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Supplemental Information

MicroRNA-34a Promotes Renal Fibrosis by Downregulation of Klotho in Tubular Epithelial Cells

Yong Liu, Xianjin Bi, Jiachuan Xiong, Wenhao Han, Tangli Xiao, Xinli Xu, Ke Yang, Chi Liu, Wei Jiang, Ting He, Yanlin Yu, Yan Li, Jingbo Zhang, Bo Zhang, and Jinghong Zhao

Supplemental Figure

Figure S1

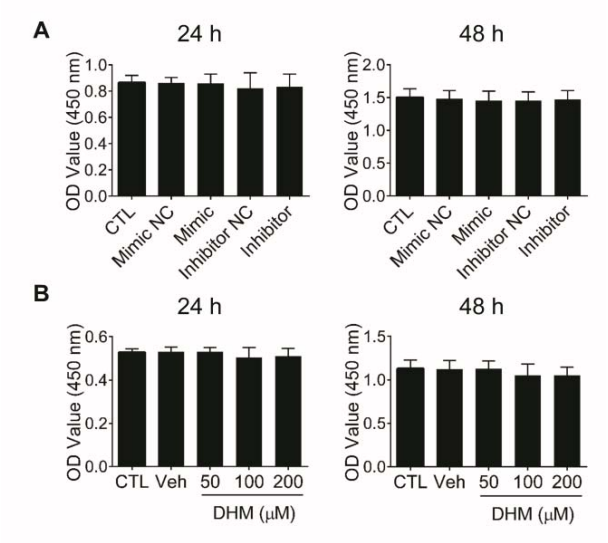

Figure legends

Figure S1. miR-34a or DHM had no obvious effect on the cell viability of HK-2 cells. (A) Cell viability of HK-2 cells transfected with miR-34a mimic (100 nM), a mimic control, inhibitor (100 nM) or an inhibitor control for 24 h or 48 h assayed by CCK-8. (B) Cell viability of HK-2 cells treated with DHM at different concentrations for 24 h or 48 h assayed by CCK-8. These data are expressed as the mean ± SEM of three independent experiments.

Supplemental Methods

Cell viability assay

Cell counting kit-8 (CCK-8, MCE, USA) was used to detect cell viability. 5000 cells per well were seeded into 96-well plates in 100 ml of cell culture medium and transfected with miR-34a mimic, mimic negative control (mimic-nc), inhibitor, or inhibitor negative control (inhibitor-nc) or treated with DHM at different concentrations, and then subjected to a cell viability analysis at 24 h or 48 h. The

medium was mixed with 10 ml of CCK-8 reaction solution for 1 h at 37 ℃. The optical density was measured at 450 nm by a microplate reader (SpectraMax M2; Molecular 145 Devices, Sunnyvale, CA, USA). Each measurement was repeated 3 times.

Supplemental Table

Table 1. Information on the control subjects and CKD patients enrolled in the study.

Clinical Feature	Control (n=5)	Renal fibrosis (n=8)
Age (years)	45.23±3.23	46.38±4.96
Gender (male/female)	3/2	5/3
Serum creatinine (mg/dL) [a]	0.58±0.13	3.06±1.47[b]
BUN (mmol/L) [a]	4.86±1.76	14.03±7.62 [b]
eGFR (ml/min per 1.73 m^2) [a]	142.00±28.26	25.20±14.51 [b]

[a]Mean±SD, [b]$P<0.01$.