SirT7-Mediated Transcription of fascin in Hyperglycemic Glomerular Endothelial Cells Contributes to EndMT in Diabetic Nephropathy

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<td>Complete List of Authors:</td>
<td>Wu, Mengchen; Shanghai General Hospital Hao, Yingxiang; Shanghai General Hospital Wu, Xinwan; Shanghai General Hospital zhu, minmin; Shanghai Cancer Hospital, chen, xiangyuan; Shanghai General Hospital Qi, Jie Yu, Zhuang; Shanghai General Hospital Xu, Hongjiao; Shanghai General Hospital</td>
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<td>Diabetic nephropathy, EndMT, SirT7</td>
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Original Article

SirT7-mediated transcription of fascin in hyperglycemic glomerular endothelial cells contributes to EndMT in diabetic nephropathy

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Abstract

Diabetic nephropathy (DN) is the main cause of end-stage renal disease worldwide. It
was reported that the endothelial-to-mesenchymal transition (EndMT) in glomerular endothelial cells plays an important role in DN. As a specific form of epithelial-to-mesenchymal transition, EndMT may involve common regulators of epithelial-to-mesenchymal transition. Fascin has been shown to mediate epithelial-to-mesenchymal transition. In addition, SirT7 has been confirmed to contribute to inflammation in hyperglycemic endothelial cells via the modulation of gene transcription. In this study, we speculate that SirT7 modulates fascin transcription and is thus involved in EndMT in hyperglycemic glomerular endothelial cells. Our data indicate that α-smooth muscle actin (α-SMA) and fascin levels are increased, while CD31 levels are decreased in the kidneys of DN rats. Consistently, our cellular experiments reveal that high glucose treatment elevates fascin levels and induces EndMT in human glomerular endothelial cells (HGECs). Moreover, silencing fascin inhibits EndMT in hyperglycaemic HGECs. Next, SirT7 is found to be decreased in hyperglycemic cells and in the kidneys of DN mice. Moreover, the inhibition of SirT7 increases fascin levels and mediates EndMT. An increase in SirtT7 expression decreases fascin expression, inhibits EndMT and improves renal function in hyperglycemic cells and DN mice. In addition, SirT7 is bound to the promoter region of fascin. In summary, the present study indicates that SirT7 transcribes fascin to contribute to hyperglycemia-induced EndMT in DN patients.

**Keywords:** endothelial-to-mesenchymal transition, EndMT; diabetic nephropathy, DN; glomerular endothelial cells, GECs
Introduction

Diabetic nephropathy (DN), which is the main cause of end-stage renal disease, is the most serious complication of diabetes [1,2]. Once the disease progresses to end-stage renal disease, the mortality increases and the cost of treatment increases [3,4]. Moreover, current treatments can delay the progression of DN, and effective treatment approaches are limited. Therefore, studies to explore the potential mechanisms of DN are urgently needed.

DN is characterized by impaired glomerular filtration capacity. The glomerular filtration barrier is constructed by human glomerular endothelial cells (HGECs), the glomerular basement membrane and podocytes. Damage to any part of the glomerular filtration barrier enhances glomerular permeability and leads to proteinuria [5]. Glomerular endothelial-to-mesenchymal transition (EndMT) was shown to play an important role in DN [6]. EndMT is defined as a reduction in the endothelial phenotype and an increase of mesenchymal phenotype [7]. EndMT in HGECs is considered the initial process of HGEC injury and is the origin of collagen-generating myofibroblasts contributing to fibrosis in DN [8,9]. In addition, blocking EndMT relieves fibrosis and improves renal dysfunction in DN [10].

As a specific form of epithelial-to-mesenchymal transition, EndMT may involve common regulators of epithelial-to-mesenchymal transition [11]. Fascin was reported to play a crucial role in epithelial-to-mesenchymal transition [12] and to play a key role in renal fibrosis [13]. However, whether fascin participates in EndMT in DN is still unknown.

Epigenetic modifications play important roles in DN [14], and histone modifications play the most important role in DN [15]. Histone modification performs physiological functions by regulating downstream gene transcription. Our previous studies have indicated that histone methylation participates in the occurrence and progression of DN via the modulation of alpha-enolase, perforin-2, protein tyrosine phosphatase 1B and phosphatase and tensin homologous transcription [16–19]. Moreover, SirT7-mediated histone acetylation is involved in hyperglycemia-mediated endothelial inflammation via modulation of death-associated protein kinase 3.
transcription [20]. However, whether SirT7 also participates in EndMT in DN is still not well known.

In the present study, we explored the underlying mechanism by which SirT7 participates in EndMT in DN. Our results indicated that SirT7 participates in EndMT in DN via modulation of fascin transcription.

Materials and Methods

Rat model

The present study complied with the Guidelines for the Care and Use of Laboratory Animals issued by the Committee on the Management and Use of Laboratory Animals of Fudan University Shanghai Cancer Center (licence number: FUSCC-IACUC-S20210456). Male Sprague Dawley rats weighing 300‒400 g were used in the present study. The rats were raised under a 12/12-h light/dark cycle and in a temperature-controlled environment (22°C–25°C). The animals in the present study underwent unilateral nephrectomy under anaesthesia (isoflurane 3%-4% induction and 1.5%-2.5% maintenance). After unilateral nephrectomy, the rats were raised for 9 weeks. The animals that received a single intraperitoneal injection of citrate buffer (0.1 M, pH 4.5) three weeks after unilateral nephrectomy were defined as the control group (Con). Animals that received a high-sugar and high-fat diet after unilateral nephrectomy and an intraperitoneal injection of streptozotocin (STZ, 50 mg/kg) three weeks after unilateral nephrectomy were defined as the DN group. To determine the therapeutic effect of Sirt7 against DN, control vector- or AAV-SirT7-treated animals were injected into the contralateral kidney at the time of unilateral nephrectomy.

Immunohistochemistry (IHC)
In this study, rat kidney tissue samples were paraffin-embedded, and IHC was subsequently performed. The paraffin sections were incubated with primary antibodies at 4°C overnight, and the antibodies used in the present study are shown in Table 1. After incubating with secondary antibody at room temperature for 1.5 h, the paraffin sections were stained with a DAB Detection kit (GeneTech, Shanghai, China). Finally, haematoxylin was used for counterstaining in the present study.

Cell culture and treatment

HGECs were obtained from Procell (Wuhan, China) and incubated in DMEM supplemented with 10% fetal bovine serum (FBS) and 1% penicillin–streptomycin solution (PS) at 37°C in a 5% CO\textsubscript{2} atmosphere. HGECs were cultured in 25 mM glucose (high glucose) DMEM for 3 days to establish a cell model of DN. Mannitol was added to normal medium (5 mM) DMEM to achieve the same osmotic pressure as high glucose medium was used to exclude the effect of osmotic pressure.

shRNA and plasmid treatments

After the HGECs were inoculated to reach 70%–80% confluence, they were transfected with the SirT7 overexpression plasmid (Sirt7-OE), SirT7 shRNA or fascin1 shRNA via Lipofectamine 2000 (Invitrogen, Carlsbad, USA). The percentage of plasmid and shRNA via Lipofectamine 2000 reagent was 1 mg/1.2 mL. Table 2 shows the shRNA sequences used in this study.
qPCR analysis

Total RNA was extracted with the use of an EZ-press RNA Purification kit (EZBioscience, Roswell, USA). Hifair® II 1st Strand cDNA Synthesis SuperMix (Yeasen, Shanghai, China) was used to synthesize cDNA for qPCR. Then, we performed qPCR with Hieff UNICON® qPCR TaqMan Probe Master Mix (Yeasen) on an ABI7500 Real-Time PCR system (Applied Biosystems, Foster City, USA). Table 3 shows the sequences of the qPCR primers used in this study.

Hematoxylin and eosin (HE) staining

The paraffin sections were placed in an oven at 60°C for 1–2 h and dewaxed with xylene (National Pharmaceutical Group, Shanghai, China) and ethanol. Hematoxylin (Sigma-Aldrich, Saint Louis, USA) was used to stain the nucleus for about 10 min and eosin (Sigma-Aldrich) was used to stain the cytoplasm for 30 s. Finally, the sections were sealed with neutral balsam (National Pharmaceutical Group), dried at room temperature and observed under a direct optical microscope (Nikon, Tokyo, Japan).

Masson trichrome staining

After paraffin sections are dewaxed in the above way, weigert iron hematoxylin (1:1 mixture of liquid A and liquid B) (National Pharmaceutical Group) is first stained for 10 min, rinsed with running water, and differentiated by 1% hydrochloric acid alcohol (National Pharmaceutical Group). Then the tissues were stained with acid
fuhong-ponceau solution (National Pharmaceutical Group) for about 8 min. After washing again, phosphomolybdate solution (OKA Biotechnology, Beijing, China) was used to differentiate and dye for 3–5 min. Until the tissues were observed under the microscope with varying degrees of red, they are dyed with aniline blue solution (National Pharmaceutical Group) for 5 min. After the last washing, dehydrated with anhydrous alcohol, transparent with xylene (National Pharmaceutical Group), sealed for microscopic examination.

Western blot analysis

Whole-cell extracts were prepared using cell lysis buffer (Cell Signaling Technology, Danvers, USAA). The protein samples were boiled in loading buffer at 100°C for 10 min. Different groups of HGECs containing the same amount of protein (50 μg) were isolated and transferred to PVDF membranes containing 8%–10% SDS–PAGE. The membrane was blocked with protein-free rapid blocking buffer (Beyotime Biotechnology, Shanghai, China) for 1 h, after which all the membranes were incubated with specific primary antibodies at 4°C overnight. After washing 5 times, the membranes were incubated with secondary antibodies at room temperature for 1 h. The membranes were subsequently washed with PBST 5 additional times. An ECL system (Beyotime Biotechnology) was used to detect the protein signals. The mean densities of the protein bands were analyzed using ImageJ.

Chromatin immunoprecipitation (ChIP) assay

ChIP assays were performed with a Simple ChIP Kit (CHIP Kit ab500; Abcam,
Cambridge, UK) according to the manufacturer’s directions. Briefly, the cells (1×10⁷) were fixed with 1% formaldehyde for 10 min at room temperature to crosslink the DNA and the proteins. The cross-linking reaction was then stopped with the use of 2.5 mM glycine. Chromatin was sheared with the use of ultrasound. After centrifugation, the supernatant was incubated with specific primary antibodies or IgG at 4°C overnight. Agarose beads were applied to immunoprecipitate the proteins. The mixture was incubated at 65°C for 4–5 h to reverse transcribe the DNA and protein. Finally, the DNA was purified by centrifugation and verified by nucleic acid gel electrophoresis. The oligonucleotide sequences of primers used for Fascin1 are listed in Table 4.

Statistical analysis

In this study, sample sizes of 8 in vivo and 5 in vitro were ascertained by the mRNA levels of fascin modulated by hyperglycemia or high glucose treatment in our pilot in vivo or in vitro experiments. The data are shown as the mean ± standard deviation. The comparison of the means of two groups was conducted by two-tailed unpaired t tests. This study employed one-way ANOVA followed by Bonferroni-corrected pairwise comparisons to compare the means of more than 2 groups. P<0.05 was considered to indicate statistical significance.

Results

Occurrence of EndMT and augmentation of fascin levels in vivo

The characteristics of the rats in this study are shown in Table 5. Hematoxylin and
eosin (HE) staining and Masson trichrome staining revealed renal damage and interstitial fibrosis in the glomeruli of DN rats (Figure 1A). Moreover, IHC staining of renal biopsy specimens from DN rats indicated that the expression of α-smooth muscle actin (α-SMA) was increased, while the expression of CD31 was decreased (Figure 1A). Fascin has been reported to play an important role in renal fibrosis [13]; therefore, we examined the levels of fascin in the renal biopsy specimens of DN animals. IHC staining revealed that fascin expression was increased in the kidneys of DN animals (Figure 1A). Consistently, western blot and qPCR assays indicated that the levels of α-SMA and fascin were increased, while the level of CD31 was decreased in DN rats (Figure 1B–E). Our results demonstrated that fascin may regulate EndMT in DN.

**High glucose induced EndMT in hyperglycemic HGECs via upregulation of fascin levels**

To further determine whether fascin participates in EndMT in DN, we constructed a cell model in this study with the use of HGECs. Our data indicated that high glucose treatment increased α-SMA and fascin expression but decreased CD31 expression at both the protein (Figure 2A–D) and mRNA levels (Figure 2E–G). These data were quite similar to those obtained for DN rats. Next, we silenced fascin expression in hyperglycaemic HGECs, and the effect of sh-fascin was confirmed via western blot (Figure 3A) and qPCR (Figure 3B) analysis. Our results indicated that the inhibition of fascin expression increased CD31 expression but decreased α-SMA levels in hyperglycemic HGECs (Figure 3). Our data revealed high glucose-induced EndMT via an increase in fascin levels in HGECs.

**SirT7 expression was reduced in DN animals and hyperglycemic HGECs**

Histone modification reportedly plays an important role in DN [14,15]. Our previous studies demonstrated that histone methylation participates in the occurrence and progression of DN [16–19]. Moreover, SirT7-mediated histone acetylation participates in hyperglycemia-mediated endothelial inflammation [20]. However,
whether SirT7 participates in EndMT in DN is still unknown. The present study demonstrated that high glucose treatment decreased SirT7 protein (Figure 4A,B) and mRNA (Figure 4C) levels in HGECs. Consistently, SirT7 expression was also inhibited in the kidneys of DN animals (Figure 4D–F). This study indicated that the level of SirT7 decreased in DN rats and hyperglycemic HGECs and may participate in EndMT in DN patients.

**SirT7 participated in EndMT in hyperglycaemic HGECs via modulation of fascin transcription**

To determine whether SirT7 modulates EndMT via modulation of fascin transcription, both loss-of-function and gain-of-function approaches were used in this study. Our data indicated that SirT7 overexpression decreased α-SMA and fascin levels but increased CD31 expression at the protein (Figure 5A–E) and mRNA (Figure 5F–I) levels. Moreover, a ChIP assay revealed that SirT7 bound to the promoter of fascin (Figure 5J). Furthermore, SirT7 silencing decreased CD31 expression and increased fascin and α-SMA protein (Figure 6A–E) and mRNA (Figure 6F–I) levels. These data demonstrated that SirT7 participated in EndMT in hyperglycaemic HGECs via modulation of fascin transcription.

**Sirt7 overexpression inhibited EndMT and improved renal dysfunction in DN animals.**

To determine the inhibitory effect of sirt7 overexpression on EndMT in vivo, we used AAV-SirT7 in this study. The effectiveness of AAV-SirT7 is shown in Figure 7A–C. Our results demonstrated that Sirt7 upregulation reduced renal injury and fibrosis (Figure 7A). Moreover, an IHC assay revealed that SirT7 overexpression reduced fascin (Figure 7C) and α-SMA levels but increased CD31 expression in the kidneys of DN rats (Figure 7A). Consistently, Western blotting and qPCR assays indicated that an increase in SirT7 expression decreased fascin expression and inhibited EndMT in DN rats (Figure 7B–F). In addition, SirT7 upregulation improved renal dysfunction in DN animals (Table 5). These results suggested that SirT7 augments the inhibition of
EndMT in DN rats, thus improving renal dysfunction. A diagram of the mechanism of action of the present study is shown in Figure 8.

**Discussion**

The core findings of the present study were that hyperglycemia was involved in the modulation of EndMT via an increase in fascin expression, thus contributing to the occurrence and progression of DN. Moreover, SirT7 was decreased in hyperglycemic HGECs and in the kidneys of DN rats. Mechanistic studies indicated that SirT7 regulated fascin transcription to induce EndMT in hyperglycemic HGECs.

The epithelial-to-mesenchymal transition, which involves intricate cell phenotypic reconstruction, plays an important role in tissue and organ damage [21]. Previous studies have demonstrated that epithelial-to-mesenchymal transition (EMT) of renal epithelial cells plays an important role in kidney fibrosis [22]. Recently, glomerular EndMT was indicated to be involved in DN [5,6]. The present study revealed that CD31 expression was reduced and that α-SMA expression was augmented in the kidneys of DN rats and hyperglycemic HGECs. The present study was quite similar to a recent study that indicated that EndMT plays a crucial role in DN [23]. It was deduced that epithelial-to-mesenchymal transition and the EndMT may be associated with other factors [11]. Fascin acts as an actin-binding protein that is enriched in the actin bundles of spikes and filopodia [24,25]. Moreover, fascin is involved in filopodia construction to increase cell migration [26]. In addition, fascin promotes epithelial-to-mesenchymal transition [27,28]. In the present study, fascin was reported to be augmented in the kidneys of DN rats and hyperglycemic HGECs. Additionally, fascin silencing enhanced CD31 levels and reduced α-SMA levels, thus suppressing EndMT in hyperglycemic HGECs. These data indicated that fascin plays a crucial role in the modulation of EndMT in DN patients.

Epigenetic modifications have been found to play an important role in DN [14], and histone modifications play the most important role in DN [15]. Our previous studies indicated that lysine methyltransferase 5A-mediated histone methylation regulates enolase 1 [16] and perforin-2 [17], thus playing a crucial role in EndMT in DN.
Moreover, SET domain containing lysine methyltransferase 8-mediated histone methylation modulates protein tyrosine phosphatase 1B [18] and phosphatase and tensin homolog [26] transcription to induce endothelial inflammation in diabetes. Furthermore, our study indicated that SirT7-mediated histone acetylation participated in endothelial inflammation via modulation of death-associated protein kinase 3 expression [20]. In the present study, we found that SirT7 overexpression inhibited α-SMA levels but elevated CD31 expression in hyperglycaemic HGECs. Moreover, silencing SirT7 upregulated α-SMA expression and decreased CD31 expression. Furthermore, AAV-SirT7 increased CD31 expression, inhibited α-SMA levels, and improved renal function in DN rats. These data indicated that SirT7 was involved in the regulation of EndMT in DN patients. In addition, SirT7 bound to the promoter of fascin, which indicated that SirT7 modulates EndMT via the regulation of fascin transcription. Our research and that of other scholars indicated that SirT7 participates in endothelial inflammation [20, Podulus apoptosis [29] and EndMT in DN. Therefore, SirT7 plays an important role in DN.

This study has several limitations. First, HGECs were used to construct a cellular model, and other primary endothelial cells were used to confirm the results of the present study. Second, the potential mechanism by which fascin induces EndMT in hyperglycaemic HGECs is still not well known and deserves further research.

In conclusion, this study demonstrated that SirT7 expression decreased, fascin expression increased, and that EndMT occurred in DN rats. In addition, this study indicated that high glucose concentrations induce EndMT via an increase in fascin levels in hyperglycemic HGECs. Moreover, SirT7 was found to negatively regulate fascin transcription to participate in the modulation of EndMT in DN. However, upregulation of SirT7 expression decreased fascin transcription, thus inhibiting EndMT and improving renal function in hyperglycemic HGECs and DN mice. Our study revealed that SirT7 may be an underlying therapeutic target for DN.

**Funding**
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References


epithelial–mesenchymal transition, Int J Oncol 2018, 52:1455-1464


Figure legends

Figure 1. EndMT and fascin levels in Con and DN rats
(A) HE staining, Masson staining, and IHC staining of α-SMA, CD31 and fascin in the kidneys of DN animals. (B) Western blot results showing the expressions of α-SMA, CD31 and fascin in the kidneys of DN animals. (C) qPCR results indicated that the mRNA levels of α-SMA was increased in the kidneys of DN rats. (D) qPCR results indicated that the mRNA level of CD31 increased in the kidneys of DN rats. (E) qPCR results indicating that the mRNA levels of fascin increased in the kidneys of DN rats (*P<0.05 vs Con; **P<0.01 vs Con, ***P<0.001 vs Con, ****P<0.0001 vs Con, n=5).

Figure 2. High glucose conditions upregulated fascin expression and induced EndMT in HGECs
(A) Western blot results showed that high glucose treatment increased α-SMA and fascin levels and decreased CD31 expression. (B) Quantification of the α-SMA band density. (C) Quantification of the CD31 band density. (D) Quantification of fascin band density. (E) qPCR analysis showed that high glucose treatment enhanced α-SMA expression. (F) qPCR analysis indicated that high glucose treatment reduced CD31 levels. (G) qPCR analysis showed that high glucose treatment augmented fascin expression (*P<0.05 vs Con; **P<0.01 vs Con; n=5).

Figure 3. Fascin silencing inhibited EndMT in hyperglycaemic HGECs
(A) Western blot analysis indicated that fascin silencing inhibited α-SMA levels and
increased CD31 levels in hyperglycemic HGECs. (B) Quantification of the α-SMA band density. (C) Quantification of the CD31 band density. (D) Quantification of fascin band density. (E) The effectiveness of the combinations was verified via a qPCR analysis. (F) qPCR analysis showed that fascin silencing decreased α-SMA levels in hyperglycaemic HGECs. (G) qPCR analysis showed that fascin silencing increased CD31 expression in hyperglycaemic HGECs (*<0.05; **P<0.01, ***P<0.001; n=5).

Figure 4. SirT7 levels were reduced in hyperglycaemic HGECs and in the kidneys of DN rats
(A) Western blot results indicated that SirT7 expression was decreased in hyperglycaemic HGECs. (B) Quantification of SirT7 band density. (C) qPCR analysis indicated that SirT7 expression was decreased in hyperglycaemic HGECs. (D) Immunostaining data indicating that SirT7 expression was decreased in the kidneys of DN rats. (E) Western blot results indicating that SirT7 expression was decreased in the kidneys of DN rats. (F) qPCR results indicating that SirT7 expression was decreased in the kidneys of DN rats (*P<0.05, **P<0.01; n=5)

Figure 5. SirT7 overexpression decreased fascin expression and inhibited EndMT in hyperglycaemic HGECs
(A) Western blot results indicated that SirT7 overexpression inhibited fascin and α-SMA levels, and increased CD31 expression in hyperglycaemic HGECs. (B) Quantification of SirT7 band density. (C) Quantification of fascin band density. (D) Quantification of the α-SMA band density. (E) Quantification of the CD31 band density. (F) The effectiveness of SirT7 overexpression was confirmed via qPCR analysis. (G) qPCR analysis indicated that SirT7 overexpression decreased fascin levels in hyperglycaemic HGECs. (H) qPCR analysis was used to determine whether SirT7 overexpression decreased α-SMA levels in hyperglycaemic HGECs. (I) qPCR analysis showed that SirT7 overexpression enhanced CD31 levels in hyperglycaemic HGECs. (J) ChIP assay showed that SirT7 binds to the promoter of
fascin (*P<0.05, **P<0.01, ***P<0.001; n=5)

Figure 6. SirT7 silencing increased fascin expression and induced EndMT in HGECs
(A) Western blot analysis indicated that SirT7 silencing increased fascin and α-SMA levels and decreased CD31 expression in HGECs. (B) Quantification of SirT7 band density. (C) Quantification of fascin band density. (D) Quantification of the α-SMA band density. (E) Quantification of the CD31 band density. (F) The effectiveness of SirT7 silencing was confirmed via qPCR. (G) qPCR analysis indicated that SirT7 silencing increased fascin expression in HGECs. (H) qPCR analysis showed that SirT7 silencing enhanced α-SMA expression in HGECs. (I) qPCR analysis showed that SirT7 silencing reduced CD31 expression in HGECs (*P<0.05, **P<0.01, ***P<0.001; n=5).

Figure 7. Overexpression of Sirt7 inhibited EndMT and improved renal dysfunction in DN rats
(A) HE staining, Masson staining, and IHC results for α-SMA, CD31, fascin and SirT7 expressions in the kidneys of the rats after the corresponding treatment. (B) Western blot results show the expressions of α-SMA, CD31, fascin and SirT7 in the kidneys of the rats after the corresponding treatment. (C) qPCR results showing SirT7 expression in the kidneys of the rats given the corresponding treatment. (D) qPCR results of fascin in the kidneys of the rats with the corresponding treatment. (E) qPCR results showing CD31 expression in the kidneys of the rats given the corresponding treatment. (F) qPCR analysis of α-SMA expression in the kidneys of the rats given the corresponding treatment (*P<0.05, **P<0.01, ***P<0.001; n=5)

Figure 8. Diagram of the potential mechanism of this experiment
The expression of SirT7, which inhibits the transcription of Fascin1, is down-regulated in hyperglycemic HGECs and the kidneys of DN rats. Fascin1 with
increased expression in hyperglycemia further induces EMT.
Table 1. The information of antibodies used in this study

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Table 2. The sequences of shRNAs

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### Table 3. Sequences of primers used for the real-time RT-PCR analysis

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### Table 4. Sequences of primers used for fascin1 promoters

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Table 5. Characteristics of rats in control (Con), diabetic nephropathy (DN), DN with empty vector (SIRT7+AVV), and DN with SirT7 overexpression (DN+AVV-SirT7) groups

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<td>11.92±1.54***</td>
<td>11.7±1.95</td>
<td>6.23±1.65###</td>
</tr>
<tr>
<td>TG (mM)</td>
<td>0.961±0.25</td>
<td>4.36±1.46**</td>
<td>4.14±1.32</td>
<td>3.23±1.15#</td>
</tr>
<tr>
<td>UA (μM)</td>
<td>232.5 (196.8, 256.2)</td>
<td>316.9 (312.3, 365.8)***</td>
<td>319.1 (289.1,386.6)</td>
<td>280.8 (279.7,284.4)</td>
</tr>
<tr>
<td>Scr (μM)</td>
<td>50.39±4.28</td>
<td>52.78±7.17</td>
<td>54.4±10.1</td>
<td>51.0±8.36#</td>
</tr>
<tr>
<td>BUN (mM)</td>
<td>6.88±0.98</td>
<td>9.32±0.70***</td>
<td>10.0±1.29</td>
<td>8.16±1.14###</td>
</tr>
<tr>
<td>TC (mM)</td>
<td>1.30 (1.25, 1.45)</td>
<td>3.10 (2.69, 3.88)***</td>
<td>3.11 (2.73,3.32)</td>
<td>2.12 (1.72,2.46)###</td>
</tr>
<tr>
<td>LDL (mM)</td>
<td>0.485 (0.460, 0.555)</td>
<td>1.195 (0.945,1.320)***</td>
<td>1.06 (0.81,1.15)</td>
<td>0.700 (0.61,0.72)###</td>
</tr>
<tr>
<td>HDL (mM)</td>
<td>0.625±0.58</td>
<td>1.023±0.21**</td>
<td>1.04±0.16</td>
<td>0.79±0.96###</td>
</tr>
</tbody>
</table>

Data were expressed as the mean ± SD and compared using an independent sample t test. Data that were not normally distributed were expressed as median (IQR) and compared using the Mann-Whitney U test.

*Compared to the control (Con) group; #Compared to the DN with SIRT7 empty vector (SIRT7+AVV) group.

*P<0.05, **P<0.01, ***P<0.001; n=8 per group.

FBS, fasting blood sugar; HbA1c, glycosylated hemoglobin; TG, triglyceride; UA, uric acid; Scr, serum creatinine; BUN, blood urea nitrogen; TC, total cholesterol; LDL, low density lipoprotein; HDL, high density lipoprotein.
Endothelial cell hyperglycemia (EndMT) may be a key factor in the development of diabetic complications. High glucose levels lead to the expression of endomembrane markers (CD31, α-SMA) and the transcriptional mediator (Fascin). This process involves SirT7 in the nucleus, which regulates Fascin mRNA expression, leading to the formation of the cytoplasmic complex.
Diabetic nephropathy (DN) has become the foremost cause of end-stage renal disease worldwide. Epigenetics plays an important role in the pathogenesis of DN. The present study indicated that SirT7 down-regulation is involved in the progression of DN by promoting endothelial-to-mesenchymal transition (EndMT).

1 SirT7 is found to mediate EndMT via modulating transcription of fascin.

2 Increased expression of SirT7 down-regulates fascin, inhibits EndMT and alleviates renal function in hyperglycemic endothelial cells and DN mice.

3 SirT7 may be a potential target for the treatment of diabetic nephropathy.