Connecting the dots

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Crosstalk between aging and the epigenome

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Epigenetic states help maintain cell identity but they are also dynamic entities that respond to signals. Indeed, cells undergoing developmental changes are characterized by global rearrangements of the epigenetic landscape. Recent studies suggest that aging is one such epigenome-shifting developmental event. What’s more, epigenetic regulators seem to influence the aging process. Aging can occur in different contexts. Here we discuss the emerging evidence that both organismal and cellular aging as well as histone protein aging have intimate connections to the epigenome.

Striking links between organismal aging and epigenome alterations have recently been identified in humans and metazoan model organisms (Feser and Tyler, 2011). For example, tissues and cells of aging organisms show increased levels or redistribution of heterochromatic marks such as trimethylation of lysine 9 on histone H3 (H3K9me3) (Kreiling et al., 2011; Wood et al., 2010). The cause and biological significance of these changes are still unclear; however, a driver for epigenome changes could be an aging program that changes the expression of chromatin modifying or demodifying enzymes. For instance, loss of H3K27me3 in (prematurely) aging human brains is accompanied by increased expression of the H3K27-specific demethylase UTX (Jin et al., 2011). Aging somatic cells in C. elegans show a similar decline in H3K27me3 and increase in UTX-1 expression (Jin et al., 2011; Maures et al., 2011). Importantly, genetic inactivation of C. elegans UTX-1 prevents the age-induced loss of H3K27me3 and extends life span of the worm via the insulin signaling-pathway, a major life span regulator (Jin et al., 2011; Maures et al., 2011). These findings suggest that loss of H3K27me3, a repressive mark associated with regions of facultative heterochromatin, is not only associated with aging but may in fact be causally involved in the aging process. Recent genetic studies in flies and worms have implicated several additional histone modifiers in life span determination (Greer et al., 2011; Siebold et al., 2010).

In addition to the organismal aging program, stochastic events can also induce epigenomic changes over time. For example, human monozygotic twins have very similar DNA methylation patterns at birth, but they also show differences that can increase as they get older (Bell and Spector, 2011). These changes may be attributed to environmental influences. Another possibility is that unavoidable errors occur during the maintenance of epigenetic patterns due to the disruptive nature of transactions at the genome. Transcription by RNA polymerases has the potential to reshuffle or even erase epigenetic signals because it requires the transient eviction and subsequent re-assembly of histones in the wake of the polymerase. Indeed, several studies suggest that transcription destabilizes chromatin and leads to partial eviction of (modified) histones and replacement by newly synthesized (unmodified) ones (Deal and Henikoff, 2010). It is worth noting, however, that the introduction of many of the histone modifications in active regions of the genome is promoted by the process of transcription initiation or elongation. Therefore, these marks can be maintained even when the histones carrying the marks are evicted (Henikoff and Shilatifard, 2011). DNA replication is another potential source of epigenetic
rearrangements. Ahead of the replication fork old modified histones dissociate from the DNA and behind the replication fork histones reassemble on the two daughter strands. The chromatin gaps on the duplicated DNA are complemented by a set of newly synthesized unmodified histones. In order to maintain an epigenetic identity, the cells need to reestablish the modification pattern of the mother cell by modifying the newly synthesized histones in the daughter cells. The speed at which this occurs can substantially differ between types and sites of modifications, and may even span multiple cell divisions (see below). Reestablishment can be facilitated by the recruitment of modifying enzymes to the replication fork or to neighboring histones that already carry the modified state and serve as templates from which a mark can be copied (Scharf et al., 2009a; Sweet et al., 2010; Xu et al., 2011). The dynamic nature of histones during transcription, replication, and repair suggests that epigenetic states in aging organisms are continuously being challenged.

From a cellular perspective, organismal aging is determined by chronological aging and replicative aging of cells (Kaeberlein, 2010). Cells that do not divide undergo chronological aging (Kaeberlein, 2010). Without cell division, modified histones are not diluted out by duplication of the genome. Therefore, maintenance of the epigenome after withdrawal from the cell cycle is dependent on adjusting the balance between modifying and demodifying enzymes or on replication-independent histone turnover. How the epigenetic landscape changes under conditions of chronological cell aging has not been extensively determined yet but several chromatin changes have been observed between yeast cells replicating in rich media and non-replicating yeast cells arrested by nutrient starvation (Zhang et al., 2011).

Replicative aging occurs due to the progressive loss of essential components such as the shortening of telomeres in the absence of telomerase or due to the asymmetric segregation of detrimental cellular factors (Kaeberlein, 2010). In cultured human fibroblasts undergoing replicative aging, DNA damage signaling due to telomere shortening has been identified as a signal that can cause global alterations of several histone modifications (O’Sullivan et al., 2010). Recent studies show that also in yeast the chromatin of replicatively aged cells is different from that of young cells. Yeast cells of old replicative age show low expression of the conserved histone deacetylase Sir2 and high levels of acetylation of its substrate H4K16 (Dang et al., 2009). The opposing roles of Sir2 and the H4K16 acetyltransferase Sas2 in life span determination suggest that H4K16 acetylation is causally linked to the yeast aging process (Dang et al., 2009). In addition, aging yeast cells show a global reduction in histone occupancy and low acetylation of H3K56, a histone deposition-related mark. Together, these studies suggest that replicative aging of cells is not only linked to global changes in histone modification but also to quantitative changes in histone assembly and chromatin packing (Feser et al., 2010).

In addition to organismal and cellular aging, recent studies have uncovered links between aging of histone proteins and the epigenome. Histones are long-lived proteins. Once synthesized in the cell they can exist for many generations...
and aging histone proteins could therefore potentially contribute to chromatin structure over long periods of time and over many cell generations. In general, old and new histones can be distinguished from each other by pulse-chase methods such as stable isotope labeling with amino acids in cell culture (SILAC). SILAC methods combined with quantitative mass spectrometry showed that methylation of H3K79, a conserved mark associated with transcription, relatively slowly accumulates with time in cultured human cells (Sweet et al., 2010). Furthermore, methylation of H3K79 was not restricted to new histones. Instead, old and new histones were found to be methylated with similar efficiency (Sweet et al., 2010). Apparently, on a bulk level, new histones do not rapidly receive a copy of the H3K79 methyl mark from the neighboring old histones after replication, which challenges some of the general models of epigenetic memory. A gradual re-establishment of parental histone modification levels in daughter cells was recently also observed for other methyl marks (Scharf et al., 2009a; Xu et al., 2011). Evidence for progressive H3K79 methylation during successive cell divisions was also found in yeast when the 'genetic pulse-chase' method Recombination-Induced Tag Exchange (RITE) was employed to biochemically purify aging histones (De Vos et al., 2011). Histones of increasing average age purified from ‘young’ asynchronously cultured yeast cells showed an increase in H3K79me3 (De Vos et al., 2011). The accumulation of methyl groups on H3K79 on aging histones is in agreement with the lack of known H3K79-specific histone demethylases in yeast and other organisms.

What is the consequence of the progressive build up of H3K79 methyl marks on aging histones? In the absence of a demethylase, the main mechanism that counteracts the H3K79 methyltransferase Dot1 is dilution of modified histones by unmodified ones during cell growth and division, suggesting that this mark may increase during chronological aging of post-replicative cells (De Vos et al., 2011). The build-up of methyl marks on aging histones may also have its consequences in replicating cells. In slow-growing yeast cells H3K79me3 is increased, indicating that the mark could function as a timer mechanism to couple cell cycle progression to global changes in chromatin structure (De Vos et al., 2011). In addition to global changes, the genomic location of the old histones may also matter. Using the RITE assay, aging histones in replicating yeast cells were found to be retained non-randomly with a preference for the 5’ ends of long poorly transcribed genes (Radman-Livaja et al., 2011). In agreement with the biochemical analysis of bulk histones, the genomic location of old histones roughly correlates with the location of H3K79me3 (Radman-Livaja et al., 2011). Together, these findings identify the inheritance of aging histones in replicating cells as a potential mechanism to modulate the epigenetic landscape.

Recent observations in various organisms show that there is a striking interdependence between the epigenetic landscape and aging of organisms, chronological and replicative aging of cells, and aging of histone proteins. However, it is often still poorly understood through which mechanisms aging alters the epigenetic state of a cell, or how an epigenetic state can affect aging. Obviously,
the aging of organisms, cells, and proteins do not occur independently. In the future, it will be important to determine the mechanistic relationship between these different aging processes in epigenome control by uncoupling the intrinsic aging of proteins from overall aging of the cell. One major challenge will be to identify the upstream signals that cause the changes in enzyme activity responsible for the loss of the ‘young’ epigenetic state in aging cells. Given the reversible nature of epigenetic states, understanding the signals and pathways of chromatin aging may provide opportunities for the development of strategies to modulate aging processes.

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