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REVIEW ARTICLE

Progress in the Mechanism of Hyperhomocysteinemia-Induced Renal Injury

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SUMMARY

Background: **Hcy is a sulfhydryl amino acid in the metabolism of methionine. It has been recognized as an independent risk factor for cardiovascular disease. In recent years, the relationship between hyperhomocysteinemia and renal disease has received attention from many researchers. However, the specific mechanisms by which Hcy plays a role in cardiovascular pathology in patients with chronic kidney disease are complex.**
Methods: **We consulted the relevant literature and sorted and summarized it.**
Results: **Multiple mechanisms of hyperhomocysteinemia-induced renal injury are summarized in detail from different perspectives, including oxidative stress, vascular endothelial damage, inflammatory response, cellular autophagy, apoptosis, fibrosis, and epigenetic regulation.**
Conclusions: **Hyperhomocysteinemia acts synergistically through multiple pathways, leading to glomerulosclerosis, tubular atrophy, and interstitial fibrosis, and ultimately accelerating renal failure. These mechanisms are complex and interrelated, suggesting that a comprehensive intervention strategy may achieve the ultimate goal of reducing renal injury.**
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KEYWORDS

homocysteine, hyperhomocysteinemia, renal injury

LIST OF ABBREVIATIONS

Hcy - Homocysteine
HHcy - Hyperhomocysteinemia
CSE - Cystathionine-γ-lyase
CBS - Cystathionine-beta-synthase
MTHFR - Methylenetetrahydrofolate reductase
CKD - Chronic kidney disease
ESRD - End-stage renal disease
SAM - S-adenosyl-methionine
SAH - S-adenosyl-hcy
eGFR - Estimated glomerular filtration rate
NOX - Nicotinamide adenine dinucleotide phosphate oxidase
HuR - Human antigen R
HO-1 - Heme oxygenase-1
ROS - Reactive oxygen species

P13K - Phosphoinositide 3-kinase
eNOS - Endothelial nitric oxide synthase
NO - Nitric oxide
NOD2 - Nucleotide binding oligomerization domain 2
TRPC6 - Transient receptor potential cation channel protein 6
PGRN - Progranulin
GSK-3β - Glycogen synthase kinase 3 beta
L-Hcy - L-homocysteine
VEGF - Vascular endothelial growth factor
IL-1β - Interleukin-1β
NLRP3 - Nucleotide-binding oligomerization domain-like receptor containing pyrin domain 3
Rac1 - RAS-related C3 botulinum toxin substrate 1
Vav2 - Vav guanine nucleotide exchange factor 2
LC3 - Light chain 3
TFEB - Transcription factor EB
ECM - Extracellular matrix
FOXO - Forkhead box O
H2S - Hydrogen sulfide
MMP-2 - Matrix metallopeptidase 2
MMP-9 - Matrix metallopeptidase 9
MMP-14 - Matrix metallopeptidase 14
Col I - Collagen type I
Col IV - Collagen type IV
Fbn-1 - Fibrillin 1
ATP - Adenosine triphosphate

REVIEW

Homocysteine (Hcy) is a sulfhydryl amino acid in the metabolism of methionine. The prevalence of hyperhomocysteinemia (HHcy) has been reported to be on the rise in recent years [1]. Hcy has been recognized as an independent risk factor for cardiovascular and cerebrovascular diseases and is highly correlated with the onset of peripheral vascular pathology, Alzheimer's disease, neoplasms, diabetes mellitus, and complications of pregnancy [2-4]. In recent years, the relationship between HHcy and renal diseases has also received attention from many researchers [5,6]. In this paper, we review the mechanism of hyperhomocysteinemia-induced renal injury to provide new ideas for the clinical treatment of HHcy and related diseases.

Overview of homocysteine

Hcy is metabolically cleared by two main pathways: 1) the transsulfuration pathway: Hcy is first condensed with serine to form cystathionine-thionine complex. Then, it is broken down into cysteine with the assistance of cystathionine-γ-lyase (CSE) and cystathionine-beta-synthase (CBS). This process is mainly catalyzed by both CBS and CSE. 2) Remethylation pathway: By using Hcy as a substrate and folate and vitamin B12 as cofactors, methionine is regenerated by methylenetetrahydrofolate reductase (MTHFR), a process that occurs mainly in the liver and kidney [7,8]. When the body is deficient in important vitamin cofactors (folic acid, vitamin B6, vitamin B12) or enzymes (MTHFR, CBS, CSE), or when methionine is ingested in excess, this will result in elevated serum or plasma Hcy concentrations [2]. In addition, alcohol consumption, cigarette smoking, coffee intake, age, gender, and altered hormone levels can also increase Hcy levels [9].

Homocysteine and kidney disease

Renal function and Hcy metabolism

Organs such as the liver, kidney, intestine, and pancreas contain enzymes required for Hcy metabolism, with MTHFR and CBS being the most active in renal tissues [10,11]; thus, the kidneys play an important role in Hcy clearance. In addition, choline is oxidized by choline dehydrogenase to betaine aldehyde, and subsequently, by betaine aldehyde dehydrogenase, oxidized to betaine [12], and both betaine aldehyde and betaine aldehyde dehydrogenase are predominantly present/localized in the liver and kidney [13,14]. Therefore, when renal function is impaired, one of the major enzymes for betaine production, betaine aldehyde dehydrogenase, is reduced or absent, resulting in reduced betaine synthesis. Betaine metabolism is critical as a methyl donor for methionine synthesis and regulation of Hcy levels, which increase when betaine concentrations are reduced or when folate and vitamin B12 are deficient [15]. Hyperhomocysteine has been reported to be present in 36% - 89% of patients with chronic kidney disease, with a prevalence of hyperhomocysteine as high as 85% to 100% in patients with end-stage renal disease [16,17].

Hyperhomocysteine and kidney disease

Chronic kidney disease (CKD) is a global health problem that can progress to advanced renal failure, end stage renal disease (ESRD), and even death, but it still lacks effective treatment. Studies have shown that elevated Hcy levels are independently associated with reduced glomerular filtration rate, suggesting that hyperhomocysteine increases the risk of developing CKD [18]. Hyperhomocysteine is associated with the pathophysiology of glomerulosclerosis and interstitial fibrosis, leading to progressive decline in renal function [19]. Hyperhomocysteine has been suggested to be an independent predictor of the development of CKD, as well as the progression of CKD, and a risk or causative factor for cardiovascular complications [20,21]. Several studies have shown that hyperhomocysteine accelerates CKD progression by modulating oxidative stress, endoplasmic reticulum stress, and inflammatory responses [22-24].

Patients with ESRD have higher rates of hospitalization, cardiovascular events, and all-cause mortality compared with patients with normal renal function, as well as serious economic burden on the country and the population [25]. Elevated Hcy is a potential risk factor for atherosclerosis in patients with preserved renal function and is particularly important in patients with ESRD [26]. Prospective clinical studies have shown that an elevation of serum Hcy by 5 pmol/L increases the probability of mortality by 7% and cardiovascular events by 9% in ESRD patients and that Hcy levels may be a risk factor for car-

diovascular disease and mortality in ESRD patients [27]. Although there is a direct correlation between Hcy levels and cardiovascular events in patients with chronic kidney disease, clinical studies have shown that elevated Hcy levels, a factor that may contribute to accelerated atherosclerosis in patients with ESRD, are not fully ameliorated by statins [28]. It has been shown by others that although high-dose intravenous B vitamins were effective in reducing Hcy levels in dialysis patients, there was limited improvement in the body's methylation potential (S-Adenosyl-Hcy (SAH), S-adenosyl-methionine (SAM)). Whereas elevated blood levels of SAH and its imbalance in proportion to SAM are closely associated with vascular injury, further exploration is needed to determine whether the combination of long-term vitamin therapy and antioxidants can improve the overall prognosis of patients with chronic kidney disease, especially with regard to the potential role of targeting cardiovascular risk [29]. Similarly, the results of 13 randomized trials involving 11,049 patients have shown that folic acid supplementation may not reduce the risk of acute myocardial infarction or stroke in patients with chronic kidney disease [30]. These results suggest that the specific mechanisms by which Hcy plays a role in cardiovascular pathology in patients with chronic kidney disease are complex.

Researchers from the Takahata region of Japan investigated the synergistic risks associated with HHcy and mild-to-moderate chronic kidney disease. An 18.6-year follow-up of 3,377 residents aged ≥ 40 revealed that individuals with both conditions exhibited significantly higher all-cause mortality risk (HR = 2.49) and cardiovascular event risk (HR = 2.11) compared to those with a single condition. This study provides the first evidence in an Asian general population that even early-stage CKD patients enter a ‘high-risk alert zone’ when co-affected by HHcy [31].

Regulatory mechanisms of Hyperhomocysteinemia-induced renal injury

Oxidative stress

Although it has been shown that hyperhomocysteine increases proteinuria in hypertensive patients, the mechanism by which hyperhomocysteine exacerbates renal damage in hypertension remains unclear. Podocyte injury is the key to proteinuria formation, and a variety of pathogenic factors can lead to abnormal expression of nephrin on the surface of podocytes, resulting in abnormal signaling inside and outside the affected podocytes, which in turn leads to dysregulation of cytoskeletal protein structure and disruption of normal podocyte structure, and ultimately leads to impaired glomerular filtration function [32-34]. A study [35] found that NOX2 and NOX4 were significantly elevated in the kidneys of spontaneously hypertensive rats with high homocysteine and higher than those of the hyperhomocysteine alone group, suggesting that hyperhomocysteine enhances localized oxidative stress in the kidneys, and this effect is enhanced in the presence of hypertension; there is a synergistic effect between hyperhomocysteine and hypertension in exacerbating oxidative stress injury. Podocyte injury is one of the earliest signs of oxidative stress [36,37], and this study also found that the nephrin expression level was significantly reduced in the hyperhomocysteine group and the spontaneous hypertension + hyperhomocysteine group, which is consistent with enhanced oxidative stress; whereas, the reduction of nephrin expression was prevented when mice were deficient in either the NOX2 or NOX4 genes, suggesting that hyperhomocysteine and hypertension can act on podocyte nephrin via NOX. These findings suggest that hyperhomocysteine can synergize with hypertension to induce proteinuria and aggravate glomerular injury and that these processes are driven by a combination of enhanced oxidative stress and reduced nephrin expression in podocytes induced by HHcy and hypertension.

Li et al. [38] found that Hcy downregulates the expression of the RNA-binding protein human antigen R (HuR), decreases the binding of HuR to the 3'-untranslated region of heme oxygenase-1 (HO-1), and affects the renal tubular cells in the HO-1 mRNA stability, thus decreasing the expression levels of HO-1 itself and its downstream substances bilirubin and iron; whereas, after the addition of HO-1 inducer, HO-1 activity and protein levels increased, while the levels of reactive oxygen species (ROS) in renal tissues were significantly reduced, and the morphological damage was alleviated in the mice, suggesting that Hcy mediates cisplatin-induced acute kidney injury in mice by disrupting the oxidative stress-antioxidant balance involving HO-1 itself and its downstream substances, while HO-1 agonists significantly attenuate acute kidney injury and CKD progression. These findings suggest that lowering Hcy levels or inducing HO-1 expression may be a potential therapeutic strategy to improve the prognosis of acute kidney injury.

Vascular endothelial damage

The phosphoinositide 3-kinase/protein kinase B (PI3K/Akt) pathway is widespread in cells and is a signaling pathway involved in the regulation of cell growth, proliferation, and differentiation [39,40]. Studies have shown that the PI3K/Akt/endothelial nitric oxide synthase (eNOS) pathway plays an important role in the abnormal endothelial function due to HHcy [41]. HHcy was induced via oral administration of L-methionine. Following four consecutive weeks of L-methionine supplementation, vascular endothelium dysfunction was assessed through impaired acetylcholine-induced endothelium-dependent relaxation, decreased serum nitrite levels, reduced eNOS mRNA expression, and compromised vascular endothelial integrity. Injection of insulin and atorvastatin significantly improved acetylcholine-induced endothelium-dependent relaxation, serum nitrate/nitrite levels, eNOS mRNA expression, and vascular endothelial integrity. This ameliorative effect of insulin was blocked by PI3K inhibitors, Akt inhibitors, and eNOS inhibitors. These findings suggest that activating PI3K and its downstream pathways may ameliorate HHcy-induced vascular endothelium dysfunction. Therapeutic interventions targeting these pathways may offer potential thera-

peutic strategies for managing vascular complications [42].

Inflammatory response

Han et al. [43] found that nucleotide binding oligomerization domain 2 (NOD2) expression was significantly increased in the renal cortex of hyperhomocysteine mice, and the levels of inflammatory factors and chemokines were increased, suggesting that the inflammatory response was enhanced in the renal cortex of hyperhomocysteine mice. When the NOD2 gene was knocked down, both hyperhomocysteinemia-induced glomerular podocyte injury and renal dysfunction were attenuated, suggesting that NOD2 was involved in hyperhomocysteine-induced renal injury. It was also found that the high expression of transient receptor potential cation channel protein 6 (TRPC6) and the production of inflammatory factors and chemokines in podocytes caused by hyperhomocysteine were suppressed when NOD2 was knocked down, suggesting that hyperhomocysteinemia-induced podocyte TRPC6 expression was NOD2-dependent, and NOD2 was involved in regulating the hyperhomocysteinemia-induced TRPC6 channel-dependent calcium signaling pathway, which resulted in increased calcium ion concentration in podocytes, and ultimately led to apoptosis induced by the remodeling of podocyte cytoskeleton proteins and the development of glomerular diseases. The abovementioned results suggest that NOD2 has the function of regulating TRPC6 ion channels, and the TRPC6-mediated calcium ion signaling pathway is an important signaling pathway connecting the intrinsic immune receptor NOD2 and kidney injury. TRPC6 expression mediated by NOD2 may be the main cause of hyperhomocysteinemia-induced kidney injury, and the intrinsic immune inflammatory response connected by the TRPC6 channel-dependent calcium signaling pathway is involved in hyperhomocysteinemia-induced kidney injury.

Progranulin (PGRN) is an autocrine growth factor found in a wide range of tissues and is involved in a variety of physiological and pathological processes such as tissue development, host defense response, insulin resistance, and inflammatory regulation [44-46], and it is particularly protective of the body in chronic inflammation. In acute kidney injury, PGRN negatively regulates the immune response [47]; however, the role of PGRN in the pathogenesis of hyperhomocysteine is unclear. Fu et al. [48] found that PGRN levels were reduced in the renal cortex, isolated glomeruli, and tubules of hyperhomocysteine mice, with glomeruli being the most markedly reduced, suggesting that hyperhomocysteine can cause a reduction of renal PGRN in mice. Compared with controls, PGRN-deficient mice fed a folate-free diet for 10 weeks had an elevated urinary protein/urinary creatinine ratio, aggravated glomerulosclerosis, thickening of the glomerular basement membrane, and loss of pedunculated cell peduncles; at the molecular level, expression of the pedunculated cell markers nephrin, podocin, and synaptopodin was reduced, and tight junction protein ZO-1 and occludin loss was exacerbated; PGRN deficiency also exacerbated the levels of proinflammatory factors in the renal cortex, suggesting that PGRN deficiency exacerbates renal insufficiency in hyperhomocysteine mice. In contrast, 6 hours after administration of recombinant human PGRN (rPGRN), the increase in proteinuria was slowed, and renal proinflammatory factors were decreased, suggesting that rPGRN has a protective effect against renal insufficiency in hyperhomocysteine mice. Subsequent analysis by mRNA and Western Blot revealed that Wnt1 gene expression was upregulated in the kidneys of hyperhomocysteine mice, which in turn led to an increase in β-catenin levels, which correlated with an increase in the level of phosphorylation of glycogen synthase kinase 3 beta (GSK-3β), and it revealed that the lack of PGRN further augmented these changes, suggesting that PGRN negatively regulates Wnt/β-catenin signaling pathway. In in vitro experiments, when podocytes were treated with rPGRN or an inhibitor of the Wnt signaling pathway, β-catenin levels were reduced, podocyte nephrin and podocin expression was increased, and podocyte apoptosis was reduced, suggesting that PGRN ameliorates hyperhomocysteinemia-induced podocyte dysfunction by inhibiting the Wnt/β-catenin signaling pathway. The administration of rPGRN attenuates disease progression and ameliorates hyperhomocysteinemia-induced renal injury and may be a novel therapeutic strategy for hyperhomocysteine patients.

It has been reported that nucleotide-binding oligomerization domain-like receptor containing pyrin domain 3 (NLRP3) inflammasome activation plays an important role in the pathogenesis of hyperhomocysteinemia-induced glomerular disease and end-stage renal disease [49-51]. Zhang et al. [23] found that L-homocysteine (L-Hcy) stimulation resulted in a significant increase in the expression of junctional proteins, the marker of podocyte injury, and a decrease in the markers of podocyte function and vascular endothelial growth factor (VEGF) secretion; F-actin alignment was disturbed, and caspase-1 activity and interleukin-1β (IL-1β) production were enhanced. Whereas pretreatment of podocytes with RAS-related C3 botulinum toxin substrate 1 (Rac1) inhibitor reduced the L-Hcy-induced increase in junctional protein expression and decrease in podocin levels and also attenuated the reduction and rearrangement of F-actin, caspase-1 activity, and IL-1β production. In contrast, in L-Hcy-treated podocytes, inhibition of Rac1 activity attenuated the formation of NLRP3 inflammasome, decreased caspase-1 activation, and consequently reduced IL-1β production. Conversely, Rac1 activation enhanced the formation and activation of NLRP3 inflammasome in these cells. The Rac1 inhibitors may inhibit the activation of NLRP3 inflammasome in podocytes, thereby reducing hyperhomocysteinemia-induced podocyte injury and glomerulosclerosis. It was also found that hyperhomocysteine triggers NLRP3 inflammasome activation, podocyte dysfunction, and glomerular injury through the activation of vav guanine nucleotide exchange factor 2 (Vav2), which increases Rac1 activity and NOX activity, leading to an increase in ROS production [22]. How,

then, is the NLRP3 inflammasome released into the extracellular space? This process may be related to the release of exosomes of NLRP3 inflammasome [52-57]. Without exosome release, NLRP3 inflammasome activation in podocytes may not trigger the development of glomerular inflammation and sclerosis during hyperhomocysteine, whereas blocking exosome secretion and preventing hyperhomocysteinemia-induced immune cell infiltration may eliminate glomerular injury [58]. Thus, modulation targeting exosome release from podocytes may be a novel therapeutic strategy for hyperhomocysteinemia-induced glomerular inflammation and injury.

Mechanisms of cellular autophagy

Autophagy has become an important mechanism required to ensure life extension and maintain homeostasis in the body [59,60]. Autophagic insufficiency in podocytes leads to oxidized protein accumulation, endoplasmic reticulum stress, and proteinuria, and Hcy can directly lead to podocyte injury and glomerulosclerosis [23, 61]. To investigate the role of autophagy in hyperhomocysteinemia-induced renal aging, Zhang et al. [62] found that the levels of aging-related proteins p53, p21, and p16 were increased in the kidneys of hyperhomocysteine rats, suggesting that renal aging occurs in hyperhomocysteine rats. Hyperhomocysteine inhibited the mRNA of autophagosome lysosome-associated marker protein in renal tissues and the production of protein light chain 3 (LC3), thereby reducing autophagy, suggesting that hyperhomocysteine induces the downregulation of renal autophagy. By using Hcy to stimulate the podocytes of mice given drugs to upregulate autophagy, a decrease in the levels of aging-related proteins was detected, suggesting that autophagic insufficiency participates in hyperhomocysteinemia-induced renal aging. Compared to Hcy, the expression of autophagosome lysosome-associated marker protein and LC3 was increased by the treatment of the drug that upregulated autophagy, suggesting that Hcy can directly lead to podocyte aging by downregulating autophagy.

Helical transcription factor EB (TFEB) controls the expression of autophagosome lysosome-associated genes, and TFEB-mediated transcriptional regulation is a key regulatory mechanism of autophagy system [63-65]. Zhang et al. [62] found that hyperhomocysteine resulted in a significant decrease in TFEB expression in renal tissues, which was significantly increased after treatment with autophagy upregulating drugs, suggesting that the expression of TFEB, a key regulator of autophagy, was decreased in hyperhomocysteinemia-induced renal aging, and the nitrative stress was involved in this process. The abovementioned findings suggest that nitrative stress-related autophagy insufficiency is involved in hyperhomocysteinemia-induced renal aging, providing new ideas for the mechanism and therapeutic strategy of renal aging.

Mechanisms of fibrillation

Cellular damage and accumulation of glomerular interstitial extracellular matrix (ECM) proteins are characteristics of CKD [66], and Akt regulates the transcription factor forkhead box O (FOXO) under normal conditions [67]; however, the role of the Akt/FOXO1 pathway in hyperhomocysteinemia-induced thylakoid membrane cell injury remains unclear, and whether hydrogen sulfide (H2S) plays a protective role remains to be clarified. Suravi Majumder et al. [68] found that Hcy inactivates Akt by inhibiting Akt phosphorylation, and inactivated Akt in turn dephosphorylates FOXO1. After dephosphorylation, FOXO1 becomes active and translocates to the nucleus to activate its target genes. Upon treatment with H2S donors or FOXO1 inhibitors, FOXO1 nuclear translocation was blocked, suggesting that H2S can modulate the Akt/FOXO1 cascade reaction. It was also found that Hcy increased the expression of matrix metallopeptidase 2 (MMP-2), matrix metallopeptidase 9 (MMP-9), and matrix metallopeptidase 14 (MMP-14) and increased the expression of thylakoid matrix proteins collagen type I (Col I), collagen type IV (Col IV), and fibrillin 1 (Fbn-1) in thylakoid cells compared to control cells, which were normalized after treatment by H2S donors or FOXO1 inhibitors, suggesting that FOXO1 signaling is involved in this process. In addition, Hcy also significantly increased apoptosis and reactive oxygen species production, and the levels of apoptosis and reactive oxygen species were reduced in thylakoid cells after treatment by H2S donors or FOXO1 inhibitors, suggesting that the Akt/FOXO1 signaling pathway is involved in hyperhomocysteinemia-induced apoptosis and reactive oxygen species. These findings suggest that Akt/FOXO1 signaling pathway participates in hyperhomocysteinemia-induced thylakoid apoptosis and matrix remodeling, and H2S donors can alleviate the Akt/FOXO1 cascade response by reducing excess ROS production, ameliorating the loss of mitochondrial membrane potential, maintaining the intracellular adenosine triphosphate (ATP) concentration, and decreasing the activation of caspases, which in turn attenuates the hyperhomocysteinemia-induced apoptosis and ECM remodeling.

Abnormalities in epigenetic regulation

miR-30a-5p is shown to be a biomarker in the urine of patients with focal segmental glomerulosclerosis and is specifically expressed in cells of the collecting ducts and podocytes [69,70]. Ding et al. [71] explored the role of miR-30a-5p and its potential mechanism in hyperhomocysteinemia-induced apoptosis of glomerulopodial podocytes. The results showed that elevated Hcy downregulated miR-30a-5p expression in Cbs+/- mice and Hcy-treated podocytes, and the level of DNA methylation of the miR-30a-5p promoter was increased by quantitative methylation analysis, indicating that the level of miR-30a-5p transcripts was regulated by DNA methylation. Meanwhile, dual luciferase reporter analysis indicated that the region between -1,400 and -921 bp of the miR-30a-5p promoter might be a regulatory element for its transcription. Mechanistic studies showed that DNA methyltransferase 1-mediated DNA hypermethylation downregulated miR30a-5p expression, and downregulated expression of miR-30a-5p promoted Hcy-induced glomerular podocyte injury by targeting FOXA1, sug-

gesting that epigenetic modification of miR-30a-5p may be involved in hyperhomocysteinemia-induced glomerular podocyte injury and there may be a new pathogenic pathway in hyperhomocysteinemia-induced CKD.

Apoptosis

Liu et al. [72] found that the expression of apoptosis-related proteins (Bax, caspase12) was upregulated in mouse kidney podocytes with hcy intervention, suggesting that podocytes apoptosis and kidney injury was caused by HHcy, which is consistent with the findings of Cao et al. [73]. After feeding methionine to CBS-deficient mice (Cbs+/-), Wu et al. [74] found that serum Hcy levels and podocytes apoptosis were significantly increased in the Cbs+/- group of mice, which exacerbated kidney injury. Conversely, the reduction of renal function led to further accumulation of Hcy, which aggravated renal injury and caused the occurrence and development of a series of nephropathies. Zhang et al. [9] found that in the cisplatin-induced acute kidney injury model, the expression of γH2AX-positive cells in the kidneys of HHcy mice increased, suggesting that HHcy aggravated DNA damage in the kidney. And DNA damage will induce the activation of mitochondrial damage pathway. On the basis of this theory, the study found that the expression of mitochondrial damage markers (ATP β and Cyt c) was increased, the number of TUNEL-positive cells in renal tissues was significantly increased, and the expression levels of caspase 3 and caspase 12 proteins were markedly elevated in the mouse model of cisplatin-induced acute kidney injury with HHcy, which indicated that HHcy could aggravate mitochondrial damage in the kidneys of mice and exacerbated apoptosis of renal tubular epithelial cells through the mitochondrial damage pathway. *In vitro*, Zhang et al. [9] used Hcy and mitochondrial inhibitors to intervene in rat renal tubular epithelial cells (NRK-52E) and found that HHcy promotes apoptosis in NRK-52E cells through the mitochondrial damage signaling pathway, whereas mitochondrial inhibitors effectively ameliorate mitochondrial damage. These findings suggest that activation of the mitochondrial damage pathway due to DNA damage may be one of the major pathways causing apoptosis in renal tubular epithelial cells.

In summary, HHcy is closely related to the progression of renal diseases. HHcy acts synergistically through multiple pathways, including oxidative stress, vascular endothelial damage, inflammatory response, fibrosis, and epigenetic regulation, leading to glomerulosclerosis, tubular atrophy, and interstitial fibrosis, and ultimately accelerating renal failure. These mechanisms are complex and interrelated, suggesting that a comprehensive intervention strategy may achieve the ultimate goal of reducing renal injury.

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Declaration of Interest:

The authors have no conflicts of interest to declare.

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