ONTOGENY OF HISTONE METHYLATION IN MOUSE KIDNEY.

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Histone H3 and H4 modifications are known to exert critical effects on gene expression. Histone methylation on lysine or arginine creates different platforms for interactions with various enzymes and transcriptional regulators. This in turn induces differential transcriptional responses of target genes. We hypothesize that histone methylation plays an important role in the epigenetic control of organogenesis. We characterized the temporal changes in histone methylation (H3- K4, K9, K27, K79) and corresponding histone lysine methyltransferases (ASH1L, SUV39, EZH1, EZH2, SUZ12, Dot1L) during mouse kidney development. Western blot analysis and RT-QPCR were performed on chromatin preparations and RNA extracted from embryonic kidney day 13.5, 15.5, 17.5 and postnatal day 1, 10, 20 and 90. The results revealed that H3K4me3 (activation mark) and H3K9me3 and H3K27me3 (repression marks) were maintained during kidney organogenesis. Interestingly, the H3K9 methyltransferase, SUV39, and enhancer of zeste 2 (EZH2), which is a methyltransferase specific to H3K27, declined significantly during renal development at the protein and RNA levels. In contrast, H3K79me3 and its methyltransferase, Dot1, increased significantly with maturation both at protein and RNA levels. Dot1l, as the only found H3K79me3 methyltransferase, has been shown to interact with Af9 to repress the epithelial sodium channel, ENac, in collecting duct cells. Dot1/Af9-mediated repression of ENac is opposed by aldosterone. Immunofluorescence revealed co-localization of ENac and H3K79me3 in embryonic and mature kidney cell lines and embryonic mouse kidneys. CONCLUSIONS: 1) global histone modification levels are tightly regulated during development and differentiation; 2) The apparent discrepancy between methyltransferase and histone methylation levels on H3K9 and K27 probably reflects redundancy by related methyltransferases and/or concomitant changes in demethylases; 3) Dot1-mediated methylation of ENac H3K79 may play a role in modulating the response of the maturing collecting duct to aldosterone.