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Recent developments in epigenetics of acute and chronic kidney diseases

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The growing epidemic of obesity and diabetes, the aging population as well as prevalence of drug abuse has led to significant increases in the rates of the closely associated acute and chronic kidney diseases, including diabetic nephropathy. Furthermore, evidence shows that parental behavior and diet can affect the phenotype of subsequent generations via epigenetic transmission mechanisms. These data suggest a strong influence of the environment on disease susceptibility and that, apart from genetic susceptibility, epigenetic mechanisms need to be evaluated to gain critical new information about kidney diseases. Epigenetics is the study of processes that control gene expression and phenotype without alterations in the underlying DNA sequence. Epigenetic modifications, including cytosine DNA methylation and covalent post-translational modifications of histones in chromatin, are part of the epigenome, the interface between the stable genome and the variable environment. This dynamic epigenetic layer responds to external environmental cues to influence the expression of genes associated with disease states. The field of epigenetics has seen remarkable growth in the past few years with significant advances in basic biology, contributions to human disease, as well as epigenomics technologies. Further understanding of how the renal cell epigenome is altered by metabolic and other stimuli can yield novel new insights into the pathogenesis of kidney diseases. In this review, we have discussed the current knowledge on the role of epigenetic mechanisms (primarily DNAme and histone modifications) in acute and chronic kidney diseases, and their translational potential to identify much needed new therapies.

Kidney International (2015) **88,** 250–261; doi:10.1038/ki.2015.148; published online 20 May 2015

KEYWORDS: AKI; CKD; epigenetics; fibrosis; inflammation; metabolic memory

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Received 30 January 2015; revised 22 March 2015; accepted 30 March 2015; published online 20 May 2015

The rates of acute and chronic kidney diseases (CKDs) are growing worldwide and in the United States, which in turn has increased the incidence of end-stage renal disease (ESRD) requiring painful dialysis. This is projected to escalate further owing to increased longevity, and the growing epidemic of diabetes and obesity, which can lead to diabetic nephropathy (DN), a widespread CKD that can progress to ESRD. Chemical pollutants, aging, over nutrition and under nutrition, sedentary lifestyles, and other environmental stressors are implicated in many diseases, including CKD, a common complex gene–environmental disease.[1–4] Thus, environmental factors can affect phenotypes via 'epigenetic' transmission mechanisms to alter disease susceptibility, suggesting that, apart from genetic traits, epigenetic variations might yield valuable information about the initiation and progression of various kidney diseases.[5–7] Several genetic studies including genome-wide association studies have identified susceptibility loci and candidate genes for renal impairment and CKDs, including DN, albeit with relatively small effects that explain only a small proportion of heritability.[8] These observations, along with studies on disease-discordant monozygotic twins, provide a strong rationale to examine how epigenetic modifications may orchestrate the pathology of various kidney diseases.

Epigenetics refers to the study of heritable changes in gene expression and phenotype that are not mediated by alterations in the underlying DNA sequence of the genome. Although heritable traits refer to transmission from parent to offspring, in the context of the cell, it can refer to memories and patterns of gene expression and cellular states being passed on dynamically during replication to daughter cells.[9,10] Epigenetic modifications include cytosine methylation of DNA (DNA methylation, DNAme), histone post-translational modifications (PTMs), and noncoding RNAs.[11–14] Together, they form an epigenetic layer over the genetic layer that can respond to environmental cues and external stimuli to alter the expression of genes associated with normal or disease phenotypes. Epigenetics has critical roles in cell identity, development, genomic imprinting, X-inactivation, and differential disease susceptibility between monozygotic twins.[10,11,13]

Recently, there has been remarkable progress in epigenetics research partly owing to technological breakthroughs in next-generation sequencing (NGS) and epigenomics (genome-wide

study of epigenetics).[15–17] The importance of epigenetic alterations in fibrosis, inflammation, and immunity associated with various renal disorders, as well as in kidney development, is increasingly being appreciated.[5,18–20] This review summarizes the current knowledge on the role of epigenetics (primarily DNAme and histone PTMs) in acute and CKDs, and its translational potential to identify much needed new therapies.

EPIGENETIC MODIFICATIONS AND THE EPIGENOME: RATIONALE FOR STUDIES IN THE KIDNEY

Epigenetic marks including DNAme, chromatin histone tail PTMs, and noncoding RNAs collectively form the 'epigenome'. Nucleosomes, the building blocks of chromatin, are made up of chromosomal DNA wrapped around core histones (H2A, H2B, H3, and H4).[11,21] The epigenome is the interface between the stable genome and the variable environment, and it dictates cell type–specific gene expression despite similarities in genetic DNA sequence.[5,6,12,13] Perturbations in the epigenome have been implicated in various pathological conditions including cancer and diabetes.[6,22,23]

Methylation at the fifth position of cytosine (5 mC) in DNA (DNAme) is a well-established epigenetic mark.[13,24] It is distributed throughout the genome generally at CpG dinucleotides, which can occur in clusters known as CpG islands. DNAme patterns are established during development by *de novo* DNA methyl transferases (DNMT) 3A and DNMT3B, whereas DNMT1 acts as a maintenance MT in later stages.[13] Promoter DNAme can repress transcription via interference with transcription factor binding or recruiting repressor complexes consisting of methyl-DNA binding proteins.[25] However, the regulatory effects of DNAme can vary from gene repression to activation depending on genomic contexts such as promoters, gene bodies, enhancers, and repeat sequences.[13] DNAme is a relatively stable epigenetic modification, and DNA demethylation was thought to occur mainly by passive mechanisms during development and cell division. However, more recently, enzymes of Ten eleven translocation (TET) family (TET1/TET2/TET3) and thymine DNA glycosylases have been shown to mediate active DNA demethylation through oxidation of 5 mc to 5-hydroxymethyl cytosine (5 hmc), which is involved in diverse biological processes.[26,27] Overall, the role of DNAme in gene regulation appears to be more complex than previously thought.

Covalent PTMs of core histones in chromatin, including histone lysine acetylation (HKAc) and methylation (HKme), are also epigenetic marks that regulate chromatin structure and gene expression.[11,21] H3KAc is generally associated with relaxed chromatin and active gene expression. On the other hand, HKme can serve as an active or repressive mark depending on the lysine residue modified and the extent of methylation (mono-, di-, or tri-). HKAc is catalyzed by histone acetyl transferases (HATs) and HKMe by histone methyltransferases. In contrast, histone deacetylases (HDACs) and histone demethylases erase HKAc and HKme, respectively. Histone modifications serve as docking sites for coactivators, co-repressors, chromatin remodeling proteins, and proteins that bind to modified histones.[11] Combinatorial effects of various histone PTMs form a 'Histone code' that dictates transcriptional outcomes by controlling compact (heterochromatin) or relaxed (euchromatin) states of chromatin.[28] Epigenomic profiling revealed that specific histone PTMs can mark and define distinct regulatory regions of the genome.[15,21,29] Histone H3 lysine 4 trimethylation (H3K4me3) is mostly associated with transcription start sites at promoters, and H3K36me3 with transcribed regions/gene bodies. H3/H4KAc and H3K4me2 are generally associated with active promoters, whereas promoters of repressed genes are enriched with H3K9me3, H3K27me3, and H4K20me3. Enhancers are associated with H3K4me1 and H3K27Ac (Figure 1).

Noncoding RNAs such as short microRNAs (about 22 nucleotides in length) and long noncoding RNAs (>200 nucleotides long) are also part of the epigenetic layer and have been demonstrated to work via epigenetic mechanisms.[30–33]

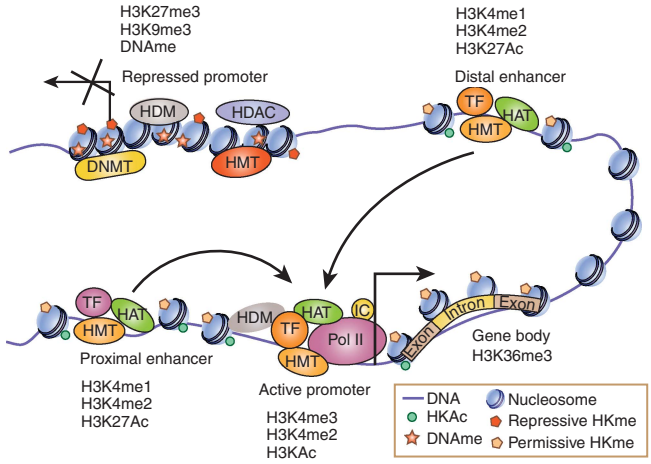

Figure 1 | Schematic of epigenetic modifications associated with chromatin function. Active promoters are enriched with permissive histone modifications (H3KAc and H3K4me), whereas transcribed gene bodies are enriched with H3K36me3. In contrast, inactive promoters are associated with repressive epigenetic modifications including DNA methylation (DNAme) and histone modifications (H3K9me3 and H3K27me3) and reduced H3KAc owing to actions of histone deacetylases (HDACs). In addition, inactive promoters could be enriched with specific histone demethylases (HDMs) that erase permissive marks, whereas active promoters could be enriched with specific HDMs that erase repressive marks. Enhancers are enriched with H3K4me1 and H3K4me2, and active enhancers are marked with H3K27Ac and histone acetyl transferases (HATs) such as P300. Enhancers can regulate the transcription of genes located several kilobases away by promoting chromatin conformation changes (bending) to promote long-range interactions between enhancer-bound transcription factors (TF) and coactivators with target gene promoters. Post-translational modifications of other core histones, including histone H4, such as H4KAc and H4K20me3, as well as on other amino acids like arginine, are also important for chromatin function (not shown). DNMT, DNA methyl transferase; HMT, histone methyl transferase; H3KAc, histone H3 lysine acetylation; IC, transcription initiation complexes; Pol II, RNA polymerase II.