DIABETIC NEPHROPATHY - NEW THERAPIES

Su0022 MICRORNA-21 AS THERAPEUTIC TARGET IN DIABETIC NEPHROPATHY

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Introduction and Aims: MicroRNAs (miRs) are important mediators of diabetic nephropathy (DN). In addition, PKC-β loss is associated with amelioration of long-term complications of DN, including mesangial expansion and tubulointerstitial fibrosis. We aimed to identify PKC-β related microRNAs in the induction of DN and the potential of their therapeutic modulation to prevent diabetic nephropathy.

Methods: DN was induced by streptozotocin in PKC-β knock out- and wildtype mice. Microarray analysis revealed several deregulated miRs. Mesangial cells (MC) and renal fibroblasts were subjected to high glucose (HG) and TGF-β treatment. PKC-β was activated by PMA. Ruboxistaurin inhibited PKC-β. The synthetic retinoid 11302 inhibited AP-1 and was analyzed by Luciferase Assay. Real time PCR and Western Blotting were performed. Electrophoretic mobility shift assay revealed transcriptional activation. Proliferation was examined by BrdU Cell Proliferation Assay. Elevated miR-21 was silenced in vivo in streptozotocin-induced diabetic mice by locked-nucleic acid (LNA) treatment targeting miR-21.

Results: Several miRs, including miR-21, were upregulated in diabetic mice. In mesangial cells and renal fibroblasts, HG, TGF-β and PMA increased miR-21 and fibrotic gene expressions (all p<0.05). Ruboxistaurin normalized miR-21 levels (p=0.001) and CTGF (p=0.05) in MC, confirming PKC-β as key factor. AP-1 was activated by TGF-β. Inhibition of AP-1 rescued miR-21-, CTGF- and Colla2 upregulation (all p<0.0001) after TGF-β-stimulation in MC, indicating AP-1 as a key transcriptional activator. TGF-β induced phosphorylation of AKT, ERK and GSK-3β in MC. Overexpression of miR-21 in MC resulted in upregulated inflammatory genes, including Il-6 and MCP-1. Furthermore, overexpression of miR-21 increased the proliferation of renal fibroblasts. MiR-21 was successfully silenced in vivo by LNA-21 treatment.

Conclusions: This study elucidates a pathway including PKC-β and miRs. Interfering with this pathway results in less glomerular and interstitial injury. Thus, miRs may serve as novel therapeutic targets to ameliorate diabetic nephropathy.