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Research article

Transcriptome analysis provides insights into high fat diet-induced kidney injury and moderate intensity continuous training-mediated protective effects

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ABSTRACT

Although physics exercise has been utilized to prevent and treat a variety of metabolic diseases, its role in obesity-related kidney diseases remains poorly understood. In this study, we assessed the protective potential of moderate intensity continuous training (MICT) against high fat diet (HFD)-induced kidney injury and found that MICT could significantly reduce obesity indexes (body weight, serum glucose, total cholesterol, high density lipoprotein cholesterol, low density lipoprotein cholesterol) and kidney injury indexes (serum creatinine and the expression of *Kim*-1 mRNA) in HFD-fed mice. PAS staining and Masson staining displayed that MICT maintained the morphological structure of kidney subunits and reduced kidney fibrosis in HFD-fed mice. By kidney RNA-seq, we identified several genes and pathways (*Cd9*, *Foxq1*, *Mier3*, TGF-β signaling pathway etc.) that might underlie HFD-induced kidney injury and MICT-mediated protective effects. In conclusion, this study revealed the protective role of MICT in HFD-induced kidney injury and suggested potential targets for the prevention and treatment of obesity-related kidney diseases.

1. Introduction

Obesity is a global epidemic associated not only with increased mortality and shortened life expectancy but also recognized as a major risk factor for various chronic diseases, including type-2 diabetes, hypertension, cardiovascular diseases, dyslipidemia, non-alcoholic fatty liver and kidney diseases [1–3]. Kidney injury induced by obesity generally manifested as glomerular hypertrophy, focal segmental glomerulosclerosis, renal tubular hypertrophy, renal tubular interstitial inflammation and fibrosis [4]. The obese state alters secretome of fat cells, resulting in the release of excess pro-inflammatory adipokines (tumor necrosis factor-α and interleukin-6 etc.) and a decrease in beneficial adipokines (leptin and adiponectin etc.), leading to chronic inflammation and insulin resistance [5–7]. In addition, renal hemodynamic changes, the progressive activation of renin–angiotensin–aldosterone system, and lipotoxicity caused by lipid metabolism disorder are all implicated in the development of obesity-related kidney diseases [8,9].

Currently, treatments for obesity-related kidney diseases remain unsatisfactory. Although weight loss through caloric restriction is effective, strictly controlling food intake is unfavorable and challenging. Studies have indicated that drugs such as renin-angiotensin

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system inhibitors, sodium glucose co-transporter 2 inhibitors and melatonin might be used to treat obesity-related kidney diseases [10–12]. However, because these drugs are not kidney-specific, a considerable number of trials are still required to assess their efficacy and potential side effects.

Emerging evidence highlights the promising role of physical exercise in preventing and treating several chronic diseases, including cardiovascular disease, diabetes, hypertension, and certain types of cancer etc. [13–17]. In terms of kidney, physical exercise has been suggested to have positive effects on a range of health-related outcomes in chronic kidney disease (CKD) [18–20]. Uchiyama et al. reported that 6 months home-based exercise improved aerobic capacity and health-related quality of life in patients with Stage 4 CKD, with possible beneficial effects on kidney function and CKD-related parameters [21]. Additionally, in diabetic kidney disease (DKD), physical exercise has been demonstrated to reduce urinary albumin levels and metabolic dysfunction, maintain the number of podocytes, and attenuate oxidative damage and inflammation, thereby slowing down the progression of nephropathy [22,23]. Long term high fat diet (HFD) feeding has been reported to induce obesity and kidney injury in mice [4,24,25]. However, whether physical exercise can attenuate HFD-induced kidney injury remains poorly understood. In this study, we present evidence that 8 weeks moderate-intensity continuous training (MICT) on treadmill remarkably alleviated long term HFD-induced obesity and kidney injury. Furthermore, we performed kidney RNA-seq to explore the underlying mechanisms by which HFD leads to nephropathy and how MICT exerts its protective effects. Our findings suggest that several genes and pathways (*Cd9*, *Foxq1*, *Mier3*, TGF-β signaling pathway etc.) might underlie HFD-induced kidney injury and MICT-mediated protective effects, and MICT could be a low-cost, low-side effect, and highly acceptable method for the prevention and treatment of obesity-related kidney diseases.

2. Materials and methods

2.1. Animals

3-week-old male C57BL/6 mice (n = 48) were housed under 22 °C ± 2 °C and 50% ± 5% humidity with a 12 h light-dark cycle. All mice had free access to water and food. After adapting for 1 week, mice were randomly divided into four groups (each n = 12, see Fig. 1 for details). Standard chow diet (D12450J, with 10 kcal% fat) and HFD (D12492, with 60 kcal% fat) were purchased from Research Diets. All animals were purchased form Tsinghua Laboratory Resource Center.

2.2. Exercise protocol

MICT were conducted on animal treadmill after 12-weeks feeding. Mice in exercise groups were adaptively trained at 7 m/min for 1 week, 10 min/day, and then formally trained for 8 weeks, 5 days/week, 45 min/day after a 10min 10 m/min warm-up. The training pace was 13 m/min in 1–2 weeks, 15 m/min in 3–4 weeks, 17 m/min in 5–8 weeks (Fig. 1). Every two weeks, maximal O_2 uptake test was performed with TSE Systems Phenomaster, and the exercise intensity was maintained with 60% VO_2 max intensity.

2.3. Body weight and serum parameter measurements

Body weight was measured and recorded every week. Blood samples were collected after fasting for 12 h and serum was separated by centrifugation at 3000 rpm for 15 min at 4 °C. Serum total cholesterol (TC), triglyceride (TG), high density lipoprotein cholesterol (HDL), low density lipoprotein cholesterol (LDL), glucose and creatinine were measured by automated chemistry analyzer (Kehua ZY KHB1280).

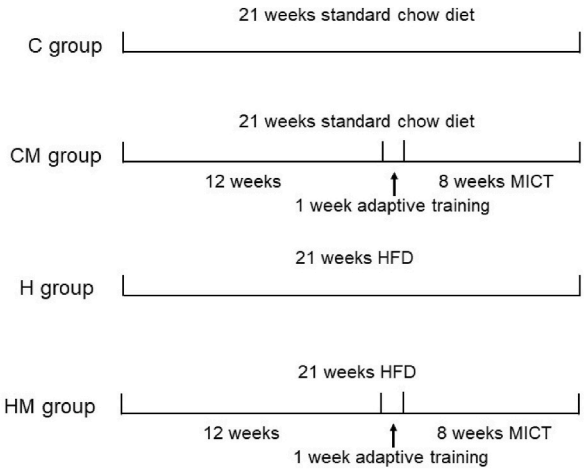

Fig. 1. Diet and training program.

2.4. Kidney histology and morphometric analysis

After harvested, kidneys were fixed in 4% paraformaldehyde and embedded in paraffin. Tissue sections (4 μm) were then stained with periodic acid schiff (PAS) and Masson's trichrome staining according to the manufacturer's instructions (Solarbio life sciences).

2.5. Quantitative real-time PCR

Total RNA extraction was performed with Trizol (Invitrogen) according to the manufacturer's instructions. cDNA was synthesized using HiScript II Q RT SuperMix (Vazyme). Quantitative RT-PCR was performed using AceQ qPCR SYBR Green Master Mix (Vazyme).

The following primers were used:

Gapdh Forward: AGAAGGTGGTGAAGCAGGCATCT.

Reverse: CGGCATCGAAGGTGGAAGAGTG.

Kim-1 Forward: GCGTGTCACCTATCAGAAGAGCAGTC.

Reverse: CCAGGAATCTCCACTCGACAACAAT.

2.6. RNA-seq data processing

Kidney RNA-seq was performed in Metware (Wuhan, China). Trim-galore (v.0.6.0) were used to trim paired-end reads and check quality. Reads were aligned to mouse genome reference (GRCm38.p6) from GENCODE using STAR (v.2.7.3a). featureCounts (v.1.6.3) counted reads matched to exon sites of genes included in GTF files from GENCODE. Differentially expressed genes (DEGs) was analyzed using DESeq2 (v.1.22.2) with raw counts as input. KEGG analysis and GO analysis of differential genes were performed by clusterProfiler (v3.10.1).

2.7. Statistical analysis

Statistical significance was assessed by Student's t-test using GraphPad Prism 7 (Graphpad software).

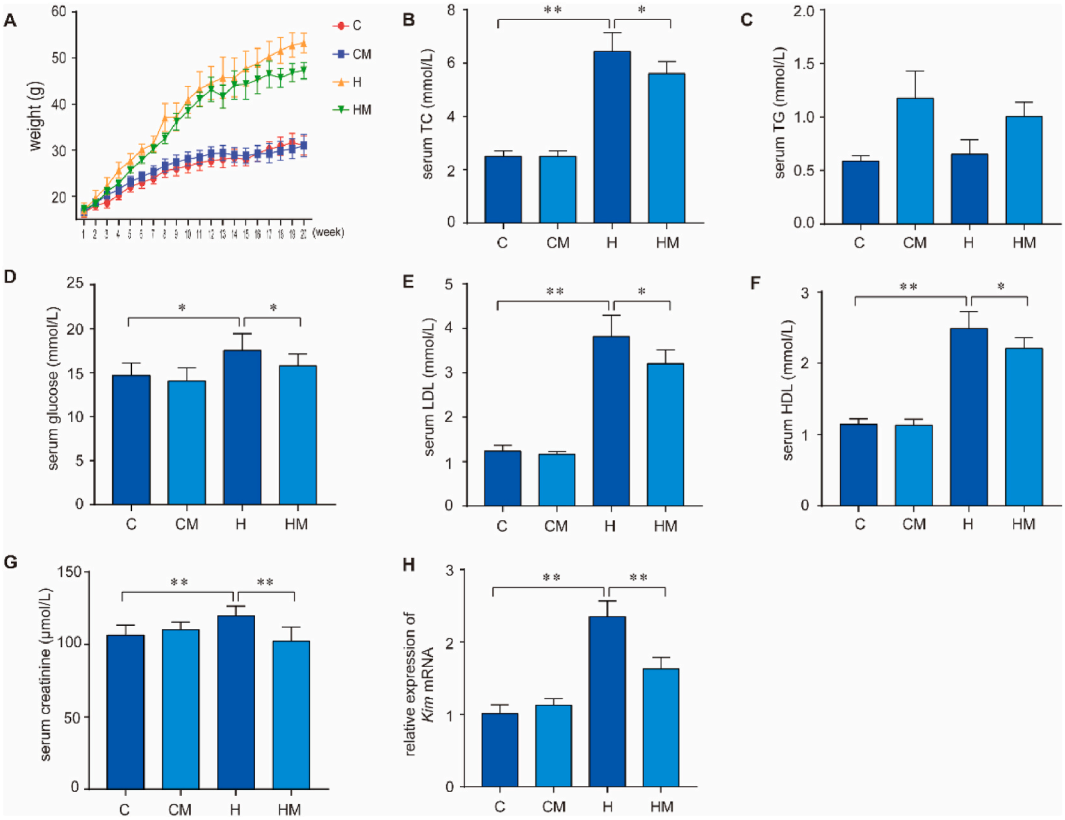

Fig. 2. 8 weeks MICT attenuates HFD-induced obesity and kidney injury in mice. (A) body weight. (B) TC, (C) TG, (D) glucose, (E) LDL, (F) HDL, (G) CRE levels in C, CM, H, HM groups' serum. (H) Expression of *Kim*-1 mRNA in C, CM, H, HM groups' kidneys; results are shown as relative expression normalized to that of *Gapdh* mRNA. **, $p < 0.01$, *, $p < 0.05$.

3. Results

3.1. MICT attenuated HFD-induced obesity and kidney injury

To assess whether MICT can attenuate the detrimetal effects of HFD on kidney, 48 Male C57BL/6J mice were averagely divided into standard chow diet (C), standard chow diet + MICT (CM), HFD (H), HFD + MICT (HM) groups; detailed diet and training programs are showed in Fig. 1. After 21 weeks HFD feeding, the body weight of H group was significantly higher than that of C group. Serum parameters, including glucose, TC, TG, HDL and LDL, were also increased in H group. After 8 weeks of MICT, both body weight and serum parameters (except for TG) decreased in the HM group compared to H group, indicating that MICT can mitigate HFD-induced obesity (Fig. 2A–F).

To evaluate kidney injury, serum creatinine and the expression of kidney injury factor-1 (KIM-1, a kidney injury biomarker) in kidney were measured. After 21 weeks of HFD feeding, the levels of creatinine and *Kim*-1 mRNA were significantly elevated in H group, but decreased after MICT (HM group) (Fig. 2G and H). As shown in Fig. 3A, PAS staining revealed the severe kidney injury in H group. Glomeruli of H group were hypertrophic, characterized by dilated renal sacs, increased mesangial matrix and thickened glomeruli. The tubular area of H group, especially the proximal convoluted tubule, displayed severe brush border disruption and epithelial cell shedding. Consistently, Masson staining showed increased collagen deposition in the glomeruli and interstitial cells of H group (Fig. 3B). However, these damages caused by HFD were remarkably alleviated by 8 weeks MICT (HM group).

Taken together, these data demonstrate that long term HFD-feeding induced obesity and kidney injury in mice, and these adverse effects could be attenuated by 8 weeks MICT.

3.2. Kidney transcriptome analysis

To investigate the underlying mechanisms of HFD-induced kidney injury and MICT-mediated protective effects, we analyzed

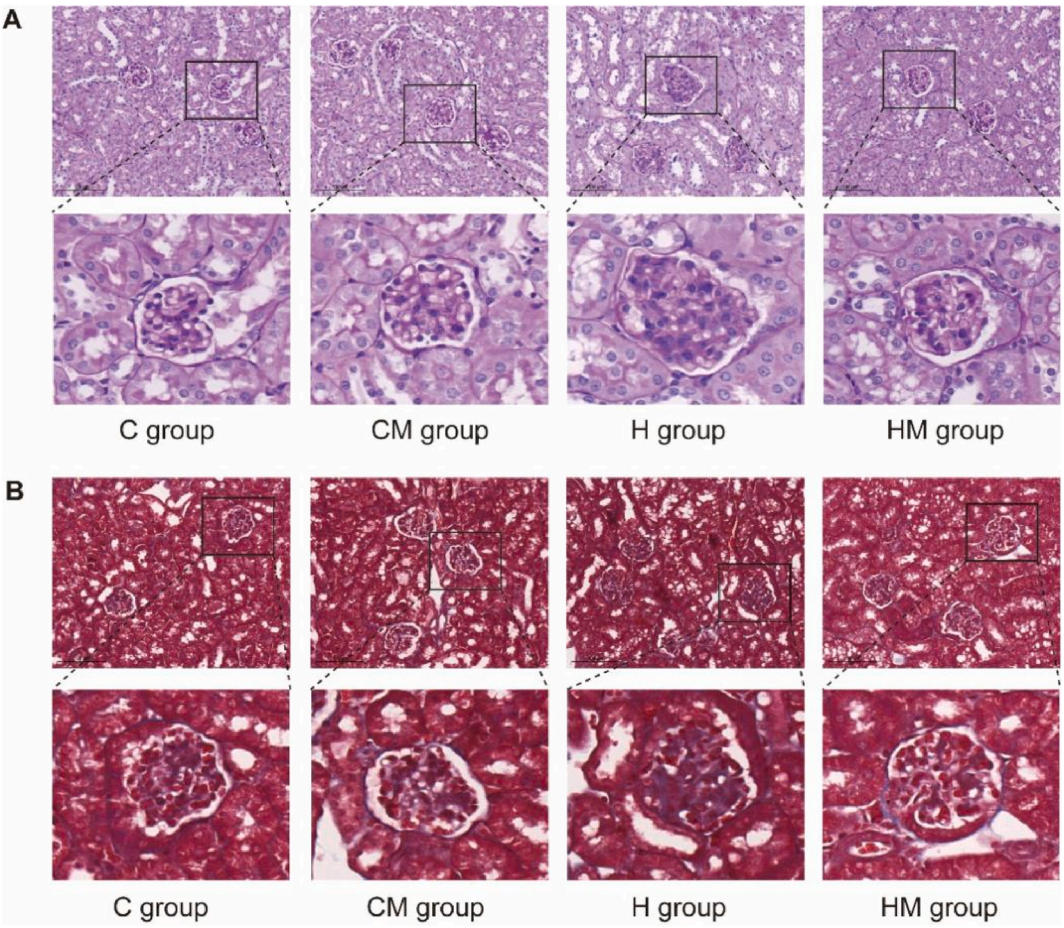

Fig. 3. (A) Representative photomicrographs of PAS-stained C, CM, H, HM groups' kidney sections. (B) Representative photomicrographs Masson's trichrome-stained C, CM, H, HM groups' kidney sections.

kidney transcriptome in C, H, HM groups. Principal components analysis (PCA) of global gene expression profiles showed that C and HM groups were clustered separately from H group in the first principal component, explaining 72% of sample variation (Fig. 4A).

DEGs among samples were screened with criteria of p adjusted value < 0.05 and $|\log2FC| \geq 1$. In total, 2297 (1074 up-regulated, 1223 down-regulated) and 3152 (1692 up-regulated, 1460 down-regulated) DEGs were identified between C and H groups (H vs C), and between H and HM groups (HM vs H), respectively (Fig. 4B). For these two DEGs datasets, 1839 DEGs were in the intersection, and about 80% DEGs in H vs C were contained in HM vs H (Fig. 4C). Most genes that were dysregulated after HFD feeding were rescued by MICT (Fig. 4 D). The top 10 significantly up- and down-regulated genes in H vs C were *Slc12a2*, *Foxq1*, *Mier3*, *Cd9*, *Nbeal1*, *Ptp4a2*, *Cd2ap*, *Hif1a*, *Galm*, *Hsph1*, and *Slc22a7*, *Ttc36*, *Mcrip2*, *Pcyt2*, *Ass1*, *Osgin1*, *Tmem259*, *Prodh*, *Uqcr11*, *Ggt1*, respectively. The top 10 significantly up- and down-regulated genes in HM vs H were *Akap8l*, *Tmem259*, *Osgin1*, *CT0104671*, *Safb2*, *Mapk15*, *Lgals4*, *Pcyt2*, *Ttc36*, *Khk*, and *Foxq1*, *Slc12a2*, *Lamp2*, *Rpl39*, *Mier3*, *Hsph1*, *Tmem64*, *Itm2b*, *Cav2*, *Cd9*, respectively (Supplementary Table 1 and Supplementary Table 2).

3.3. GO functional classification of DEGs

In order to excavate the biological function of these DEGs, we performed gene ontology (GO) functional classification for DEGs in H vs C and HM vs H. In H vs C, a total of 1570 terms were significantly enriched (q value < 0.01, 1257 up-regulated and 313 down-regulated). The top 10 significantly enriched up-regulated terms were regulation of supramolecular fiber organization, regulation of protein-containing complex assembly, positive regulation of catabolic process, membrane raft, membrane microdomain, guanyl nucleotide binding, guanyl ribonucleotide binding, GTP binding, regulation of cellular component size, protein localization to plasma membrane (Fig. 5A, Supplementary Table 3). The top 10 significantly enriched down-regulated terms were active transmembrane transporter activity, mitochondrial inner membrane, anion transport, organelle inner membrane, cellular amino acid metabolic process, small molecule catabolic process, secondary active transmembrane transporter activity, alpha-amino acid metabolic process, organic anion transport, organic acid catabolic process (Fig. 5B–Supplementary Table 4).

In HM vs H, a total of 1780 terms were significantly enriched (q value < 0.01, 312 up-regulated and 1468 down-regulated). The top 10 significantly enriched up-regulated terms were active transmembrane transporter activity, secondary active transmembrane transporter activity, anion transport, cellular amino acid metabolic process, apical plasma membrane, apical part of cell, mitochondrial

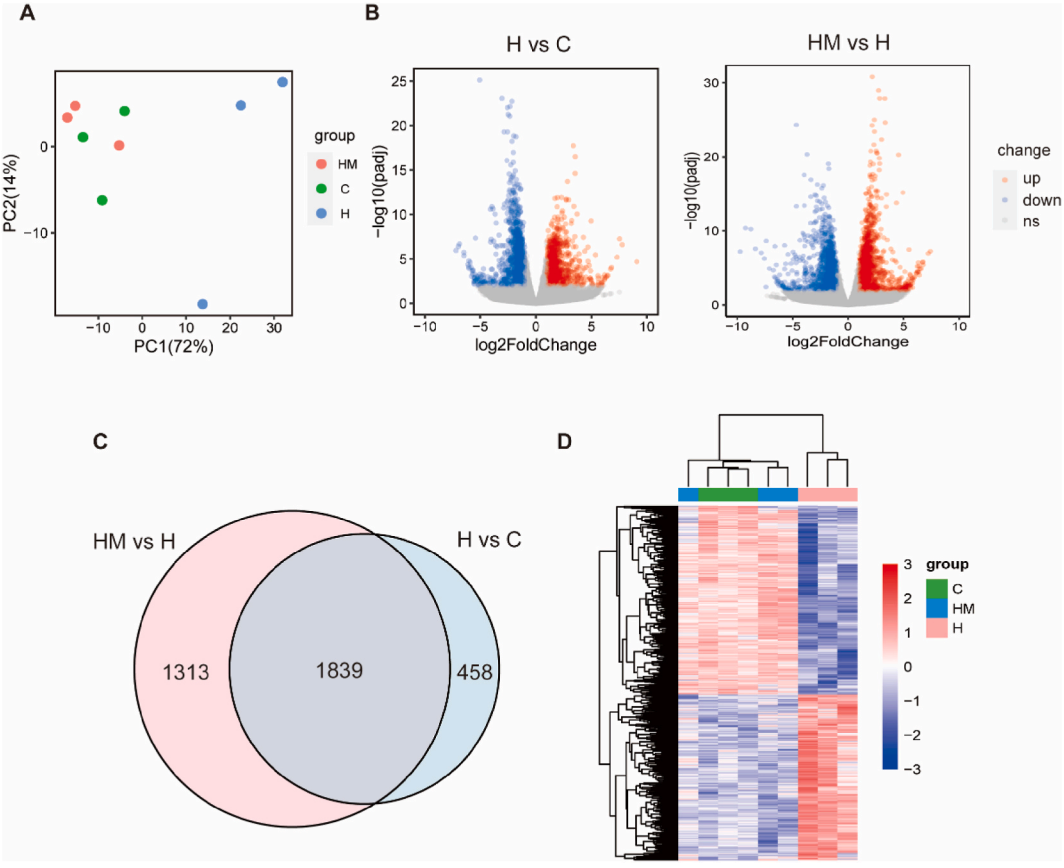

Fig. 4. Kidney transcriptome analysis. (A) PCA of total genes in C, H, HM groups. (B) Volcano plots show DEGs between C and H groups, and between H and HM groups (C). Hierarchical clustering analysis of DEGs (H vs C) in C, H, HM groups. (D) Venn diagram of the two DEGs datasets.

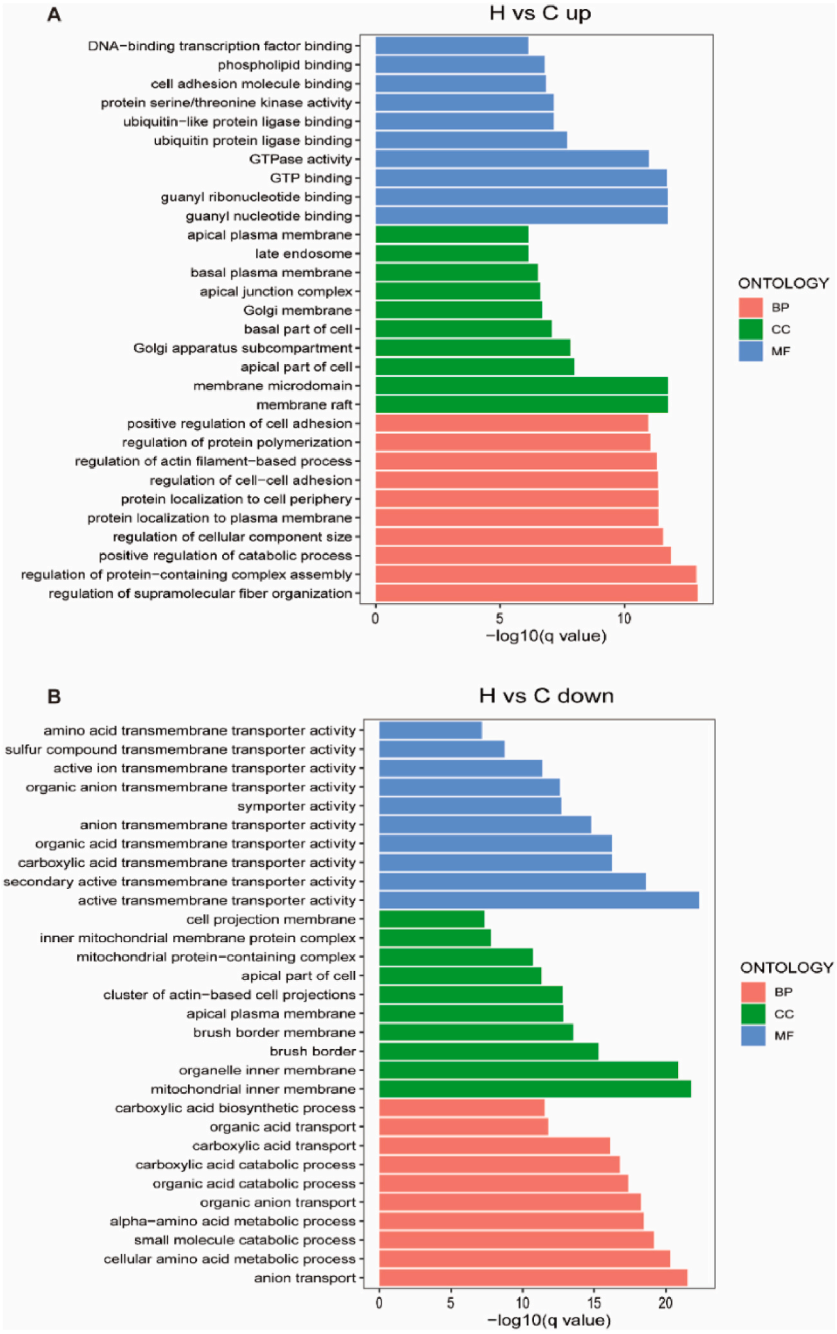

Fig. 5. GO function classification of DEGs (H vs C). (A) Up-regulated terms. (B) Down-regulated terms. BP, biological process. CC, cell component. MF, molecular function.

inner membrane, organic anion transport, organelle inner membrane, anion transmembrane transporter activity (Fig. 6A, Supplementary Table 5). The top 10 significantly enriched down-regulated terms were response to endoplasmic reticulum stress, regulation of supramolecular fiber organization, protein localization to cell periphery, protein localization to plasma membrane, establishment of protein localization to organelle, positive regulation of catabolic process, regulation of protein-containing complex assembly, regulation of cellular response to growth factor stimulus, Golgi vesicle transport, intrinsic component of organelle membrane (Fig. 6B–Supplementary Table 6).

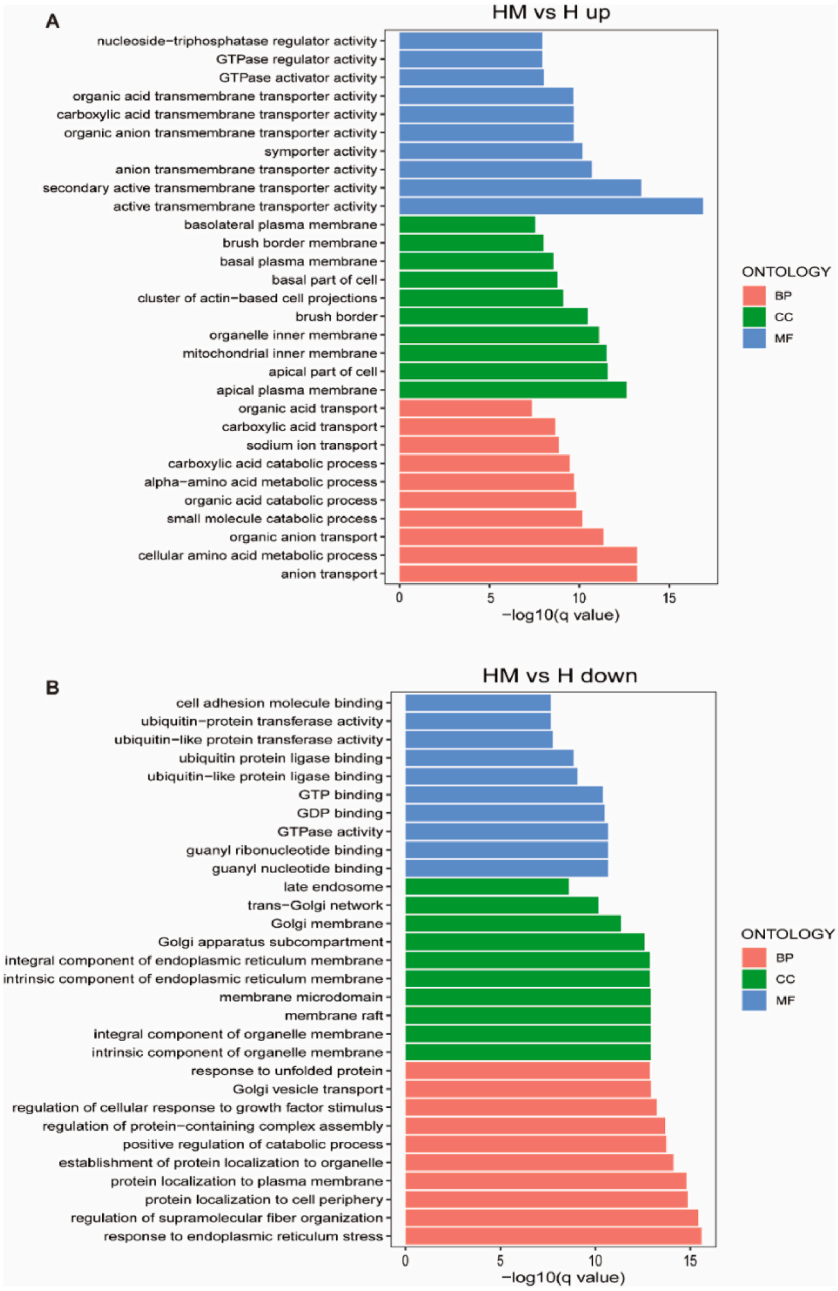

Fig. 6. GO function classification of DEGs (HM vs H). (A) Up-regulated terms. (B) Down-regulated terms.

3.4. KEGG pathway analysis of DEGs

Kyoto encyclopedia of genes and genomes (KEGG) pathway analysis was further performed to gain more insight into these DEGs. In H vs C, DEGs were significantly enriched in 115 pathways (q value < 0.05, 94 up-regulated and 21 down-regulated). The top 10 significantly enriched up-regulated pathways were tight junction, endocytosis, focal adhesion, Relaxin signaling pathway, proteoglycans in cancer, dopaminergic synapse, Hippo signaling pathway, regulation of actin cytoskeleton, adherens junction, Estrogen signaling pathway (Fig. 7A and Supplementary Table 7). The top 10 significantly enriched down-regulated pathways were peroxisome, oxidative phosphorylation, lysosome, mineral absorption, Parkinson disease, glycine, serine and threonine metabolism, sulfur metabolism, chemical carcinogenesis, arginine and proline metabolism, diabetic cardiomyopathy (Fig. 7B and Supplementary Table 8).

In HM vs H, DEGs were significantly enriched in 66 pathways (q value < 0.05, 5 up-regulated and 61 down-regulated). The

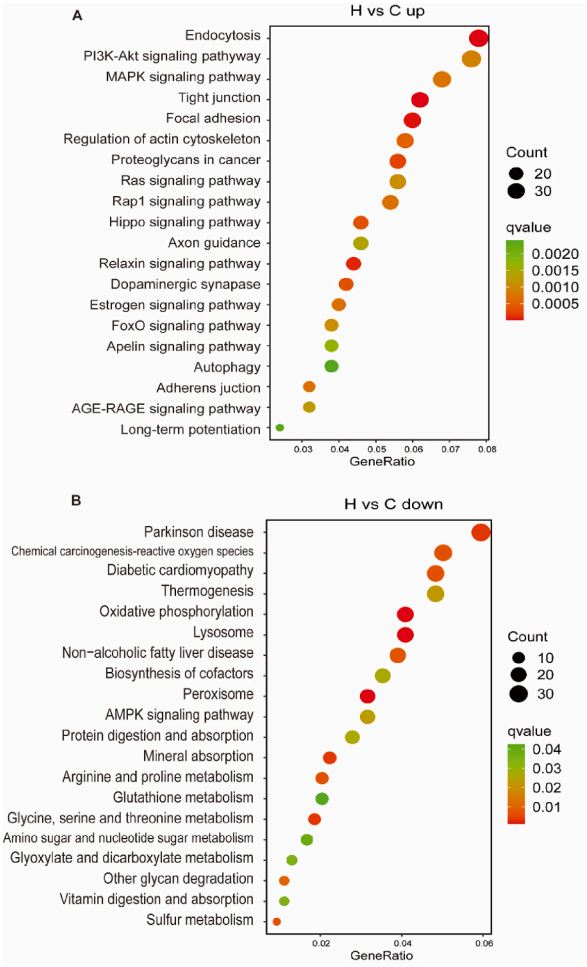

Fig. 7. KEGG pathway analysis of DEGs (H vs C). (A) Up-regulated pathways. (B) Down-regulated pathways.

significantly enriched up-regulated pathways were ABC transporters, biosynthesis of cofactors, arginine and proline metabolism, Glycine, serine and threonine metabolism, peroxisome (Fig. 8A and Supplementary Table 9). The top 10 significantly enriched down-regulated pathways were protein processing in endoplasmic reticulum, proteoglycans in cancer, lipid and atherosclerosis, Hippo signaling pathway, TGF-beta signaling pathway, adherens junction, axon guidance, tight junction, focal adhesion, autophagy (Fig. 8B and Supplementary Table 10).

4. Discussion

Diets rich in saturated fat in modern lifestyles are believed to be a major driver of the global obesity epidemic, with obesity now recognized as a risk factor for the development of kidney diseases. Physics exercise has been applied as an effective method to prevent and treat a variety of metabolic diseases, however, the role of physics exercise in obesity-related kidney diseases remains poorly understood. In this study, we found that 8 weeks MICT significantly reduced obesity indexes (body weight, serum glucose, TC, HDL, LDL, except for TG) and kidney injury indexes (creatinine and the expression of *Kim*-1 mRNA) in HFD-fed mice. Consistently, PAS staining and Masson staining displayed that MICT maintained the morphological structure of kidney subunits and reduced kidney fibrosis in HFD-fed mice. Thus, these data demonstrate a protective role of MICT in HFD-induced kidney injury.

Furthermore, we performed RNA-seq to explore the underlying mechanism by which HFD leads to nephropathy and MICT exerts its protective effects. Using the criteria of p adjusted value < 0.05 and $|\log2FC| \geq 1$, we screened 2297 and 3152 DEGs in H vs C and HM vs H, respectively. Among the most significant DEGs, several genes that were dysregulated after HFD-feeding and were rescued by MICT caught our attention:

CD9 (up-regulated in H vs C, while down-regulated in HM vs H) is a 24 kDa tetraspanin membrane protein, known to regulate cell adhesion and migration, cancer progression and metastasis [26,27]. Lazareth et al. reported that the expression of CD9 increased significantly in parietal epithelial cells in mouse models of crescentic glomerulonephritis and focal segmental glomerulosclerosis, and in kidneys from individuals diagnosed with these diseases. Targeting *Cd9* gene in parietal epithelial cells prevents the oriented

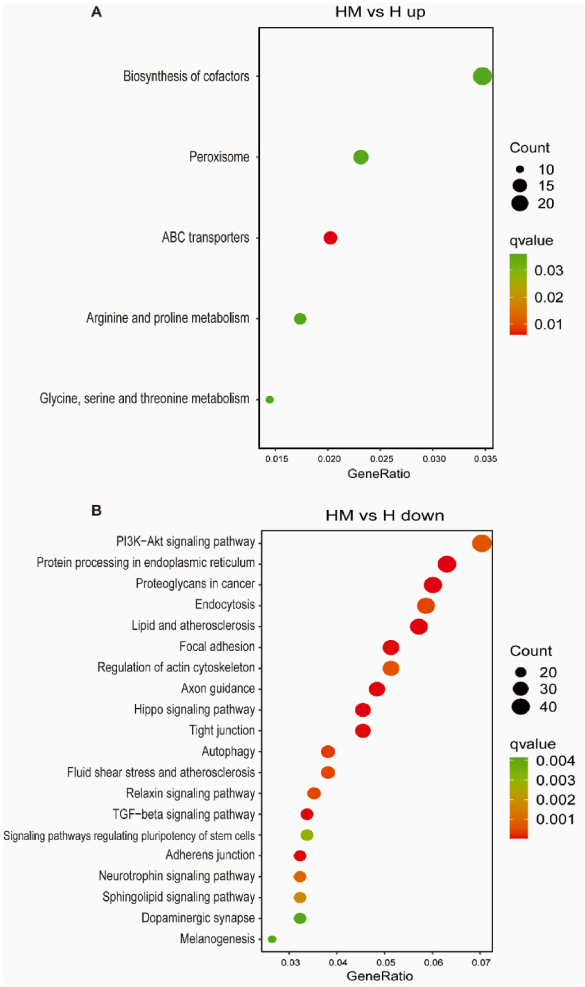

Fig. 8. KEGG pathway analysis of DEGs (HM vs H). (A) Up-regulated pathways. (B) Down-regulated pathways.

migration of parietal epithelial cells into the glomerular tuft, thereby alleviating glomerular damage in crescentic glomerulonephritis and focal segmental glomerulosclerosis mouse models [28].

FoxQ1 (up-regulated in H vs C, while down-regulated in HM vs H) is a member of forkhead box transcription factor family [29]. Liang et al. found that expression of FoxQ1 was elevated in renal tubular epithelial cells under high-glucose, high-lipid conditions, and FoxQ1 inhibited Sirt4 expression, leading to mitochondrial dysfunction and promoting the development of DKD [30]. In addition, FoxQ1 is recognized as a driver of epithelial-mesenchymal transition (EMT) and metastasis in multiple cancer types [31,32]. EMT is widely accepted as a mechanism for the transformation of injured renal tubular cells into mesenchymal cells, and is involved in the development of fibrosis in kidney diseases [33]. Hence, potential linkage between FoxQ1 and kidney fibrosis might exist.

Mier3 (up-regulated in H vs C, while down-regulated in HM vs H) was identified as a frequently mutated gene in hyper-mutated colorectal cancer, and higher expression of Mier3 was detected in breast cancer [34]. Huang et al. reported that Mier3 promoted the proliferation and migration of breast cancer cells, and Mier3 interacted with histone deacetylase 1/2 and Snail to form an inhibitory complex, which could bind to E-cadherin promoter and promoted EMT by silencing E-cadherin [35]. Therefore, elevated Mier3 might contribute to the kidney fibrosis in HFD-fed mice.

Moving beyond individual genes, GO and KEGG analysis screened 1570 terms and 115 pathways in H vs C, and 1780 terms and 66 pathways in HM vs H. Several previous studies have reported that HFD disrupted lipid metabolic homeostasis in the kidney. Kume et al. showed that HFD stimulated lipogenesis enzymes in fatty acid synthesis pathway but inhibited lipolysis, resulting in excess accumulation of lipid in the kidney [25]. Changes in lipid metabolism eventually caused kidney injury. Yamamoto et al. demonstrated that HFD caused lysosomal phospholipid accumulation in proximal renal tubule cells, which impairs autophagy flux, leading to lipotoxicity and kidney injury [24]. As expected, in H vs C, a wide range of terms and pathways involved in metabolism were down-regulated, including alpha−amino acid metabolic process, cellular amino acid metabolic process, protein digestion and absorption, arginine and proline metabolism, glutathione metabolism, vitamin digestion and absorption, threonine metabolism etc. These results indicate that long-term HFD feeding led to systemic metabolism disorders, not limited to lipid metabolism. On the contrary, in HM vs H,

alpha−amino acid metabolic process, cellular amino acid metabolic process, arginine and proline metabolism, glycine, serine and threonine metabolism were up-regulated, while fluid shear stress and atherosclerosis, lipid and atherosclerosis were down-regulated, suggesting that MICT rescued HFD-induced lipid accumulation and metabolism disorders in kidney to some extent.

Kidney fibrosis is a common pathophysiological mechanism contributing to the progression of several kidney diseases [36,37]. Among various contributors, TGF-β1 is considered the key factor driving fibrosis. TGF-β1 activates both canonical and non-canonical TGF-β signaling pathways, which induces the activation and proliferation of myofibroblasts and subsequent accumulation of extracellular matrix [38,39]. In this study, we found that TGF-β signaling pathway was up-regulated in H vs C and down-regulated in HM vs H, which is consistent with the results of Masson staining showing increased kidney fibrosis in H group and decreased fibrosis in HM group. It was reported that exercise training increased the level of fibroblast growth factor 21, which inactivated TGF-β1-Smad2/3-matrix metalloproteinase 2/9 signaling pathway, thereby alleviating myocardial fibrosis [40]. As well, similar mechanism might also underlie MICT-mediated protective effects on HFD-induced kidney fibrosis.

5. Conclusions

In this study, we determine the protective role of MICT in HFD-induced kidney injury. We identified several genes and pathways (*Cd9*, *Foxq1*, *Mier3*, TGF-β signaling pathway etc.) that might underlie HFD-induced kidney injury and MICT-mediated protective effects, and might serve as potential targets for the treatment of obesity-related nephropathy.

Ethics statement

Animal experiments were approved by the Institutional Animal Care and Use Committee of Tsinghua University (THU-LARC-2023-004) and were conducted in accordance with the ARRIVE guidelines.

Data availability statement

The data that support the findings of this study are available from the corresponding authors upon reasonable request.

CRediT authorship contribution statement

Weihao Hong: Writing – review & editing, Writing – original draft, Visualization, Validation, Methodology, Investigation, Formal analysis, Conceptualization. **Yisheng Luan:** Writing – review & editing, Software, Methodology, Investigation. **Yixuan Ma:** Writing – review & editing, Software, Methodology, Investigation. **Bing Zhang:** Writing – review & editing, Supervision, Project administration, Funding acquisition. **Yingzhe Xiong:** Writing – review & editing, Supervision, Project administration, Investigation, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e27157.

References

[1] N.O. Câmara, K. Iseki, H. Kramer, Z.H. Liu, K. Sharma, Kidney disease and obesity: epidemiology, mechanisms and treatment, Nat. Rev. Nephrol. 13 (2017) 181–190.
[2] A.E. Declèves, K. Sharma, Obesity and kidney disease: differential effects of obesity on adipose tissue and kidney inflammation and fibrosis, Curr. Opin. Nephrol. Hypertens. 24 (2015) 28–36.
[3] M. Wang, Z. Wang, Y. Chen, Y. Dong, Kidney damage caused by obesity and its feasible treatment drugs, Int. J. Mol. Sci. 23 (2022).
[4] Y. Sun, X. Ge, X. Li, J. He, X. Wei, J. Du, J. Sun, X. Li, Z. Xun, W. Liu, et al., High-fat diet promotes renal injury by inducing oxidative stress and mitochondrial dysfunction, Cell Death Dis. 11 (2020) 914.
[5] T. Miyamoto, J.J. Carrero, P. Stenvinkel, Inflammation as a risk factor and target for therapy in chronic kidney disease, Curr. Opin. Nephrol. Hypertens. 20 (2011) 662–668.
[6] S.D. Navaneethan, J.P. Kirwan, E.M. Remer, E. Schneider, B. Addeman, S. Arrigain, E. Horwitz, J.C. Fink, J.P. Lash, C.A. McKenzie, et al., Adiposity, physical function, and their associations with insulin resistance, inflammation, and adipokines in CKD, Am. J. Kidney Dis. 77 (2021) 44–55.
[7] R.L. Amdur, H.I. Feldman, J. Gupta, W. Yang, P. Kanetsky, M. Shlipak, M. Rahman, J.P. Lash, R.R. Townsend, A. Ojo, et al., Inflammation and progression of CKD: the CRIC study, Clin. J. Am. Soc. Nephrol. : CJASN 11 (2016) 1546–1556.
[8] J.E. Hall, J.M. do Carmo, A.A. da Silva, Z. Wang, M.E. Hall, Obesity-induced hypertension: interaction of neurohumoral and renal mechanisms, Circ. Res. 116 (2015) 991–1006.
[9] H. Nishi, T. Higashihara, R. Inagi, Lipotoxicity in kidney, heart, and skeletal muscle dysfunction, Nutrients 11 (2019).

[10] K. Jaikumkao, A. Pongchaidecha, V. Chatsudthipong, S.C. Chattipakorn, N. Chattipakorn, A. Lungkaphin, The roles of sodium-glucose cotransporter 2 inhibitors in preventing kidney injury in diabetes, Biomed. Pharmacothe. = Biomedecine & pharmacotherapie 94 (2017) 176–187.
[11] E. Ozbek, Y.O. Ilbey, M. Ozbek, A. Simsek, M. Cekmen, A. Somay, Melatonin attenuates unilateral ureteral obstruction-induced renal injury by reducing oxidative stress, iNOS, MAPK, and NF-kB expression, J. Endourol. 23 (2009) 1165–1173.
[12] N. Tsuboi, Y. Okabayashi, A. Shimizu, T. Yokoo, The renal pathology of obesity, Kidney Int. Rep. 2 (2017) 251–260.
[13] B.K. Pedersen, B. Saltin, Exercise as medicine - evidence for prescribing exercise as therapy in 26 different chronic diseases, Scand. J. Med. Sci. Sports 25 (Suppl 3) (2015) 1–72.
[14] C. Zhu, H. Ma, A. He, Y. Li, C. He, Y. Xia, Exercise in cancer prevention and anticancer therapy: efficacy, molecular mechanisms and clinical information, Cancer Lett. 544 (2022) 215814.
[15] C. Fiuza-Luces, A. Santos-Lozano, M. Joyner, P. Carrera-Bastos, O. Picazo, J.L. Zugaza, M. Izquierdo, L.M. Ruilope, A. Lucia, Exercise benefits in cardiovascular disease: beyond attenuation of traditional risk factors, Nat. Rev. Cardiol. 15 (2018) 731–743.
[16] S. Balducci, M. Sacchetti, J. Haxhi, G. Orlando, V. D'Errico, S. Fallucca, S. Menini, G. Pugliese, Physical exercise as therapy for type 2 diabetes mellitus, Diabetes/Metabolism Res. Rev. 30 (Suppl 1) (2014) 13–23.
[17] A. Sabbahi, R. Arena, A. Elokda, S.A. Phillips, Exercise and hypertension: uncovering the mechanisms of vascular control, Prog. Cardiovasc. Dis. 59 (2016) 226–234.
[18] T.J. Wilkinson, N.F. Shur, A.C. Smith, "Exercise as medicine" in chronic kidney disease, Scand. J. Med. Sci. Sports 26 (2016) 985–988.
[19] K.L. Johansen, P. Painter, Exercise in individuals with CKD, Am. J. Kidney Dis. 59 (2012) 126–134.
[20] N.C. Bishop, J.O. Burton, M.P.M. Graham-Brown, D.J. Stensel, J.L. Viana, E.L. Watson, Exercise and chronic kidney disease: potential mechanisms underlying the physiological benefits, Nat. Rev. Nephrol. 19 (2023) 244–256.
[21] K. Uchiyama, K. Adachi, K. Muraoka, T. Nakayama, T. Oshida, M. Yasuda, A. Hishikawa, H. Minakuchi, K. Miyashita, H. Tokuyama, et al., Home-based aerobic exercise and resistance training for severe chronic kidney disease: a randomized controlled trial, J. Cachexia, Sarcopenia Muscle 12 (2021) 1789–1802.
[22] L. Yang, D.X. Li, B.Q. Cao, S.J. Liu, D.H. Xu, X.Y. Zhu, Y.J. Liu, Exercise training ameliorates early diabetic kidney injury by regulating the H(2) S/SIRT1/p53 pathway, Faseb. J. 35 (2021) e21823.
[23] J. Wadén, H.K. Tikkanen, C. Forsblom, V. Harjutsalo, L.M. Thorn, M. Saraheimo, N. Tolonen, M. Rosengård-Bärlund, D. Gordin, H.O. Tikkanen, et al., Leisure-time physical activity and development and progression of diabetic nephropathy in type 1 diabetes: the FinnDiane Study, Diabetologia 58 (2015) 929–936.
[24] T. Yamamoto, Y. Takabatake, A. Takahashi, T. Kimura, T. Namba, J. Matsuda, S. Minami, J.Y. Kaimori, I. Matsui, T. Matsusaka, et al., High-fat diet-induced lysosomal dysfunction and impaired autophagic flux contribute to lipotoxicity in the kidney, J. Am. Soc. Nephrol. : JASN (J. Am. Soc. Nephrol.) 28 (2017) 1534–1551.
[25] S. Kume, T. Uzu, S. Araki, T. Sugimoto, K. Isshiki, M. Chin-Kanasaki, M. Sakaguchi, N. Kubota, Y. Terauchi, T. Kadowaki, et al., Role of altered renal lipid metabolism in the development of renal injury induced by a high-fat diet, J. Am. Soc. Nephrol. : JASN (J. Am. Soc. Nephrol.) 18 (2007) 2715–2723.
[26] A. Lorico, M. Lorico-Rappa, J. Karbanová, D. Corbeil, G. Pizzorno, CD9, a tetraspanin target for cancer therapy? Exp. Biol. Med. (Maywood, N.J.) 246 (2021) 1121–1138.
[27] R. Reyes, B. Cardeñes, Y. Machado-Pineda, C. Cabañas, Tetraspanin CD9: a key regulator of cell adhesion in the immune system, Front. Immunol. 9 (2018) 863.
[28] H. Lazareth, C. Henique, O. Lenoir, V.G. Puelles, M. Flamant, G. Bollée, C. Fligny, M. Camus, L. Guyonnet, C. Millien, et al., The tetraspanin CD9 controls migration and proliferation of parietal epithelial cells and glomerular disease progression, Nat. Commun. 10 (2019) 3303.
[29] Y. Li, Y. Zhang, Z. Yao, S. Li, Z. Yin, M. Xu, Forkhead box Q1: a key player in the pathogenesis of tumors (Review), Int. J. Oncol. 49 (2016) 51–58.
[30] L. Liang, C. Wo, Y. Yuan, H. Cao, W. Tan, X. Zhou, D. Wang, R. Chen, M. Shi, F. Zhang, et al., miR-124-3p improves mitochondrial function of renal tubular epithelial cells in db/db mice, Faseb J. 37 (2023) e22794.
[31] H. Zhang, F. Meng, G. Liu, B. Zhang, J. Zhu, F. Wu, S.P. Ethier, F. Miller, G. Wu, Forkhead transcription factor foxq1 promotes epithelial-mesenchymal transition and breast cancer metastasis, Cancer Res. 71 (2011) 1292–1301.
[32] J. Feng, L. Xu, S. Ni, J. Gu, H. Zhu, H. Wang, S. Zhang, W. Zhang, J. Huang, Involvement of FoxQ1 in NSCLC through regulating EMT and increasing chemosensitivity, Oncotarget 5 (2014) 9689–9702.
[33] R.M. Carew, B. Wang, P. Kantharidis, The role of EMT in renal fibrosis, Cell Tissue Res. 347 (2012) 103–116.
[34] M. Peng, Y. Hu, W. Song, S. Duan, Q. Xu, Y. Ding, J. Geng, J. Zhou, MIER3 suppresses colorectal cancer progression by down-regulating Sp1, inhibiting epithelial-mesenchymal transition, Sci. Rep. 7 (2017) 11000.
[35] W. Huang, J. Chen, X. Liu, X. Liu, S. Duan, L. Chen, X. Liu, J. Lan, Y. Zou, D. Guo, et al., MIER3 induces epithelial-mesenchymal transition and promotes breast cancer cell aggressiveness via forming a co-repressor complex with HDAC1/HDAC2/Snail, Exp. Cell Res. 406 (2021) 112722.
[36] L. Li, H. Fu, Y. Liu, The fibrogenic niche in kidney fibrosis: components and mechanisms, Nat. Rev. Nephrol. 18 (2022) 545–557.
[37] T.T. Ma, X.M. Meng, TGF-β/Smad and renal fibrosis, Adv. Exp. Med. Biol. 1165 (2019) 347–364.
[38] Y. Isaka, Targeting TGF-β signaling in kidney fibrosis, Int. J. Mol. Sci. 19 (2018).
[39] X.M. Meng, D.J. Nikolic-Paterson, H.Y. Lan, TGF-β: the master regulator of fibrosis, Nat. Rev. Nephrol. 12 (2016) 325–338.
[40] Y. Ma, Y. Kuang, W. Bo, Q. Liang, W. Zhu, M. Cai, Z. Tian, Exercise training alleviates cardiac fibrosis through increasing fibroblast growth factor 21 and regulating TGF-β1-smad2/3-MMP2/9 signaling in mice with myocardial infarction, Int. J. Mol. Sci. 22 (2021).