Research progress of MicroRNA in podocytes autophagy in diabetic nephropathy

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Abstract: Diabetic nephropathy (DN) is one of the most common microvascular complications of diabetes, and is a kind of abnormal microangiopathy of kidney structure, function or clinical indicators caused by diabetes. Podocyte injury has been considered as a major contributor to the progression of diabetic nephropathy(DN). microRNA can participate in podocytes injury through autophagy.In this paper, the mechanism of microRNA involved in DN podocytes autophagy was reviewed to provide reference for the treatment of DN in the future.

Keywords: Diabetic Nephropathy; Podocytes; Microrna; Autophagy

Introduction

Diabetic nephropathy (DN), a microvascular complication of diabetes, has a high incidence in many countries[1].At present, the pathogenesis of DN is still being explored. Studies have shown that podocyte loss or injury is one of the earliest observed features in the pathogenesis of diabetic nephropathy[2].Podocytes, which are visceral epithelial cells of the renal capsule, are attached to the outside of the glomerular basement membrane. This membrane, together with podocytes and the capillary endothelium, forms the glomerular filtration barrier。 Podocyte viability and apoptosis as well as autophagy can affect glomerular function.MicroRNAs (miRNAs or miRs) are a class of short noncoding, highly conserved RNAs with 19–25 nucleotides in length, which can bind to the 30-untranslated region (30UTR) of target mRNAs, and thereby promote mRNA degradation or mRNA translation inhibition.Autophagy[3]refers to the process involving the decomposition of intracellular components via lysosomes. Autophagy plays an important role in maintaining and regulating cell homeostasis by degrading intracellular components and providing degradation products to cells.In pathological environment, cells usually repair damage by forming autophagosomes to remove damaged proteins and organelles. Thus, disruption of autophagy disturbs cellular homeostasis and contributes to the development of various diseases. This paper reviews the signaling pathways related to miRNA in DN podocytes autophagy.

1. SOX2OT induces podocyte autophagy through miR9/SIRT1 axis

Long non-coding RNAs (lncRNAs) play an important role in the pathogenesis of various human diseases, A study showed[4] that the lncRNA SOX2-overlapping transcript (SOX2OT) is significantly down-regulated in DN mice and high glucose (HG)-treated human podocytes cells (HPCs).SOX2OT overexpression significantly promoted cell proliferation and inhibited cell apoptosis und.r HG stimulation. Furthermore, SOX2OT overexpression notably decreased protein levels of pro-apoptotic Caspase-3 and Bax, whereas increased levels of anti-apoptotic Bcl-2 under HG stimulation. In contrast, SOX2OT knockdown exerted the opposite effect. These data indicate that SOX2OT overexpression alleviates the

HG-induced HPCs injury. SIRT1 is a deacetylase and can induce autophagy via deacetylation of autophagy-related marker Beclin-1 and other autophagy mediators, Other studies have shown[5] that SIRT1 has been shown to promote cell survival by suppressing p53-dependent apoptosis in response to DNA damage and oxidative stress, and recent data suggests that the interplay of SIRT1-p53 pathway controls cellular senescence.Furthermore, SIRT1 was also shown to modulate PGC-1a activity and to attenuate aldosterone-induced mitochondri damage and podocyte injury.bioinformatics analysis revealed that SOX2OT harbors predicted binding sites of miR-9 and sirtuin 1 (SIRT1) might act as a putative target of miR-9. Data revealed that SOX2OT overexpression significantly decreased miR-9 expression, but notably increased SIRT1 mRNA and protein levels.By contrast, SOX2OT knockdown exerted the opposite effect. The protective effects of SOX2OT on podocytes injury were mediated through autophagy induction by the miR-9/SIRT1 axis.

2. CASC2 induces podocyte autophagy through miR-9-5P/ PPARγ axis

LncRNA cancer susceptibility candidate 2 (CASC2),located on chromosome 10q26, plays a regulatory role as an anti-cancer factor in various cancers. Recently[6], Wang et al. revealed that CASC2 was specifically reduced in serum and renal tissues of type 2 diabetes patients with chronic renal failure, and follow-up identified that the serum of patients with low CASC2 expression had higher incidence of chronic renal failure. there were complementary sites between miR-9-5p and CASC2 by bioinformatics website starBase v2.0.The Peroxisome proliferator-activated receptor gamma (PPARγ) protein, located in the cellular nucleus, contains 505 amino acids and has a molecular weight of 57.6kDa. PPAR-γ is best known for its abilities to regulate pathways linked to adipocyte differentiation and metabolism .PPARγ is implicated in several metabolic syndromes, including DN. Down-regulated PPARγ could activate β-catenin signaling to destroy podocyte architectural integrity and increase cell apoptosis in DN. Data revealed[7] that CASC2 mainly up-regulated the expression of PPARγ by acting as the ceRNA of miR-9-5p, thus alleviating HG-induced podocytes injury through increasing cell viability, autophagy and reducing cell apoptosis.

3. MiR-21 inhibits podocyte autophagy through the PTEN-PI3K/ Akt/m TOR pathway

MiR-21 can regulate cell differentiation, proliferation and apoptosis, and the serum concentration of DN patients is significantly higher than that of normal people, [7] Tensin homolog gene (PTEN) is a tumor suppressor gene on chromosome 10q23.3, which has lipid and protein diphosphate activity and plays an important role in cell growth, proliferation, survival, apoptosis, angiogenesis, cell migration and invasion. Phosphatidylinositol 3 kinase (PI3K)/protein kinase B (Akt)/Mammalian target of rapamycin (mTOR) signaling pathway is involved in the regulation of glucose metabolism, cell proliferation, cell transcription and apoptosis, and is closely related to the occurrence and development of DN.microRNA-21 (miR-21) [8] as the molecular link between high glucose and PTEN suppression. Renal cortices from OVE26 type 1 diabetic mice showed significantly elevated levels of miR-21 associated with reduced PTEN and increased fibronectin content. In renal mesangial cells, high glucose increased the expression of miR-21, which targeted the 3'-UTR of PTEN mRNA to inhibit PTEN protein expression. Overexpression of miR-21 mimicked the action of high glucose, which included a reduction in PTEN expression and a concomitant increase in Akt phosphorylation. In contrast, expression of miR-21 Sponge, to inhibit endogenous miR-21, prevented down-regulation of PTEN and phosphorylation of Akt induced by high glucose. Other studies[9] have shown that ursolic acid (UA)can lead to the recovery of autophagic function in podocytes through the inhibition of miR-21 expression, which leads to up-regulated PTEN expression and decreased abnormal activation of the PI3K/Akt/mTOR pathway.

4. MiR-21 inhibits podocellular autophagy through the FOXO1 axis

Cell autop-hagic activity was obviously reduced in podocytes after HG treatment,as evidenced by reduced LC3II/LC3I ratio and increased p62 protein expression in HG. Also, the depletion of miR-21 weakened HG-mediated autophagy inhibition in podocytes. these data showed that miR-21 loss alleviated HG-induced podocyte injury.The Forkhead Box O1 (FOXO1) is mainly expressed in adipocytes, muscle cells, hepatocytes and islet cells, and is involved in the physiological processes of glucose and lipid metabolism, proliferation, differentiation and apoptosis of islet B cells. FOXO1 can alleviate inflammation and induce autophagy at the cellular level. showed that hepatic expression levels of microRNA-21 (miR-21) were decreased in high-fat diet (HFD)-induced diabetic mice. Adenovirus-mediated overexpression of miR-21 decreased the expression ofphosphoenolpyruvate carboxykinase (PEPCK) and glucose-6-phosphatase (G6Pase) and inhibited glucose production in primary mouse hepatocytes. Furthermore, overexpression of miR-21 in mouse hepatocytes and mouse livers decreased the protein levels of FOXO1 and increased hepatic insulin sensitivity. By contrast, silencing of miR-21 increased the protein levels of FOXO1, subsequently leading to a decrease in insulin sensitivity and impaired glucose intolerance in mice fed with high-fat diet for 4 weeks. These results suggest that FOXO1 was a potential target of miR-21[10] MiR-21 exerted its pro-apoptosis and anti-autophagy effects by targeting FOXO1 in HG-cultured podocytes. Atr enhanced FOXO1 expression by downregulating miR-21 in HG-cultured podocytes. We concluded that Atr mitigated kidney injury in DN mice and alleviated HG-mediated apoptosis increase and autophagy inhibition in podocytes by regulating miR-21/FOXO1 axis.

5. The p53/miR-34a/SIRT1 axis inhibits podocyte autophagy

MiR-34a, a p53-regulated miRNA, directly targets SIRT1 and contributed to DN progression. MiR-34a represses SIRT1 to activate p53 and establish a positive feedback loop[11]. we observed that serum miR-34a level was positively correlated with podocyte injury in DN patients. The expression of acetylated p53 and miR-34a was upregulated, SIRT1was downregulated in glomeruli from patients with DN and STZ induced diabetic mice, as well as in human podocytes treated with advanced glycation end (AGE). MiR-34a antagonism in vitro and vivo in STZ induced diabetic mice developed alleviated glomerulus injury as reflected by attenuated albuminuria, reduced podocyte loss and restored autophagic flux. In human podocyte, inhibition of AGE formation by pyridoxamine prevented miR-34a dependent repression of SIRT1, p53 acetylation and activate podocyte autophagy in a dose-dependent manner. MiR-34a overexpression increases acetylation of p53 by translational repression of SIRT1. SIRT1 overexpression also impacts AGE induced apoptosis through deacetylating p53,whereas silencing of SIRT1 by EX527 attenuated the cytoprotective functions of miR-34a knockdown.Moreover, blockade of p53 acetylation significantly rescued miR-34a-induced apoptosis through SIRT1 restoration. Collectively, Targeting modulation of p53/miR-34a/SIRT1 feedback by miR-34a knockdown or overexpression of SIRT1 could rescue podocyte injury during DN.

6. Expectation

DN appears to be strictly related to some mirnas in its pathophysiological processes. miRNA can be involved in cell differentiation, pyrodeath, oxidative stress and so on[12].As a result, DN appears to be strictly associated with certain miRNAs during its pathophysiological process. DN patients may benefit from therapy targeting DN-associated miRNAs.In addition, miRNA is specifically expressed in both urine and blood, and is easy to obtain and detect, which also has great potential in the early diagnosis of DN, observation of disease progression and prognosis.

References

[1] Fan Y, Lau ESH, Wu H, et al.Incidence of long-term diabetes complications and mortality in youth-onset type 2 diabetes: A systematic review. Diabetes Res Clin Pract. 2022 Sep;191:110030.

[2] Lay AC, Hale LJ, Stowell-Connolly H, et al. IGFBP-1 expression is reduced in human type 2 diabetic glomeruli and modulates β1-integrin/FAK signalling in human podocytes. Diabetologia. 2021 Jul;64(7):1690-1702.

[3] Ichimiya T, Yamakawa T, Hirano T, et al.Autophagy and Autophagy-Related Diseases: A Review. Int J Mol Sci. 2020 Nov 26;21(23):8974.

[4] Zhang Y, Chang B, Zhang J,et al. LncRNA SOX2OT alleviates the high glucose-induced podocytes injury through autophagy induction by the miR-9/SIRT1 axis. Exp Mol Pathol. 2019 Oct;110:104283.

[5] Hong Q, Zhang L, Das B, et al.Increased podocyte Sirtuin-1 function attenuates diabetic kidney injury. Kidney Int. 2018 Jun;93(6):1330-1343.

[6] Platt C, Coward RJ. Peroxisome proliferator activating receptor-γ and the podocyte. Nephrol Dial Transplant. 2017 Mar 1;32(3):423-433.

[7] Li F, Dai B, Ni X. Long non-coding RNA cancer susceptibility candidate 2 (CASC2) alleviates the high glucose-induced injury of CIHP-1 cells via regulating miR-9-5p/PPARγ axis in diabetes nephropathy. Diabetol Metab Syndr. 2020 Aug 6;12:68.

[8] Dey N, Das F, Mariappan MM, et al.MicroRNA-21 orchestrates high glucose-induced signals to TOR complex 1, resulting in renal cell pathology in diabetes. J Biol Chem. 2011 Jul 22;286(29):25586-25603.

[9] Xu L, Fan Q, Wang X, et al.Ursolic acid improves podocyte injury caused by high glucose. Nephrol Dial Transplant. 2017 Aug 1;32(8):1285-1293.

[10] Luo A, Yan H, Liang J, et al. MicroRNA-21 regulates hepatic glucose metabolism by targeting FOXO1. Gene. 2017 Sep 5;627:194-201.

[11] Liang Y, Liu H, Zhu J,et al.Inhibition of p53/miR-34a/SIRT1 axis ameliorates podocyte injury in diabetic nephropathy. Biochem Biophys Res Commun. 2021 Jun 25;559:48-55.

[12] Ding X, Jing N, Shen A, et al.MiR-21-5p in macrophage-derived extracellular vesicles affects podocyte pyroptosis in diabetic nephropathy by regulating A20. J Endocrinol Invest. 2021 Jun;44(6):1175-1184.

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