Table S1. The primer sequences of targeted genes used for qRT-PCR.

Name	Forward Primer (5'-3')	Reverse Primer (5'-3')
circSAMD4	CCAGCACAAGTACAAGAATCATTA	AGAGTGAGCCAGGATTTTGGG
circSamd4	GCAAGCACGAGAATCATTAACCA	TTCGATAGAGTGTGCCAGGAT
mmu_circRNA_005186	GAGGATGCCTGCCGAGTTGT	CTTTGCCATAGGGGTGCGTG
mmu_circRNA_000706	CGACTAGCAACAGGGAGTGATG	CAGCAATAGCAGGGTTACCAAT
mmu_circRNA_30398	ATACTCAAACCCAAACGAAGG	CATTGTTACTGGTGCCCTCA
mmu_circ_28799	ACAAGGGCAAGAAGAGGCAC	GTGACTTGCGGCCAGGA
mmu_circRNA_28239	GCTCAATGAGGAGGATTACTGG	AAGCGTAGTCTCCACTGGTCAT
SAMD4A	TCGAGGCTTTGGGCAATCC	GAGCTGACGAATCCACTGGT
SRSF3	ATGCATCGTGATTCCTG	CTGCGACGAGGTGGAGG
GAPDH	CTGGGCTACACTGAGCACC	AAGTGGTCGTTGAGGGCAATG
Gapdh	CACTGAGCAAGAGAGGCCCTAT	GCAGCGAACTTTATTGATGGTATT
CIRBP	TTTGGGTTTGTCACCTTTG	CTGCCTGGTCTACTCGGAT
Cirbp	GGACTCAGCTTCGACACCAAC	ATGGCGTCCTTAGCGTCATC
Cirbp$^{fl/fl}$	TTTTGGATTCTGTTCCTTTGCCTC	CTTCAAGTGGGGTTTCTTTCACAC
Ggt1-Cre	CATCACATCAGGCACCCCAGAA	GAACATCTTCAGGTTCTGCGGGA

SAMD4A: sterile alpha motif domain-containing protein 4A; SRSF3: serine-rich splicing factor 3; CIRBP: cold-inducible RNA-binding protein

Table S2. The siRNA sequences of targeted genes used in this study.

Name	Sequences
si-NC	UUCUCCGAACGUGUCACGUdTdT
si-*circSAMD4*	AGCACAAGUACAAGAAUCAUUdTdT
si-*CIRBP* 1	CUUCUCAAAGUACGGACAGAU
si-*CIRBP* 2	GCCAUGAAUGGGAAGUCUGUA
si-*CIRBP* 3	CCUACAGAGACAGUUAUGATT
si-*SRSF3* 1	CCUGUCCAUUGGACUGUAATT
si-*SRSF3* 2	UGGAACUGUCGAAUGGUGAAA
si-*SRSF3* 3	CCCUCGAGAUGAUUAUCGUTT
si-*HNRNPM* 1	CUGUGCAAGCUAUAUCUAUGU
si-*HNRNPM* 2	ACAAGCAUAGUCUGAGCGGAA
si-*RBM3* 1	AGUGGCAGGUAUUAUGACAGU
si-*RBM3* 2	GGACGUUCCAGAGACUAUATT

CIRBP: cold-inducible RNA-binding protein; SRSF3: serine-rich splicing factor 3; HNRNPM: heterogeneous nuclear ribonucleoprotein M; RBM3: RNA binding motif protein 3

Table S3. Clinical characteristics in the normal control subjects.

Number	Age (year)	Sex	SCr (μmol/L)	BUN (mmol/L)
1	56	Female	59.4	3.52
2	50	Female	55.9	3.11
3	71	Male	123.3	5.9
4	56	Male	81	3.64
5	60	Female	61.5	4.41

SCr: Serum creatinine; BUN: blood urea nitrogen

Table S4. Clinical characteristics in the subjects with ATI.

Number	Age (year)	Sex	SCr (μmol/L)	BUN (mmol/L)	Diagnosis
1	54	Male	321.1	14.4	Nephrotic syndrome; ATI
2	37	Male	312.1	24.16	Nephrotic syndrome; ATI
3	42	Male	225.7	15.44	Nephrotic syndrome; ATI
4	52	Male	215	15.5	Nephrotic syndrome; ATI
5	67	Male	213.7	14.54	Nephrotic syndrome; ATI
6	32	Female	206	7.3	Henoch-Schönlein nephritis; ATI
7	19	Male	202.6	9.11	Nephrotic syndrome; ATI
8	17	Male	162.5	12.78	Nephrotic syndrome; ATI

SCr: Serum creatinine; BUN: blood urea nitrogen; ATI: acute tubular injury

Table S5. Top six circRNAs ranked by sequence conservation between human and mouse species.

Mmu_circ_RNA			Homologous hsa_circ_RNA			Conservation [a]		
Seqname	Length (nt)	Gene symbol	Seqname	Length (nt)	Gene symbol	Query Cover	Identity	circBank database
mmu_circRNA_005305	519	*Samd4*	*has_circ_0004846*	519	*SAMD4A*	99%	93.00%	conserved
mmu_circRNA_28799	509	*Myh9*	*has_circ_0004470*	509	*MYH9*	99%	92.11%	conserved
mmu_circRNA_28239	290	*Npr3*	*has_circ_0072107*	290	*NPR3*	100%	91.72%	conserved
mmu_circRNA_000706	420	*Wdr1*	*has_circ_0003550*	420	*WDR1*	99%	88.54%	conserved
mmu_circRNA_005186	744	*Epha2*	*has_circ_0010132*	738	*EPHA2*	98%	86.84%	conserved
mmu_circRNA_30398	529	*Fkbp5*	*has_circ_0001599*	527	*FKBP5*	99%	85.98%	conserved

a Conservation metrics (query coverage and identity) were determined using NCBI BLAST (https://blast.ncbi.nlm.nih.gov/Blast.cgi). Higher identity scores indicate greater homology with human circRNAs.

Table S6. Predicted interaction between SRSF3 protein and *circSAMD4* using the CATRAPID website.

Gene Symbol	RNA ID	Interaction Propensity	RNA Binding Protein Propensity	RNA-Binding Domains	RNA-Binding Motifs	RNA-Binding Motifs_IDs
SRSF3	*has_circ_0004846* (*circSAMD4*)	11.72	1	1	1	P84103|SRSF3|UGGAC|26876937|PAR-CLIP

Supplemental Figures

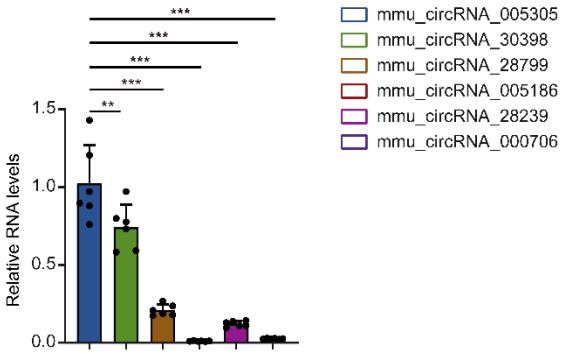

Figure S1. Relative circRNA levels in renal cortex tissues by qRT-PCR. All quantitative data are presented as mean ± SD. Statistical analysis was performed using one-way ANOVA with Tukey's multiple comparisons test. $^{**}P < 0.01$, $^{***}P < 0.001$.

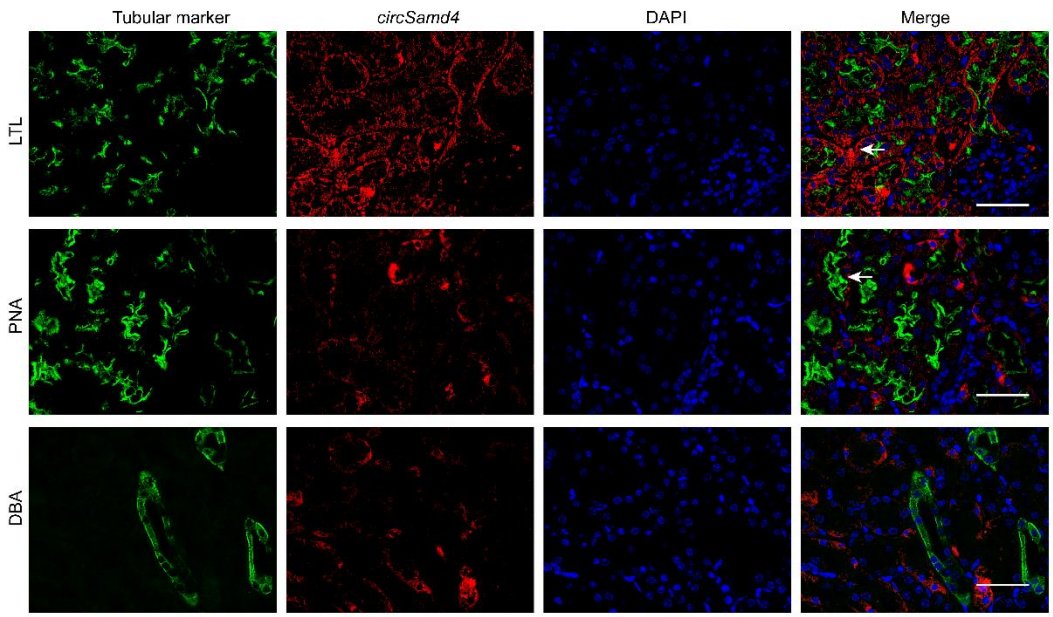

Figure S2. Co-localization staining of *circSamd4* and various segment-specific tubular markers in the kidneys of the CI-AKI model. *CircSamd4* (red) and various segment-specific tubular markers (green), including lotus tetragonolobus lectin (LTL), peanut agglutinin (PNA), and dolichos biflorus agglutinin (DBA), were detected by RNA FISH-immunofluorescence. Scale bar, 50 μm.

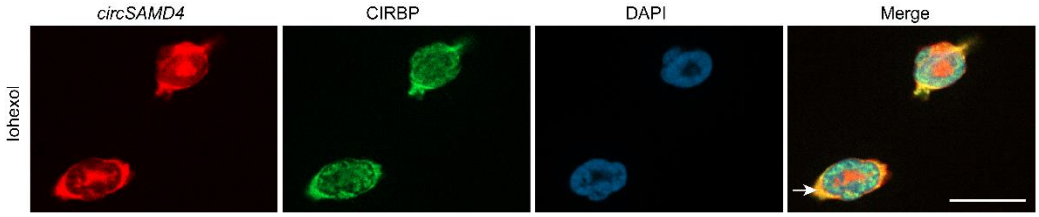

Figure S3. Co-localization staining of *circSAMD4* and CIRBP in iohexol-induced HK-2 cells by RNA FISH-immunofluorescence. Arrows indicate positive co-localization area. Scale bar, 20 μm.

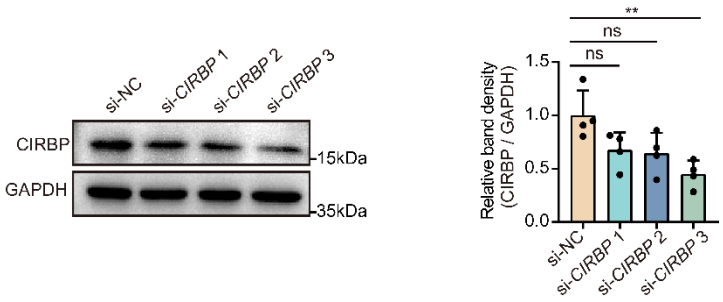

Figure S4. Immunoblot and densitometric analyses of CIRBP expression in HK-2 cells transfected with three different *CIRBP*-targeting siRNAs. All quantitative data are presented as mean ± SD. Statistical analysis was performed using one-way ANOVA with Tukey's multiple comparisons test (A). $^{**}P < 0.01$, ns, not significant.

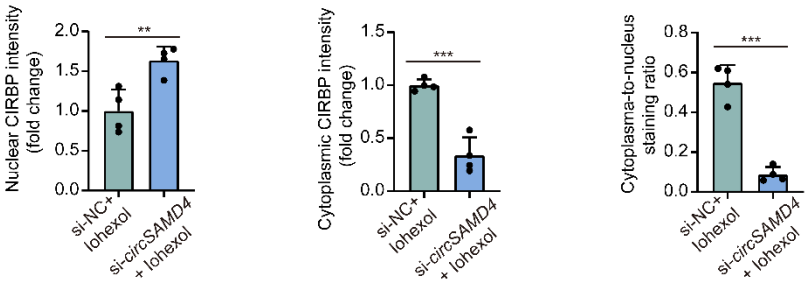

Figure S5. Quantification of nuclear CIRBP, cytoplasmic CIRBP, and cytoplasmic-to-nuclear CIRBP ratio, respectively (n = 4). All quantitative data are presented as mean ± SD. Statistical analysis was performed using unpaired two-tailed Student's t-test. $^{**}P < 0.01$, $^{***}P < 0.001$.

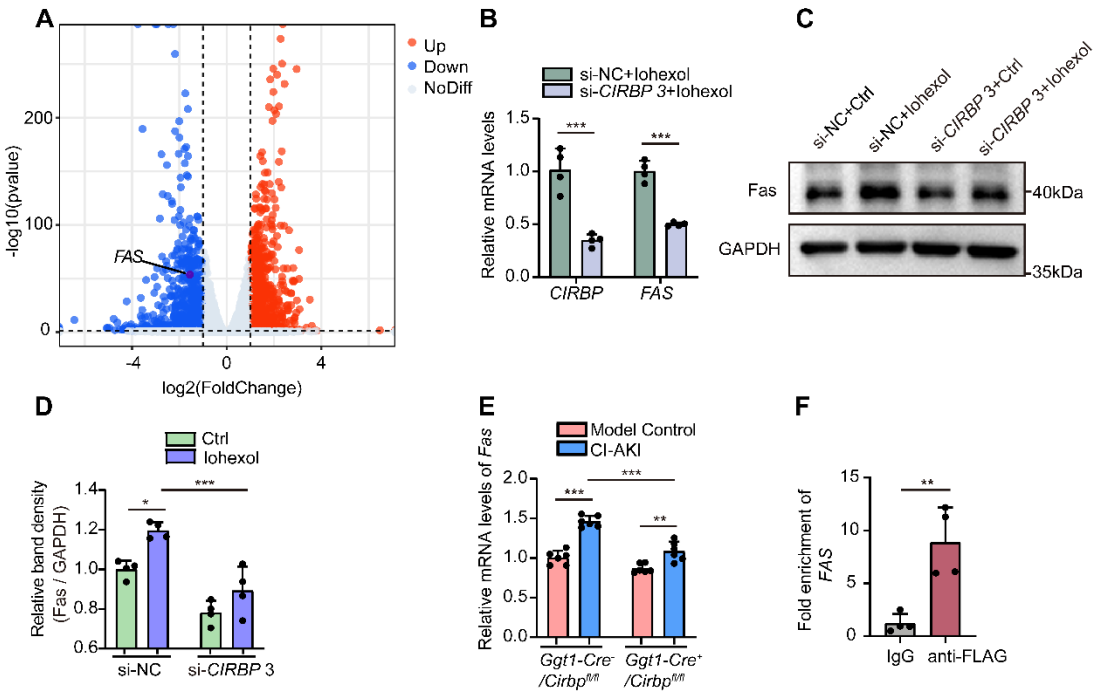

Figure S6. CIRBP suppression attenuated apoptosis through downregulating *FAS* mRNA levels in iohexol-induced HK-2 cells.

(A) Volcano plot of transcriptomics in iohexol-induced HK-2 cells transfected with *CIRBP* siRNA 3 or control siRNA. (B) qRT-PCR analysis of *CIRBP* and *FAS* mRNA levels in HK-2 cells transfected with *CIRBP* siRNA 3 or control siRNA for 48 hours, followed by iohexol treatment (200 mg iodine/mL, 6 hours) (n = 4). (C and D) Immunoblot and densitometric analyses of Fas in HK-2 cells. Cells were transfected with *CIRBP* siRNA 3 or control siRNA for 48 hours, followed by 6-hour treatment with iohexol (200 mg iodine/mL) (n = 4). (E) Relative *Fas* mRNA levels in kidney cortex from *Ggt1-Cre*$^{+}$*/Cirbp*$^{fl/fl}$ and *Ggt1-Cre*$^{-}$*/Cirbp*$^{fl/fl}$ mice with CI-AKI (n = 6). (F) HK-2 cells were transfected with CIRBP-FLAG plasmids for 48 hours, followed by 6-hour treatment with iohexol (200 mg iodine/mL). The RIP assay was performed using anti-FLAG antibody followed by qRT-PCR to detect the enrichment of *FAS*. Anti-IgG antibody was served as a control (n = 4). All quantitative data are

presented as mean ± SD. Statistical analysis was performed using unpaired two-tailed Student's t-test (B and F), or two-way ANOVA with Tukey's multiple comparisons test (D and E). *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

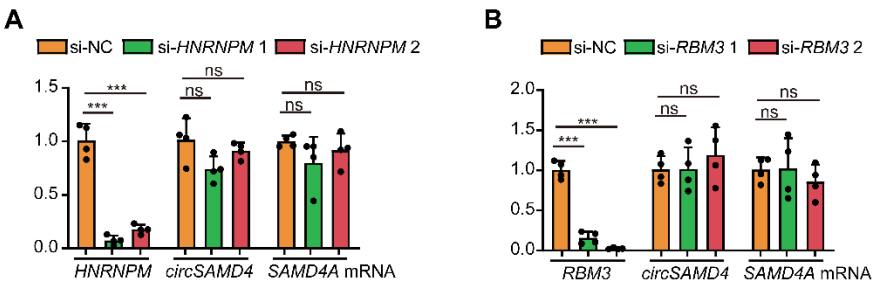

Figure S7. Neither *HNRNPM* nor *RBM3* knockdown affected *circSAMD4* expression

(A) Relative expression of *circSAMD4*, linear *SAMD4A* and *HNRNPM* in HK-2 cells transfected with two different *HNRNPM*-targeting siRNAs. The expression levels of these genes were normalized to *GAPDH* mRNA levels (n = 4). (B) Relative expression of *circSAMD4*, linear *SAMD4A* and *RBM3* in HK-2 cells transfected with two different *RBM3*-targeting siRNAs. The expression levels of these genes were normalized to *GAPDH* mRNA levels (n = 4). All quantitative data are presented as mean ± SD (A-B). Statistical analysis was performed using one-way ANOVA with Tukey's multiple comparisons test (A-B). ***$P < 0.001$, ns, not significant.

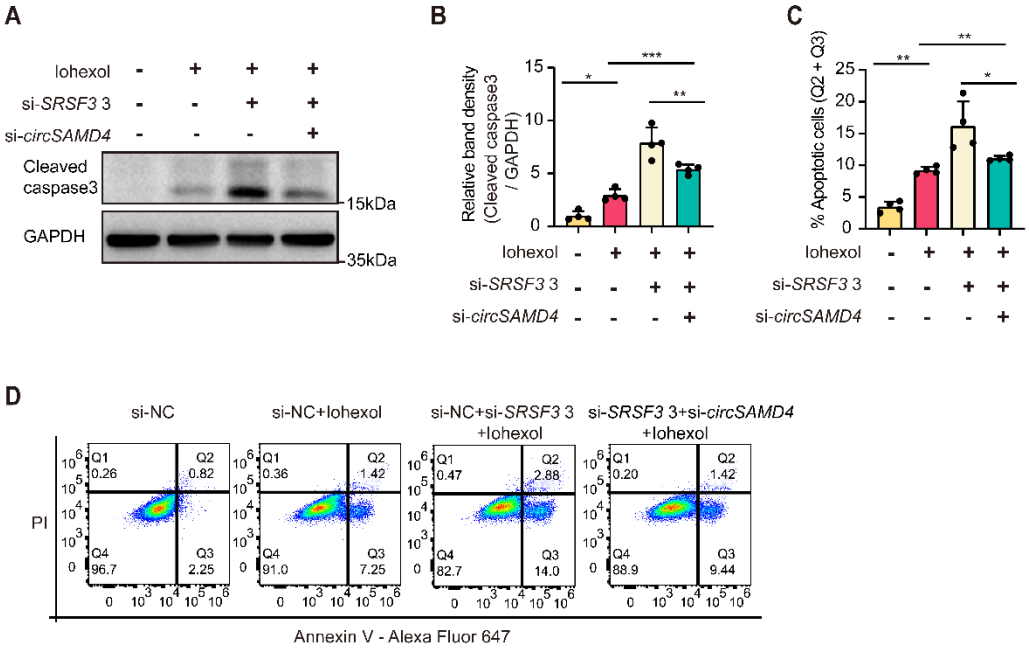

Figure S8. *CircSAMD4* knockdown abrogated the pro-apoptotic effects by SRSF3 suppression in iohexol-treated RTECs *in vitro*

Cells were co-transfected with *SRSF3* siRNA 3 (or control siRNA) and *circSAMD4* siRNA (or control siRNA) for 48 hours, followed by 6-hour treatment with iohexol (200 mg iodine/mL) (n = 4). (A and B) Immunoblot and densitometric analyses of cleaved caspase-3 expression in HK-2 cells (n = 4). (C and D) Flow cytometric analysis of apoptosis in HK-2 cells (n = 4). All quantitative data are presented as mean ± SD (B and D). Statistical analysis was performed using one-way ANOVA with Tukey's multiple comparisons test (B and D). *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

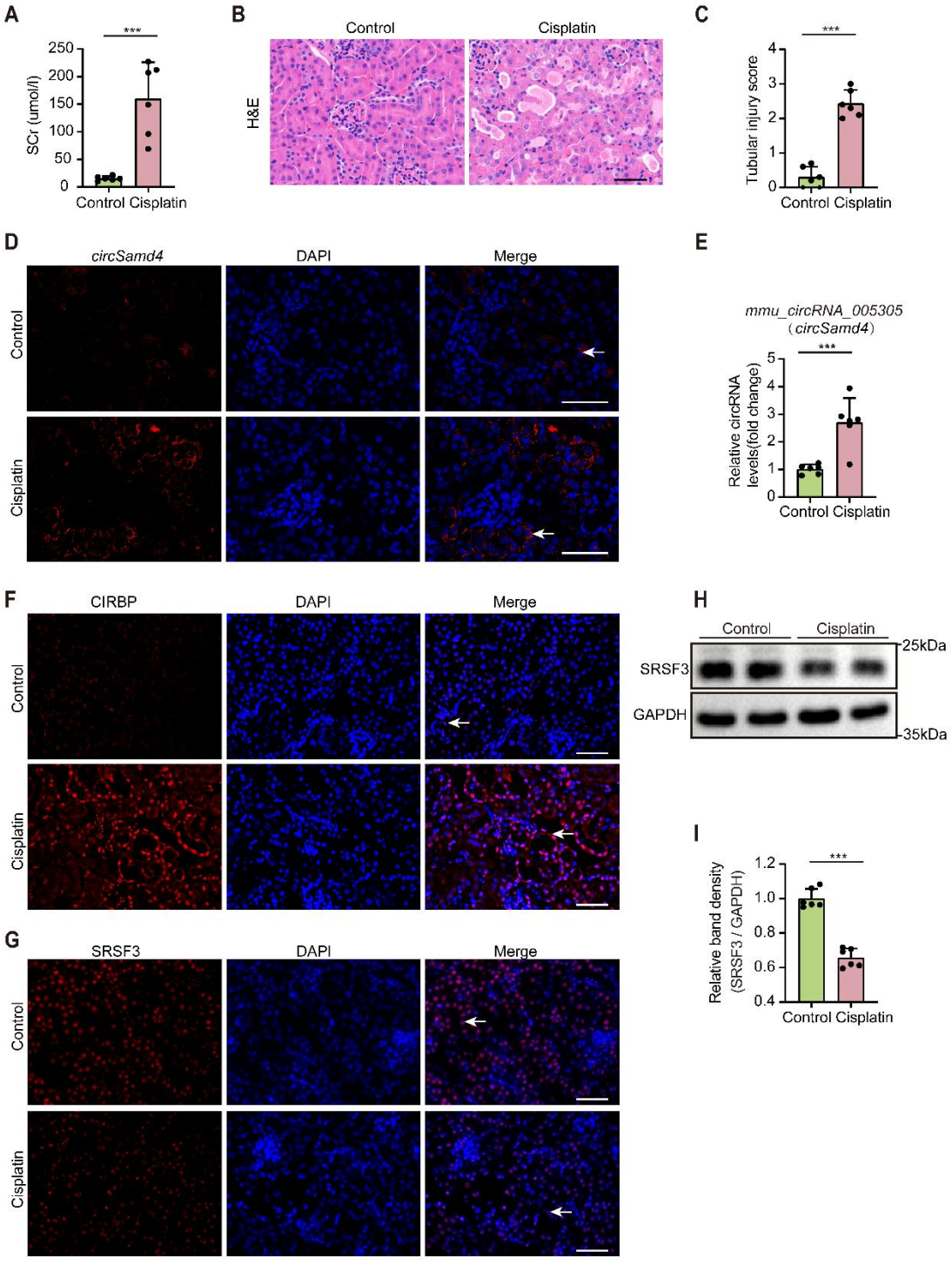
A
***
SCr (umol/l)
250
200
150
100
50
0
Control Cisplatin
B
Control
Cisplatin
H&E
C
***
Tubular injury score
4
3
2
1
0
Control Cisplatin
D
circSamd4
DAPI
Merge
Control
Cisplatin
E
mmu_circRNA_005305
(circSamd4)
***
Relative circRNA
levels(fold change)
5
4
3
2
1
0
Control Cisplatin
F
CIRBP
DAPI
Merge
Control
Cisplatin
H
Control
Cisplatin
SRSF3
GAPDH
25kDa
35kDa
G
SRSF3
DAPI
Merge
Control
Cisplatin
I
***
Relative band density
(SRSF3 / GAPDH)
1.2
1.0
0.8
0.6
0.4
Control Cisplatin

Figure S9. Altered expression of SRSF3, *circSamd4* and CIRBP in RTECs in cisplatin-induced AKI.

Male C57BL/6 mice were injected with one dose of cisplatin (30 mg/kg). Control mice were injected with normal saline. Serum and kidney tissues were collected at 48 hours after cisplatin injection. (A) Serum creatinine (SCr) levels (n = 6). (B and C) Representative images of hematoxylin and eosin (H&E) staining and pathological score in kidney tissues (n = 6). Scale bar: 50 μm. (D) Representative RNA FISH staining images of *circSamd4* in kidney tissues. The white arrows indicated *circSamd4*-positive signals. Scale bar: 50 μm. (E) *CircSamd4* (*mmu_circ_005305*) expression in kidney tissues, measured by qRT-PCR (n = 6). (F) Representative immunofluorescence images of CIRBP in kidney sections from mice. Scale bar: 50μm. The arrows point to nuclear staining of CIRBP in control or nuclear and cytoplasmic staining in RTECs in cisplatin-induced AKI. (G) Representative immunofluorescence images of SRSF3 in kidney sections from mice. Scale bar: 50μm. Arrows point to representative tubule cells with nuclear SRSF3. (H-I) Immunoblot and densitometric analyses of SRSF3 expression in kidney cortex (n = 6). All quantitative data are presented as mean ± SD (A, C, E and I). Statistical analysis was performed using unpaired two-tailed Student's t-test (A, C, E and I). ***$P < 0.001$.

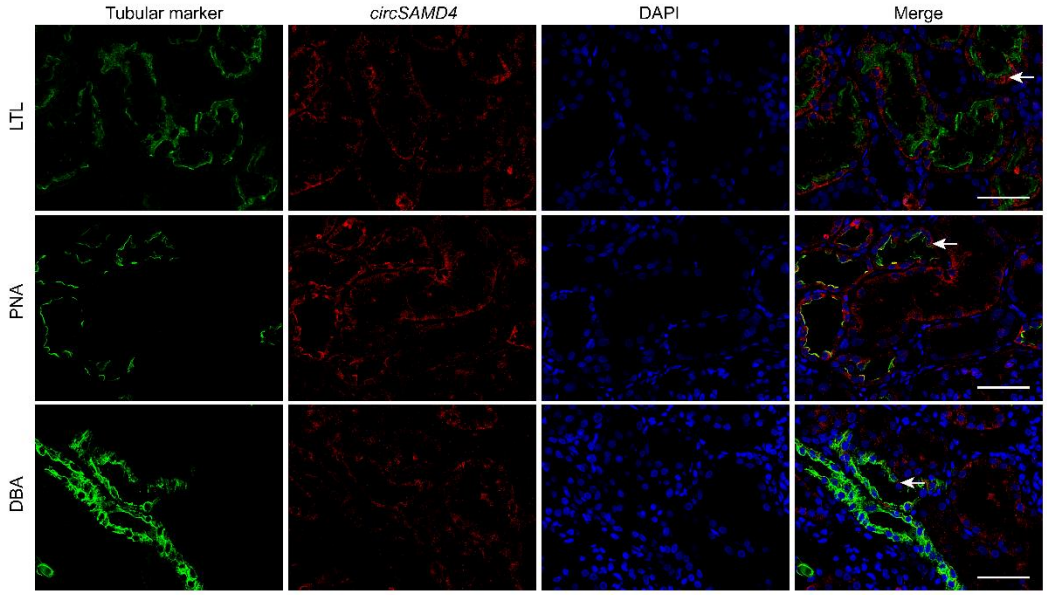

Figure S10. Co-localization staining of *circSAMD4* and various segment-specific tubular markers in renal biopsy specimens from patients pathologically diagnosed with ATI. *CircSAMD4* (red) and various segment-specific tubular markers (green), including lotus tetragonolobus lectin (LTL), peanut agglutinin (PNA), and dolichos biflorus agglutinin (DBA), were detected by RNA FISH-immunofluorescence. Scale bar, 50 μm.

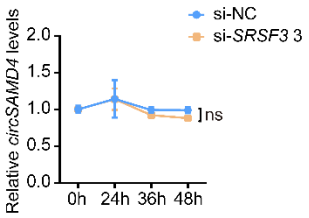

Figure S11. qRT-PCR analysis for the expression of *circSAMD4* after treatment with Actinomycin D (10 μg/mL) at the indicated time points in HK-2 cells (n = 4). All quantitative data are presented as mean ± SD. Statistical analysis was performed using two-way ANOVA with Tukey's multiple comparisons test. ns, not significant.