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DEVELOPMENTAL EPIGENOMICS

The Polycomb landscape in mouse development

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Polycomb group proteins assemble into two primary complexes—Polycomb repressive complex (PRC) 1 and 2— that safeguard cell fate by repressing gene transcription. Two new studies explore the PRC1 landscape during the transition from gametes to embryogenesis in mice, providing insight into the intergenerational transmission of epigenetic information and gene regulation dynamics as embryos prepare for gastrulation.

PRC1 and PRC2 are two evolutionarily conserved complexes that impact cell identity and fate via an epigenetic mode of gene repression. In classical Polycomb-mediated regulation, PRC2 catalyzes histone H3 lysine 27 trimethylation (H3K27me3), a mark recognized by PRC1, which in turn deposits histone H2A lysine 119 monoubiquitination (H2AK119ub1). However, more recent data have indicated that in animals the relationship is not quite so straightforward. Variant forms of PRC1 (vPRC1) can act upstream of PRC2 and initiate the canonical pathway in flies and mice^{1–5}. Furthermore, genetic perturbation indicates that PRC2 mutant mice die during gastrulation⁶, whereas complete PRC1 mutants exhibit an even more severe arrest at the 2-cell stage⁷. Thus, the relationship between the two complexes is not unidirectional. Genome-wide patterns of H3K27me3 in mouse gametes and embryos have been described⁸, however H2AK119ub1 maps have not been so readily achieved. In this issue, Mei et al.⁹ and Chen et al.¹⁰ utilized emerging technology—as well as complementary genetic approaches—to shine light on the interplay between PRC1 and PRC2 during the earliest windows of mammalian development.

H2AK119ub1 dynamics

In order to assess allele-specific H2AK119ub1 profiles during embryogenesis, both groups employed Cleavage Under Targets and Release Using Nuclease (CUT&RUN), a method for semi-quantitative profiling of chromatin-bound factors genome-wide¹¹, in F1 hybrid mice. The technique is particularly useful for samples that are difficult to harvest, as high signal-to-noise ratios reduce the number of cells required. These H2AK119ub1 maps add to a publicly available tapestry of histone modifications, DNA polymerase, DNA methylation and transcriptional profiles in the early mouse embryo¹². It should be noted that neither group collected CUT&RUN samples derived from reciprocal crosses of F1 hybrids, which can confound parent-specific effects with strain-specific effects¹³.

Exploring these data, both groups showed widespread H2AK119ub1 establishment during oogenesis, resulting in large domains that overlap the promoters of genes (**Fig. 1a**). Integrated analysis with existing datasets⁸ revealed a strong correlation between H2AK119ub1 and H3K27me3 in mature oocytes. Dynamics between PRC1 and PRC2 during oocyte progression

were not sampled, thus it is difficult to ascertain the stepwise establishment of the Polycomb landscape in the female germline.

Conversely, both groups generated maps of H2AK119ub1 at key stages of early embryogenesis. Strikingly, following fertilization, the symmetry between H2AK119ub1 and H3K27me3 observed in oocytes is lost over gene deserts. While H2AK119ub1 is depleted, H3K27me3 is stably retained through the blastocyst stage. In contrast, a distinct set of gene-rich loci devoid of both marks in the oocyte show sequential gain of H2AK119ub1 in the early 2-cell embryo followed by H3K27me3 gain in the blastocyst and epiblast. The findings reported here provide noteworthy *in vivo* data consistent with the studies largely performed in cell culture that demonstrated that PRC1 can act upstream PRC2^{3,5}. In embryos, symmetry is restored following implantation, where H2AK119ub1 and H3K27me3 domains are restricted to their canonical targets: the promoters of developmentally important genes.

PRC1 impacts embryogenesis

To understand the biological significance underlying the H3K27me3 and H2AK119ub1 dynamics, it is paramount to interrogate the relationship between PRC1 and PRC2 through genetic means. To circumvent the severe PRC1 mutant phenotype, the two groups undertook complementary approaches: Mei et al. genetically ablated PRC1 subunits (*Pcgf1/6*) in oocytes to assess the role of PRC1 in the female germline and embryo (**Fig. 1b**). Importantly, PCGF1 and PCGF6 are components of vPRC1. Chen et al. utilized a PRC2 mutant (*Eed*) to determine the role of H3K27me3 in H2AK119ub1 establishment in oocytes (**Fig. 1c**). Additionally, Chen et al. overexpressed Polycomb repressive deubiquitinase (PR-DUB) in zygotes, which efficiently removes H2AK119ub1 (**Fig. 1d**).

Pcgf1/6 knockout (KO) oocytes exhibit substantial loss of H2AK119ub1, but still retain the vast majority (~88%) of H3K27me3 (**Fig. 1b**). RNA-seq analysis of oocytes globally devoid of H2AK119ub1 show specific upregulation of the subset of genes that had co-incidentally lost H3K27me3. Previous work demonstrated that PRC2 controls a non-canonical form of transient maternal imprinting and imprinted X-inactivation^{14,15}. Pertinently, embryos generated by fertilizing *Pcgf1/6* KO oocytes with wild-type sperm exhibit upregulation of a subset of this class imprinted genes, including *Xist*—the key upstream regulator of X-inactivation. Analysis of extra-embryonic tissues demonstrates a failure of imprinting, associated with increased placental size. In sum, vPRC1 plays a key role in establishing repressive modifications transmitted from oocyte to embryo.

Genetically deleting *Eed* (essential for PRC2 activity) revealed that while H3K27me3 is globally lost, H2AK119ub1 is largely unaffected in oocytes and morula-stage embryos (**Fig. 1c**). Exceptionally, H2AK119ub1 is lost over non-canonically imprinted regions and *Xist*^{14,15}. Hence, there appears to be notably strong link between PRC1 and PRC2 to regulate this class of genes.

Chen et al. interrogated the role of H2AK119ub1, *per se*, by directly injecting PR-DUB mRNA into fertilized zygotes (**Fig. 1d**). Interestingly, while H2AK119ub1 was lost, H3K27me3 was unaffected, and RNA-seq analysis showed no change in imprinted gene expression in 4C embryos. Taken together, it appears PRC1 regulation of imprints likely occurs in the oocyte, possibly through PRC2 recruitment, but is dispensable in the embryo. Crucially, however, depleting H2AK119ub1 results in premature activation of canonical target Polycomb-regulated genes and arrest at the 4-cell stage.

The PR-DUB overexpression experiment strongly indicates that H2AK119ub1 is immensely important for the earliest stages of embryo progression. Both groups make the striking finding that PRC1 regulation at this class of genes occurs seemingly independently of PRC2, perhaps explaining the milder PRC2 mutant phenotype. While formal confirmation awaits, this suggests that upstream vPRC1 activity is taking place to repress Polycomb-regulated genes. However, it begs the question of why PRC2 does not nucleate at these regions soon after fertilization. Are key factors that recruit PRC2 to vPRC1-marked regions absent, or perhaps antagonistic factors are present? Moreover, what is the biological rationale for limiting PRC2 activity at these stages? Finally, what is the mechanistic explanation for the sufficiency of vPRC1 for repression in this window? Future work will hopefully address these exciting questions, further elucidating Polycomb-mediated gene regulation in the context of *in vivo* mammalian development.

References

1. Lagarou, A. et al. dKDM2 couples histone H2A ubiquitylation to histone H3 demethylation during Polycomb group silencing. *Genes Dev* **22**, 2799-2810 (2008).
2. Tavares, L. et al. RYBP-PRC1 complexes mediate H2A ubiquitylation at polycomb target sites independently of PRC2 and H3K27me3. *Cell* **148**, 664-678 (2012).
3. Blackledge, N. P. et al. Variant PRC1 complex-dependent H2A ubiquitylation drives PRC2 recruitment and Polycomb domain formation. *Cell* **157**, 1445-1459 (2014).
4. Kahn, T. G. et al. Interdependence of PRC1 and PRC2 for recruitment to Polycomb Response Elements. *Nucleic Acids Res* **44**, 10132-10149 (2016).
5. Almeida, M. et al. PCGF3/5-PRC1 initiates Polycomb recruitment in X chromosome inactivation. *Science* **356**, 1081-1084 (2017).
6. O'Carroll, D. et al. The polycomb-group gene *Ezh2* is required for early mouse development. *Molecular and Cellular Biology* **21**, 4330-4336 (2001).
7. Posfai, E. et al. Polycomb function during oogenesis is required for mouse embryonic development. *Genes Dev* **26**, 920-932 (2012).
8. Zheng, H. et al. Resetting epigenetic memory by reprogramming of histone modifications in mammals. *Molecular Cell* **63**, 1066-1079 (2016).
9. Mei, H. et al. H2AK119ub1 guides maternal inheritance and zygotic installation of H3K27me3 in mouse embryos. *Nature Genetics* (2021). [NG-A55842R2 Inoue; accepted]
10. Chen, Z. et al. Distinct dynamics and functions of H2AK119ub1 and H3K27me3 in mouse preimplantation embryos. *Nature Genetics* (2021). [NG-A55846R2 Zhang; accepted]
11. Skene, P. J. & Henikoff, S. An efficient targeted nuclease strategy for high-resolution mapping of DNA binding sites. *Elife* **6**, (2017).
12. Xia, W. & Xie, W. Rebooting the Epigenomes during Mammalian Early Embryogenesis. *Stem Cell Reports* **15**, 1158-1175 (2020).
13. Wang, X. & Clark, A. G. Using next-generation RNA sequencing to identify imprinted genes. *Heredity* **113**, 156-166 (2014).

14. Inoue, A. et al. Maternal H3K27me3 controls DNA methylation-independent imprinting. *Nature* **547**, 419-424 (2017).
15. Inoue, A. et al. Maternal Eed knockout causes loss of H3K27me3 imprinting and random X inactivation in the extraembryonic cells. *Genes Dev* **32**, 1525-1536 (2018).

H2AK119ub1
H3K27me3

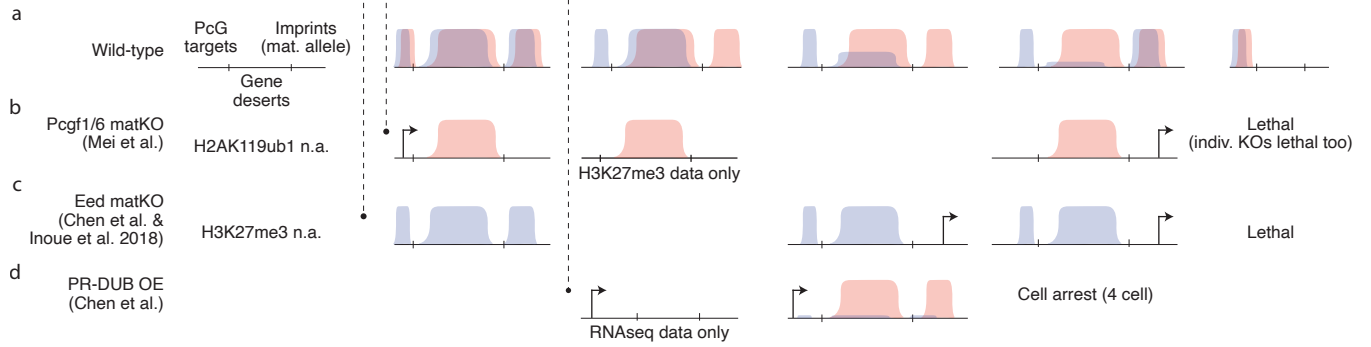
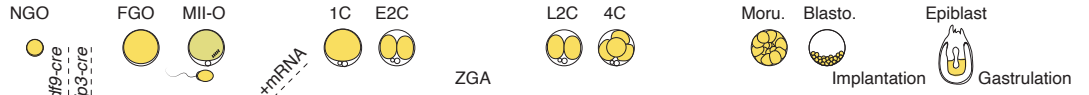


Fig. 1: Polycomb and transcriptional landscapes in normal and modified mouse embryos. H2AK119ub1 and H3K27me3 dynamics over Polycomb Group (PcG) target gene promoters, gene deserts and the maternal allele of non-canonically imprinted genes in oocytes and embryos in **a**, wild-type **b**, *Pcgf1/6* maternal knockouts **c**, *Eed* maternal knockouts **d**, PR-DUB overexpressers. PcG; Polycomb Group, NGO; non-growing oocyte, FGO; fully grown oocyte, MII-O; meiosis II oocyte, 1C; 1-cell, E2C; early 2-cell, ZGA; zygotic genome activation, L2C; late 2-cell, 4C; 4-cell, moru.; morula, blasto., blastocyst; n.a., not available. Arrows represent ectopic transcription.