H2A forms dimers with TS H2B.1 less efficiently than with canonical H2B [51]. The TS H2A.1–TS H2B.1 dimer was found to be more stable than other combinations of canonical histones [26••]. Another layer of combinatorial complexity can arise from incorporation of two different types of H2A–H2B dimers in one nucleosome. These, so-called heterotypic nucleosomes, may perform specific functions. The H2A.Z/H2A heterotypic nucleosomes, for example, were found *in vivo* in mouse trophoblast cells. These nucleosomes mark transcription start sites during the G1 phase [52]. There are some other H2A and H2B variants, like mH2A variant, known to participate in X-chromosome inactivation, which tend to form heterotypic nucleosomes with canonical histones *in vitro* [39]. Furthermore, certain histone variants such as H2A.L and subH2B [51,53] were shown to be involved in the formation of protein assemblies in spermatids, which are distinct from nucleosomes, and whose exact structure is still unclear.

Finally it should be noted that *in vivo* chromatin remodeling via histone variants goes hand in hand with histone post-translational modifications, which, together with the variants, may affect nucleosome stability and structure. Depletion of histone variants in a cell sometimes may be rescued by specific post-translational modifications of canonical histones (e.g. shown for TS H2B.1 in spermatogenic cells [54••]).

Concluding remarks and future challenges

Emerging experimental evidence highlights a delicate regulation of cellular functions through the growing number of known histone variants, which can be either universal to eukaryotes or species, tissue or cell cycle specific. H2A and H2B histones are the most sequence and structurally variable among all histones giving rise to additional variability and complexity upon H2A–H2B dimer and octamer formations. While data for H2B variants started to accumulate fairly recently, these variants have now been shown to regulate processes such as spermatogenesis, inheritance, genome reprogramming, enhancement of reprogramming in induced pluripotent stem cells, and the regulation of neuronal lifespan. It was shown that functional specificity of H2A and H2B variants might be coupled with their structure, sequence and distinct variant-specific post-translational modification patterns. In addition, the substitution of canonical histones by variants may alter nucleosome structure and stability and thus affect transcription factor binding and transcription kinetics. Given the growing appreciation of nucleosome as a dynamic entity, the ultimate goal is to understand the relation between sequence, structural variability and dynamics in nucleosomes, this in turn would shed light on their specific function.

Conflict of interest statement

Nothing declared.

Acknowledgements

We would like to thank Victor Zhurkin, Paul Talbert and Vasily Studitsky for inspiring discussions and comments. This work was supported by the Intramural Research Program of the National Library of Medicine NIH. AS is supported by the US-Russia Collaboration in the Biomedical Sciences NIH visiting fellows program.

Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at http://dx.doi.org/10.1016/j.sbi.2015.02.004.

References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- • of special interest
- •• of outstanding interest

1. Kornberg RD: **Chromatin structure: a repeating unit of histones and DNA**. *Science* 1974, **184**:868-871.

2. Luger K, Mader AW, Richmond RK, Sargent DF, Richmond TJ: **Crystal structure of the nucleosome core particle at 2.8 A resolution**. *Nature* 1997, **389**:251-260.

3. Cui F, Cole HA, Clark DJ, Zhurkin VB: **Transcriptional activation of yeast genes disrupts intragenic nucleosome phasing**. *Nucleic Acids Res* 2012, **40**:10753-10764.

4. Teif VB, Vainshtein Y, Caudron-Herger M, Mallm JP, Marth C, Hofer T, Rippe K: **Genome-wide nucleosome positioning during embryonic stem cell development**. *Nat Struct Mol Biol* 2012, **19**:1185-1192.

5. Zlatanova J, Bishop TC, Victor JM, Jackson V, van Holde K: **The nucleosome family: dynamic and growing**. *Structure* 2009, **17**:160-171.

6. Luger K, Dechassa ML, Tremethick DJ: **New insights into nucleosome and chromatin structure: an ordered state or a disordered affair?** *Nat Rev Mol Cell Biol* 2012, **13**:436-447.

7. Talbert PB, Henikoff S: **Environmental responses mediated by histone variants**. *Trends Cell Biol* 2014 http://dx.doi.org/10.1016/j.tcb.2014.07.006.

8. Lesburgueres E, Gobbo OL, Alaux-Cantin S, Hambucken A, Trifilieff P, Bontempi B: **Early tagging of cortical networks is required for the formation of enduring associative memory**. *Science* 2011, **331**:924-928.

9. Zovkic IB, Paulukaitis BS, Day JJ, Etikala DM, Sweatt JD: **Histone H2A.Z subunit exchange controls consolidation of recent and remote memory**. *Nature* 2014 http://dx.doi.org/10.1038/nature13707.

10. • Santoro SW, Dulac C: **The activity-dependent histone variant H2BE modulates the life span of olfactory neurons**. *Elife* 2012, **1**:e00070.

H2B.E is a newly characterized replication-independent histone variant found to be expressed in olfactory neurons of mice. The H2B.E expression anticorrelates with sensory activity and cellular life span, suggesting that it is used in adaptation of olfactory receptors population to the environment.

11. Hu G, Cui K, Northrup D, Liu C, Wang C, Tang Q, Ge K, Levens D, Crane-Robinson C, Zhao K: **H2A.Z facilitates access of active**

and repressive complexes to chromatin in embryonic stem cell self-renewal and differentiation. *Cell Stem Cell* 2013, **12**:180-192.

12. Simonet NG, Reyes M, Nardocci G, Molina A, Alvarez M: **Epigenetic regulation of the ribosomal cistron seasonally modulates enrichment of H2A.Z and H2A.Zub in response to different environmental inputs in carp (*Cyprinus carpio*)**. *Epigenetics Chromatin* 2013, **6**:22.

13. Tims HS, Gurunathan K, Levitus M, Widom J: **Dynamics of nucleosome invasion by DNA binding proteins**. *J Mol Biol* 2011, **411**:430-448.

14. Kulaeva OI, Hsieh FK, Chang HW, Luse DS, Studitsky VM: **Mechanism of transcription through a nucleosome by RNA polymerase II**. *Biochim Biophys Acta* 2013, **1829**:76-83.

15. Kulaeva OI, Gaykalova DA, Pestov NA, Golovastov VV, Vassylyev DG, Artsimovitch I, Studitsky VM: **Mechanism of chromatin remodeling and recovery during passage of RNA polymerase II**. *Nat Struct Mol Biol* 2009, **16**: 1272-1278.

16. Chang HW, Kulaeva OI, Shaytan AK, Kibanov M, Kuznedelov K, Severinov KV, Kirpichnikov MP, Clark DJ, Studitsky VM: **Analysis of the mechanism of nucleosome survival during transcription**. *Nucleic Acids Res* 2014, **42**:1619-1627.

17. Arimura Y, Tachiwana H, Oda T, Sato M, Kurumizaka H: **Structural analysis of the hexasome, lacking one histone H2A/H2B dimer from the conventional nucleosome**. *Biochemistry* 2012, **51**:3302-3309.

18. Sugiyama M, Arimura Y, Shirayama K, Fujita R, Oba Y, Sato N, Inoue R, Oda T, Sato M, Heenan RK *et al.*: **Distinct features of the histone core structure in nucleosomes containing the histone H2A.B variant**. *Biophys J* 2014, **106**:2206-2213.

19. Bohm V, Hieb AR, Andrews AJ, Gansen A, Rocker A, Toth K, Luger K, Langowski J: **Nucleosome accessibility governed by the dimer/tetramer interface**. *Nucleic Acids Res* 2011, **39**:3093-3102.

20. Nguyen VQ, Ranjan A, Stengel F, Wei D, Aebersold R, Wu C, Leschziner AE: **Molecular architecture of the ATP-dependent chromatin-remodeling complex SWR1**. *Cell* 2013, **154**: 1220-1231.

21. • Hondele M, Stuwe T, Hassler M, Halbach F, Bowman A, Zhang ET, Nijmeijer B, Kotthoff C, Rybin V, Amlacher S *et al.*: **Structural basis of histone H2A–H2B recognition by the essential chaperone FACT**. *Nature* 2013, **499**:111-114.
The crystal structure of H2A–H2B with Spt16M domain of chaperone FACT. The location of the binding site on H2A–H2B dimer suggests cometition between FACT and DNA binding.

22. Marino-Ramirez L, Jordan IK, Landsman D: **Multiple independent evolutionary solutions to core histone gene regulation**. *Genome Biol* 2006, **7**:R122.

23. Eirin-Lopez JM, Gonzalez-Romero R, Dryhurst D, Mendez J, Ausio J: **Long-term evolution of histone families: old notions and new insights into their mechanisms of diversification across eukaryotes**. *Evol Biol Concept Model Appl* 2009:139-162. doi 10.1007/978-3-642-00952-5_8.

24. Talbert PB, Henikoff S: **Histone variants — ancient wrap artists of the epigenome**. *Nat Rev Mol Cell Biol* 2010, **11**:264-275.

25. Marzluff WF, Wagner EJ, Duronio RJ: **Metabolism and regulation of canonical histone mRNAs: life without a poly(A) tail**. *Nat Rev Genet* 2008, **9**:843-854.

26. •• Shinagawa T, Takagi T, Tsukamoto D, Tomaru C, Huynh LM, Sivaraman P, Kumarevel T, Inoue K, Nakato R, Katou Y *et al.*: **Histone variants enriched in oocytes enhance reprogramming to induced pluripotent stem cells**. *Cell Stem Cell* 2014, **14**:217-227.
This paper characterizes histone variants TS H2A.1 and TS H2B.1 which are involved in activation of the paternal genome after fertilization. It was shown that these variants can be used to enhance induced pluripotent stem cell generation. The properties of TS H2A.1–TS H2B.1 containing nucleosomes were studied.

27. Marzluff WF, Gongidi P, Woods KR, Jin J, Maltais LJ: **The human and mouse replication-dependent histone genes**. *Genomics* 2002, **80**:487-498.

28. • Talbert PB, Ahmad K, Almouzni G, Ausio J, Berger F, Bhalla PL, Bonner WM, Cande WZ, Chadwick BP, Chan SW *et al.*: **A unified phylogeny-based nomenclature for histone variants**. *Epigenetics Chromatin* 2012, **5**:7.
This paper introduces unified nomenclature for histone variants and gives a concise overview of histone variation across species.

29. • Bonisch C, Schneider K, Punzeler S, Wiedemann SM, Bielmeier C, Bocola M, Eberl HC, Kuegel W, Neumann J, Kremmer E *et al.*: **H2A.Z.2.2 is an alternatively spliced histone H2A.Z variant that causes severe nucleosome destabilization**. *Nucleic Acids Res* 2012, **40**:5951-5964.
Characterization of a splice isoform of H2A.Z with high expression levels in brain.

30. Nishibuchi I, Suzuki H, Kinomura A, Sun J, Liu NA, Horikoshi Y, Shima H, Kusakabe M, Harata M, Fukagawa T *et al.*: **Reorganization of damaged chromatin by the exchange of histone variant H2A.Z-2**. *Int J Radiat Oncol Biol Phys* 2014, **89**:736-744.

31. Marino-Ramirez L, Levine KM, Morales M, Zhang S, Moreland RT, Baxevanis AD, Landsman D: **The Histone Database: an integrated resource for histones and histone fold-containing proteins**. *Database (Oxford)* 2011, **2011**:bar048.

32. Eirin-Lopez JM, Ishibashi T, Ausio J: **H2A.Bbd: a quickly evolving hypervariable mammalian histone that destabilizes nucleosomes in an acetylation-independent way**. *FASEB J* 2008, **22**:316-326.

33. Rogakou EP, Pilch DR, Orr AH, Ivanova VS, Bonner WM: **DNA double-stranded breaks induce histone H2AX phosphorylation on serine 139**. *J Biol Chem* 1998, **273**: 5858-5868.

34. Hashimoto K, Panchenko AR: **Mechanisms of protein oligomerization, the critical role of insertions and deletions in maintaining different oligomeric states**. *Proc Natl Acad Sci U S A* 2010, **107**:20352-20357.

35. • Macadangdang BR, Oberai A, Spektor T, Campos OA, Sheng F, Carey MF, Vogelauer M, Kurdistani SK: **Evolution of histone 2A for chromatin compaction in eukaryotes**. *Elife* 2014:e02792 http://dx.doi.org/10.7554/eLife.02792.
Using evolutionary sequence analysis and *in vitro* experiments the authors have shown that H2A N-terminal tail arginines are important for chromatin compaction and were aquired in evolution as the genomes got bigger.

36. Suto RK, Clarkson MJ, Tremethick DJ, Luger K: **Crystal structure of a nucleosome core particle containing the variant histone H2A.Z**. *Nat Struct Biol* 2000, **7**:1121-1124.

37. Horikoshi N, Sato K, Shimada K, Arimura Y, Osakabe A, Tachiwana H, Hayashi-Takanaka Y, Iwasaki W, Kagawa W, Harata M *et al.*: **Structural polymorphism in the L1 loop regions of human H2A.Z.1 and H2A.Z.2**. *Acta Crystallogr D Biol Crystallogr* 2013, **69**:2431-2439.
Crystal structures of nucleosomes containing H2A.Z.1 and H2A.Z.2 histone variants reveal unexpected structural variability in the L1 loop region despite no sequence differences in this region.

38. Chakravarthy S, Gundimella SK, Caron C, Perche PY, Pehrson JR, Khochbin S, Luger K: **Structural characterization of the histone variant macroH2A**. *Mol Cell Biol* 2005, **25**:7616-7624.

39. Chakravarthy S, Luger K: **The histone variant macro-H2A preferentially forms ''hybrid nucleosomes''**. *J Biol Chem* 2006, **281**:25522-25531.

40. Panchenko AR, Wolf YI, Panchenko LA, Madej T: **Evolutionary plasticity of protein families: coupling between sequence and structure variation**. *Proteins* 2005, **61**:535-544.

41. Bonisch C, Hake SB: **Histone H2A variants in nucleosomes and chromatin: more or less stable?** *Nucleic Acids Res* 2012, **40**:10719-10741.

42. Fenley AT, Adams DA, Onufriev AV: **Charge state of the globular histone core controls stability of the nucleosome**. *Biophys J* 2010, **99**:1577-1585.

43. Bao Y, Konesky K, Park YJ, Rosu S, Dyer PN, Rangasamy D, Tremethick DJ, Laybourn PJ, Luger K: **Nucleosomes containing the histone variant H2A.Bbd organize only 118 base pairs of DNA**. *EMBO J* 2004, **23**:3314-3324.

44. Doyen CM, Montel F, Gautier T, Menoni H, Claudet C, Delacour-Larose M, Angelov D, Hamiche A, Bednar J, Faivre-Moskalenko C *et al.*: **Dissection of the unusual structural and functional properties of the variant H2A.Bbd nucleosome**. *EMBO J* 2006, **25**:4234-4244.

45. Syed SH, Boulard M, Shukla MS, Gautier T, Travers A, Bednar J, Faivre-Moskalenko C, Dimitrov S, Angelov D: **The incorporation of the novel histone variant H2AL2 confers unusual structural and functional properties of the nucleosome**. *Nucleic Acids Res* 2009, **37**:4684-4695.

46. Chodaparambil JV, Barbera AJ, Lu X, Kaye KM, Hansen JC, Luger K: **A charged and contoured surface on the nucleosome regulates chromatin compaction**. *Nat Struct Mol Biol* 2007, **14**:1105-1107.

47. Materese CK, Savelyev A, Papoian GA: **Counterion atmosphere and hydration patterns near a nucleosome core particle**. *J Am Chem Soc* 2009, **131**:15005-15013.

48. Fan JY, Rangasamy D, Luger K, Tremethick DJ: **H2A.Z alters the nucleosome surface to promote HP1alpha-mediated chromatin fiber folding**. *Mol Cell* 2004, **16**:655-661.

49. Zhou J, Fan JY, Rangasamy D, Tremethick DJ: **The nucleosome surface regulates chromatin compaction and couples it with transcriptional repression**. *Nat Struct Mol Biol* 2007, **14**:1070-1076.

50. Soboleva TA, Nekrasov M, Pahwa A, Williams R, Huttley GA, Tremethick DJ: **A unique H2A histone variant occupies the transcriptional start site of active genes**. *Nat Struct Mol Biol* 2012, **19**:25-30.

51. Govin J, Escoffier E, Rousseaux S, Kuhn L, Ferro M, Thevenon J, Catena R, Davidson I, Garin J, Khochbin S *et al.*: **Pericentric heterochromatin reprogramming by new histone variants during mouse spermiogenesis**. *J Cell Biol* 2007, **176**:283-294.

52. Nekrasov M, Amrichova J, Parker BJ, Soboleva TA, Jack C, Williams R, Huttley GA, Tremethick DJ: **Histone H2A.Z inheritance during the cell cycle and its impact on promoter organization and dynamics**. *Nat Struct Mol Biol* 2012, **19**:1076-1083.

53. Tran MH, Aul RB, Xu W, van der Hoorn FA, Oko R: **Involvement of classical bipartite/karyopherin nuclear import pathway components in acrosomal trafficking and assembly during bovine and murid spermiogenesis**. *Biol Reprod* 2012, **86**:84.

54. •• Montellier E, Boussouar F, Rousseaux S, Zhang K, Buchou T, Fenaille F, Shiota H, Debernardi A, Hery P, Curtet S *et al.*: **Chromatin-to-nucleoprotamine transition is controlled by the histone H2B variant TH2B**. *Genes Dev* 2013, **27**:1680-1692.
TS H2B.1 histone variant participates in the final stages of chromatin to nucleoprotamine transition in male germ cells, and reassembles back after fertilization.

55. Rizzo PJ: **Those amazing dinoflagellate chromosomes**. *Cell Res* 2003, **13**:215-217.

56. Xiao A, Li H, Shechter D, Ahn SH, Fabrizio LA, Erdjument-Bromage H, Ishibe-Murakami S, Wang B, Tempst P, Hofmann K *et al.*: **WSTF regulates the H2A.X DNA damage response via a novel tyrosine kinase activity**. *Nature* 2009, **457**:57-62.

57. Millar CB: **Organizing the genome with H2A histone variants**. *Biochem J* 2013, **449**:567-579.

58. Ismail IH, Hendzel MJ: **The gamma-H2A.X: is it just a surrogate marker of double-strand breaks or much more?** *Environ Mol Mutagen* 2008, **49**:73-82.

59. Zlatanova J, Thakar A: **H2A.Z: view from the top**. *Structure* 2008, **16**:166-179.

60. Soboleva TA, Nekrasov M, Ryan DP, Tremethick DJ: **Histone variants at the transcription start-site**. *Trends Genet* 2014, **30**:199-209.

61. Gamble MJ, Kraus WL: **Multiple facets of the unique histone variant macroH2A: from genomics to cell biology**. *Cell Cycle* 2010, **9**:2568-2574.

62. Gonzalez-Romero R, Mendez J, Ausio J, Eirin-Lopez JM: **Quickly evolving histones, nucleosome stability and chromatin folding: all about histone H2A.Bbd**. *Gene* 2008, **413**:1-7.

63. Chadwick BP, Willard HF: **Histone H2A variants and the inactive X chromosome: identification of a second macroH2A variant**. *Hum Mol Genet* 2001, **10**:1101-1113.

64. Wu F, Caron C, De Robertis C, Khochbin S, Rousseaux S: **Testis-specific histone variants H2AL1/2 rapidly disappear from paternal heterochromatin after fertilization**. *J Reprod Dev* 2008, **54**:413-417.

65. Pentakota SK, Sandhya SAPS, Chandra N, Satyanarayana Rao MR: **Mapping post-translational modifications of mammalian testicular specific histone variant TH2B in tetraploid and haploid germ cells and their implications on the dynamics of nucleosome structure**. *J Proteome Res* 2014 http://dx.doi.org/10.1021/pr500597a.

66. Boulard M, Gautier T, Mbele GO, Gerson V, Hamiche A, Angelov D, Bouvet P, Dimitrov S: **The NH2 tail of the novel histone variant H2BFWT exhibits properties distinct from conventional H2B with respect to the assembly of mitotic chromosomes**. *Mol Cell Biol* 2006, **26**:1518-1526.

67. Churikov D, Siino J, Svetlova M, Zhang K, Gineitis A, Morton Bradbury E, Zalensky A: **Novel human testis-specific histone H2B encoded by the interrupted gene on the X chromosome**. *Genomics* 2004, **84**:745-756.

68. Aul RB, Oko RJ: **The major subacrosomal occupant of bull spermatozoa is a novel histone H2B variant associated with the forming acrosome during spermiogenesis**. *Dev Biol* 2002, **242**:376-387.

69. Collart D, Romain PL, Huebner K, Pockwinse S, Pilapil S, Cannizzaro LA, Lian JB, Croce CM, Stein JL, Stein GS: **A human histone H2B.1 variant gene, located on chromosome 1, utilizes alternative 3′ end processing**. *J Cell Biochem* 1992, **50**:374-385.

70. Wang D, Ulyanov NB, Zhurkin VB: **Sequence-dependent Kink-and-Slide deformations of nucleosomal DNA facilitated by histone arginines bound in the minor groove**. *J Biomol Struct Dyn* 2010, **27**:843-859.

71. Shoemaker BA, Zhang D, Tyagi M, Thangudu RR, Fong JH, Marchler-Bauer A, Bryant SH, Madej T, Panchenko AR: **IBIS (Inferred Biomolecular Interaction Server) reports, predicts and integrates multiple types of conserved interactions for proteins**. *Nucleic Acids Res* 2012, **40**:D834-D840.

72. Beitz E: **TEXshade: shading and labeling of multiple sequence alignments using LATEX2 epsilon**. *Bioinformatics* 2000, **16**:135-139.

73. Pettersen EF, Goddard TD, Huang CC, Couch GS, Greenblatt DM, Meng EC, Ferrin TE: **UCSF Chimera — a visualization system for exploratory research and analysis**. *J Comput Chem* 2004, **25**:1605-1612.

74. Sali A, Blundell TL: **Comparative protein modelling by satisfaction of spatial restraints**. *J Mol Biol* 1993, **234**:779-815.