Germline histone dynamics and epigenetics
Siew Loon Ooi¹ and Steven Henikoff¹,²

Germ cells have the same DNA sequence as somatic cells, but the processes that act on their chromatin are different. Germline chromatin undergoes a series of dramatic remodeling events during the life cycle of an organism. Different aspects of germline chromatin have been dissected in recent years, such as differences between the sex chromosomes and autosomes in histone variants and modifications. Excitingly, histone dynamics have recently been implicated in imprinted X inactivation and genomic imprinting processes that are independent of DNA methylation. Taken together with observations of core histone retention in mature sperm of diverse animals, histones have become prime candidates for mediating germline epigenetic inheritance.

Addresses
¹ Division of Basic Sciences, Fred Hutchinson Cancer Research Center, 1100 Fairview Ave N., Seattle, WA 98109
²Howard Hughes Medical Institute

Corresponding author: Henikoff, Steven (steveh@fhcrc.org)

Introduction
The human genome comprises ~2 m of DNA, which fits into a cell that is only 10 μm in diameter. Such a high degree of compaction is achieved by histones, which wrap DNA to form chromatin. The basic unit of chromatin is the nucleosome, containing 146 base pairs (bp) of DNA wrapped around the histone core, an octamer consisting of two copies each of H2A, H2B, H3 and H4. Canonical histones are encoded by clusters of genes that are transcriptionally and post-transcriptionally regulated to be tightly expressed during S phase. In contrast, histone variants are encoded by unlinked genes that are expressed throughout the cell cycle. Thus, S-phase histones are incorporated primarily during DNA replication, whereas histone variants can be incorporated throughout the cell cycle. Not only is chromatin structure important for packaging DNA, but chromatin composed of different histone variants or with different post-translational histone modifications has been implicated in diverse biological processes, such as transcriptional silencing and mitosis. For example, centromeres are defined by where centromeric H3 (CenH3), a histone H3 variant, assembles [1–3]. Another histone H3 variant, H3.3, marks active chromatin and is enriched in histone modifications associated with transcriptional activity [4–8]. Histones are thus prime candidates for transmission of epigenetic information — phenotypes that can be stably inherited via cell division without changes in DNA sequence.

Germ cells have the same DNA sequence as somatic cells. However, unlike somatic cells, they are totipotent and can give rise to an entire organism. During the life cycle of an organism, germ-cell chromatin undergoes numerous chromatin remodeling events (Figure 1). During mammalian spermatogenesis, concomitant with its condensation, sperm chromatin undergoes a series of remodeling events that are mediated by S-phase histones, histone variants, transition proteins and protamines [9,10]. The result of these elaborate processes is transcriptionally inert mature sperm DNA, the most highly condensed DNA structure known. During oogenesis, oocyte DNA also becomes more condensed and transcriptionally quiescent. Upon fertilization, the paternal DNA is decondensed, and protamines are replaced by histones to generate chromatin structures compatible with somatic cellular processes, such as transcription. Epigenetic reprogramming events such as genome-wide demethylation of the paternal genome take place after fertilization.

Chromatin remodeling occurs very rapidly upon fertilization. In mice, cytological observations have revealed that most protamines are removed from sperm chromatin within 30 minutes after fertilization, and active demethylation of the paternal genome begins at about this time [11⁺,12]. The time between fertilization and the completion of chromatin remodeling events presents a narrow, but crucial, window of opportunity for the zygote to mark or retain the identity of the parental genome and its associated epigenetic information. Understanding chromatin dynamics during this narrow window of opportunity is crucial for our understanding of how epigenetic information can be transmitted via the germline and has important implications for epigenetic events such as genomic imprinting and X-chromosome inactivation. In this review, we focus on recent progress in understanding germline histone dynamics and its possible implications for epigenetic inheritance.

Histone dynamics during gametogenesis
Histone dynamics in the mouse germline
Important progress in recent years has shed light on the basis for differences between the sex chromosomes and
the autosomes during gametogenesis. In mice, during oogenesis, genes on both X chromosomes are transcriptionally active. During spermatogenesis, the single X chromosome pairs at its pseudoautosomal region with the Y chromosome to form the sex (or XY) body, which is transcriptionally silenced during the pachytene stage of meiotic prophase in a process termed meiotic sex chromosome inactivation (MSCI).

A prominent marker for the sex body is the phosphorylated form of the histone variant H2AX (γ-H2AX) [13]. In response to DNA double-strand breaks, H2AX is phosphorylated at the ‘SQ’ motif at its C terminus [14,15], allowing efficient accumulation of DNA repair proteins. From the leptotene stage of spermatogenesis onwards, γ-H2AX can be detected as punctate staining throughout the nucleus [13]. In late meiotic prophase (pachytene–diplotene), γ-H2AX is detected exclusively on the sex body prior to MSCI [13]. The ubiquitous γ-H2AX pattern at early prophase requires ataxia-telangiectasia mutated (ATM), a kinase that mediates responses to DNA double-strand breaks [16,17]. In contrast, sex-chromosome-specific H2AX phosphorylation is ATM-independent [16,18] but requires BRCA1 (breast cancer 1, an early onset gene) [19], a tumor suppressor protein involved in coordinating cellular responses to DNA damage. BRCA1 itself and ATR (ATM and Rad3-related gene), a candidate protein kinase for H2AX phosphorylation, are also specifically localized to the unpaired region of the XY chromatin in late meiotic prophase [19].

γ-H2AX accumulation on the sex chromosome is crucial for sex body formation, with H2AX-deficient male mice failing to form a sex body [20]. H2AX-deficient male, but not female, mice are infertile, exhibit severe defects in XY chromosome pairing and do not initiate MSCI. Moreover,
other sex body proteins, such as the histone variant macroH2A1.2, do not preferentially localize with the sex chromosomes in H2AX-deficient male mice. Currently, it is unclear why γ-H2AX is required for sex body formation; it may aid in the condensation of the sex body during its establishment or promote efficient accumulation of factors crucial for sex body maintenance.

In addition, the ubiquitous γ-H2AX accumulation at early prophase requires SPO11, a topoisomerase-like protein required for meiotic DNA double-strand breaks [13,16*]. This finding suggests that the early prophase wave of H2AX phosphorylation is related to meiotic recombination. γ-H2AX staining could be observed in SPO11-deficient zygotene-like spermatocytes; 44%, 31% and 25% of spermatocytes contain one, two, or more than two γ-H2AX signals, respectively [16*]. However, X- and Y-chromosomal DNA hybridization signals do not colocalize with the γ-H2AX signal [16*]. In fact, in ~80% of SPO11-deficient zygotene-like spermatocytes, X and Y chromosomes are physically separated and do not synapse. Because the sex body is not formed in SPO11-deficient nuclei, it has not been possible to probe the relationship between γ-H2AX accumulation and meiotic DNA double strand breaks. It is interesting that many DNA damage-related proteins, such as ATR, BRCA1 and RAD51, accumulate on the sex body; however, the function of their accumulation on the sex body remains to be determined.

To determine if unpaired DNA or other features of the sex chromosomes trigger MSCI, artificial situations that result in unpaired DNA during meiosis were created [18**]. Like the sex body, unsynapsed chromosomes, including autosomes, were found to acquire BRCA1, ATR and γ-H2AX during both male and female meiosis and to become transcriptionally silent [18**]. Furthermore, the localization of ATR and γ-H2AX onto unsynapsed chromosomes requires BRCA1 [18**]. These results suggest that unpaired DNA or chromosome asynapsis can drive MSCI. If so, then MSCI might be prevented by providing the X and Y chromosomes with artificial synapsis partners during meiosis [21**]. Indeed, in 12% of the nuclei of XYY male mice, fully synapsed YY chromosomes that are physically separated from the X chromosomes were observed; these did not accumulate γ-H2AX, and transcription could be detected [21**]. A synapsis partner was also provided for an otherwise unpaired X chromosome using male mice carrying the reciprocal X and chromosome 16 translocation (Figure 2). In this case, the X chromosome with the translocation underwent full, but nonhomologous, synapsis with chromosome 16 about half of the time (Figure 2a). Interestingly, even though only part of each chromosome was homologous, full synapsis was observed between the X chromosome with the translocation and the endogenous chromosome 16. These fully synapsed chromosomes do not accumulate γ-H2AX, and the normally silenced X-linked gene becomes active (Figure 2c).

Thus, synapsis of chromosomes, regardless of homology, regulates meiotic silencing, a phenomenon termed meiotic silencing of unsynapsed chromatin (MSUC) [21**]. This was suggested to be the mechanism that leads to MSCI [21**]. As long as full synapsis is achieved, even if the DNA sequence is not homologous, chromosomes do not acquire BRCA1, ATR or γ-H2AX and are not transcriptionally silenced [13,18**]. MSUC differs from meiotic silencing by unpaired DNA (MSUD), described in Neurospora, where DNA homology, not chromosome synapsis, drives the process. MSUC demonstrates that a general silencing mechanism operates during both male and female meiosis that senses and silences unsynapsed chromosome segments. MSUC may have evolved to defend the genome from foreign DNA or to help detect and eliminate cells with synaptic errors prior to late pachytenes [18**,22].

Other chromatin differences between the sex chromosome and autosomes have also been reported. The sex body is depleted of S-phase histone H3 from the mid or late pachytene stage onwards, concomitant with incorporation of the constitutive histone H3.3 variant, suggesting that nucleosome replacement occurs during MSCI [23*]. In contrast to the phosphorylation of H2AX, the depletion of S phase H3 is a result of MSCI — its depletion is not detected in the spermatocytes of Setp1−/− mice that do not synapse and fail to form a sex body. During the pachytene stage of spermatogenesis, the X and Y chromosomes also acquire histone H3 dimethyl lysine 9 (H3K9me2), a histone modification associated with transcriptional silencing, and undergo extensive histone H3 and H4 deacetylation that persist into early spermiogenesis, consistent with observations that the sex body undergoes MSCI [24].

Histone dynamics in the C. elegans germline

How widespread is MSCI/MSUC? Interestingly, differences between X chromosomes and autosomes have been documented in C. elegans [25,26,27**]. During meiosis, the single X chromosome in males (XO) and the two X chromosomes in hermaphrodites (XX) are deficient in H3.3 [27**] and in histone modifications associated with transcriptional activity, such as histone H3 K4 dimethyl (H3K4me2) [25,26]. Only during the postpachytene stages of oogenesis, coincident with a burst of expression of X-linked genes involved in oogenesis [28], do the X chromosomes begin to accumulate H3.3 and histone modifications associated with transcriptional activity [25,26,27**]. In addition, the single X chromosome of C. elegans male is enriched for H3K9me2 and condenses earlier than autosomes during the pachytene stage [26]. Interestingly, H3K9 dimethylation can occur during both oogenesis and spermatogenesis, and is determined by the pairing status of the X chromosome, but not by germline

www.sciencedirect.com

Author's personal copy

Like MSCI in mice, the lack of a pairing partner causes the single X chromosome to acquire H3K9me2 during male worm meiosis and become transcriptionally silent. H3.3 deficiency on the X chromosome is in contrast to the incorporation of H3.3 onto the sex body during MSCI in mice. The X chromosome of *C. elegans* is depleted of germline-specific genes, so perhaps the X chromosome deficiency in histone variants and modifications associated with transcriptional activity is simply a consequence of the lack of X-linked genes expressed in germ cells rather than the result of meiotic silencing. H3K9me2 is associated with silent chromatin; however, it is unclear if H3K9me2 occurs by default on histones prior to nucleosome assembly or results from methyltransferases acting on nucleosomes. Nor is it known if the *C. elegans* X chromosomes also acquire DNA repair/damage-related proteins as occurs on the sex body during meiosis in mouse. Thus, it remains to be determined if MSUC is a conserved feature among organisms.

**Germline histone dynamics and epigenetics**

How long does MSCI persist and does it have consequences beyond meiosis? By the spermatid stage in mice the X and Y chromosomes display RNA polymerase II staining, suggesting that at least some X and Y chromosome genes are being reactivated. However, 87% of X-linked genes remain suppressed post-meiotically, and meiotically active autosomal genes, when inserted into X chromosomes, remain suppressed in spermatids. In female mouse embryos, it is always the paternal X chromosome that is inactivated in the placenta. It has been proposed that the paternal X chromosome is inherited in a pre-inactivated form, on the basis of the absence...
of active transcript staining on the paternal X chromosome in two-cell embryos [31]. However, MSCI was found not to be required for imprinted inactivation of a Xist-containing transgene [32]. MSCI persists into spermatids, but whether it has consequences beyond meiosis remains an interesting possibility.

A study in C. elegans pointed to the possibility that the history of X chromosomes during meiosis may have detectable effects in the next generation [22]. An imprint has been described in C. elegans early embryos, whereby histone H3 modifications associated with transcriptional activity are deficient specifically on the paternal X chromosome [22]. Interestingly, the stability of this imprint in the early embryos was affected by the pairing status of the X chromosome during meiosis. The imprint persists longer in embryos generated from fertilization of a hermaphrodite by an XO male worm than in those generated from self-fertilization of a hermaphrodite, in which the paternal X chromosome has a pairing partner during meiosis. The functional significance of this imprint remains unknown.

Histone retention in mature sperm

The final product of spermatogenesis is highly condensed, transcriptionally inert protamine-packaged mature sperm. The replacement of histones by transition proteins followed by protamines was once thought to be complete. However, this might simply reflect the fact that the detection of histones in mature sperm using immunofluorescence in cytological preparations has been a challenge, because the highly condensed nature of sperm can render the epitope inaccessible. Nevertheless, over the past 20 years, a few studies have used other techniques, such as high-performance liquid chromatography and western blotting, to document the presence of histones in mature sperm. Histones, including the histone variants H2AX and H3.3, have been detected in human sperm [33,34]. CenH3 (CENP-A) has been purified from bull sperm and found to be restricted to discrete spots that are likely to correspond to centromeres [35,36].

In recent years, modern technologies have been used to analyze the proteome of sperm in C. elegans and Drosophila. Proteomic analysis was performed for chromatin-associated proteins of spermatogenic germ cells in C. elegans [37]. Three small sperm-specific nuclear basic proteins were identified, in addition to all four S-phase histones. Interestingly, the universal replication-independent histone variants H3.3 and H2AZ, and a sperm-specific histone H2A variant, HTAS-1, were also identified. Furthermore, fluorescence analysis of a worm strain expressing H3.3 fused to GFP showed that H3.3 is a component of mature sperm in C. elegans [27**]. The Drosophila proteomic analysis was performed using purified mature sperm, and this study also identified core histones as components of mature sperm [38]. Taken together with earlier observations, these studies demonstrated that core histones are retained in mature sperm.

In human sperm, ~15% of nuclear basic proteins are histones [33]. Because gene transcription units occupy <3% of the human genome, in theory the low amount of histones retained in mature sperm is sufficient to package all coding regions in the genome. This possibility raises the intriguing question of whether histone retention in mature sperm serves a positive function or is merely the result of incomplete replacement by transition proteins or protamines. Two early studies addressed whether histone retention in mature sperm occurs in specific or random genomic regions. Human sperm DNA was separated into histone- or protamine-bound fractions and sequences from both fractions were cloned [39]. Size-selected single-copy clones derived from these two fractions were found to not cross-hybridize, but rather to hybridize preferentially to histone- or protamine-bound sperm DNA, respectively [39]. In another study, the chromatin structure of the β-globin gene family was analyzed in human sperm. Genes coding for ε and γ globin, which are active in the embryonic yolk sac, preferentially associate with histones, while genes coding for β and δ globin, which are silent in the embryonic yolk sac but expressed later during development, preferentially associate with protamines [40]. These results suggest that histones are retained in mature sperm non-randomly across the genome and that their retention might be linked to gene activity. Considering that spermatogenesis involves multiple chromatin remodeling events, histone retention in mature sperm raises the question of how histones resist replacement by transition proteins and protamines.

Histone retention in mature sperm might serve multiple roles. First, during spermatogenesis, as sperm DNA becomes more condensed, genes transcribed at later stages of spermatogenesis might need to be free of transition proteins and protamines. Second, zygotic transcription can be detected at the one-cell stage in mice, but it is not clear how rapid and complete chromatin remodeling is upon fertilization [41]. Prepacking of very early developmental genes with histones in sperm might prevent their tight condensation by protamines and thus facilitate their transcription in early embryos. Third, genomic regions packaged by histones in mature sperm might serve as nucleation sites for chromatin remodeling upon fertilization. Finally, histones are the best candidates for mediating epigenetic information, and their retention in mature sperm raises the possibility that they do so via the germline.

Chromatin remodeling upon fertilization

Upon fertilization, chromatin undergoes dramatic remodeling, and recent studies have elucidated the role of histone variants in this process. An important advance in understanding the mechanism of chromatin remodeling...
upon fertilization was the characterization of the *Drosophila sesame* mutant [42**]. Upon fertilization of *sesame* eggs, the paternal DNA does not decondense. Molecular cloning showed that *ssm* encodes a loss-of-function point mutation in HIRA, the chaperone for H3.3/H4 nucleosome assembly. In addition, FLAG-tagged-H3.3 was found to be specifically assembled onto paternal chromatin upon fertilization prior to the first round of DNA replication, suggesting that only H3.3 is used to remodel paternal chromatin [42**]. Similar incorporation of H3.3 into the paternal chromatin was also reported for mice, where, in the pre-S phase zygote, the paternal chromatin stains positive for HIRA, but is devoid of S-phase H3 [11*]. Preferential incorporation of H3.3 into the male pronucleus upon fertilization occurs prior to genome activation in mice [43*]. These results showed that, upon fertilization, the H3.3 histone variant is used to remodel paternal chromatin. Incorporation of H3.3 into the paternal chromatin upon fertilization was also reported in *C. elegans* [27**], where, in contrast to the situation for flies and mice, H3.3 is imported into both the paternal and maternal pronuclei upon fertilization. The realization that histone variants are used to remodel chromatin upon fertilization provides an opportunity to use them as tools to directly probe the chromatin remodeling process upon fertilization.

**Histones dynamics and imprinting**

Genomic imprinting is the mono-allelic expression of genes from only one of the parental chromosomes. Imprints are established during gametogenesis and are often associated with DNA methylation. Genomic imprinting occurs in both the embryonic and extraembryonic (placental) tissues, although some genes are imprinted only in the placenta. All exclusively placental imprinted genes known to date are silenced paternally and expressed from the maternal alleles.

Recent advances have uncovered similarities between placental genomic imprinting and imprinted X inactivation, which is the specific inactivation of the paternal X chromosome. Imprinted X inactivation occurs in pre-implantation embryos, and is later maintained only in the placenta [44,45]. In contrast, the choice of which X chromosome to inactivate in the post-implantation embryo is random. Both placental genomic imprinting and imprinted X inactivation utilize non-coding RNAs *in cis*, such as XIST RNA for imprinted X inactivation and Kcnq01 RNA for chromosome 7 IC2 locus imprinting. Interestingly, neither process requires the DNA maintenance methyltransferase DNMT1 [46]. Instead, as with imprinted X inactivation, these placental imprinted genes are marked by histone modifications associated with transcriptional repression — H3K9me and histone H3 methylated lysine 27 (H3K27me) — as well as the Polycomb H3K27 methyltransferase complex, which is involved in the maintenance of transcriptional repression [46–51]. It has been suggested that histone modification is the more ancient imprinting system, whereas DNA methylation, a more stable mark, would have evolved later to maintain imprinting [45,52].

Placental genomic imprinting also resembles genomic imprinting in plants. Plant imprinting has been described only in the endosperm, the seed component that provides nutrients to the embryo during seed development and that is analogous to the mammalian placenta. Most endosperm-imprinted genes are silenced paternally and expressed from the maternal alleles only. Studies of *MEDEA (MEA)* gene imprinting in plants have led to surprises [53*]. The maternal allele of *MEA* is kept active by DEMETER (DME), a DNA glycosylase/lyase that excises 5-methylcytosine, which is followed by DNA repair, resulting in DNA demethylation [53*]. In *dme* mutant endosperm, the maternal allele of *MEA* is methylated and not expressed. *MEA* encodes the plant ortholog of E(z), a Polycomb complex protein. Surprisingly, the silencing of the maternal *MEA* paternal allele is not maintained by DNA methylation [53*]. Instead, the paternal *MEA* allele is enriched in H3K27me, and its silencing is controlled by Polycomb complex proteins expressed from the maternal genome, including MEA itself. It would be interesting to determine if an analogous DNA glycosylase is also used for active DNA demethylation in mammals. Like placental-imprinted genes and imprinted X inactivation, silencing of the paternal allele is independent of the DNA maintenance methyltransferase (*MET1*, which is orthologous to mammalian DNMT1), but is marked by ‘repressive’ histone modifications including H3K27me. It will be important to determine the role of histone dynamics in both the establishment and maintenance of genomic imprinting.

**Histones as candidates for germline epigenetic inheritance**

Histone variants and modifications are potentially capable of transmitting epigenetic information through mitosis. However, a role for histones in transmitting epigenetic information through the germline or meiosis has yet to be demonstrated. In fact, examples of germline epigenetic inheritance in animals are scarce (reviewed in [54]). Most such examples are manifested through transgene expression, for example in *C. elegans* [55*], *Drosophila* [56] and mice [57]. Other examples include the paternal X chromosome imprint in *C. elegans* [22] and the imprints observed in the dominant *agouti viable yellow* (A*) and axin-fused alleles in mice (reviewed in [58]).

Now that histone dynamics have been implicated in imprinting and that many examples of histone retention in mature sperm have emerged, there has been renewed interest in probing whether histones transmit epigenetic information through the germline. How is the ‘memory’ transmitted via the germline? The occurrence of genomic
imprinting and imprinted X inactivation implies that the zygote retains ‘memory’ of which genes or chromosomes are paternally derived. The basis of this memory could be DNA methylation, but, if so, it must resist the active genome-wide DNA demethylation that occurs in the mammalian paternal genome upon fertilization [12]. What makes DNA methylation of imprinted genes resistant? Perhaps this resistance is mediated by histones.

That memory can be mediated by histones is best exemplified by the retention of cenH3 in mammalian sperm [35]. In higher eukaryotes, centromere location appears to be determined by where CenH3s assemble into centromeric nucleosomes [2,3]. The retention of CenH3 in mammalian sperm suggested that CenH3 helps the next generation maintain the centromere locus, and we propose that a similar epigenetic process involving other histone variants and modifications underlies imprinting. Histone H3 variants are prime candidates for this. Histone H3.3 marks active genes and is enriched in histone modifications associated with transcriptional activity [4–8]. According to our proposal, the paternal placental imprinted genes should be packaged in S-phase activity [4–8]. According to our proposal, the paternal placental imprinted genes should be packaged in S-phase activity [4–8]. According to our proposal, the paternal placental imprinted genes should be packaged in S-phase activity [4–8].

Conclusions
Given the growing evidence that histone modifications and variants are crucial for epigenetic memory in the soma, it is surprising that so little is known about the significance of histone retention in sperm. Where in sperm genomes are histones localized? What processes might be responsible for the transmission of epigenetic information by histones via mature sperm? With advances in high-resolution technologies such as microarrays and proteomic analyses, the time is now ripe to probe the possibility that histones may be carriers of germline epigenetic information.

Acknowledgements
We would like to thank Diana Chiu, Yamini Dalal, Omar Fernandez-Capetillo, Mary Gehring, Andre Nussenzweig and Godfried van der Heijden for discussions. SLQ is supported by the Damon Runyon Cancer Research Foundation (DRG-1818-04).

References and recommended reading
Papers of particular interest, published within the period of review, have been highlighted as:
● of special interest
○ of outstanding interest
In the pre-S phase mouse zygote, the paternal, but not the maternal, pronucleus is found to be specifically devoid of S phase histones. In addition, the histone H3.3/H4 nucleosome assembly chaperone, HIIRA, can be detected only in the paternal chromatin, suggesting that H3.3 is preferentially incorporated into the paternal chromatin upon fertilization. This work shows that in the early mouse zygote, the paternal and maternal chromatin can be distinguished from each other.
15. Rogakou EP, Pilch DR, Orr AH, Ivanova VS, Bonner WM, Burgoyne PS: Recombination DNA double-strand breaks in mice 'moonlight' in the formation of chromosomes. Therefore, some of the machinery involved in meiotic double-strand break formation and repair 'moonlights' in the formation of the sex body.
sex body, could be induced on unsynapsed chromosomes during both male and female meiosis. Therefore, lack of synapsis appears to trigger SSC.


Unsynapsed chromosomes become meiotically silenced during both male and female meiosis. The authors show that meiotic silencing can be prevented by providing the sex chromosome with a synopsis partner. Surprisingly, synopsis alone prevents meiotic silencing, even when the chromosomes are not homologous. This meiotic silencing of unsynapsed chromatin or MSUC is proposed to be the basis for SSC.


24. Khalil AM, Boyar FZ, Driscoll DJ: Histone H3.3 incorporation into the sex body and concomitant depletion of H3 are found to occur during the induction of SSC, suggesting that chromosome-wide nucleosome replacement occurs during SSC. In contrast to γ-H2AX accumulation, H3 variant replacement is a consequence of SSC. H3 variant replacement also occurs during MSUC.


45. Reik W, Dean W, Walter J, Lewis A, Mitsuya K, Umlauf D, Smith P: The Authors demonstrate that maternal H3.3 is preferentially incorporated into the paternal X chromosome upon fertilization, and this incorporation occurs prior to zygotic transcription. H3.3 was also detected in the nuclei of early embryos, suggesting that the epigenetic mechanism in the early embryo involves histone replacement.


Active demethylation of DNA is shown to occur via the action of the DEMETER glycosylase/lyase. DEMETER is required for expression of the maternal allele of MEDEA, a Polycomb complex gene. The authors find that silencing of the maternal allele of MEDEA is independent of DNA methylation. Instead, the paternal allele of MEDEA is enriched in repressive histone modifications, and its silencing is maintained by maternally expressed Polycomb group proteins, including MEDEA itself.


The authors show that the C. elegans germline is capable of being imprinted. Certain transgenes, when transmitted via the sperm, display 1.5–2-fold higher expression than when transmitted via the egg. Interestingly, the imprint resulting from sperm transmission could be reset when transmitted through the oogenic germline.

