*db/db* mice.[8] This discrepancy could be attributed to a distinct level or period, persistent or intermittent, of elevation of the circulating protein. To determine whether increasing serum ANGPTL2 affects renal fibrosis, Morinaga *et al.*[1] compared skin-specific transgenic *Angptl2*–expression mice to wild-type mice in a UUO model instead of vascularly injecting recombinant ANGPTL2 protein, which may be an unstable form. This transgenic model showed elevated levels of serum ANGPTL2 and comparable levels of tissue ANGPTL2. They successfully illustrated that the degree of UUO-mediated fibrosis is comparable between wild-type and transgenic mice, implicating that systemic additional administration of ANGTPTL2 itself is not of immediate therapeutic value against kidney fibrosis. Instead, small molecule inhibition or antibody-mediated neutralization of ANGPTL2 is deemed logical as a possible antifibrotic approach.

From a clinical viewpoint, the value of ANGPTL2 as a biomarker may also be warranted. One cohort study reported that the human serum concentration of ANGPTL2 is strongly linked to albuminuria but not to a decreased estimated glomerular filtration rate in patients with chronic kidney disease.[9] Intriguingly, Morinaga *et al.*[1] also found significantly increased serum ANGPTL2 levels in UUO mice compared with sham mice. Because a genetically forced release of ANGPTL2 into the bloodstream did not affect tissue damage, as mentioned before, high levels of serum ANGPTL2 in patients with chronic kidney disease or UUO mice could be due to it being released from the damaged organ. In addition to independent cohort studies, further experimental investigations with different animal models are needed to define the detailed mechanisms underlying the increase in serum ANGPTL2.

In summary, Morinaga *et al.*[1] identified ANGPTL2 as an important activator of kidney TGF-β signaling and demonstrated a molecular link between fibrosis and inflammation in the kidney. Because targeting the kidney fibrogenic pathway has been under intensive clinical investigation, ANGPTL2 inhibition could be beneficial for various kidney diseases in which interstitial fibrosis is histologically notable—such as diabetic nephropathy, hypertensive kidney diseases—and also for transplanted renal graft dysfunction.

**DISCLOSURE**

The author declared no competing interests.

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# Epigenetic memory in kidney diseases

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**Epigenetic mechanisms have been the focus of intensive research. De Marinis *et al.* demonstrated that high glucose levels exert stimulatory effects on activation histone marks, leading to the upregulation of thioredoxin-interacting protein (*TXNIP*) gene expression, which is proinflammatory. They also showed that the effect was reversed by the inhibition of histone acetyltransferase, suggesting a new therapeutic approach for improving diabetic kidney disease. Epigenetic changes are memorized as epigenetic memory that could exacerbate diabetic complications.**

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Epigenetic changes accumulate as cell memory, and this epigenetic memory plays a crucial role in the long-term consequences of adult-onset diseases and aging. Epidemiologic and clinical data support the idea that early metabolic control has a lasting influence on the clinical outcomes of diabetic complications; this phenomenon is called "metabolic memory."[1] Intensive glucose therapy in patients with type 2 diabetes mellitus showed a lower risk of microvascular complications according to the results of UKPDS 80 (the United Kingdom

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Prospective Diabetes Study).[2] Similarly, stringent control on blood glucose levels of patients with type 1 diabetes was effective in inhibiting microalbuminuria and microvascular complications in the DCCT/EDIC (Diabetes Control and Complications Trial/Epidemiology of Diabetes Interventions and Complications) study. In addition, over the 10-year follow-up period of the DCCT/EDIC study, the albumin excretion rate was controlled and the glomerular filtration rate remained within normal limits in patients with type 1 diabetes mellitus. It is reasonable to speculate that this long-term effect, such as metabolic memory, may be mediated by epigenetic changes.

De Marinis *et al.*[3] (2016) aimed to elucidate a mechanism of metabolic memory and demonstrated that high glucose levels had inhibitory effects on repressive histone marks in the promoter of the thioredoxin-interacting protein (*TXNIP*) gene. Using both a mouse model prone to diabetes and an *in vitro* model of mesangial cells exposed to high glucose, they showed that histone modifications increase the expression of this proinflammatory gene in the kidney.

*TXNIP* is reported to be augmented by high glucose levels and to promote oxidative stress. Shah *et al.* demonstrated that *Txnip*-deficient mouse mesangial cells showed protection from high-glucose-induced reactive oxygen species.[4] They also showed that *TXNIP* knockdown with small, interfering RNA abolished the increased mitochondrial oxygen generation and apoptosis in cultured human podocytes exposed to high glucose levels. Siddiqi *et al.* demonstrated that a histone methyltransferase enzyme, the enhancer of zeste homolog 2 (EZH2), regulated the expression of *TXNIP* in attenuating oxidative injury in podocytes under high-glucose conditions.[5] EZH2 is known to catalyze the addition of methyl groups to histone 3 at lysine 27 (H3K27), an inhibitory histone mark. They showed that the depletion of EZH2 augmented *TXNIP* expression and oxidative stress while it decreased H3K27me3, thereby inducing podocyte injury and proteinuria in diabetic rats. EZH2 upregulation through the inhibition of microRNA-101 resulted in the downregulation of *TXNIP* and the attenuation of oxidative stress. These findings suggested that epigenetic processes performed by EZH2 play important roles as antioxidant effects in the diabetic kidney.

An increasing number of reports now show that histone modifications are profoundly associated with metabolic memory. For example, De Marinis *et al.*[3] showed that TXNIP is regulated by EZH2, which decreases the repressive histone mark H3K27me3, whereas Villeneuve *et al.* demonstrated that the repressive histone mark H3K9me3 (lysine-9 tri-methylation) plays an important role in metabolic memory.[6] H3K9me3 levels were decreased at the promoters of inflammatory genes in cultured *db/db* vascular smooth muscle cells relative to control *db/+* cells. One of the H3K9me3 methyltransferases, Suv39h1 (a suppressor of variegation 3-9 homolog 1), was also shown to be reduced in *db/db* vascular smooth muscle cells, while its overexpression reversed the diabetic phenotype. These results showed the protective roles of H3K9me3 and Suv39h1 against inflammatory phenotype of vascular smooth muscle cells in diabetes. Thus, histone modification changes are maintained in diabetic mouse kidneys as metabolic memory, suggesting that Suv39h1 could be a candidate for therapeutic targets in diabetic nephropathy.

Epigenetic modifications include cytosine DNA methylation and covalent posttranslational modifications of histones in chromatin. The dynamic epigenetic layer responds to the external environment to influence the expression of genes associated with renal disease states. For example, DNA methylation in kidney disease is also reported to be a factor in fibrogenesis, and profibrotic transforming growth factor-β inhibited the expression of *Rasal1* (RAS protein activator like 1), a negative regulator of Ras signaling, by promoting DNA methylation at its promoter via DNA (cytosine-5)-methyltransferase-1 (DNMT1), which is a methyltransferase of DNA cytosine-5.[7] The methylation of CpG island promoters effectively silences transcription, and the hypermethylation of the *Rasal1* promoter by DNMT1 contributes to fibroblast activation and renal fibrosis. This fibrotic change is reportedly reversed by bone morphogenetic protein, which upregulates *Rasal1* by inducing TET3 (ten-eleven translocation methylcytosine dioxygenase 3)–mediated hydroxymethylation.[8]

De Marinis *et al.*[3] have, therefore, demonstrated that changes to histone modification at downstream loci of target genes are associated with renal dysfunction and fibrosis. Following this, they performed chromatin immunoprecipitation–quantitative polymerase chain reaction to examine various histone modifications, including H3K4me1, H3K4me3, H3K9ac, and H3K27me3. Their results showed not only that there were age-related increases in H3K9ac, H3K4me1, and H3K4me3, and a decrease in H3K27me3, but that these changes were also much more pronounced in diabetic mice. This suggested that age- and glucose-related increases in *TXNIP* expression may be associated with histone modification *in vivo*. However, the authors examined histone modification changes using mouse mesangial cells and demonstrated that the activation mark H3K4me3 significantly decreased in response to high glucose levels, despite H3K9ac and H3K4me1 increasing significantly at the promoter region of *TXNIP*. In addition, no changes were observed in H3K27me3, the repression mark. In human mesangial cell lines, they demonstrated that H3K4me3 levels tended to increase in response to high glucose levels. In their Discussion, they mention that there is no obvious explanation for this paradoxical finding except for the duration of hyperglycemia. It is possible that