Knocking off their Sox: lineage-specific repression by Polycomb in epidermal stem cells

Tudorita Tumber

Molecular Biology and Genetics, Cornell University, Ithaca, NY, USA. Correspondence to: tt252@cornell.edu

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A recent publication in *The EMBO Journal* (Bardot *et al*, 2013) provides novel insights into lineage specification during the development of the mouse skin. Ezhkova and colleagues demonstrate that Ezh1 and Ezh2, core enzymes of the Polycomb Repressive Complex (PRC), restrict differentiation of Merkel cells, a specialized population of mechanosensory cells by directly repressing the cell fate determinant transcription factor (TF) Sox2 in epidermal progenitors.

Generally, lineage determination starts with morphogenetic gradients, triggering signalling events that converge on specific TFs, which in turn remodel one cellular state into another. With the exception of a few cases, such as Oct4 and Nanog in embryonic stem cells (ESCs) and MyoD in muscle cells, lineage determinant TFs are not cell type specific. Instead, unique combinations confer certain differentiation states (Boyer *et al*, 2006). Given the right combination of TFs, many cells to work with, and sufficient time, it might actually be possible to convert any cell type into another. Examples range from transdifferentiation of fibroblasts into muscle cells or neurons (Pournasr *et al*,

2011) to turning terminally differentiated cells into pluripotent stem cells (Yamanaka and Blau, 2010). However, such dramatic fate conversions occur with low efficiency, which might be due to the requirement to change the epigenetic landscape that determines the transcriptional potential of each cell.

Studies using cultured ESCs investigated the interplay between the chromatin context, often defined as the status of covalent modifications of histone proteins, and TFs. These studies demonstrated that Ezh1/2, the enzymes that catalyse trimethylation of lysine 27 on histone H3 (H3K27Me3), play an essential role in repressing lineage determinant TFs, while ensuring maintenance of the pluripotent stem cell state. Upon differentiation of ESCs, lineage-specific genes lose H3K27Me3 marks and are activated, while genes not required for lineage commitment remain repressed (Boyer *et al*, 2006; Shen *et al*, 2008).

However, the role of Polycomb-mediated gene repression in tissue stem/progenitor cell differentiation during morphogenesis and homeostasis *in vivo* is not well understood. Perturbing H3K27Me3 patterns during development was

Figure 1 In tissue progenitors, PRC complexes promote stem cell self-renewal by inhibiting various cell cycle regulators, such as INK4A/ARF/INK4B, E2F/RB, P21 and P57. In addition, in conjunction with other epigenetic pathways they keep transcription factors essential for lineage determination (depicted here as XY, Z and QT) repressed. Certain lineages (in this case Lineage 1) depend exclusively upon loss of PRC repression to upregulate fate determinants. However, most lineages are correctly specified even in the absence of PRC, suggesting that either they are independent of PRC or that they depend on additional epigenetic factors.

predicted to have catastrophic effects, and indeed, ablation of *Ezh2* resulted in early embryonic lethality (O'Carroll *et al*, 2001). Yet, tissue-specific deletion of *Ezh2* has less dramatic phenotypes than initially expected. For example, loss of *Ezh2* results in the acceleration of developmental steps, such as early stratification and acquisition of barrier function in the skin with no apparent effect in postnatal life (Ezhkova *et al*, 2009). Similarly, in the brain, *Ezh2* loss induces a premature switch from neurogenesis to gliogenesis (Pereira *et al*, 2010). In addition, *Ezh2* deletion seems to affect the rate of cell differentiation, rather than the lineage choice, as seen in adult muscle (Juan *et al*, 2011) or upon hormonal induction in the mammary gland (Pal *et al*, 2013). *Ezh2* function during development seems to be mediated in part by promoting tissue progenitor cell proliferation, in particular by repressing the *Ink4a/Arf/Ink4b* tumour suppressor locus (Sauvageau and Sauvageau, 2010); (Figure 1).

How about the regulation of lineage determining TFs? The first surprise came from the observation that in *Ezh2*-deficient tissue progenitors, de-repression of mixed lineage genes was mild, if it occurred at all (Shen *et al*, 2008; Ezhkova *et al*, 2009, 2011; Pereira *et al*, 2010; Juan *et al*, 2011; Pal *et al*, 2013). Still more surprising was that even lineage-specific factors that need to be turned on during subsequent differentiation steps cannot be fully de-repressed in the progenitors upon loss of the H3K27Me3 mark.

The current study (Bardot *et al*, 2013) is the first to demonstrate the direct functional repression of lineage determinant TFs by H3K27Me3 in tissue progenitor cells *in vivo*. Explicitly, the authors investigate specification

of Merkel cells, a specialized subtype of post-mitotic epithelial-derived cell, which are involved in mechano-transduction and are innervated by sensory neurons. They identify Sox2 as an important lineage determinant of Merkel cells in the adult skin. This function is mediated by direct transcriptional activation of *Atoh1*, a TF already known to be required for Merkel cell specification. Sox2 and *Atoh1*, as well as promoters of other TF, such as *Isl1* are marked with H3K27Me3 in epidermal progenitors, and *Ezh1/2* loss in epithelial skin results in the activation of these genes, which leads to increased Merkel cell differentiation. Importantly, in *Ezh1/2/Sox2* triple knockout skin, Merkel cell numbers are normal, demonstrating an essential role for Sox2 downstream of *Ezh1/2* in promoting Merkel cell fate. Therefore, these results suggest that in addition to ESCs, Polycomb-mediated repression of lineage determinant TFs also occurs in skin progenitors, leading to the maintenance of the epidermal stem cell state. Given the redundancy of *Ezh1* and *Ezh2* in the skin, it is possible that simultaneous targeting in other tissue stem cells will uncover more H3K27Me3-dependent lineage determinant TFs. Although it is obvious that Polycomb-mediated gene regulation does not act alone during lineage determination in normal development, the data presented by Ezhkova and colleagues indicate that at least some specialized lineages may exclusively depend on de-repression of H3K27Me3-marked gene loci for cell fate specification *in vivo* (Figure 1).

Conflict of interest

The author declares that she has no conflict of interest.

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