© 2021. Published by The Company of Biologists Ltd | Journal of Cell Science (2021) 134, jcs244749. doi:10.1242/jcs.244749

CELL SCIENCE AT A GLANCE

SUBJECT COLLECTION: EXPLORING THE NUCLEUS

Histone variants at a glance

Paul B. Talbert* and Steven Henikoff

ABSTRACT

Eukaryotic nucleosomes organize chromatin by wrapping 147 bp of DNA around a histone core particle comprising two molecules each of histone H2A, H2B, H3 and H4. The DNA entering and exiting the particle may be bound by the linker histone H1. Whereas deposition of bulk histones is confined to S-phase, paralogs of the common histones, known as histone variants, are available to carry out functions throughout the cell cycle and accumulate in post-mitotic cells. Histone variants confer different structural properties on nucleosomes by wrapping more or less DNA or by altering nucleosome stability. They carry out specialized functions in DNA repair, chromosome segregation and regulation of transcription initiation, or perform tissue-specific roles. In this Cell Science at a Glance article and the accompanying poster, we briefly examine new insights into histone origins and discuss variants from each of the histone families, focusing on how structural differences may alter their functions.

KEY WORDS: Chromatin, Epigenetics, Replacement histones

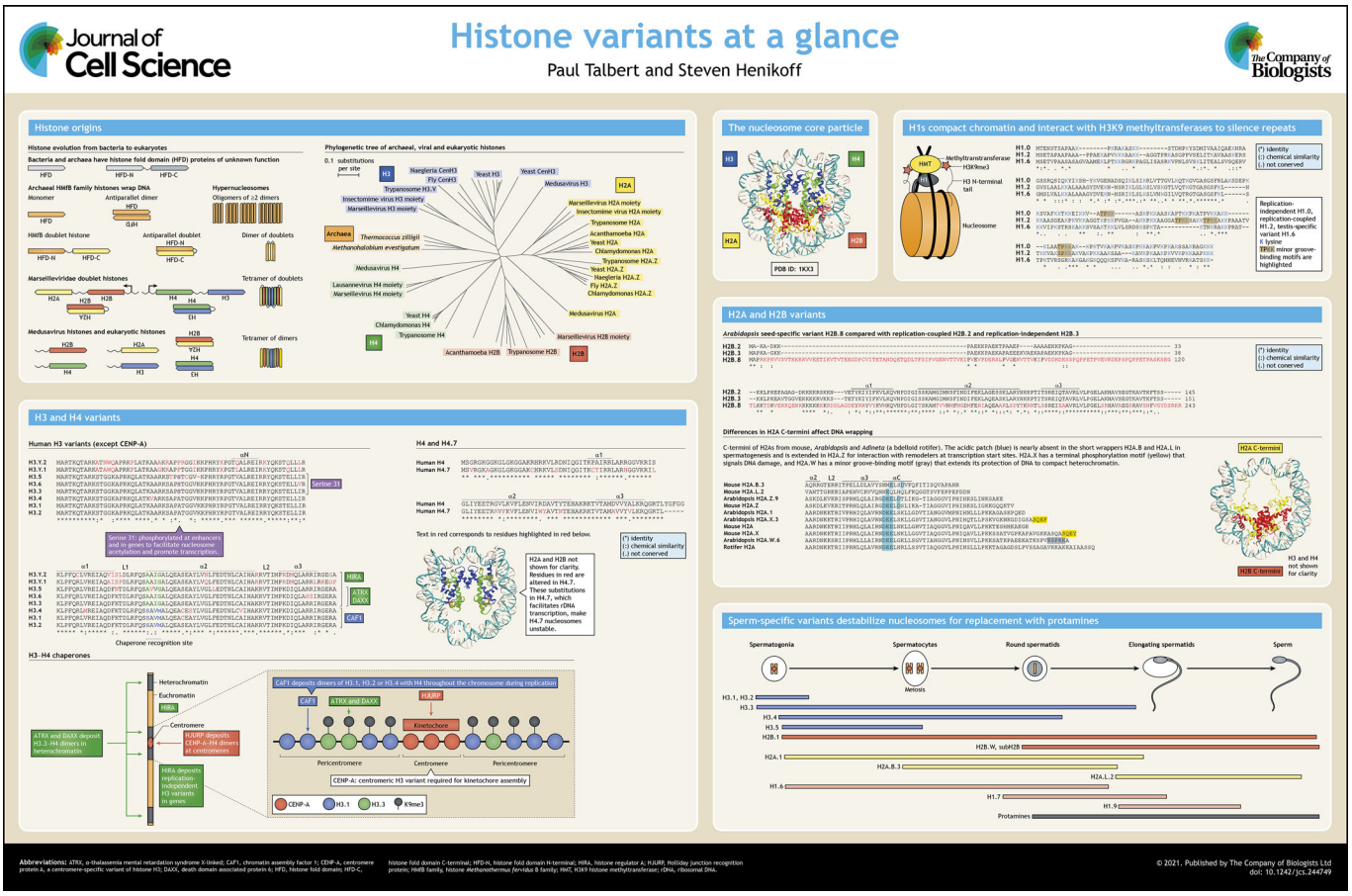
Howard Hughes Medical Institute, Fred Hutchinson Cancer Research Center, 1100 Fairview Avenue N, Seattle, WA 98109, USA.

*Author for correspondence (ptalbert@fredhutch.org)

P.B.T., 0000-0003-2563-3878; S.H., 0000-0002-7621-8685

Introduction

Eukaryotic chromatin is organized by nucleosomes, which package and regulate access to DNA and whose primary role may be to globally repress transcription (Kornberg and Lorch, 2020). Nucleosomes are formed from an octameric core particle of two molecules each of the histones H2A, H2B, H3 and H4, which wraps 147 bp of DNA (Luger et al., 1997) (see poster). Histone fold domain proteins (HFDs) have a long history in all domains of life (Box 1), but a eukaryotic innovation is their ability to heterodimerize in specific pairs, H3 with H4, and H2A with H2B, which can further associate through four-helix bundles, H3 with H3,



Journal of Cell Science (2021) 134, jcs244749. doi:10.1242/jcs.244749

Glossary

+1 nucleosome: the first nucleosome of a gene, immediately downstream of the nucleosome-depleted region at the transcription start site.
Azoospermia: absence of motile sperm in the semen. It can be due to an obstruction of the reproductive tract or to the failure to produce sperm.
Blastopore: a mouth-like indentation or opening into the developing gastrula. In vertebrates, it becomes the anus.
Cohesins: ring-like protein complexes that hold loops of DNA or sister chromatids together.
HMfB family: a family of HFD proteins in archaea (named for histone *Methanothermus fervidus* B) that can wrap DNA but have no significant primary sequence similarity with to eukaryotic histones. They can dimerize, form tetramers or form more extended oligomers.
Protamines: small arginine-rich proteins that replace histones in sperm of many vertebrates to further condense the chromatin, presumably to improve sperm motility and DNA protection. Similar proteins are found in many invertebrates.
Rapid response genes: genes typically activated within minutes of some stimulus.
Sex body: the condensed X and Y chromosomes in mammalian spermatocytes, as the visual manifestation of undergoing meiotic sex chromosome inactivation.

Box 1. Histone origins

Histone fold domain (HFD) proteins are found in all three cellular domains and in some viruses (see poster), although two families of HFD proteins that are found in both bacteria and archaea have unknown functions and DNA-binding abilities (Alva and Lupas, 2019). Many archaea additionally have one or more histones of the histone *Methanothermus fervidus* B (HMfB) family, which bind to DNA and are more similar in structure to eukaryotic histones, although they are not specifically related in sequence and mostly lack the unstructured tails of eukaryotic histones (Henneman et al., 2018). HMfB family histones fold together, usually in homomeric dimers, which can be further polymerized through four-helix bundles to form tetrameric HFD particles that wrap 60 bp of DNA or to form more extended DNA-wrapping polymers (Mattiroli et al., 2017), termed hypernucleosomes (Henneman et al., 2018) (see poster). Some also have tandemly coupled, diverged HFDs that fold together and then dimerize into particles with four HFDs. A similar organization of pairs of tandem HFDs has recently been found in Marseilleviridae, where the tandem HFDs have tails and have distant similarity with H4–H3 and H2B–H2A (Erives, 2017). Medusavirus (Yoshikawa et al., 2019), which belongs to the same large clade of nucleo-cytoplasmic large DNA viruses as the Marseilleviridae, encodes individual core histones specifically related to H2A, H2B, H3 and H4. The individual viral histone domains branch in a phylogenetic tree at the base of all eukaryotic histones of the corresponding histone family (see poster), suggesting these viral histones represent an intermediate state in the evolution of histones from archaeal-like HFDs to modern eukaryotic histones. It seems likely that these viruses captured histones at different times in the evolution of proto-eukaryotic histones and adapted them to package their own genomes. Medusavirus also has an H1 linker histone, but it is most closely related (43% identical) to the H1 of its protist host *Acanthamoeba castellani*, suggesting a more recent derivation than the core histones, which do not show a specific relationship to *Acanthamoeba*.

and H4 with H2B, to form a central H3–H4 tetramer flanked by two H2A–H2B dimers (see poster). The HFDs have unstructured tails that are the sites of post-translational modifications. A nucleosome may be further stabilized by the H1 'linker' histone, which interacts with the DNA that links adjacent nucleosomes. 'Bulk' or 'replication-coupled' (RC) histones are primarily deposited during replication. In animals, RC histones are encoded by multiple genes with special mRNAs that lack introns and polyadenylated (polyA) tails, but instead have a 3′ stem-loop structure (Marzluff, 2005). A subset of these genes can also form transcripts with polyA tails in differentiated tissues that no longer replicate, and therefore are available to replace evicted histones (Lyons et al., 2016). Some histone genes, however, encode distinct paralogs that differ in amino acid sequence from their RC counterparts. These histone 'variants' are usually encoded by single genes that have both introns and polyA tails, and are typically available for deposition throughout the cell cycle [replication-independent (RI)], replacing evicted RC histones. Histone variants often confer different structural properties on nucleosomes and often have distinct functions in cell division, transcription, DNA repair, differentiation and chromatin remodeling. We have previously reviewed the phylogenomic scope and dynamics of histone variants (Talbert and Henikoff, 2010, 2014, 2017). In this Cell Science at a Glance article, and the accompanying poster, we instead focus on recent insights into histone origins, and present an overview of variants in each histone class, emphasizing mammalian variants.

H3 variants

Most multicellular and some unicellular eukaryotes have separate RC and RI H3 paralogs (Postberg et al., 2010; Waterborg, 2012), although in organisms with only one form of H3, such as yeast, that form is deposited both during and outside of replication and more closely resembles RI variants in sequence (Ahmad and Henikoff, 2002a). In most animals, the RC and RI H3s correspond to human H3.2 and H3.3, respectively. Human H3.1, which differs in a single amino acid from H3.2, is a mammal-specific RC paralog (Postberg et al., 2010). Differences between RC and RI H3s in residues 87–90 in the N-terminal end of α-helix 2 (SAVM versus AAIG in animals) specify different modes of chromatin assembly (Ahmad and Henikoff, 2002a,b) mediated by different chaperones: the chromatin assembly factor 1 (CAF1; CHAF1A, CHAF1B and RBBP4) complex for RC H3.1–H4 dimers, and histone regulator protein A (HIRA) (Tagami et al., 2004), or α-thalassemia mental retardation syndrome X-linked (ATRX) and death domain-associated protein 6 (DAXX) (Elsässer et al., 2012) for RI H3.3–H4 dimers (see poster). Whereas CAF1 distributes H3.1 and H3.2 throughout the genome during replication, HIRA replaces nucleosomes lost during transcription with H3.3 nucleosomes, thereby depositing H3.3 in active genes, promoters, enhancers, transcription termination sites and other locations of histone turnover (Mito et al., 2005, 2007; Wirbelauer et al., 2005). ATRX and DAXX deposit H3.3 in telomeres, imprinted genes and other heterochromatic loci, where it is modified with the trimethlyated lysine 9 heterochromatic mark (denoted H3K9me3) to maintain heterochromatin at these locations (Drane et al., 2010; Elsässer et al., 2015; Goldberg et al., 2010; Voon et al., 2015; Wong et al., 2010). The difference between deposition pathways for RC H3.1 and RI H3.3 underlies the more-severe effects of pediatric diffuse midline gliomas caused by H3.1K27M mutations than those caused by H3.3K27M mutations, both of which inhibit global formation of H3K7me3, which normally prevents tumorigenesis (Sarthy et al., 2020).

Although *Caenorhabditis* H3.3 is not required for viability (Delaney et al., 2018), in H3.3-deficient *Drosophila* males, chromosomes fail to condense properly for meiosis and undergo segregation defects (Sakai et al., 2009). In mice with H3.3 knockout mutations, failure to maintain heterochromatin leads to mitotic abnormalities and embryonic lethality (Jang et al., 2015). In H3.3-reduced male mice, apoptosis of spermatogonia and spermatocytes occurs, and the transition from histones to

Journal of Cell Science (2021) 134, jcs244749. doi:10.1242/jcs.244749

protamines (see Glossary) during spermatogenesis is incomplete (Yuen et al., 2014).

RI H3.3s also typically differ from RC H3s in having a serine or threonine residue in the N-terminal tail (serine 31 in animals) (Waterborg and Robertson, 1996), which is phosphorylated in the pericentromere (Hake et al., 2005) and at telomeres (Wong et al., 2009) during metaphase by checkpoint kinase 1 (CHK1; also known as CHEK1) and Aurora B kinases (Chang et al., 2015; Li et al., 2017). In euchromatin of mouse embryonic stem cells, phosphorylation of H3.3S31 (H3.3S31ph) by CHK1 promotes p300-dependent acetylation at enhancers, which facilitates differentiation of these cells (Martire et al., 2019). In *Xenopus* embryos, H3.3S31ph is necessary for blastopore (see Glossary) closure, and nucleosomes with the phosphomimic H3.3S31D are enriched for H3.3K27 acetylation, which is permissive for gene activation (Sitbon et al., 2020). In addition, in mouse cells, such as macrophages, stimulated to rapid response (see Glossary), H3.3S31 is co-transcriptionally phosphorylated over stimulation-induced gene bodies and interacts directly with the histone lysine N-methyltransferase protein SETD2 to promote H3K36me3 formation in genes and to eject the co-repressor ZMYND11, enhancing transcription (Armache et al., 2020).

In *Arabidopsis*, H3.3T31 inhibits the H3K27 methyltransferases *Arabidopsis* trithorax-related proteins 5 and 6 (ATXR5 and ATXR6). ATXR5 and ATXR6 themselves recognize H3.1A31 and methylate K27 to assure this heterochromatic mark is inherited through replication while avoiding silencing active chromatin containing H3.3 (Jacob et al., 2014). Similarly, the H3.3-like sperm-specific variant H3.10 is altered near K27 and not recognized by ATXR5 or ATXR6, contributing to the loss of H3K27me3 and the expression of spermatogenesis genes (Borg et al., 2020). This suggests that a serine or threonine at residue 31 of H3.3 may be conserved across eukaryotic kingdoms to facilitate nucleosome acetylation and enhanced access for transcriptional machinery and to prevent silencing.

Human H3.Y.1 and H3.Y.2 (H3.X) are H3.3-like variants expressed in early cleavage-stage embryos, where they are induced by brief expression of the double homeobox protein 4 (DUX4) transcription factor (Resnick et al., 2019). They become incorporated into DUX4-inducible genes and promote perdurance of expression of these genes. Similar to what is found for the H3R42H change in mouse H3.4 (Table 1), the H3R42K change in H3.Y.1 and H3.Y.2 create more flexible DNA ends that bind histone H1 less efficiently in both homotypic H3.Y.1 nucleosomes and heterotypic H3.Y.1-H3.3 nucleosomes (Kujirai et al., 2016). Despite having a chaperone recognition sequence identical to H3.3 in the α2-helix, H3.Y.1 and H3.Y.2 nucleosomes are only deposited by HIRA and not by DAXX, which requires the C-terminus of H3.3 (Zink et al., 2017).

Other human variants, H3.6, H3.7 and H3.8, are tissue-specific (Taguchi et al., 2017). H3.6 forms nucleosomes that seem to have a deposition pattern similar to H3.3 nucleosomes, but are less stable because of an I62V change that reduces hydrophobic contact with H4. H3.7 does not form nucleosomes *in vitro* and H3.8 has a very low but detectable expression level in ovary, colon and kidney (Taguchi et al., 2017). Tissue-specific H3 variants are fairly common in ciliates, plants and animals (Ingouff et al., 2007; Maehara et al., 2015; Moosmann et al., 2011; Postberg et al., 2010), and are often expressed during animal spermatogenesis, where they may destabilize nucleosomes in the transition from histones to protamines (Table 1).

The most divergent and universal H3 variant is the centromere-specific variant cenH3, known as Cse4 in budding yeast, CENP-A in animals and CENH3 in plants. cenH3 serves as a foundational protein required to build the kinetochore in most eukaryotes, but surprisingly it has been lost in trypanosomes (Akiyoshi and Gull, 2014), the fungus *Mucor* (Navarro-Mendoza et al., 2019) and in four clades of holocentric insects (Drinnenberg et al., 2014), which comprise thousands of species. The HFDs of cenH3s are generally only ∼50% conserved with those of other H3s and their N-terminal tails and loop 1 are typically longer than H3 tails and cannot be

Table 1. Mammalian sperm- and oocyte-specific histone variants

Name	Expression	Comments	Reference(s)
H3.4 (H3t)	Spermatogonia, prior to entry into meiosis; RC	Less stable nucleosomes than H3.1 (human); absence leads to azoospermia (see Glossary)	Tachiwana et al., 2008; Ueda et al., 2017
H3.5	Spermatogonia to leptotene (human)	Unstable; reduction associated with non-obstructive azoospermia	Shiraishi et al., 2018; Urahama et al., 2016
H2B.1 (TH2B)	Spermatogonia, persists into embryo; also in oocytes	Knockout has no defect; tagged version is sterile	Beedle et al., 2019; Montellier et al., 2013; Shinagawa et al., 2014
H2B.W	Round spermatid to sperm, at telomeres (human, Chinese hamster cells)	Some alleles associated with increased infertility; divergent tail fails to recruit condensation factors	Boulard et al., 2006; Churikov et al., 2004; Gineitis et al., 2000; Teimouri et al., 2018
subH2B	Extranuclear subacrosome (bull, mouse)	Mediates nuclear docking of acrosomal vescicle	Aul and Oko, 2002; Tran et al., 2011
H2A.1 (TH2A)	Spermatogonia, oocytes, embryos; co-expressed with H2B.1	Disruption of H2A.1 and H2B.1 causes sterility in males, maternal effect on zygotic activation	Shinagawa et al., 2015; Shinagawa et al., 2014
H2A.B.3	Pachytene to round spermatid; oocytes, early embryos	Short wrapper; knockout has altered splicing, reduced fertility	Anuar et al., 2019; Soboleva et al., 2017; Wu et al., 2014
H2A.L.2	Elongating and condensing spermatids	Short wrapper; with H2B.1 loads transition proteins, which recruit protamines	Barral et al., 2017
H1.6 (H1T)	Primary spermatocytes to round spermatids, ESCs, cancer cells (human)	Binds to rDNA, LINEs, SINEs, LTRs; unnecessary for fertility	Mahadevan et al., 2020; Tani et al., 2016
H1.7 (H1T2)	Apical pole of round and elongating spermatids	Needed for elongation, condensation, protamine assembly, fertility	Martianov et al., 2005; Tanaka et al., 2005
H1.8 (H1oo)	Oocyte germinal vescicle through to 2–4-cell embryo	Homologous to 'cleavage stage' H1s of frogs and sea urchins	Tanaka et al., 2001
H1.9 (HILS1)	Elongating and condensing spermatids	Binds LINEs, LTRs and satellite repeats	Mishra et al., 2015; Yan et al., 2003

LINE, long interspersed nuclear elements; SINE, short interspersed nuclear element; LTR, long terminal repeat. Sperm-specific variants often are less stable, probably to facilitate replacement of histones with protamines in mature sperm. Oocyte histones are important for zygotic gene activation. Results are from mouse, except as noted.

Journal of Cell Science (2021) 134, jcs244749. doi:10.1242/jcs.244749

aligned between divergent species (Talbert et al., 2008). Distinct chaperones exist for cenH3s (Camahort et al., 2007; Chen et al., 2014; Dunleavy et al., 2009; Mizuguchi et al., 2007; Stoler et al., 2007), which, at least in *Drosophila*, recognize loop 1 and co-evolve with it (Rosin and Mellone, 2016). Unlike most other histones, cenH3s in animals and plants evolve rapidly, presumably because competition between centromeres for inclusion in the egg in female meiosis drives rapid evolution of centromeres (Henikoff et al., 2001). This phenomenon, known as centromere drive, can result in the expansion of centromeres (reviewed in Kursel and Malik, 2018; Rosin and Mellone, 2017) and/or in centromeres with a 10 bp periodicity of A/T dinucleotides, which favor stable wrapping of nucleosomes (Talbert and Henikoff, 2020). cenH3s are thought to then evolve to restore equal segregation of unequal centromeres. This conflict highlights the evolutionary tension between genetic (DNA-dependent) and epigenetic (DNA-sequence-independent) pressures on cenH3 evolution (Dawe and Henikoff, 2006).

In summary, from a likely single H3.3-like ancestor, H3 paralogs diversified to specialize for roles in replication, kinetochore formation and spermatogenesis and for differential modification to promote or resist gene silencing. H3 variants can differ in their stability, their modifications, the enzymes or structures that interact with them, or, through their chaperones, differ in where and when they are assembled into nucleosomes.

H4 variants

H4 variants are infrequent, but humans have H4.7 (H4.G), which has a truncated C-terminus and only 85% amino acid identity with RC H4 (Long et al., 2019). The *H4C7* gene is encoded in the histone 1 gene cluster along with RC histones, but forms a polyadenylated transcript that is expressed at low, but elevated, levels in breast and colon cancer cell lines relative to normal breast tissue (Long et al., 2019). H4.7 localizes to the nucleolus through an interaction of its α3-helix with nucleophosmin 1 and appears to form unstable nucleosome-like structures and less compact chromatin on rDNA, which promotes rRNA transcription (Pang et al., 2020).

H2B variants

Whereas H3 and H4 RC genes usually encode only one or a few distinct proteins, RC H2A and H2B genes often encode several different proteins. In *Arabidopsis*, ten H2Bs differ mostly in the lysine-rich N-terminal tails and in tissue-specific expression (Bergmuller et al., 2007). Although most appear to be RC H2Bs, H2B.3 is enriched in mature leaves and in nucleosomes containing H3.3 and/or H2A.Z, consistent with it being a RI variant, and H2B.8 is enriched in dry seed (Jiang et al., 2020) (see poster). A similar heterogeneity of RC H2B and H2A proteins is found in humans, mice and sea urchins (Marzluff et al., 2002, 2006). In mice, the mRNA for H2B.21 (H2B.E), which differs in five amino acids from the H2B consensus, is transcribed from the HIST2 gene cluster of RC histones but has a polyA tail, typical of RI variants, and is expressed exclusively in the main olfactory epithelium and the vomeronasal organ (Santoro and Dulac, 2012). A model of H2B.21 function proposes that increased neuronal activity due to exposure to olfactory stimulants reduces H2B.21 in the corresponding olfactory receptor cells and increases neuronal longevity, whereas stimulant deprivation leads to increased H2B.21 levels and shorter neuronal life span. Other H2B variants are mainly restricted to roles in spermatogenesis (Table 1).

H2A variants

H2A variants occupy the entry and exit positions along the wrap of nucleosomal DNA, making them ideal components to control access to the DNA. Bulk RC H2As in humans differ mostly at their C-termini (Marzluff et al., 2002). The mammalian RI variant H2A.22 (H2A.J) differs from bulk H2As because of an A11V change in the N-terminus and by several residues at its C-terminus, including a potential SQ phosphorylation site, and promotes senescence-associated inflammatory gene expression in cells with persistent DNA damage (Contrepois et al., 2017). Other RI variants also differ at their C-termini, which can alter the extent of DNA wrapping (Doyen et al., 2006b; Osakabe et al., 2018) (see poster). They may also differ at the acidic patch, which interacts with chromatin remodelers, and in loop 1, where the two copies of H2A in a nucleosome contact each other to stabilize nucleosomes (Osakabe et al., 2018).

H2A.X – DNA damage and beyond

H2A.X differs from bulk H2As in possessing a C-terminal SQD/EΦ phosphorylation motif (where Φ indicates a hydrophobic residue, often phenylalanine or tyrosine). In response to DNA damage, the serine (S139 in humans) becomes phosphorylated (then known as γH2A.X) and recruits repair enzymes to double-strand breaks (reviewed in Talbert and Henikoff, 2014). In single-celled eukaryotes, such as yeast, H2A.X can be the primary form of H2A, whereas in multicellular eukaryotes H2A.X is usually closely related to RC H2As in the same group of multicellular organisms. Because of its conserved function in the DNA damage response, it seems probable that H2A.X is ancestral, and that the various RC H2As are derived from it (Talbert and Henikoff, 2010). In humans, H2A.X differs from other H2A variants in producing mRNAs with either a stem-loop structure typical of RC H2As or a polyadenylated transcript like RI variants (Mannironi et al., 1989). The stem-loop structure may be relevant to deposition of H2A.X during UVC-induced DNA damage repair (Piquet et al., 2018), where it may serve to augment the amount of H2A.X available to be phosphorylated. Sites prone to DNA damage in cycling cells become enriched with H2A.X, whereas resting cells do not show such enrichment (Seo et al., 2012). γH2A.X stimulates the polymerase activity of poly-ADP-ribose polymerase 1 (PARP1), which detects double-strand breaks and recruits additional repair factors (Sharma et al., 2019).

The functions of H2A.X are not limited to DNA repair. In mouse embryonic stem cells, H2A.X is deposited at rDNA promoters and recruits the nucleolar remodeling complex to repress rDNA transcription and limit cell proliferation (Eleuteri et al., 2018), independently of S139 phosphorylation. In *Xenopus*, a variant of H2A.X (H2A.X-F) is abundant and is phosphorylated in oocytes, eggs and early embryos in the absence of DNA damage (Shechter et al., 2009), suggesting it has a role in promoting the rapid early divisions or in activating the zygotic genome. H2A.X-knockout mice are viable but show repair defects and genomic instability, and males are infertile (Celeste et al., 2002), presumably because the X and Y chromosomes in spermatocytes fail to form the sex body (see Glossary) or initiate meiotic sex chromosome inactivation (Fernandez-Capetillo et al., 2003).

H2A.Z – a transcriptional regulator

The RI variant H2A.Z has a prominent and complex role in transcriptional regulation (reviewed in Giaimo et al., 2019). It is absent from the early-diverging metamonads *Giardia* and *Trichomonas* (Dalmasso et al., 2011), but is strongly conserved in nearly all other eukaryotes, where it commonly occurs in the +1 nucleosome position (see Glossary) of genes (Raisner et al., 2005) and appears to poise genes for transcription and promote RNA

Journal of Cell Science (2021) 134, jcs244749. doi:10.1242/jcs.244749

polymerase II (RNAPII) recruitment (Adam et al., 2001). H2A.Z has an extended acidic patch that stimulates ATP-dependent remodelers (Dann et al., 2017; Goldman et al., 2010). The yeast SWR1 complex replaces H2A–H2B dimers with H2A.Z–H2B dimers in the +1 nucleosome of transcribed genes (Ranjan et al., 2013) and even in transcribed upstream antisense noncoding RNAs (Bagchi et al., 2020). *In vitro*, H2A.Z nucleosomes have a lower breaking force in an optical tweezer assay (Rudnizky et al., 2016) than bulk H2A nucleosomes. This may underlie the ability of H2A.Z to lower the barrier to transcription of the +1 nucleosome in *Drosophila* cells (Weber et al., 2014), which occurs through the loss of an H2A.Z–H2B dimer and its DNA contacts (Ramachandran et al., 2017), as well as its lower thermal stability (Osakabe et al., 2018). This presumably facilitates the eviction of H2A.Z without loss of H3 in the thermal response of *Arabidopsis* (Cortijo et al., 2017). In yeast, eviction of H2A.Z is dependent on the transcription pre-initiation complex (Tramantano et al., 2016) and serine-5-phophorylated RNAPII (Wu et al., 2009). Apparently contradictory effects of H2A.Z on gene activation or silencing in different contexts are mediated at least in part by acetylation of the N-terminus or monoubiquitylation of the C-terminus, respectively (Giaimo et al., 2019). H2A.Z is also enriched at enhancers, where it is necessary for recruitment of RNAPII and cohesins (see Glossary), which mediate enhancer–promoter interaction, and for the transcription of enhancer RNAs (Brunelle et al., 2015).

Chordates encode two H2A.Z proteins (Eirín-López et al., 2009), H2A.Z.1 and H2A.Z.2, which differ by three conserved amino acids (reviewed in Cheema et al., 2020). H2A.Z.1-knockout animals die during early development (Faast et al., 2001) and H2A.Z.2 is required for melanocyte development in zebrafish (Raja et al., 2020). In humans, H2A.Z.1 and H2A.Z.2 have qualitatively similar, but quantitatively different, expression patterns, with a subset of H2A.Z.2-biased enhancers affecting genes that are downregulated in the cranio-facial abnormality disease floating harbor syndrome (Greenberg et al., 2019). In primates, H2A.Z.2 has two splice variants, H2A.Z.2.1 and H2A.Z.2.2 (Bonisch et al., 2012). H2A.Z.2.2 destabilizes nucleosomes due to its shorter C-terminus, which resembles short H2As in length (Table 1). Despite the requirements for H2A.Z.1 in embryonic development, and the role of H2A.Z.2 in melanocyte development and cranio-facial formation, a double knockout of these two genes in mouse skeletal muscle has little effect on either basal or induced transcription, calling into question whether H2A.Z plays any necessary part in transcription or simply has a replacement function (Belotti et al., 2020). The strong conservation of H2A.Z at transcription start sites across diverse eukaryotic kingdoms is hard to rationalize if it has no role in transcription, since other replacement H2As, such as H2A.X are readily available, in most cases. However, if the role of H2A.Z is to help attract RNAPII or reduce the barrier of the +1 nucleosome, there are likely redundant pathways and cofactors for accomplishing this.

H2A.W and macroH2A

Arabidopsis thaliana has four H2A variants – bulk H2A, H2A.X, H2A.Z and the plant-specific H2A.W – all of which form homotypic nucleosomes (Osakabe et al., 2018). With its extended C-terminus (which has a putative minor groove-binding motif KSPKK), H2A.W protects an additional 10–15 bp of linker DNA beyond the 147 bp of most nucleosomes from micrococcal nuclease. H2A.W is found in heterochromatin (Yelagandula et al., 2014), where it may serve a silencing function. Like H2A.X, it can be phosphorylated during the DNA damage response (Lorković et al., 2017). Extended C-terminal tails are also found in H2As of bdelloid rotifers, freshwater microorganisms, which replace conventional H2A, H2A.X and H2A.Z, and are speculated to help protect against DNA damage from desiccation (Van Doninck et al., 2009). In animals, the macroH2A variant acts like H2A.W in protecting 10 bp of extranucleosomal DNA and is distinguished from other H2As in that the HFD is connected to a separate macrodomain by a basic protein linker region (Chakravarthy et al., 2012). Heterotypic macroH2A–H2A nucleosomes form a more stable octamer (Bowerman et al., 2019), and the linker region facilitates condensation (Muthurajan et al., 2011). MacroH2A nucleosomes have reduced recruitment of chromatin remodelers, inhibit acetylation by p300 (also known as EP300) (Chang et al., 2008; Doyen et al., 2006a) and present a barrier to reprogramming cells (Pliatska et al., 2018) by stabilizing both active and inactive gene expression patterns.

In contrast to H2A.W and macroH2A, four families of short H2As (H2A.B, H2A.L, H2A.P and H2A.Q), which wrap only 110–130 bp of DNA and have shortened docking domains, reduced DNA-binding capability and smaller acidic patches, are encoded on the X chromosome of placental mammals (Bao et al., 2004; Dai et al., 2018; Molaro et al., 2018). All these families have stage-specific expression in testes, where they have roles in splicing and the transition to protamines, and are evolving rapidly (Table 1).

H1 variants

Linker histone H1 lacks a HFD and has a different origin than the other histones (Kasinsky et al., 2001). It is absent in the early-diverging metamonads (Dalmasso et al., 2011) and it is unclear whether it was present in the last eukaryotic common ancestor. In multicellular eukaryotes, H1s have a tripartite structure in which a globular domain with a winged helix motif separates basic N-terminal and C-terminal domains that are variable. The lysine-rich C-terminus, which often contains S/TPKK minor groove-binding motifs, is similar to lysine-rich DNA-binding proteins in bacteria and is necessary for chromatin compaction (Healton et al., 2020; Kasinsky et al., 2001). The winged helix, which binds at the nucleosome dyad and interacts with the entry and exit linker DNAs (Bednar et al., 2017) (see poster), was either independently acquired in plants, mycetozoans and opisthokonts (animals, fungi and near relatives), or independently lost in kinetoplastids, alveolates and *Entamoeba* (Kasinsky et al., 2001).

Humans and other mammals have 11 H1 paralogs (seven somatic paralogs and four germline paralogs). The genes encoding the somatic paralogs H1.1–H1.5 and the ‘testis-specific’ variant H1.6 (H1t) are part of the histone gene cluster on chromosome VI, whereas RI variants H1.0 and H1.7–H1.10 (H1T2, H1oo, HILS1 and H1X) are encoded elsewhere. In ChIP-seq experiments of endogenous or HA-tagged H1.0, H1.2–H1.5 and H1.10 in a breast cancer cell line, H1s are broadly found on genes, repeats and upstream promoters, but are depleted at transcription start sites (Millán-Ariño et al., 2014). In human lung fibroblasts, H1.5 is enriched over splice sites of exons shorter than the length of a nucleosome and promotes their inclusion, apparently by stalling RNAPII (Glaich et al., 2019). In mouse embryonic stem cells, H1.2 and H1.3 (H1c and H1d) are enriched in heterochromatic domains marked with H3K9me3, and *in vitro* H1.0–H1.5 interact directly with H3K9 methyltransferases through their C-termini and promote H3K9 methylation (Healton et al., 2020) (see poster). Knockout of individual H1s generally has little effect on mouse development, but triple knockout of H1.2, H1.3 and H1.4 (H1e) results in embryonic lethality (Fan et al., 2003). In an embryonic cell line derived from

the triple knockout, with only 20% of global H1 expression, strong de-repression of transcription of the major satellite and other repeat classes occurs together with loss of H3K9me3 on the affected sequences (Healton et al., 2020). Although H1 variants have at least partially redundant functions, they show different effects on nucleosome spacing when introduced to H1-free *Xenopus* oocytes, with H1.2 and H1.3 increasing nucleosome repeat length by only 5–7 bp, whereas H1.4 and H1.0 from *Xenopus* and chicken (H5) increase nucleosome repeat length by 13–20 bp (Öberg et al., 2012). Short repeat length results in greater compaction and silencing (Healton et al., 2020).

The somatic RI variants H1.0 and H1.10 are enriched at nucleolus-associated domains and at RNAPII-enriched domains, respectively (Mayor et al., 2015). H1.0 is conserved in vertebrates and invertebrates, and is found primarily in differentiated tissues (González-Romero et al., 2009). H1.0-binding sites positively correlate with the presence of H3K27me3 (a mediator of developmental silencing), high nucleosome density and GC-rich genes in fibroblasts, and are at low density in AT-rich regions (Torres et al., 2016). H1.0 is often heterogeneously expressed in tumor cells, with H1.0 levels correlating with tumor cell differentiation and patient survival, whereas silencing of H1.0 favors self-renewing cells. Similarly, lower levels of H1.10 are an adverse prognosticator for astrocytic gliomas (Sepsa et al., 2015).

Perspective

From an ancestral set of five proteins, four of which are among the most conserved proteins known, histone variants continue to diversify and innovate to respond to the necessity of regulating access to DNA in all the contexts in which organisms find themselves. Histone variants greatly expand the roles and dynamics of nucleosomes by wrapping more or less DNA, by having greater or lesser stability, by having unique post-translational modifications or by interacting with other chromatin components. Although ancient variants, such as H2A.Z and H3.3, are well-studied, they continue to raise questions – are their effects on transcription and silencing cellular adaptations to their chromatin maintenance functions in non-dividing cells, when RC histones are unavailable? Similar questions arise in considering the DNA repair-independent functions of H2A.X. More recently evolved variants raise additional questions, such as what is the role of the enigmatic macroH2A domain, the only globular domain fused to a histone, and which is unique to animals. Especially intriguing are the sperm-specific variants in every histone class, which are collectively involved in the process of packaging the genome into protamines, but their individual roles are only starting to become clear. Other challenges include understanding how dysregulation or mutation of histone variants and their chaperones promote tumorigenesis (Bennett et al., 2019; Lowe et al., 2019; Nacev et al., 2019; Skene and Henikoff, 2013). New profiling and gene editing techniques promise to address these challenges.

Acknowledgements

We thank the members of our lab and anonymous reviewers for helpful comments. Structural models shown in the poster were made using iCn3D on the NCBI server (Wang et al., 2020).

Competing interests

The authors declare no competing or financial interests.

Funding

This work was funded by Howard Hughes Medical Institute. Deposited in PMC for immediate release.

Cell science at a glance

A high-resolution version of the poster and individual poster panels are available for downloading at http://jcs.biologists.org/lookup/doi/10.1242/jcs.244749.supplemental

References

Adam, M., Robert, F., Larochelle, M. and Gaudreau, L. (2001). H2A.Z is required for global chromatin integrity and for recruitment of RNA polymerase II under specific conditions. *Mol. Cell. Biol.* **21**, 6270-6279. doi:10.1128/MCB.21.18.6270-6279.2001

Ahmad, K. and Henikoff, S. (2002a). Histone H3 variants specify modes of chromatin assembly. *Proc. Natl. Acad. Sci. USA* **99** Suppl. 4, 16477-16484. doi:10.1073/pnas.172403699

Ahmad, K. and Henikoff, S. (2002b). The histone variant H3.3 marks active chromatin by replication-independent nucleosome assembly. *Mol. Cell* **9**, 1191-1200. doi:10.1016/S1097-2765(02)00542-7

Akiyoshi, B. and Gull, K. (2014). Discovery of unconventional kinetochores in kinetoplastids. *Cell* **156**, 1247-1258. doi:10.1016/j.cell.2014.01.049

Alva, V. and Lupas, A. N. (2019). Histones predate the split between bacteria and archaea. *Bioinformatics* **35**, 2349-2353. doi:10.1093/bioinformatics/bty1000

Anuar, N. D., Kurscheid, S., Field, M., Zhang, L., Rebar, E., Gregory, P., Buchou, T., Bowles, J., Koopman, P., Tremethick, D. J. et al. (2019). Gene editing of the multi-copy H2A.B gene and its importance for fertility. *Genome Biol.* **20**, 23. doi:10.1186/s13059-019-1633-3

Armache, A., Yang, S., Martínez de Paz, A., Robbins, L. E., Durmaz, C., Cheong, J. Q., Ravishankar, A., Daman, A. W., Ahimovic, D. J., Klevorn, T. et al. (2020). Histone H3.3 phosphorylation amplifies stimulation-induced transcription. *Nature* **583**, 852-857. doi:10.1038/s41586-020-2533-0

Aul, R. B. and Oko, R. J. (2002). The major subacrosomal occupant of bull spermatozoa is a novel histone H2B variant associated with the forming acrosome during spermiogenesis. *Dev. Biol.* **242**, 376-387. doi:10.1006/dbio.2002.0575

Bagchi, D. N., Battenhouse, A. M., Park, D. and Iyer, V. R. (2020). The histone variant H2A.Z in yeast is almost exclusively incorporated into the +1 nucleosome in the direction of transcription. *Nucleic Acids Res.* **48**, 157-170. doi:10.1093/nar/gkz1075

Bao, Y., Konesky, K., Park, Y. J., Rosu, S., Dyer, P. N., Rangasamy, D., Tremethick, D. J., Laybourn, P. J. and Luger, K. (2004). Nucleosomes containing the histone variant H2A.Bbd organize only 118 base pairs of DNA. *EMBO J.* **23**, 3314-3324. doi:10.1038/sj.emboj.7600316

Barral, S., Morozumi, Y., Tanaka, H., Montellier, E., Govin, J., de Dieuleveult, M., Charbonnier, G., Coute, Y., Puthier, D., Buchou, T. et al. (2017). Histone variant H2A.L.2 guides transition protein-dependent protamine assembly in male germ cells. *Mol. Cell* **66**, 89-101.e8. doi:10.1016/j.molcel.2017.02.025

Bednar, J., Garcia-Saez, I., Boopathi, R., Cutter, A. R., Papai, G., Reymer, A., Syed, S. H., Lone, I. N., Tonchev, O., Crucifix, C. et al. (2017). Structure and dynamics of a 197 bp nucleosome in complex with linker histone H1. *Mol. Cell* **66**, 384-397.e8. doi:10.1016/j.molcel.2017.04.012

Beedle, M.-T., Topping, T., Hogarth, C. and Griswold, M. (2019). Differential localization of histone variant TH2B during the first round compared with subsequent rounds of spermatogenesis. *Dev. Dyn.* **248**, 488-500. doi:10.1002/dvdy.33

Belotti, E., Lacoste, N., Simonet, T., Papin, C., Padmanabhan, K., Scionti, I., Gangloff, Y. G., Ramos, L., Dalkara, D., Hamiche, A. et al. (2020). H2A.Z is dispensable for both basal and activated transcription in post-mitotic mouse muscles. *Nucleic Acids Res.* **48**, 4601-4613. doi:10.1093/nar/gkaa157

Bennett, R. L., Bele, A., Small, E. C., Will, C. M., Nabet, B., Oyer, J. A., Huang, X., Ghosh, R. P., Grzybowski, A. T., Yu, T. et al. (2019). A mutation in histone H2B represents a new class of oncogenic driver. *Cancer Discov.* **9**, 1438-1451. doi:10.1158/2159-8290.CD-19-0393

Bergmuller, E., Gehrig, P. M. and Gruissem, W. (2007). Characterization of post-translational modifications of histone H2B-variants isolated from Arabidopsis thaliana. *J. Proteome Res.* **6**, 3655-3668. doi:10.1021/pr0702159

Bonisch, C., Schneider, K., Pünzeler, S., Wiedemann, S. M., Bielmeier, C., Bocola, M., Eberl, H. C., Kuegel, W., Neumann, J., Kremmer, E. et al. (2012). H2A.Z.2.2 is an alternatively spliced histone H2A.Z variant that causes severe nucleosome destabilization. *Nucleic Acids Res.* **40**, 5951-5964. doi:10.1093/nar/gks267

Borg, M., Jacob, Y., Susaki, D., LeBlanc, C., Buendía, D., Axelsson, E., Kawashima, T., Voigt, P., Boavida, L., Becker, J. et al. (2020). Targeted reprogramming of H3K27me3 resets epigenetic memory in plant paternal chromatin. *Nat. Cell Biol.* **22**, 621-629. doi:10.1038/s41556-020-0515-y

Boulard, M., Gautier, T., Mbele, G. O., Gerson, V., Hamiche, A., Angelov, D., Bouvet, P. and Dimitrov, S. (2006). 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.* **26**, 1518-1526. doi:10.1128/MCB.26.4.1518-1526.2006

Bowerman, S., Hickok, R. J. and Wereszczynski, J. (2019). Unique dynamics in asymmetric macroH2A-H2A hybrid nucleosomes result in increased complex stability. *J. Phys. Chem. B* **123**, 419-427. doi:10.1021/acs.jpcb.8b10668

Brunelle, M., Nordell Markovits, A., Rodrigue, S., Lupien, M., Jacques, P.-E. and Gévry, N. (2015). The histone variant H2A.Z is an important regulator of enhancer activity. *Nucleic Acids Res.* **43**, 9742-9756. doi:10.1093/nar/gkv825

Camahort, R., Li, B., Florens, L., Swanson, S. K., Washburn, M. P. and Gerton, J. L. (2007). Scm3 is essential to recruit the histone h3 variant cse4 to centromeres and to maintain a functional kinetochore. *Mol. Cell* **26**, 853-865. doi:10.1016/j.molcel.2007.05.013

Celeste, A., Petersen, S., Romanienko, P. J., Fernandez-Capetillo, O., Chen, H. T., Sedelnikova, O. A., Reina-San-Martin, B., Coppola, V., Meffre, E., Difilippantonio, M. J. et al. (2002). Genomic instability in mice lacking histone H2AX. *Science* **296**, 922-927. doi:10.1126/science.1069398

Chakravarthy, S., Patel, A. and Bowman, G. D. (2012). The basic linker of macroH2A stabilizes DNA at the entry/exit site of the nucleosome. *Nucleic Acids Res.* **40**, 8285-8295. doi:10.1093/nar/gks645

Chang, E. Y., Ferreira, H., Somers, J., Nusinow, D. A., Owen-Hughes, T. and Narlikar, G. J. (2008). MacroH2A allows ATP-dependent chromatin remodeling by SWI/SNF and ACF complexes but specifically reduces recruitment of SWI/SNF. *Biochemistry* **47**, 13726-13732. doi:10.1021/bi8016944

Chang, F. T. M., Chan, F. L., McGhie, J. D. R., Udugama, M., Mayne, L., Collas, P., Mann, J. R. and Wong, L. H. (2015). CHK1-driven histone H3.3 serine 31 phosphorylation is important for chromatin maintenance and cell survival in human ALT cancer cells. *Nucleic Acids Res.* **43**, 2603-2614. doi:10.1093/nar/gkv104

Cheema, M. S., Good, K. V., Kim, B., Soufari, H., O'Sullivan, C., Freeman, M. E., Stefanelli, G., Casas, C. R., Zengeler, K. E., Kennedy, A. J. et al. (2020). Deciphering the enigma of the histone H2A.Z-1/H2A.Z-2 isoforms: novel insights and remaining questions. *Cells* **9**, 1167. doi:10.3390/cells9051167

Chen, C. C., Dechassa, M. L., Bettini, E., Ledoux, M. B., Belisario, C., Heun, P., Luger, K. and Mellone, B. G. (2014). CAL1 is the Drosophila CENP-A assembly factor. *J. Cell Biol.* **204**, 313-329. doi:10.1083/jcb.201305036

Churikov, D., Siino, J., Svetlova, M., Zhang, K., Gineitis, A., Morton Bradbury, E. and Zalensky, A. (2004). Novel human testis-specific histone H2B encoded by the interrupted gene on the X chromosome. *Genomics* **84**, 745-756. doi:10.1016/j.ygeno.2004.06.001

Contrepois, K., Coudereau, C., Benayoun, B. A., Schuler, N., Roux, P. F., Bischof, O., Courbeyrette, R., Carvalho, C., Thuret, J. Y., Ma, Z. et al. (2017). Histone variant H2A.J accumulates in senescent cells and promotes inflammatory gene expression. *Nat. Commun.* **8**, 14995. doi:10.1038/ncomms14995

Cortijo, S., Charoensawan, V., Brestovitsky, A., Buning, R., Ravarani, C., Rhodes, D., van Noort, J., Jaeger, K. E. and Wigge, P. A. (2017). Transcriptional regulation of the ambient temperature response by H2A.Z nucleosomes and HSF1 transcription factors in arabidopsis. *Mol. Plant* **10**, 1258-1273. doi:10.1016/j.molp.2017.08.014

Dai, L., Xie, X. and Zhou, Z. (2018). Crystal structure of the histone heterodimer containing histone variant H2A.Bbd. *Biochem. Biophys. Res. Commun.* **503**, 1786-1791. doi:10.1016/j.bbrc.2018.07.114

Dalmasso, M. C., Sullivan, W. J., Jr. and Angel, S. O. (2011). Canonical and variant histones of protozoan parasites. *Front. Biosci.* **16**, 2086-2105. doi:10.2741/3841

Dann, G. P., Liszczak, G. P., Bagert, J. D., Muller, M. M., Nguyen, U. T. T., Wojcik, F., Brown, Z. Z., Bos, J., Panchenko, T., Pihl, R. et al. (2017). ISWI chromatin remodellers sense nucleosome modifications to determine substrate preference. *Nature* **548**, 607-611. doi:10.1038/nature23671

Dawe, R. K. and Henikoff, S. (2006). Centromeres put epigenetics in the driver's seat. *Trends Biochem. Sci.* **31**, 662-669. doi:10.1016/j.tibs.2006.10.004

Delaney, K., Mailler, J., Wenda, J. M., Gabus, C. and Steiner, F. A. (2018). Differential expression of histone H3.3 genes and their role in modulating temperature stress response in Caenorhabditis elegans. *Genetics* **209**, 551-565. doi:10.1534/genetics.118.300909

Doyen, C. M., An, W., Angelov, D., Bondarenko, V., Mietton, F., Studitsky, V. M., Hamiche, A., Roeder, R. G., Bouvet, P. and Dimitrov, S. (2006a). Mechanism of polymerase II transcription repression by the histone variant macroH2A. *Mol. Cell. Biol.* **26**, 1156-1164. doi:10.1128/MCB.26.3.1156-1164.2006

Doyen, C.-M., Montel, F., Gautier, T., Menoni, H., Claudet, C., Delacour-Larose, M., Angelov, D., Hamiche, A., Bednar, J., Faivre-Moskalenko, C. et al. (2006b). Dissection of the unusual structural and functional properties of the variant H2A.Bbd nucleosome. *EMBO J.* **25**, 4234-4244. doi:10.1038/sj.emboj.7601310

Drane, P., Ouararhni, K., Depaux, A., Shuaib, M. and Hamiche, A. (2010). The death-associated protein DAXX is a novel histone chaperone involved in the replication-independent deposition of H3.3. *Genes Dev.* **24**, 1253-1265. doi:10.1101/gad.566910

Drinnenberg, I. A., deYoung, D., Henikoff, S. and Malik, H. S. (2014). Recurrent loss of CenH3 is associated with independent transitions to holocentricity in insects. *eLife* **3**, e03676. doi:10.7554/eLife.03676

Dunleavy, E. M., Roche, D., Tagami, H., Lacoste, N., Ray-Gallet, D., Nakamura, Y., Daigo, Y., Nakatani, Y. and Almouzni-Pettinotti, G. (2009). HJURP is a cell-cycle-dependent maintenance and deposition factor of CENP-A at centromeres. *Cell* **137**, 485-497. doi:10.1016/j.cell.2009.02.040

Eirín-López, J. M., González-Romero, R., Dryhurst, D., Ishibashi, T. and Ausió, J. (2009). The evolutionary differentiation of two histone H2A.Z variants in chordates (H2A.Z-1 and H2A.Z-2) is mediated by a stepwise mutation process that affects three amino acid residues. *BMC Evol. Biol.* **9**, 31. doi:10.1186/1471-2148-9-31

Eleuteri, B., Aranda, S. and Ernfors, P. (2018). NoRC recruitment by H2A.X deposition at rRNA gene promoter limits embryonic stem cell proliferation. *Cell Rep.* **23**, 1853-1866. doi:10.1016/j.celrep.2018.04.023

Elsässer, S. J., Huang, H., Lewis, P. W., Chin, J. W., Allis, C. D. and Patel, D. J. (2012). DAXX envelops a histone H3.3-H4 dimer for H3.3-specific recognition. *Nature* **491**, 560-565. doi:10.1038/nature11608

Elsässer, S. J., Noh, K. M., Diaz, N., Allis, C. D. and Banaszynski, L. A. (2015). Histone H3.3 is required for endogenous retroviral element silencing in embryonic stem cells. *Nature* **522**, 240-244. doi:10.1038/nature14345

Erives, A. J. (2017). Phylogenetic analysis of the core histone doublet and DNA topo II genes of Marseilleviridae: evidence of proto-eukaryotic provenance. *Epigenet. Chromatin* **10**, 55. doi:10.1186/s13072-017-0162-0

Faast, R., Thonglairoam, V., Schulz, T. C., Beall, J., Wells, J. R., Taylor, H., Matthaei, K., Rathjen, P. D., Tremethick, D. J. and Lyons, I. (2001). Histone variant H2A.Z is required for early mammalian development. *Curr. Biol.* **11**, 1183-1187. doi:10.1016/S0960-9822(01)00329-3

Fan, Y., Nikitina, T., Morin-Kensicki, E. M., Zhao, J., Magnuson, T. R., Woodcock, C. L. and Skoultchi, A. I. (2003). H1 linker histones are essential for mouse development and affect nucleosome spacing in vivo. *Mol. Cell. Biol.* **23**, 4559-4572. doi:10.1128/MCB.23.13.4559-4572.2003

Fernandez-Capetillo, O., Mahadevaiah, S. K., Celeste, A., Romanienko, P. J., Camerini-Otero, R. D., Bonner, W. M., Manova, K., Burgoyne, P. and Nussenzweig, A. (2003). H2AX is required for chromatin remodeling and inactivation of sex chromosomes in male mouse meiosis. *Dev. Cell* **4**, 497-508. doi:10.1016/S1534-5807(03)00093-5

Giaimo, B. D., Ferrante, F., Herchenröther, A., Hake, S. B. and Borggrefe, T. (2019). The histone variant H2A.Z in gene regulation. *Epigenet. Chromatin* **12**, 37. doi:10.1186/s13072-019-0274-9

Gineitis, A. A., Zalenskaya, I. A., Yau, P. M., Bradbury, E. M. and Zalensky, A. O. (2000). Human sperm telomere-binding complex involves histone H2B and secures telomere membrane attachment. *J. Cell Biol.* **151**, 1591-1598. doi:10.1083/jcb.151.7.1591

Glaich, O., Leader, Y., Lev Maor, G. and Ast, G. (2019). Histone H1.5 binds over splice sites in chromatin and regulates alternative splicing. *Nucleic Acids Res.* **47**, 6145-6159. doi:10.1093/nar/gkz338

Goldberg, A. D., Banaszynski, L. A., Noh, K. M., Lewis, P. W., Elsaesser, S. J., Stadler, S., Dewell, S., Law, M., Guo, X., Li, X. et al. (2010). Distinct factors control histone variant H3.3 localization at specific genomic regions. *Cell* **140**, 678-691. doi:10.1016/j.cell.2010.01.003

Goldman, J. A., Garlick, J. D. and Kingston, R. E. (2010). Chromatin remodeling by imitation switch (ISWI) class ATP-dependent remodelers is stimulated by histone variant H2A.Z. *J. Biol. Chem.* **285**, 4645-4651. doi:10.1074/jbc.M109.072348

González-Romero, R., Ausió, J., Mendez, J. and Eirín-López, J. M. (2009). Histone genes of the razor clam Solen marginatus unveil new aspects of linker histone evolution in protostomes. *Genome/National Research Council Canada=Genome/Conseil national de recherches Canada* **52**, 597-607. doi:10.1139/G09-034

Greenberg, R. S., Long, H. K., Swigut, T. and Wysocka, J. (2019). Single amino acid change underlies distinct Roles of H2A.Z subtypes in human syndrome. *Cell* **178**, 1421-1436.e24. doi:10.1016/j.cell.2019.08.002

Hake, S. B., Garcia, B. A., Kauer, M., Baker, S. P., Shabanowitz, J., Hunt, D. F. and Allis, C. D. (2005). Serine 31 phosphorylation of histone variant H3.3 is specific to regions bordering centromeres in metaphase chromosomes. *Proc. Natl. Acad. Sci. USA* **102**, 6344-6349. doi:10.1073/pnas.0502413102

Healton, S. E., Pinto, H. D., Mishra, L. N., Hamilton, G. A., Wheat, J. C., Swist-Rosowska, K., Shukeir, N., Dou, Y., Steidl, U., Jenuwein, T. et al. (2020). H1 linker histones silence repetitive elements by promoting both histone H3K9 methylation and chromatin compaction. *Proc. Natl. Acad. Sci. USA* **117**, 14251-14258. doi:10.1073/pnas.1920725117

Henikoff, S., Ahmad, K. and Malik, H. S. (2001). The centromere paradox: stable inheritance with rapidly evolving DNA. *Science* **293**, 1098-1102. doi:10.1126/science.1062939

Henneman, B., van Emmerik, C., van Ingen, H. and Dame, R. T. (2018). Structure and function of archaeal histones. *PLoS Genet.* **14**, e1007582. doi:10.1371/journal.pgen.1007582

Ingouff, M., Hamamura, Y., Gourgues, M., Higashiyama, T. and Berger, F. (2007). Distinct dynamics of HISTONE3 variants between the two fertilization products in plants. *Curr. Biol.* **17**, 1032-1037. doi:10.1016/j.cub.2007.05.019

Jacob, Y., Bergamin, E., Donoghue, M. T., Mongeon, V., LeBlanc, C., Voigt, P., Underwood, C. J., Brunzelle, J. S., Michaels, S. D., Reinberg, D. et al. (2014). Selective methylation of histone H3 variant H3.1 regulates heterochromatin replication. *Science* **343**, 1249-1253. doi:10.1126/science.1248357

Jang, C. W., Shibata, Y., Starmer, J., Yee, D. and Magnuson, T. (2015). Histone H3.3 maintains genome integrity during mammalian development. *Genes Dev.* **29**, 1377-1392. doi:10.1101/gad.264150.115

Jiang, D., Borg, M., Lorković, Z. J., Montgomery, S. A., Osakabe, A., Yelagandula, R., Axelsson, E. and Berger, F. (2020). The evolution and functional divergence of the histone H2B family in plants. *PLoS Genet.* **16**, e1008964. doi:10.1371/journal.pgen.1008964

Kasinsky, H. E., Lewis, J. D., Dacks, J. B. and Ausió, J. (2001). Origin of H1 linker histones. *FASEB J.* **15**, 34-42. doi:10.1096/fj.00-0237rev

Kornberg, R. D. and Lorch, Y. (2020). Primary role of the nucleosome. *Mol. Cell* **79**, 371-375. doi:10.1016/j.molcel.2020.07.020

Kujirai, T., Horikoshi, N., Sato, K., Maehara, K., Machida, S., Osakabe, A., Kimura, H., Ohkawa, Y. and Kurumizaka, H. (2016). Structure and function of human histone H3.Y nucleosome. *Nucleic Acids Res.* **44**, 6127-6141. doi:10.1093/nar/gkw202

Kursel, L. E. and Malik, H. S. (2018). The cellular mechanisms and consequences of centromere drive. *Curr. Opin. Cell Biol.* **52**, 58-65. doi:10.1016/j.ceb.2018.01.011

Li, M., Dong, Q. and Zhu, B. (2017). Aurora kinase B phosphorylates histone H3.3 at Serine 31 during mitosis in mammalian cells. *J. Mol. Biol.* **429**, 2042-2045. doi:10.1016/j.jmb.2017.01.016

Long, M., Sun, X., Shi, W., Yanru, A., Leung, S. T. C., Ding, D., Cheema, M. S., MacPherson, N., Nelson, C. J., Ausio, J. et al. (2019). A novel histone H4 variant H4G regulates rDNA transcription in breast cancer. *Nucleic Acids Res.* **47**, 8399-8409. doi:10.1093/nar/gkz547

Lorković, Z. J., Park, C., Goiser, M., Jiang, D., Kurzbauer, M. T., Schlögelhofer, P. and Berger, F. (2017). Compartmentalization of DNA damage response between heterochromatin and euchromatin is mediated by distinct H2A histone variants. *Curr. Biol.* **27**, 1192-1199. doi:10.1016/j.cub.2017.03.002

Lowe, B. R., Maxham, L. A., Hamey, J. J., Wilkins, M. R. and Partridge, J. F. (2019). Histone H3 mutations: an updated view of their role in chromatin deregulation and cancer. *Cancers (Basel)* **11**, 660. doi:10.3390/cancers11050660

Luger, K., Mäder, A. W., Richmond, R. K., Sargent, D. F. and Richmond, T. J. (1997). Crystal structure of the nucleosome core particle at 2.8 Å resolution. *Nature* **389**, 251-260. doi:10.1038/38444

Lyons, S. M., Cunningham, C. H., Welch, J. D., Groh, B., Guo, A. Y., Wei, B., Whitfield, M. L., Xiong, Y. and Marzluff, W. F. (2016). A subset of replication-dependent histone mRNAs are expressed as polyadenylated RNAs in terminally differentiated tissues. *Nucleic Acids Res.* **44**, 9190-9205. doi:10.1093/nar/gkw620

Maehara, K., Harada, A., Sato, Y., Matsumoto, M., Nakayama, K. I., Kimura, H. and Ohkawa, Y. (2015). Tissue-specific expression of histone H3 variants diversified after species separation. *Epigenet. Chromatin* **8**, 35. doi:10.1186/s13072-015-0027-3

Mahadevan, I. A., Kumar, S. and Rao, M. R. S. (2020). Linker histone variant H1t is closely associated with repressed repeat-element chromatin domains in pachytene spermatocytes. *Epigenet. Chromatin* **13**, 9. doi:10.1186/s13072-020-00335-x

Mannironi, C., Bonner, W. M. and Hatch, C. L. (1989). H2A.X. a histone isoprotein with a conserved C-terminal sequence, is encoded by a novel mRNA with both DNA replication type and polyA 3′ processing signals. *Nucleic Acids Res.* **17**, 9113-9126. doi:10.1093/nar/17.22.9113

Martianov, I., Brancorsini, S., Catena, R., Gansmuller, A., Kotaja, N., Parvinen, M., Sassone-Corsi, P. and Davidson, I. (2005). Polar nuclear localization of H1T2, a histone H1 variant, required for spermatid elongation and DNA condensation during spermiogenesis. *Proc. Natl. Acad. Sci. USA* **102**, 2808-2813. doi:10.1073/pnas.0406060102

Martire, S., Gogate, A. A., Whitmill, A., Tafessu, A., Nguyen, J., Teng, Y. C., Tastemel, M. and Banaszynski, L. A. (2019). Phosphorylation of histone H3.3 at serine 31 promotes p300 activity and enhancer acetylation. *Nat. Genet.* **51**, 941-946. doi:10.1038/s41588-019-0428-5

Marzluff, W. F. (2005). Metazoan replication-dependent histone mRNAs: a distinct set of RNA polymerase II transcripts. *Curr. Opin. Cell Biol.* **17**, 274-280. doi:10.1016/j.ceb.2005.04.010

Marzluff, W. F., Gongidi, P., Woods, K. R., Jin, J. and Maltais, L. J. (2002). The human and mouse replication-dependent histone genes. *Genomics* **80**, 487-498. doi:10.1006/geno.2002.6850

Marzluff, W. F., Sakallah, S. and Kelkar, H. (2006). The sea urchin histone gene complement. *Dev. Biol.* **300**, 308-320. doi:10.1016/j.ydbio.2006.08.067

Mattiroli, F., Bhattacharyya, S., Dyer, P. N., White, A. E., Sandman, K., Burkhart, B. W., Byrne, K. R., Lee, T., Ahn, N. G., Santangelo, T. J. et al. (2017). Structure of histone-based chromatin in Archaea. *Science* **357**, 609-612. doi:10.1126/science.aaj1849

Mayor, R., Izquierdo-Bouldstridge, A., Millán-Ariño, L., Bustillos, A., Sampaio, C., Luque, N. and Jordan, A. (2015). Genome distribution of replication-independent histone H1 variants shows H1.0 associated with nucleolar domains and H1X associated with RNA polymerase II-enriched regions. *J. Biol. Chem.* **290**, 7474-7491. doi:10.1074/jbc.M114.617324

Millán-Ariño, L., Islam, A. B., Izquierdo-Bouldstridge, A., Mayor, R., Terme, J. M., Luque, N., Sancho, M., López-Bigas, N. and Jordan, A. (2014). Mapping of six somatic linker histone H1 variants in human breast cancer cells uncovers specific features of H1.2. *Nucleic Acids Res.* **42**, 4474-4493. doi:10.1093/nar/gku079

Mishra, L. N., Gupta, N. and Rao, S. M. (2015). Mapping of post-translational modifications of spermatid-specific linker histone H1-like protein, HILS1. *J. Proteomics* **128**, 218-230. doi:10.1016/j.jprot.2015.08.001

Mito, Y., Henikoff, J. G. and Henikoff, S. (2005). Genome-scale profiling of histone H3.3 replacement patterns. *Nat. Genet.* **37**, 1090-1097. doi:10.1038/ng1637

Mito, Y., Henikoff, J. G. and Henikoff, S. (2007). Histone replacement marks the boundaries of cis-regulatory domains. *Science* **315**, 1408-1411. doi:10.1126/science.1134004

Mizuguchi, G., Xiao, H., Wisniewski, J., Smith, M. M. and Wu, C. (2007). Nonhistone Scm3 and histones CenH3-H4 assemble the core of centromere-specific nucleosomes. *Cell* **129**, 1153-1164. doi:10.1016/j.cell.2007.04.026

Molaro, A., Young, J. M. and Malik, H. S. (2018). Evolutionary origins and diversification of testis-specific short histone H2A variants in mammals. *Genome Res.* **28**, 460-473. doi:10.1101/gr.229799.117

Montellier, E., Boussouar, F., Rousseaux, S., Zhang, K., Buchou, T., Fenaille, F., Shiota, H., Debernardi, A., Hery, P., Curtet, S. et al. (2013). Chromatin-to-nucleoprotamine transition is controlled by the histone H2B variant TH2B. *Genes Dev.* **27**, 1680-1692. doi:10.1101/gad.220095.113

Moosmann, A., Campsteijn, C., Jansen, P. W., Nasrallah, C., Raasholm, M., Stunnenberg, H. G. and Thompson, E. M. (2011). Histone variant innovation in a rapidly evolving chordate lineage. *BMC Evol. Biol.* **11**, 208-2148-11-208. doi:10.1186/1471-2148-11-208

Muthurajan, U. M., McBryant, S. J., Lu, X., Hansen, J. C. and Luger, K. (2011). The linker region of macroH2A promotes self-association of nucleosomal arrays. *J. Biol. Chem.* **286**, 23852-23864. doi:10.1074/jbc.M111.244871

Nacev, B. A., Feng, L., Bagert, J. D., Lemiesz, A. E., Gao, J., Soshnev, A. A., Kundra, R., Schultz, N., Muir, T. W. and Allis, C. D. (2019). The expanding landscape of 'oncohistone' mutations in human cancers. *Nature* **567**, 473-478. doi:10.1038/s41586-019-1038-1

Navarro-Mendoza, M. I., Pérez-Arques, C., Panchal, S., Nicolás, F. E., Mondo, S. J., Ganguly, P., Pangilinan, J., Grigoriev, I. V., Heitman, J., Sanyal, K. et al. (2019). Early diverging fungus Mucor circinelloides lacks centromeric histone CENP-A and displays a mosaic of point and regional centromeres. *Curr. Biol.* **29**, 3791-3802.e6. doi:10.1016/j.cub.2019.09.024

Öberg, C., Izzo, A., Schneider, R., Wrange, Ö. and Belikov, S. (2012). Linker histone subtypes differ in their effect on nucleosomal spacing in vivo. *J. Mol. Biol.* **419**, 183-197. doi:10.1016/j.jmb.2012.03.007

Osakabe, A., Lorković, Z. J., Kobayashi, W., Tachiwana, H., Yelagandula, R., Kurumizaka, H. and Berger, F. (2018). Histone H2A variants confer specific properties to nucleosomes and impact on chromatin accessibility. *Nucleic Acids Res.* **46**, 7675-7685. doi:10.1093/nar/gky540

Pang, M. Y. H., Sun, X., Ausió, J. and Ishibashi, T. (2020). Histone H4 variant, H4G, drives ribosomal RNA transcription and breast cancer cell proliferation by loosening nucleolar chromatin structure. *J. Cell. Physiol.* **235**, 9601-9608. doi:10.1002/jcp.29770

Piquet, S., Le Parc, F., Bai, S.-K., Chevallier, O., Adam, S. and Polo, S. E. (2018). The histone chaperone FACT coordinates H2A.X-dependent signaling and repair of DNA damage. *Mol. Cell* **72**, 888-901.e7. doi:10.1016/j.molcel.2018.09.010

Pliatska, M., Kapasa, M., Kokkalis, A., Polyzos, A. and Thanos, D. (2018). The histone variant MacroH2A blocks cellular reprogramming by inhibiting mesenchymal-to-epithelial transition. *Mol. Cell. Biol.* **38**, e00669-17. doi:10.1128/MCB.00669-17

Postberg, J., Forcob, S., Chang, W.-J. and Lipps, H. J. (2010). The evolutionary history of histone H3 suggests a deep eukaryotic root of chromatin modifying mechanisms. *BMC Evol. Biol.* **10**, 259. doi:10.1186/1471-2148-10-259

Raisner, R. M., Hartley, P. D., Meneghini, M. D., Bao, M. Z., Liu, C. L., Schreiber, S. L., Rando, O. J. and Madhani, H. D. (2005). Histone variant H2A.Z marks the 5′ ends of both active and inactive genes in euchromatin. *Cell* **123**, 233-248. doi:10.1016/j.cell.2005.10.002

Raja, D. A., Subramaniam, Y., Aggarwal, A., Gotherwal, V., Babu, A., Tanwar, J., Motiani, R. K., Sivasubbu, S., Gokhale, R. S. and Natarajan, V. T. (2020). Histone variant dictates fate biasing of neural crest cells to melanocyte lineage. *Development* **147**, dev182576. doi:10.1242/dev.182576

Ramachandran, S., Ahmad, K. and Henikoff, S. (2017). Transcription and remodeling produce asymmetrically unwrapped nucleosomal intermediates. *Mol. Cell* **68**, 1038-1053.e4. doi:10.1016/j.molcel.2017.11.015

Ranjan, A., Mizuguchi, G., FitzGerald, P. C., Wei, D., Wang, F., Huang, Y., Luk, E., Woodcock, C. L. and Wu, C. (2013). Nucleosome-free region dominates histone acetylation in targeting SWR1 to promoters for H2A.Z replacement. *Cell* **154**, 1232-1245. doi:10.1016/j.cell.2013.08.005

Resnick, R., Wong, C.-J., Hamm, D. C., Bennett, S. R., Skene, P. J., Hake, S. B., Henikoff, S., van der Maarel, S. M. and Tapscott, S. J. (2019). DUX4-induced histone variants H3.X and H3.Y mark DUX4 target genes for expression. *Cell Rep.* **29**, 1812-1820.e5. doi:10.1016/j.celrep.2019.10.025

Rosin, L. and Mellone, B. G. (2016). Co-evolving CENP-A and CAL1 domains mediate centromeric CENP-A deposition across Drosophila species. *Dev. Cell* **37**, 136-147. doi:10.1016/j.devcel.2016.03.021

Rosin, L. F. and Mellone, B. G. (2017). Centromeres drive a hard bargain. *Trends Genet.* **33**, 101-117. doi:10.1016/j.tig.2016.12.001
Rudnizky, S., Bavly, A., Malik, O., Pnueli, L., Melamed, P. and Kaplan, A. (2016). H2A.Z controls the stability and mobility of nucleosomes to regulate expression of the LH genes. *Nat. Commun.* **7**, 12958. doi:10.1038/ncomms12958
Sakai, A., Schwartz, B. E., Goldstein, S. and Ahmad, K. (2009). Transcriptional and developmental functions of the H3.3 histone variant in Drosophila. *Curr. Biol.* **19**, 1816-1820. doi:10.1016/j.cub.2009.09.021
Santoro, S. W. and Dulac, C. (2012). The activity-dependent histone variant H2BE modulates the life span of olfactory neurons. *eLife* **1**, e00070. doi:10.7554/eLife.00070
Sarthy, J. F., Meers, M. P., Janssens, D. H., Henikoff, J. G., Feldman, H., Paddison, P. J., Lockwood, C. M., Vitanza, N. A., Olson, J. M., Ahmad, K. et al. (2020). Histone deposition pathways determine the chromatin landscapes of H3.1 and H3.3 K27M oncohistones. *eLife* **9**, e61090. doi:10.7554/eLife.61090
Seo, J., Kim, S. C., Lee, H.-S., Kim, J. K., Shon, H. J., Salleh, N. L. M., Desai, K. V., Lee, J. H., Kang, E.-S., Kim, J. S. et al. (2012). Genome-wide profiles of H2AX and γ-H2AX differentiate endogenous and exogenous DNA damage hotspots in human cells. *Nucleic Acids Res.* **40**, 5965-5974. doi:10.1093/nar/gks287
Sepsa, A., Levidou, G., Gargalionis, A., Adamopoulos, C., Spyropoulou, A., Dalagiorgou, G., Thymara, I., Boviatsis, E., Themistocleous, M. S., Petraki, K. et al. (2015). Emerging role of linker histone variant H1x as a biomarker with prognostic value in astrocytic gliomas. A multivariate analysis including trimethylation of H3K9 and H4K20. *PLoS ONE* **10**, e0115101. doi:10.1371/journal.pone.0115101
Sharma, D., De Falco, L., Padavattan, S., Rao, C., Geifman-Shochat, S., Liu, C.-F. and Davey, C. A. (2019). PARP1 exhibits enhanced association and catalytic efficiency with γH2A.X-nucleosome. *Nat. Commun.* **10**, 5751. doi:10.1038/s41467-019-13641-0
Shechter, D., Chitta, R. K., Xiao, A., Shabanowitz, J., Hunt, D. F. and Allis, C. D. (2009). A distinct H2A.X isoform is enriched in Xenopus laevis eggs and early embryos and is phosphorylated in the absence of a checkpoint. *Proc. Natl. Acad. Sci. USA* **106**, 749-754. doi:10.1073/pnas.0812207106
Shinagawa, T., Takagi, T., Tsukamoto, D., Tomaru, C., Huynh, L. M., Sivaraman, P., Kumarevel, T., Inoue, K., Nakato, R., Katou, Y. et al. (2014). Histone variants enriched in oocytes enhance reprogramming to induced pluripotent stem cells. *Cell Stem Cell* **14**, 217-227. doi:10.1016/j.stem.2013.12.015
Shinagawa, T., Huynh, L. M., Takagi, T., Tsukamoto, D., Tomaru, C., Kwak, H.-G., Dohmae, N., Noguchi, J. and Ishii, S. (2015). Disruption of Th2a and Th2b genes causes defects in spermatogenesis. *Development* **142**, 1287-1292. doi:10.1242/dev.121830
Shiraishi, K., Shindo, A., Harada, A., Kurumizaka, H., Kimura, H., Ohkawa, Y. and Matsuyama, H. (2018). Roles of histone H3.5 in human spermatogenesis and spermatogenic disorders. *Andrology* **6**, 158-165. doi:10.1111/andr.12438
Sitbon, D., Boyarchuk, E., Dingli, F., Loew, D. and Almouzni, G. (2020). Histone variant H3.3 residue S31 is essential for Xenopus gastrulation regardless of the deposition pathway. *Nat. Commun.* **11**, 1256. doi:10.1038/s41467-020-15084-4
Skene, P. J. and Henikoff, S. (2013). Histone variants in pluripotency and disease. *Development* **140**, 2513-2524. doi:10.1242/dev.091439
Soboleva, T. A., Parker, B. J., Nekrasov, M., Hart-Smith, G., Tay, Y. J., Tng, W.-Q., Wilkins, M., Ryan, D. and Tremethick, D. J. (2017). A new link between transcriptional initiation and pre-mRNA splicing: The RNA binding histone variant H2A.B. *PLoS Genet.* **13**, e1006633. doi:10.1371/journal.pgen.1006633
Stoler, S., Rogers, K., Weitze, S., Morey, L., Fitzgerald-Hayes, M. and Baker, R. E. (2007). Scm3, an essential Saccharomyces cerevisiae centromere protein required for G2/M progression and Cse4 localization. *Proc. Natl. Acad. Sci. USA* **104**, 10571-10576. doi:10.1073/pnas.0703178104
Tachiwana, H., Osakabe, A., Kimura, H. and Kurumizaka, H. (2008). Nucleosome formation with the testis-specific histone H3 variant, H3t, by human nucleosome assembly proteins in vitro. *Nucleic Acids Res.* **36**, 2208-2218. doi:10.1093/nar/gkn060
Tagami, H., Ray-Gallet, D., Almouzni, G. and Nakatani, Y. (2004). Histone H3.1 and H3.3 complexes mediate nucleosome assembly pathways dependent or independent of DNA synthesis. *Cell* **116**, 51-61. doi:10.1016/S0092-8674(03)01064-X
Taguchi, H., Xie, Y., Horikoshi, N., Maehara, K., Harada, A., Nogami, J., Sato, K., Arimura, Y., Osakabe, A., Kujirai, T. et al. (2017). Crystal structure and characterization of novel human histone H3 variants, H3.6, H3.7, and H3.8. *Biochemistry* **56**, 2184-2196. doi:10.1021/acs.biochem.6b01098
Talbert, P. B. and Henikoff, S. (2010). Histone variants—ancient wrap artists of the epigenome. *Nat. Rev. Mol. Cell Biol.* **11**, 264-275. doi:10.1038/nrm2861
Talbert, P. B. and Henikoff, S. (2014). Environmental responses mediated by histone variants. *Trends Cell Biol.* **24**, 642-650. doi:10.1016/j.tcb.2014.07.006
Talbert, P. B. and Henikoff, S. (2017). Histone variants on the move: substrates for chromatin dynamics. *Nat. Rev. Mol. Cell Biol.* **18**, 115-126. doi:10.1038/nrm.2016.148
Talbert, P. B. and Henikoff, S. (2020). What makes a centromere? *Exp. Cell Res.* **389**, 111895. doi:10.1016/j.yexcr.2020.111895
Talbert, P. B., Bayes, J. J. and Henikoff, S. (2008). The evolution of centromeres and kinetochores: a two-part fugue. In *The Kinetochore: From Molecular Discoveries to Cancer Therapy*, Vol. 1 (ed. P. De Wulf and W. C. Earnshaw), pp. 193-230: Springer.
Tanaka, M., Hennebold, J. D., Macfarlane, J. and Adashi, E. Y. (2001). A mammalian oocyte-specific linker histone gene H1oo: homology with the genes for the oocyte-specific cleavage stage histone (cs-H1) of sea urchin and the B4/H1M histone of the frog. *Development* **128**, 655-664.
Tanaka, H., Iguchi, N., Isotani, A., Kitamura, K., Toyama, Y., Matsuoka, Y., Onishi, M., Masai, K., Maekawa, M., Toshimori, K. et al. (2005). HANP1/H1T2, a novel histone H1-like protein involved in nuclear formation and sperm fertility. *Mol. Cell. Biol.* **25**, 7107-7119. doi:10.1128/MCB.25.16.7107-7119.2005
Tani, R., Hayakawa, K., Tanaka, S. and Shiota, K. (2016). Linker histone variant H1T targets rDNA repeats. *Epigenetics* **11**, 288-302. doi:10.1080/15592294.2016.1159369
Teimouri, M., Najaran, H., Hosseinzadeh, A. and Mazoochi, T. (2018). Association between two common transitions of H2BFWT gene and male infertility: a case-control, meta, and structural analysis. *Andrology* **6**, 306-316. doi:10.1111/andr.12464
Torres, C. M., Biran, A., Burney, M. J., Patel, H., Henser-Brownhill, T., Cohen, A. S., Li, Y., Ben-Hamo, R., Nye, E., Spencer-Dene, B. et al. (2016). The linker histone H1.0 generates epigenetic and functional intratumor heterogeneity. *Science* **353**, aaf1644. doi:10.1126/science.aaf1644
Tramantano, M., Sun, L., Au, C., Labuz, D., Liu, Z., Chou, M., Shen, C. and Luk, E. (2016). Constitutive turnover of histone H2A.Z at yeast promoters requires the preinitiation complex. *eLife* **5**, e14243. doi:10.7554/eLife.14243
Tran, M. H., Aul, R. B., Xu, W., van der Hoorn, F. and Oko, R. (2011). Involvement of Classical Bipartite/Karyopherin nuclear import pathway components in acrosomal trafficking and assembly during bovine and murid spermiogenesis. *Biol. Reprod.* **86**, 1-11. doi:10.1095/biolreprod.111.096842
Ueda, J., Harada, A., Urahama, T., Machida, S., Maehara, K., Hada, M., Makino, Y., Nogami, J., Horikoshi, N., Osakabe, A. et al. (2017). Testis-specific histone variant H3t gene is essential for entry into spermatogenesis. *Cell Rep.* **18**, 593-600. doi:10.1016/j.celrep.2016.12.065
Urahama, T., Harada, A., Maehara, K., Horikoshi, N., Sato, K., Sato, Y., Shiraishi, K., Sugino, N., Osakabe, A., Tachiwana, H. et al. (2016). Histone H3.5 forms an unstable nucleosome and accumulates around transcription start sites in human testis. *Epigenet. Chromatin* **9**, 2. doi:10.1186/s13072-016-0051-y
Van Doninck, K., Mandigo, M. L., Hur, J. H., Wang, P., Guglielmini, J., Milinkovitch, M. C., Lane, W. S. and Meselson, M. (2009). Phylogenomics of unusual histone H2A Variants in Bdelloid rotifers. *PLoS Genet.* **5**, e1000401. doi:10.1371/journal.pgen.1000401
Voon, H. P. J., Hughes, J. R., Rode, C., De La Rosa-Velazquez, I. A., Jenuwein, T., Feil, R., Higgs, D. R. and Gibbons, R. J. (2015). ATRX plays a key role in maintaining silencing at interstitial heterochromatic loci and imprinted genes. *Cell Rep.* **11**, 405-418. doi:10.1016/j.celrep.2015.03.036
Wang, J., Youkharibache, P., Zhang, D., Lanczycki, C. J., Geer, R. C., Madej, T., Phan, L., Ward, M., Lu, S., Marchler, G. H. et al. (2020). iCn3D, a web-based 3D viewer for sharing 1D/2D/3D representations of biomolecular structures. *Bioinformatics* **36**, 131-135. doi:10.1093/bioinformatics/btz502
Waterborg, J. H. (2012). Evolution of histone H3: emergence of variants and conservation of post-translational modification sites. *Biochem. Cell Biol. Biochim. Biol. Cell.* **90**, 79-95. doi:10.1139/o11-036
Waterborg, J. H. and Robertson, A. J. (1996). Common features of analogous replacement histone H3 genes in animals and plants. *J. Mol. Evol.* **43**, 194-206. doi:10.1007/BF02338827
Weber, C. M., Ramachandran, S. and Henikoff, S. (2014). Nucleosomes are context-specific, H2A.Z-modulated barriers to RNA polymerase. *Mol. Cell* **53**, 819-830. doi:10.1016/j.molcel.2014.02.014
Wirbelauer, C., Bell, O. and Schubeler, D. (2005). Variant histone H3.3 is deposited at sites of nucleosomal displacement throughout transcribed genes while active histone modifications show a promoter-proximal bias. *Genes Dev.* **19**, 1761-1766. doi:10.1101/gad.347705
Wong, L. H., Ren, H., Williams, E., McGhie, J., Ahn, S., Sim, M., Tam, A., Earle, E., Anderson, M. A., Mann, J. et al. (2009). Histone H3.3 incorporation provides a unique and functionally essential telomeric chromatin in embryonic stem cells. *Genome Res.* **19**, 404-414. doi:10.1101/gr.084947.108
Wong, L. H., McGhie, J. D., Sim, M., Anderson, M. A., Ahn, S., Hannan, R. D., George, A. J., Morgan, K. A., Mann, J. R. and Choo, K. H. A. (2010). ATRX interacts with H3.3 in maintaining telomere structural integrity in pluripotent embryonic stem cells. *Genome Res.* **20**, 351-360. doi:10.1101/gr.101477.109
Wu, W.-H., Wu, C.-H., Ladurner, A., Mizuguchi, G., Wei, D., Xiao, H., Luk, E., Ranjan, A. and Wu, C. (2009). N terminus of Swr1 binds to histone H2AZ and provides a platform for subunit assembly in the chromatin remodeling complex. *J. Biol. Chem.* **284**, 6200-6207. doi:10.1074/jbc.M808830200
Wu, B. J., Dong, F. L., Ma, X. S., Wang, X. G., Lin, F. and Liu, H. L. (2014). Localization and expression of histone H2A variants during mouse oogenesis and preimplantation embryo development. *Genet. Mol. Res.* **13**, 5929-5939. doi:10.4238/2014.August.7.8
Yan, W., Ma, L., Burns, K. H. and Matzuk, M. M. (2003). HILS1 is a spermatid-specific linker histone H1-like protein implicated in chromatin remodeling during

mammalian spermiogenesis. *Proc. Natl. Acad. Sci. USA* **100**, 10546-10551. doi:10.1073/pnas.1837812100

Yelagandula, R., Stroud, H., Holec, S., Zhou, K., Feng, S., Zhong, X., Muthurajan, U. M., Nie, X., Kawashima, T., Groth, M. et al. (2014). The histone variant H2A.W defines heterochromatin and promotes chromatin condensation in Arabidopsis. *Cell* **158**, 98-109. doi:10.1016/j.cell.2014.06.006

Yoshikawa, G., Blanc-Mathieu, R., Song, C., Kayama, Y., Mochizuki, T., Murata, K., Ogata, H. and Takemura, M. (2019). Medusavirus, a novel large DNA virus discovered from hot spring water. *J. Virol.* **93**, e02130-18. doi:10.1128/JVI.02130-18

Yuen, B. T. K., Bush, K. M., Barrilleaux, B. L., Cotterman, R. and Knoepfler, P. S. (2014). Histone H3.3 regulates dynamic chromatin states during spermatogenesis. *Development* **141**, 3483-3494. doi:10.1242/dev.106450

Zink, L.-M., Delbarre, E., Eberl, H. C., Keilhauer, E. C., Bönisch, C., Pünzeler, S., Bartkuhn, M., Collas, P., Mann, M. and Hake, S. B. (2017). H3.Y discriminates between HIRA and DAXX chaperone complexes and reveals unexpected insights into human DAXX-H3.3-H4 binding and deposition requirements. *Nucleic Acids Res.* **45**, 5691-5706. doi:10.1093/nar/gkx131