Supporting Information

Supplementary materials and methods

Patients and samples

Overall, 15 healthy controls, 15 chronic hepatitis B (CHB) patients and 15 CHC patients undergoing liver biopsy from the First Affiliated Hospital of Wenzhou Medical University (FAHWMU) from January 2011 to June 2013 were selected. Serum samples were obtained from patients attending the FAHWMU from January 2011 to December 2015. In total, 110 therapy-naive patients who had undergone liver biopsy for staging and grading of CHB as well as 110 healthy controls (normal liver biochemistry, no history of liver disease, alcohol abuse or viral hepatitis) were enrolled (Table S1). Inclusion criteria were as follows: CHB defined by detectable serum HB antigen and serum HBV DNA for more than six months. Exclusion criteria were as follows: (i) patient ages <16 years, (ii) co-infection with human immunodeficiency virus (HIV), (iii) coexistence of liver injury caused by other etiologies, including hepatitis C virus (HCV) infection, drug intake, alcohol consumption and auto-immune hepatitis, (iv) severe systematic diseases, (v) pregnancy and lactation. Demographic and clinical information was additionally obtained from all patients. Our study was approved by the Ethics Committee of the FAHWMU and informed consent for use of liver or blood samples obtained from all participants. All ethical regulations relevant to human research participants were followed.

Liver histology

A 16-gauge Menghini needle was used to performed liver biopsy. Each liver specimen, at least in 2.0 cm, was advised by physicians in care. Samples were collected for hematoxylin-eosin staining, and experienced hepatopathologists reviewed the results. Additionally, a minimum of

8-10 portal tracts in samples was necessary for patient admission. Fibrosis stage (F0 = no fibrosis – F6 = cirrhosis) was assessed using the Ishak scoring system.

Blood sampling

At the time of liver biopsy, blood samples of each patient were collected. Samples were centrifuged at 3400 g for 7 min at room temperature and at 12,000 *g* for 10 min at 4°C to remove the remaining cells. They were then stored at -80°C for further processing.

Virology

Serum HBV DNA was detected using the Artus HBV QS-RGQ Kit (Qiagen) with a lower detection limit of 10.2 IU/ml. Roche Modular E170 Immunoassay Analyzer (Roche) was uesd to quantified HBsAg, HBeAg and antibodies against HBsAg, HBeAg and hepatitis B core antigen.

Supplementary Tab.1: Characteristics of CHB patients and healthy subjects

		CHB patients (n=110)	healthy subjects (n=110)
Age (years)	Mean±SD	41.5±9.3	42.2±10.1
Sex (n)	Male	63(57.2%)	61 (55.4%)
	Female	47 (42.8%)	49 (44.6%)
ALT (U/L)	Mean±SD	115.3±96.1	
Liver HBV DNA (IU/cells)	Mean±SD	2.569±4.766	
Serum cVIM	Mean±SD	1.869±0.430	
HBeAg	+	53 (48.1%)	
	-	57 (51.9%)	
Fibrosis (n)	F0	7 (6.4%)	
	F1	18 (16.4%)	
	F2	15 (13.6%)	
	F3	16 (14.5%)	
	F4	13 (11.8%)	
	F5	27 (24.5%)	
	F6	14 (12.7%)	

Supplementary Tab.2: Primers used in this study.

Primer Name	Sequence (5'-3')
Mouse	
cVIM-F	ACGGTTGAGACCAGAGATGG
cVIM-R	AGTGAGGTCAGGCTTGGAAA
cANRIL-F	CCTCCTCATGTGGAATCACC
cANRIL-R	TTCAAGGGTCAGCCTCATCT
TGFBR1-F	AAAGCAGTCAGCTGGCCTTG
TGFBR1-R	AGGTGGTGCCCTCTGAAATG
TGFBR2-F	TGGCTTCGAACACCATGGAA
TGFBR2-R	TAGAGGGCGGTGAACAACAG
NOX4-F	TGGCCAACGAAGGGGTTAAA
NOX4-R	TCGCCCAACATTTGGTGAATG
KLF6-F	TGTGGGGTCAAATACAGGGGAA
KLF6-R	AAGGACTTTTCACCCGTTCGTTCA
TGFβ1-F	ACTGCAAGTCAGAGACGTGG
TGFβ1-R	GGAATAGGGGCGTCTGAGGA
SRF-F	AGTTGGGGTAGGGTGTCACT
SRF-R	CCCCATGAAACGTAGGCTGT
FN1-F	GGTCACCCTGTTCTGCTTCA
FN1-R	TGTCTGGGTGACTTTCCTGC
Sp1-F	CCACCATGAGCGACCAAGAT
Sp1-R	CGCTACCCCCATTATTGCCA
Col1A1-F	CGATGGATTCCCGTTCGAGT
Col1A1-R	GAGGCCTCGGTGGACATTAG
α-SMA-F	TCTTCCAGCCATCTTTCATTGGGAT
α-SMA-R	CCTGTTTTGGCTCCCTATGTCT
Vimentin-F	AGACCAGAGATGGACAGGTGA
Vimentin-R	CTGGTACTGCACTGTTGCAC
TIMP1-F	GCGGTTCTGGGACTTGTGGGCATA
TIMP1-R	GCCCCCTTTGCATCTCTGGCATC
MMP2-F	ACCTGGATGCCGTCGTGGACCTG
MMP2-R	CGCCAGGCTGCTTCACATCCTTC
IL-6-F	ACAACCACGGCCTTCCCTACTT
IL-6-R	CACGATTTCCCAGAGAACATGTG
TGF-β1-F	GCCCTGGATACCAACTATTGCTTCA
TGF-β1-R	CAGAAGTTGGCATGGT
TINCR-F	CAAGGTACACCTAGCCGACG
TINCR-R	TTCTGCGCCAAGAGCTTACA
GAPDH-F	AGGAGAGTGTTTCCTCGTCC
GAPDH-R	TGAGGTCAATGAAGGGGTCG
U6-F	GAAGATTTAGCATGGCCCCTGC
U6-R	CAGTGCAGGGTCCGAGGT
cVIM-siRNA-1	TGCCCTTAAAGGCACTAACGAGT
cVIM-siRNA-2	AGGATGAGATCCAAAACATGAAG

Human	
hsa_circ_0000221-F	TGTCGATGTAGTTGGCGAAG
hsa_circ_0000221-R	ACCTCAACGAGAAGGTGGAG
hsa_circ_0007939-F	GGTCAAAGGTATGGCATTGG
hsa_circ_0007939-R	ACTGCTCTTTCCCTGGCTTT
hsa_circ_0017865-F	CGTCACCTTCGTGAATACCA
hsa_circ_0017865-R	AAACTTCTGCAGCCTTTGGA
hsa_circ_0017866-F	TTCCAGCAAGTATCCAACCA
hsa_circ_0017866-R	AAAGCCTGTCTTTGCTCGAA
hsa_circ_0017867-F	AGATGGCCCTTGACATTGAG
hsa_circ_0017867-R	GGAGAAGAGGCGAACGAG
hsa_circ_0017868-F	GGATTCACTCCCTCTGGTTG
hsa_circ_0017868-R	GGAGAAGAGGCGAACGAG
hsa_circ_0017869-F	TTCCAGCAAGTATCCAACCA
hsa_circ_0017869-R	GGAGAAGAGGCGAACGAG
hsa_circ_0017870-F	TCTGGATTCACTCCCTCTGG
hsa_circ_0017870-R	CGCATTGTCAACATCCTGTC
hsa_circ_0017871-F	TTCCAGCAAGTATCCAACCA
hsa_circ_0017871-R	CGCATTGTCAACATCCTGTC
hsa_circ_0017872-F	TGGATTCACTCCCTCTGGTT
hsa_circ_0017872-R	ATTCCACTTTGCGTTCAAGG
hsa_circ_0017873-F	TCCAGCAAGTATCCAACCAA
hsa_circ_0017873-R	CCTCTTCGTGGAGTTTCTTCA
hsa_circ_0017874-F	GTACCGGAGACAGGTGCAGT
hsa_circ_0017874-R	GGCTTGGAAACATCCACATC
hsa_circ_0017875-F	AGATGGCCCTTGACATTGAG
hsa_circ_0017875-R	GGCTTGGAAACATCCACATC
hsa_circ_0017876-F	TCTGGATTCACTCCCTCTGG
hsa_circ_0017876-R	GTGAGGTCAGGCTTGGAAAC
hsa_circ_0017877-F	TTCCAGCAAGTATCCAACCA
hsa_circ_0017877-R	GGCTTGGAAACATCCACATC
hsa_circ_0017878-F	TCCAGCAAGTATCCAACCAA
hsa_circ_0017878-R	CTGCACCTGTCTCCGGTACT
hsa_circ_0017879-F	CCTACAGGAAGCTGCTGGAA
hsa_circ_0017879-R	GCTTCAACGGCAAAGTTCTC
hsa_circ_0017880-F	TCTGGATTCACTCCCTCTGG
hsa_circ_0017880-R	AACGGCAAAGTTCTCTTCCA
hsa_circ_0017881-F	TCCAGCAAGTATCCAACCAA
hsa_circ_0017881-R	AGGCGGCCAATAGTGTCTT
hsa_circ_0017882-F	TCCAGCAAGTATCCAACCAAC
hsa_circ_0017882-R	CCAGAGGGAGTGAATCCAGA
hsa_circ_0017883-F	CGGTTGAAACTAGAGATGGACA
hsa_circ_0017883-R	TGAGTGGGTATCAACCAGAGG
hsa_circ_0017884-F	TTCCAGCAAGTATCCAACCA
hsa_circ_0017884-R	ATTGCTGCACTGAGTGTGTG
hsa_circ_0093230-F	GCAGGAGGAGATGCTTCAGA

hsa_circ_0093230-R	ACTGGCTCCCGGAGAAGAG
hsa_circ_0093232-F	ACCTCTACGAGGAGGAGATGC
hsa_circ_0093232-R	GCAGGATCTTATTCTGCTGCTC
hsa_circ_0093233-F	ACCTCTACGAGGAGGAGATGC
hsa_circ_0093233-R	TGTCGATGTAGTTGGCGAAG

Supplementary Tab.3: Up-regulated or down-regulated circRNAs in liver fibrosis

circRNA	Regulation	GeneSymbol	P-value
mmu_circRNA_29981	up	App	0.049175159
mmu_circRNA_012164	up	Hist1h1c	0.002970874
mmu_circRNA_20588	up	Pam	0.014034001
mmu_circRNA_37780	up	Syf2	0.024078973
mmu_circRNA_007217	up	Hist1h1c	0.003261803
mmu_circRNA_40537	up	Antxr1	0.033347472
mmu_circRNA_23791	up	Spag5	0.0381213
mmu_circRNA_36840	up	Svep1	0.034132532
mmu_circRNA_018777	up	Col3a1	0.009699984
mmu_circRNA_26326	up	Sema4d	0.016765115
mmu_circRNA_34451	up	Fbn1	0.010741952
mmu_circRNA_23124	up	Rtn4	0.049617416
mmu_circRNA_29984	up	Ltn1	0.040634076
mmu_circRNA_37916	up	Pgd	0.027174359
mmu_circRNA_015902	up	Clip2	0.005204066
mmu_circRNA_24182	up	Acly	0.000201364
mmu_circRNA_19118	up	Samd4	0.048250246
mmu_circRNA_37328	up	Dhcr24	0.028201056
mmu_circRNA_30668	up	Adgre1	0.003752486
mmu_circRNA_19794	up	Lmbrd1	0.043492598
mmu_circRNA_19982	up	Map4k4	0.042358766
mmu_circRNA_27519	up	Samd4	0.038562935
mmu_circRNA_19346	up	Pgd	0.037615846
mmu_circRNA_34309	up	Rtf1	0.004720457
mmu_circRNA_32665	up	Btrc	0.031973352
mmu_circRNA_39081	up	Sparcl1	0.0101094
mmu_circRNA_34779	up	Tpx2	0.021333323
mmu_circRNA_29154	up	Ppl	0.00153531
mmu_circRNA_28239	up	Npr3	0.049582734
mmu_circRNA_005305	up	Samd4	0.049573841
mmu_circRNA_45890	up	Rps6ka3	0.04247312
mmu_circRNA_26413	up	Ntrk2	0.041744953
mmu_circRNA_39653	up	Baiap2l1	0.002078411
mmu_circRNA_22197	up	Bicc1	0.037300025
mmu_circRNA_017702	up	Slc35f5	0.042870363
mmu_circRNA_44559	up	Myo1e	0.005579929
mmu_circRNA_36691	up	Unc13b	0.005411979
mmu_circRNA_34279	up	Knstrn	0.034316795
mmu_circRNA_000298	up	Ect2	0.001133466
mmu_circRNA_32994	up	Vim	0.020330809
mmu_circRNA_34544	up	Sirpa	0.011532885
mmu_circRNA_011784	up	Arhgap10	0.008056417
mmu_circRNA_45392	up	Rp2h	0.013447414
mmu_circRNA_30260	up	Smoc2	0.001386797
mmu_circRNA_006940	down	9130011E15Rik	0.043846956
mmu_circRNA_25777	down	Serpina4-ps1	0.017909055
mmu_circRNA_21916	down	Rtn4ip1	0.049761176

mmu_circRNA_21120	down	Tnr	0.047860526
mmu_circRNA_28144	down	Ghr	0.032418353
mmu_circRNA_29063	down	Fam186b	0.004624547
mmu_circRNA_28834	down	Slc25a17	0.039413973
mmu_circRNA_22878	down	Camk2b	0.043426037
mmu_circRNA_21911	down	Sobp	0.025312431
mmu_circRNA_25732	down	Atxn3	0.046665629
mmu_circRNA_39652	down	Lmtk2	0.038077759
mmu_circRNA_37243	down	Raver2	0.043913776
mmu_circRNA_39156	down	Ep400	0.002552742
mmu_circRNA_32345	down	Kank1	0.016208401
mmu_circRNA_42397	down	Lhpp	0.018030244
mmu_circRNA_42398	down	Lhpp	0.007353568
mmu_circRNA_26977	down	Ppap2a	0.037184896
mmu_circRNA_22784	down	Tmem194	0.021444162
mmu_circRNA_36581	down	Bach2	0.046418701
mmu_circRNA_34113	down	Slc1a2	0.000341918
mmu_circRNA_25382	down	Gphn	0.02254617
mmu_circRNA_41020	down	Slco1b2	0.006157182
mmu_circRNA_38486	down	Rgs12	0.023791373
mmu_circRNA_31987	down	Dym	0.043242595
mmu_circRNA_36004	down	Slc30a7	0.047274749
mmu_circRNA_32003	down	Pias2	0.000185612
mmu_circRNA_42780	down	Dctn6	0.005768733

Supplementary Tab.4: The target miRNAs of cVIM predicted by miRanda software.

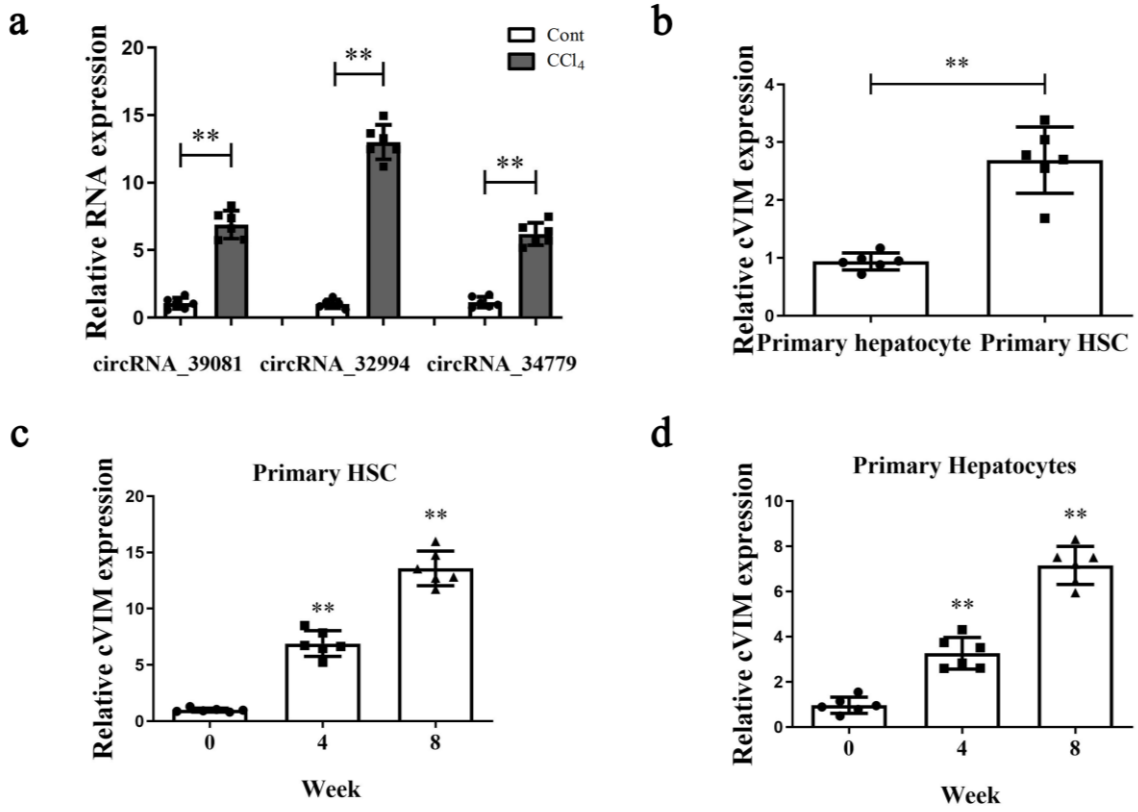
miRNA	Max Score	Max Energy	Binding position In cVIM	Predicted by RNAhybrid
mmu-miR-30a-5p	103	-21.66	40,128	Yes
mmu-miR-125a-3p	109	-26.37	57,179	Yes
mmu-miR-9-5p	97	-20.27	611	Yes
mmu-miR-149-3p	98	-24.89	520,149, 568,56	Yes
mmu-miR-187-3p	96	-24.4	342,616,179,115	Yes
mmu-miR-191-3p	99	-23.01	231,143	Yes
mmu-miR-200b-5p	95	-24.46	73,428,294	Yes
mmu-miR-143-5p	98	-29.24	236,98	Yes
mmu-miR-23a-5p	95	-22.73	466,564,1	Yes
mmu-miR-26b-3p	101	-24.94	488,222	Yes
mmu-miR-93-3p	96	-20.79	479	Yes
mmu-miR-34a-5p	87	-26.41	167,97	Yes
mmu-miR-326-5p	97	-26.33	414,272,54,96	Yes
mmu-miR-328-3p	103	-32.07	66,520,560,166,5	Yes
mmu-miR-330-3p	89	-23.59	73,172	Yes
mmu-miR-122-5p	97	-20.98	356	Yes
mmu-miR-337-5p	98	-25.57	73,596	Yes
mmu-miR-341-5p	93	-23.25	352,288	Yes
mmu-miR-345-3p	93	-28.44	12,207,129	Yes
mmu-miR-346-3p	101	-30.48	505,2,204	Yes
mmu-miR-107-3p	97	-22.24	467	Yes
mmu-miR-17-3p	110	-24.12	59,242	Yes
mmu-miR-25-5p	93	-22.73	152	Yes
mmu-miR-212-5p	87	-21.90	477	Yes
mmu-miR-320-5p	97	-23.57	491,123	Yes
mmu-miR-26a-2-3p	94	-21.47	227	Yes
mmu-miR-29b-2-5p	101	-20.51	608,404	Yes
mmu-miR-125b-1-3p	102	-24.85	57,232,116,1,169	Yes
mmu-miR-217-5p	90	-20.61	247	Yes
mmu-miR-378a-5p	96	-26.57	227,13,477,201	Yes
mmu-miR-381-3p	100	-22.24	323	Yes
mmu-miR-215-3p	103	-22.17	263	Yes
mmu-miR-196b-5p	106	-24.29	472,98,236,286	Yes
mmu-miR-412-5p	99	-22.31	566	No
mmu-miR-434-3p	103	-25.66	133	No
mmu-miR-465a-3p	97	-20.31	439	No
mmu-miR-540-3p	98	-24.08	506,353	No
mmu-miR-423-5p	99	-24.36	507,153,63,563	No
mmu-miR-681	101	-25.57	100,165	No
mmu-miR-1298-3p	99	-21.02	22	No
mmu-miR-764-3p	100	-24.73	354	No
mmu-miR-652-5p	103	-24.27	562	No
mmu-miR-490-3p	98	-25.20	557	No
mmu-miR-693-3p	111	-22.00	468,163,94,247	No
mmu-miR-146b-3p	107	-23.92	285,187,258	No
mmu-miR-698-5p	100	-23.18	459,569	No

mmu-miR-717	101	-21.47	502	No
mmu-miR-3099-3p	97	-24.89	521,163,66	No
mmu-miR-344d-1-5p	103	-21.54	100,352,6	No
mmu-miR-676-3p	97	-24.61	116	No
mmu-miR-615-3p	103	-27.21	7,612	No
mmu-miR-453	100	-22.41	156,351	No
mmu-miR-92b-3p	98	-27.08	231,299,134	No
mmu-miR-873a-5p	101	-20.90	280	No
mmu-miR-877-3p	103	-25.33	490,134	No
mmu-miR-1894-5p	98	-22.13	481	No
mmu-miR-1930-3p	99	-22.47	242,76	No
mmu-miR-1231-5p	98	-25.51	511,67,25,601,171	No

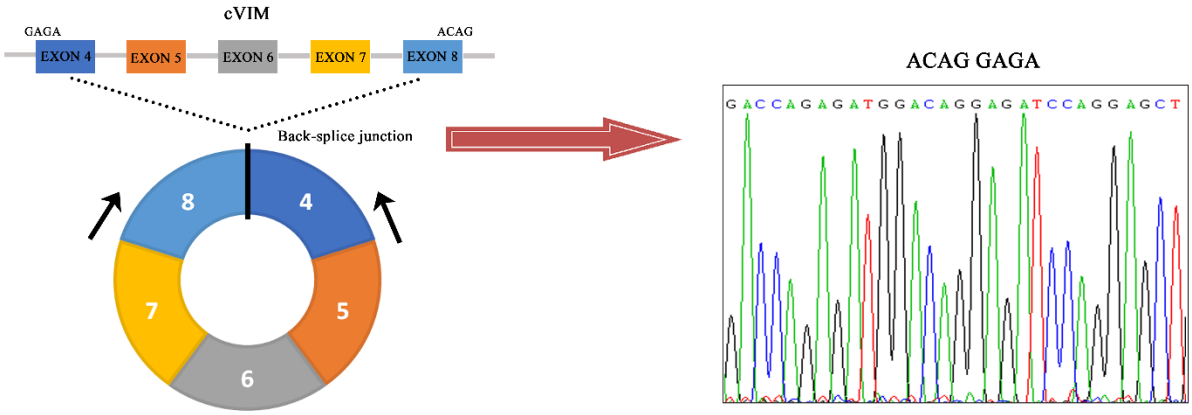
Supplementary Tab.5: TGF-β pathway-related genes which are the targets of miR-9-5p and miR-122-5p.

TGF-β pathway-related genes	miRNAs	The studies proving that they are the targets of the indicated miRNAs
TGFBR1 and TGFBR2	miR-9-5p	Epigenetically-Regulated MicroRNA-9-5p Suppresses the Activation of Hepatic Stellate Cells via TGFBR1 and TGFBR2 [1]
TGFBR2	miR-9-5p	Protective role for miR-9-5p in the fibrogenic transformation of human dermal fibroblasts [2]
TGFBR2 and NOX4	miR-9-5p	miR-9-5p suppresses pro-fibrogenic transformation of fibroblasts and prevents organ fibrosis by targeting NOX4 and TGFBR2 [3]
KLF6	miR-122-5p	NEAT1 accelerates the progression of liver fibrosis via regulation of microRNA-122 and Kruppel-like factor 6 [4]
TGF-β1	miR-122	microRNA-122 down-regulation may play a role in severe myocardial fibrosis in human aortic stenosis through TGF-β1 up-regulation [5]
TGFBR2	miR-122-5p	miR-24 and miR-122 Negatively Regulate the Transforming Growth Factor-β/Smad Signaling Pathway in Skeletal Muscle Fibrosis [6]
SRF and FN1	miR-122-5p	Identification of a novel TGF-β-miR-122-fibronectin 1/serum response factor signaling cascade and its implication in hepatic fibrogenesis [7]

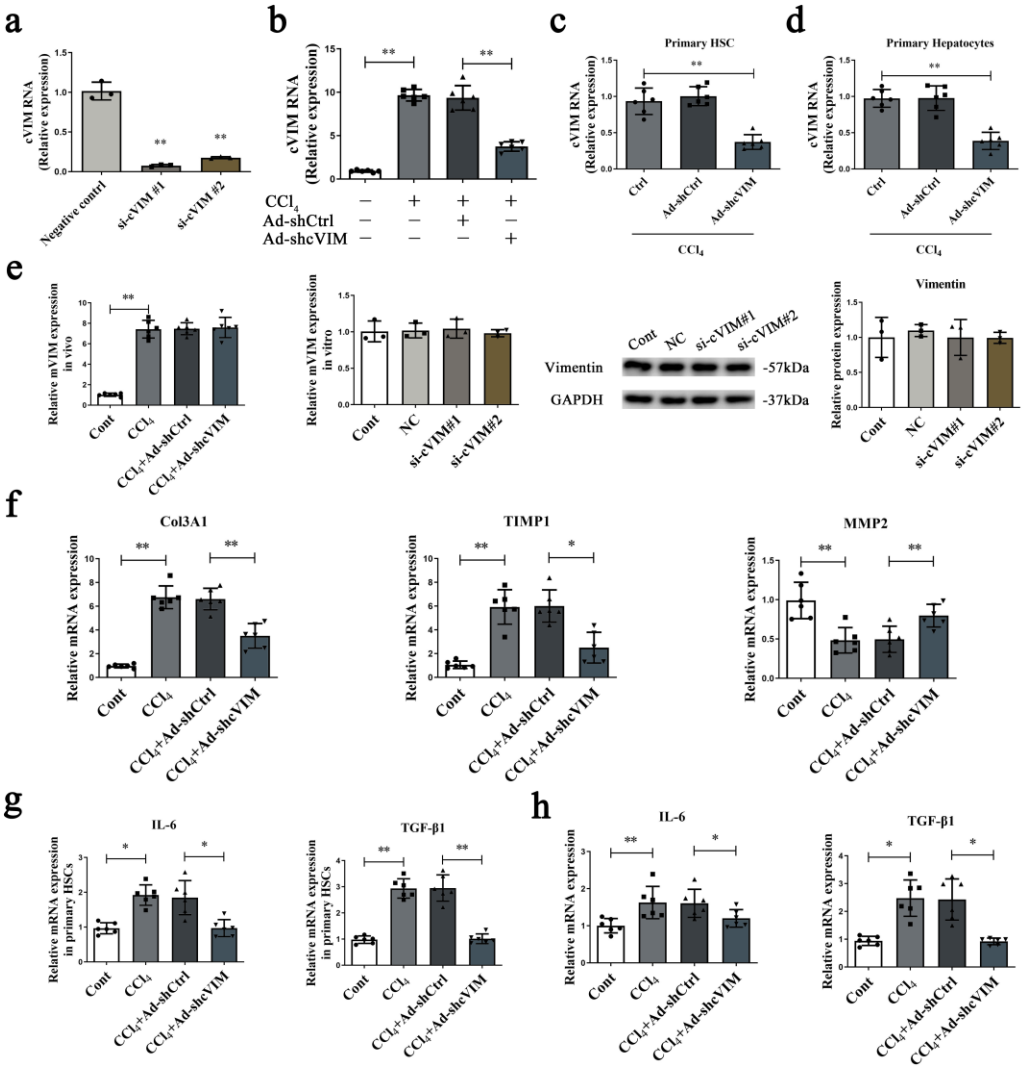
Supplementary Fig. 1 Expression of cVIM in primary HSCs and primary hepatocytes. a Expressions of circRNA_39081, circRNA_32994 (cVIM) and circRNA_34779 in primary HSCs isolated from CCl_4-treated mice as well as healthy controls (n=6 per group). **b** cVIM was detected in primary HSCs and hepatocytes, which were isolated from healthy mice (n=6 per group). **(c and d**) cVIM was detected in primary HSCs (c) and hepatocytes (d), which were isolated from CCl_4-treared mice at different weeks (n=6 per group). Each value is the mean ± SD of six independent experiments. $^{**}P<0.01$ compared with the control.



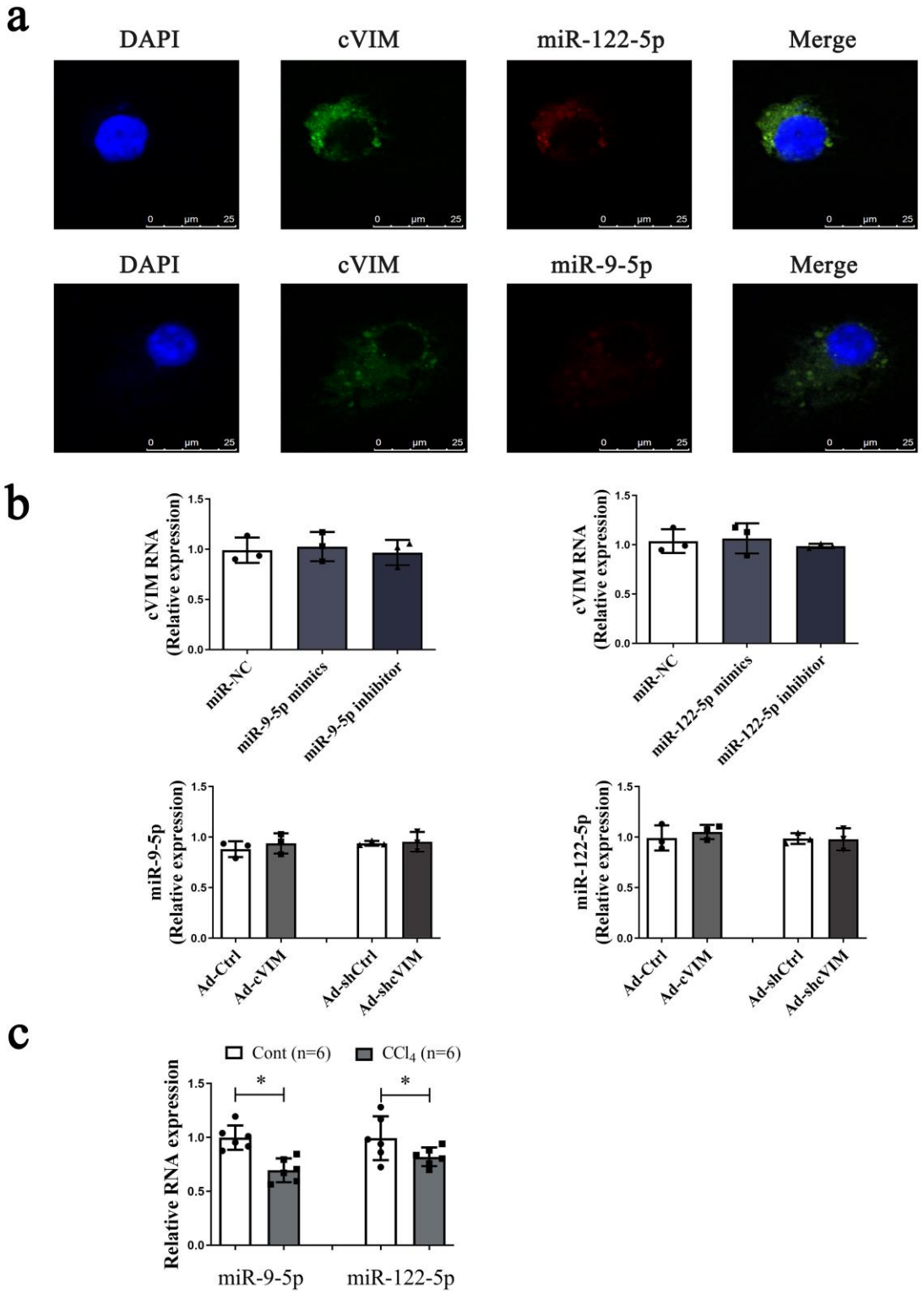
Supplementary Fig. 2 Sanger sequencing showing the back-spliced events of cVIM.



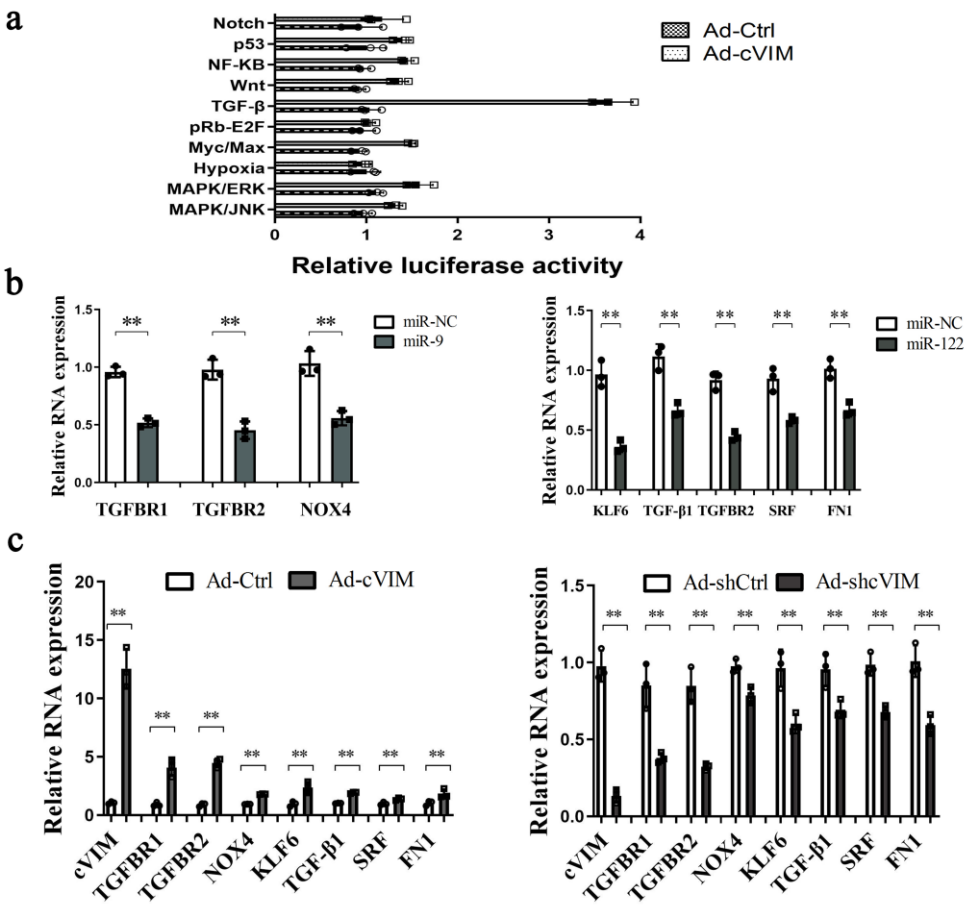
Supplementary Fig. 3 Expression of cVIM *in vitro and in vivo* after cVIM knockdown treatment. a Expression of cVIM in primary 1-day-old HSCs isolated from CCl_4-treated mice after cVIM siRNA treatment for 48 h (n=3 per group). **b** cVIM in the liver from CCl_4-treated mice after Ad-shcVIM treatment (n=6 per group). **(c and d**) cVIM in isolated primary HSCs (c) as well as primary hepatocytes (d) from CCl_4-treated mice after Ad-shcVIM treatment (n=6 per group). **e** Expression of mVIM after silencing of cVIM *in vitro* (n=3 per group) *and in vivo* (n=6 per group). **f** ECM markers in liver tissues of CCl_4-treated mouse (n=6 per group). **(g and h**) Profibrotic cytokines in HSCs (g) and in liver tissues (h) (n=6 per group). Each value is the mean ± SD of three independent experiments. **P*<0.05 and ***P*<0.01 compared with the control.



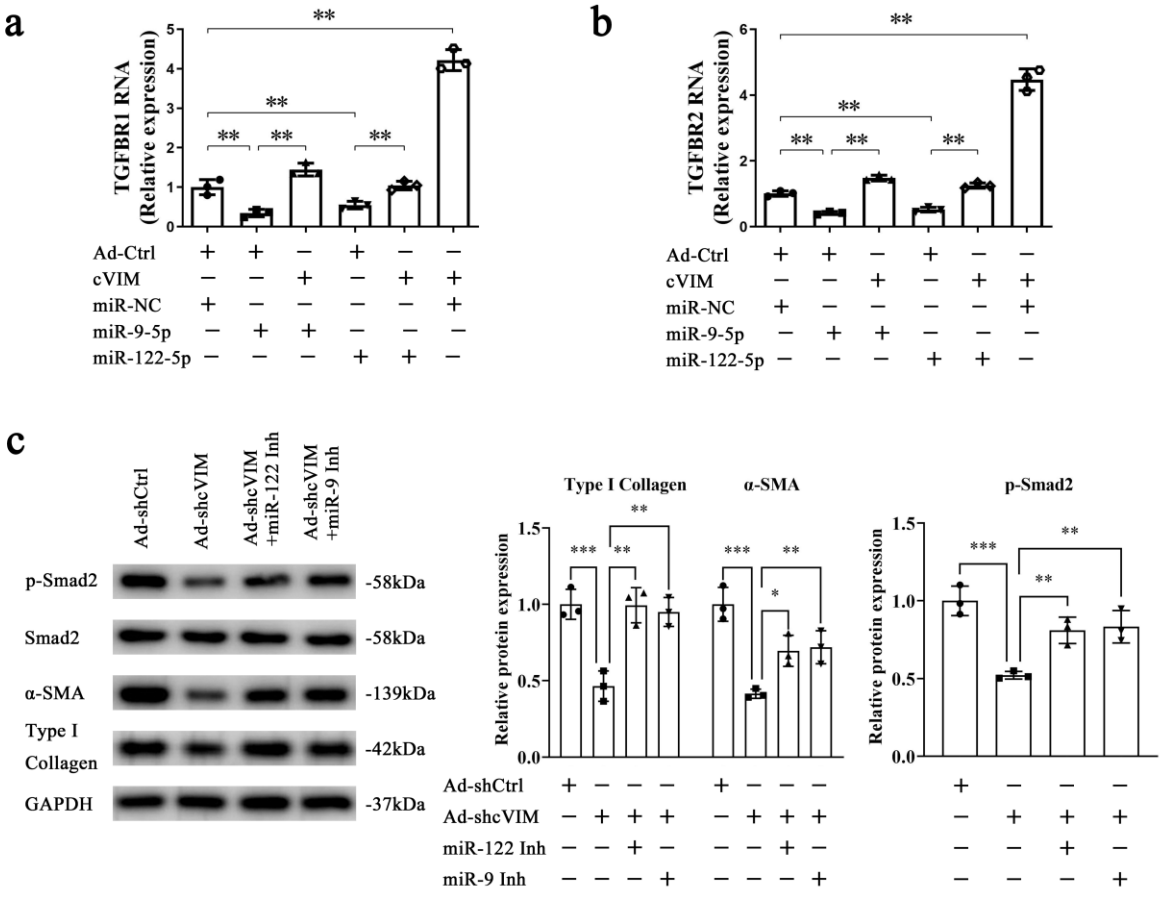
Supplementary Fig.4 cVIM functions as a sponge for miR-122-5p and miR-9-5p. a Co-localization between miR-122-5p/miR-9-5p and cVIM was observed by FISH in primary 1-day-old HSCs isolated from CCl_4-treated mice. Nuclei were stained with DAPI (n=3 per group). Scale bar, 25 μm. **b** qRT-PCR showed that cVIM did not change significantly upon miR-122-5p or miR-9-5p mimics/inhibitor in primary 1-day-old HSCs isolated from CCl_4-treated mice. qRT-PCR showed that miR-122-5p or miR-9-5p did not change significantly after overexpressing or silencing cVIM in primary 1-day-old HSCs isolated from CCl_4-treated mice (n=3 per group). **c** The expressions of miR-122-5p and miR-9-5p were decreased in the fibrotic livers (n=6 per group). Each value is the mean ± SD of three independent experiments. *P<0.05.


a
DAPI
cVIM
miR-122-5p
Merge
DAPI
cVIM
miR-9-5p
Merge
b
cVIM RNA (Relative expression)
miR-NC
miR-9-5p mimics
miR-9-5p inhibitor
cVIM RNA (Relative expression)
miR-NC
miR-122-5p mimics
miR-122-5p inhibitor
miR-9-5p (Relative expression)
Ad-Ctrl
Ad-cVIM
Ad-shCtrl
Ad-shcVIM
miR-122-5p (Relative expression)
Ad-Ctrl
Ad-cVIM
Ad-shCtrl
Ad-shcVIM
c
Cont (n=6)
CCl4 (n=6)
Relative RNA expression
miR-9-5p
miR-122-5p

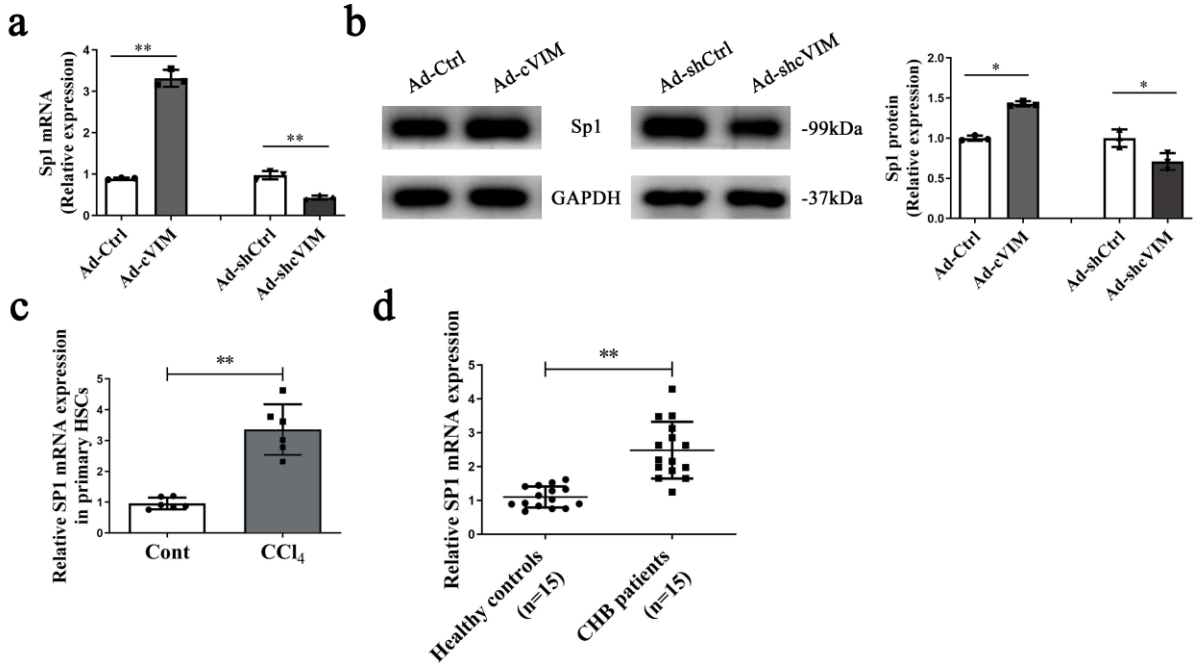
Supplementary Fig.5 cVIM inhibits liver fibrosis through TGFBR1/TGFBR 2-mediated TGF-β pathway activation. Primary 1-day-old HSCs isolated from CCl_4-treated mice were transduced with Ad-cVIM or Ad-shcVIM for 48 h. Cells were also transfected with miR-122-5p or miR-9-5p mimics for 24 h. **a** Signaling pathway reporter array was used for seeking the relative pathway associated with cVIM (n=3 per group). **b** qRT-PCR showed that the targets of miR-9-5p (TGFBR1, TGFBR2 and NOX4) were significantly downregulated upon overexpression of miR-9-5p, and the targets of miR-122-5p (KLF6, TGF-β1, TGFBR2, SRF and FN1) were significantly downregulated upon overexpression of miR-122-5p (n=3 per group). **c** The targets of miR-9-5p and miR-122-5p were examined in cells after overexpressing or silencing cVIM (n=3 per group). Each value is the mean ± SD of three independent experiments. ***P*<0.01.



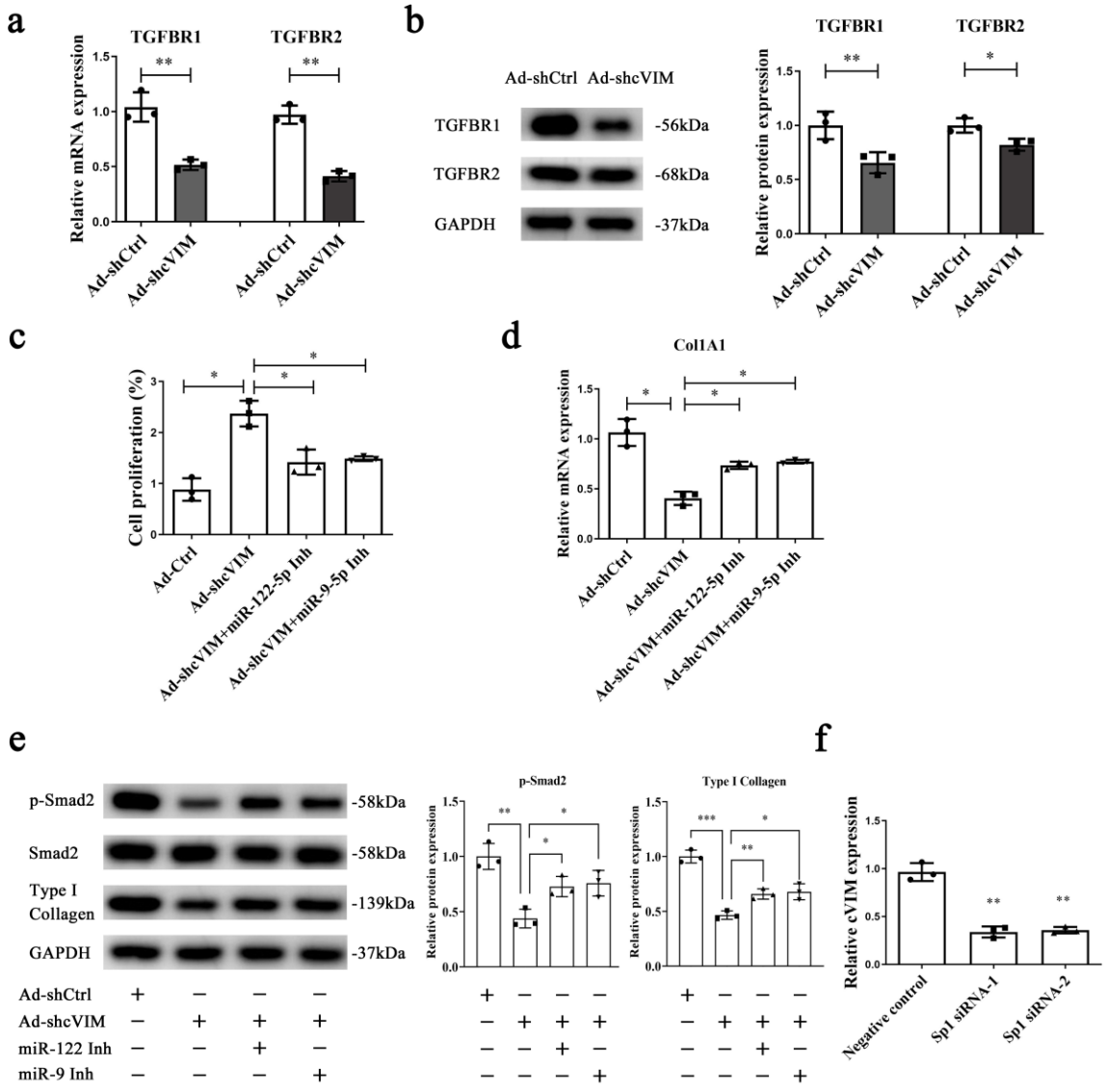
Supplementary Fig.6 cVIM accelerates liver fibrosis progression via miR-122-5p/miR-9-5p-midaited TGF-β pathway. Primary 1-day-old HSCs isolated from CCl_4-treated mice were transduced with Ad-cVIM for 48 h and then transfected with miR-122-5p/miR-9-5p mimics for additional 24 h. Moreover, cells were transduced with Ad-shcVIM for 48 h and then transfected with miR-122-5p/miR-9-5p inhibitor for additional 24 h. **a** Reduced TGFBR1 by miR-122-5p or miR-9-5p was blocked down by overexpression of cVIM (n=3 per group). **b** Reduced TGFBR2 by miR-122-5p or miR-9-5p was blocked down by overexpression of cVIM (n=3 per group). **c** Immunobolt analysis showed that miR-122-5p or miR-9-5p inhibitor inhibited the downregulation of p-smad2, α-SMA and type I collagen induced by cVIM knockdown (n=3 per group). Each value is the mean ± SD of three independent experiments. **P*<0.05, ***P*<0.01 and ****P*<0.001.



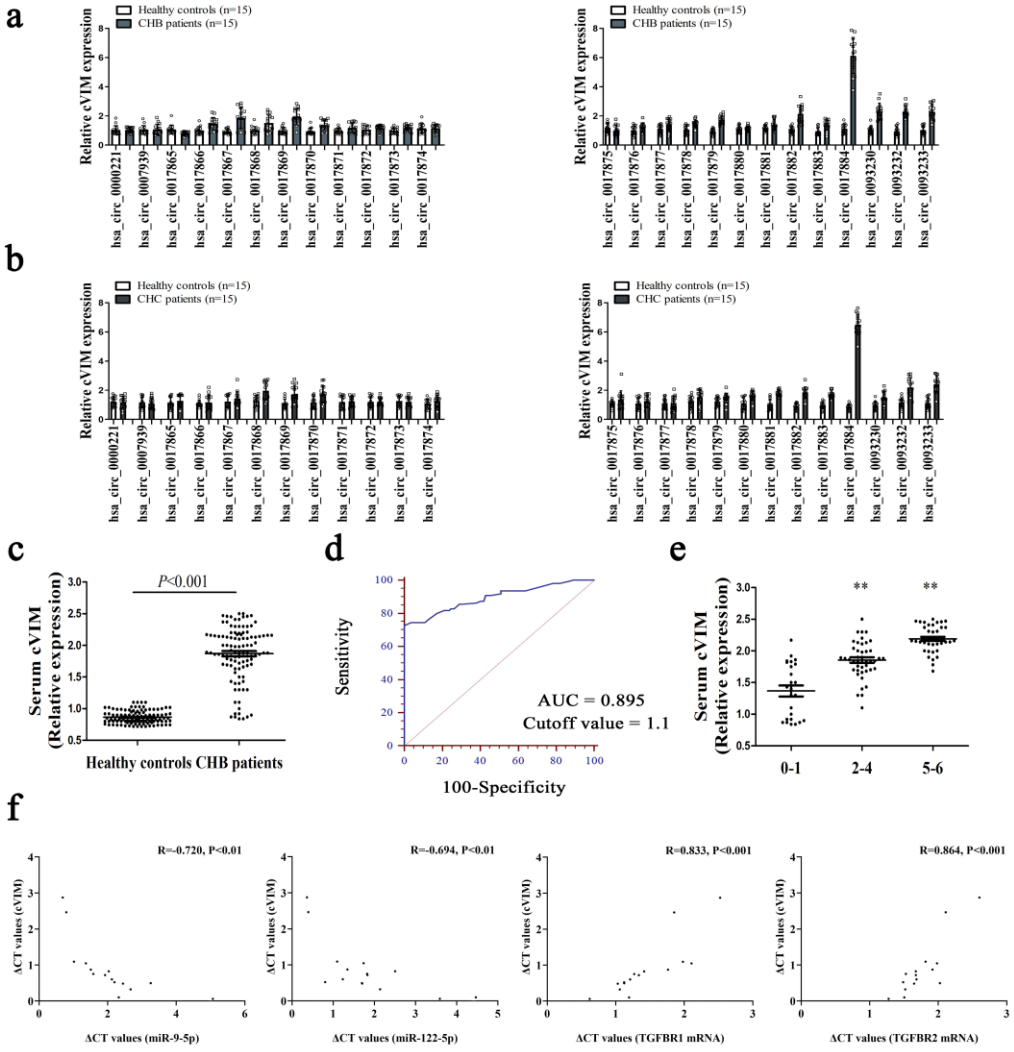
Supplementary Fig.7 Overexpression of cVIM contributes to the expression of Sp1. Primary 1-day-old HSCs isolated from CCl_4-treated mice were transduced with Ad-cVIM or Ad-shcVIM for 48 h. **a** Sp1 mRNA expression (n=3 per group). **b** Sp1 protein expression (n=3 per group). **c** Expression of Sp1 in activated HSCs isolated from CCl_4-treated mice (n=6 per group). **d** Expression of Sp1 in CHB patients with liver fibrosis (n=15 per group). Each value is the mean ± SD of three independent experiments. **P*<0.05 and ***P*<0.01.



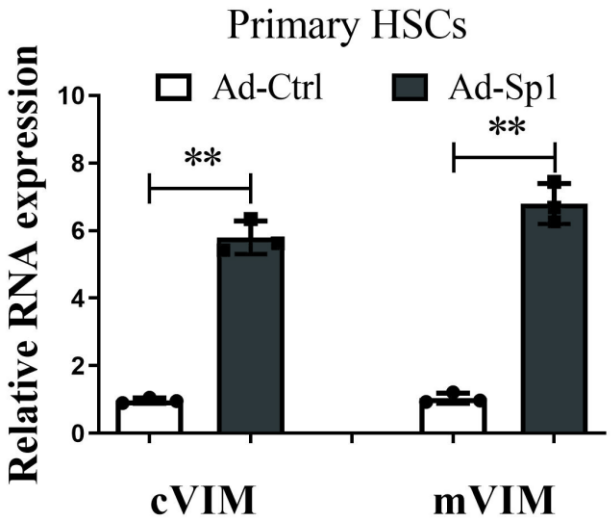
Supplementary Fig.8 Effects of loss of cVIM in hepatocytes. Primary hepatocytes were transduced with Ad-shcVIM for 48 h and then transfected with miR-122-5p/miR-9-5p inhibitor for additional 24 h. **a** mRNA expressions of TGFBR1 and TGFBR2 (n=3 per group). **b** Protein expressions of TGFBR1 and TGFBR2 (n=3 per group). **c** Cell proliferation (n=3 per group). **d** mRNA expression of Col1A1 (n=3 per group). **e** Protein expressions of p-smad2 and type I collagen (n=3 per group). **f** cVIM expression (n=3 per group). cVIM expression was examined in primary hepatocytes with Sp1 siRNA for 48 h. Each value is the mean ± SD of three independent experiments. **P*<0.05, ***P*<0.01 and ****P*<0.001.



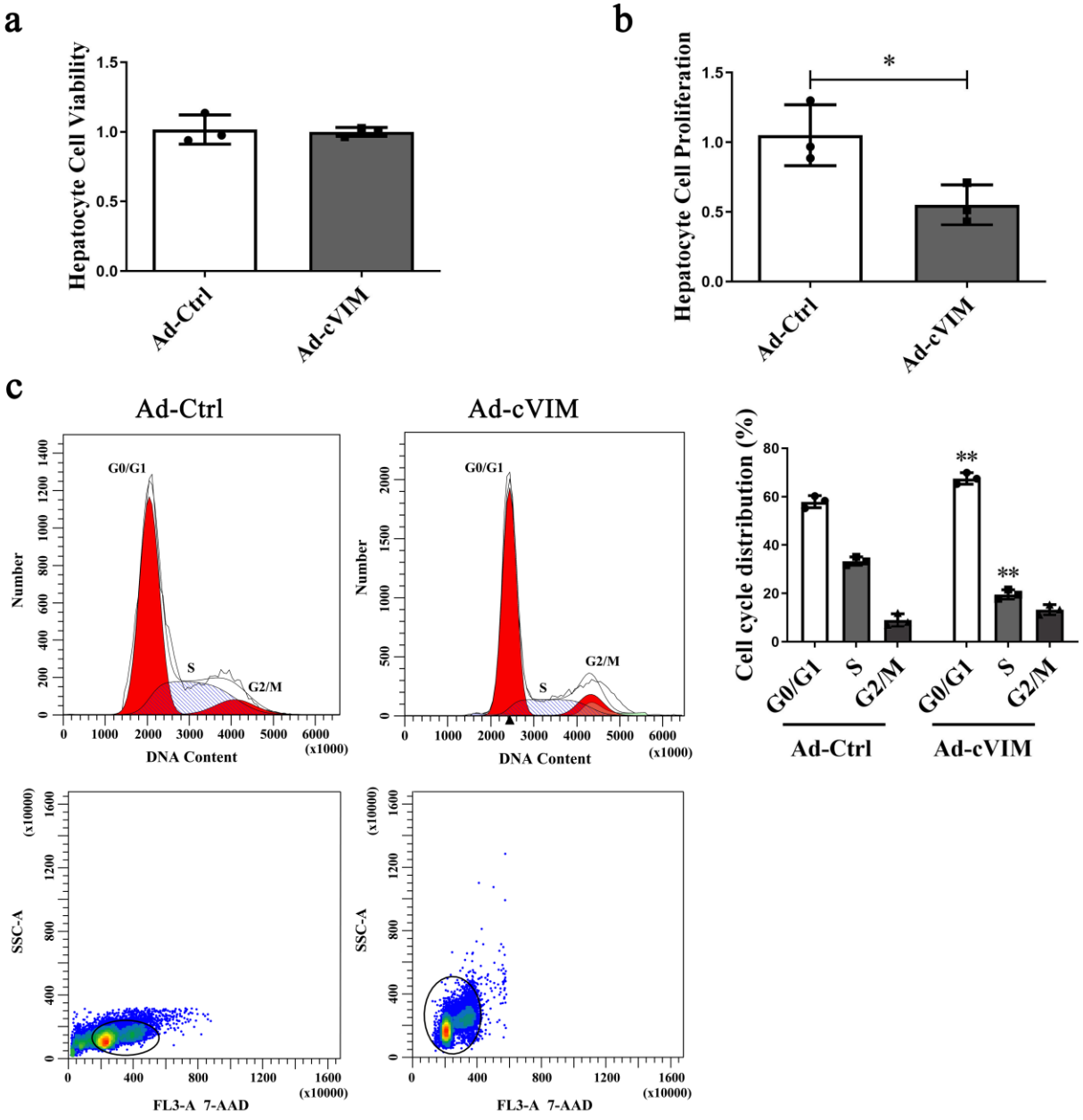
Supplementary Fig.9 Expression of liver cVIM and serum cVIM in patients with liver fibrosis. (a and b) All circRNA splices derived from human VIM gene were detected in the liver tissues from CHB patients (a, n=15 per group) and CHC patients (b, n=15 per group). **c** Serum cVIM levels in CHB patients (n=110 per group). **d** ROC curve analysis of serum cVIM for discriminating CHB patients with liver fibrosis (n=110 per group) from healthy controls (n=110 per group). **e** Serum cVIM levels in CHB patients with different fibrosis scores (n=110 per group). **f** The expression levels of miR-9-5p, miR-122-5p, TGFBR1 or TGFBR2 in liver samples from CHB patients (n=15 per group). Each value is the mean ± SD of three independent experiments. ***P*<0.01 compared with the control.



Supplementary Fig.10 Expression of cVIM and mVIM after Sp1 overexpression. Each value is the mean ± SD of three independent experiments. ***P*<0.01.



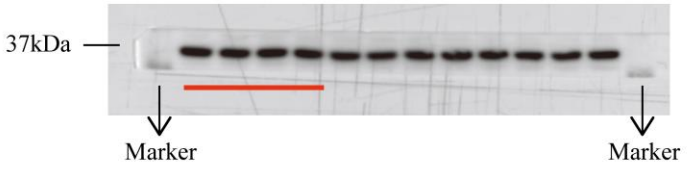
Supplementary Fig.11 Roles of cVIM in hepatocytes. a Trypan blue staining analysis (n=3 per group). **b** CCK-8 analysis (n=3 per group). **c** Cell cycle analysis (n=3 per group). Each value is the mean ± SD of three independent experiments. *P<0.05 and **P<0.01.



Supplementary Fig.12 Unedited blot/gel images.

Fig.3g

Type I Collagen



GAPDH

Fig.5b

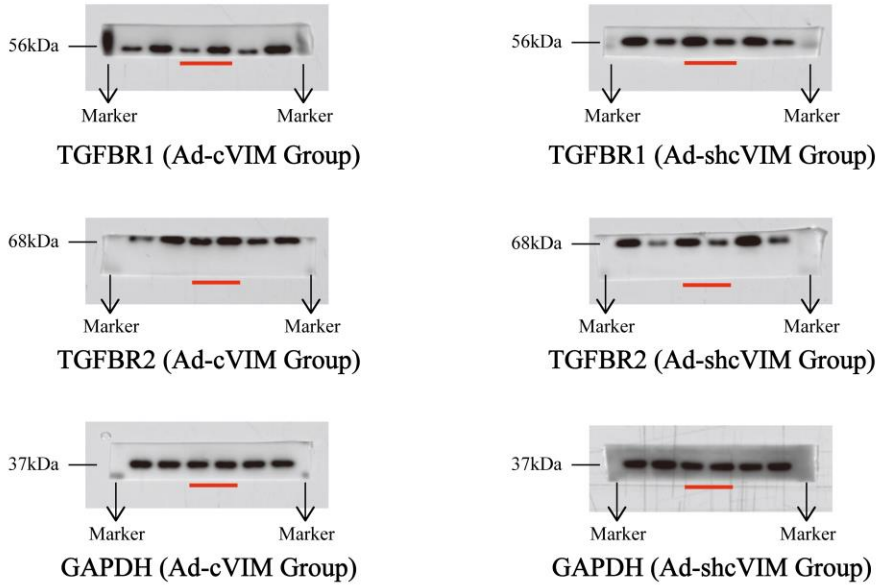


Fig.5c

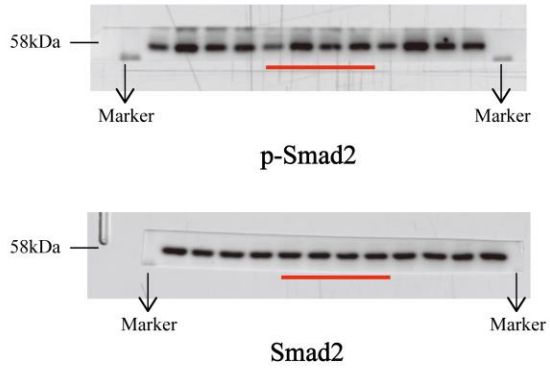


Fig.5f

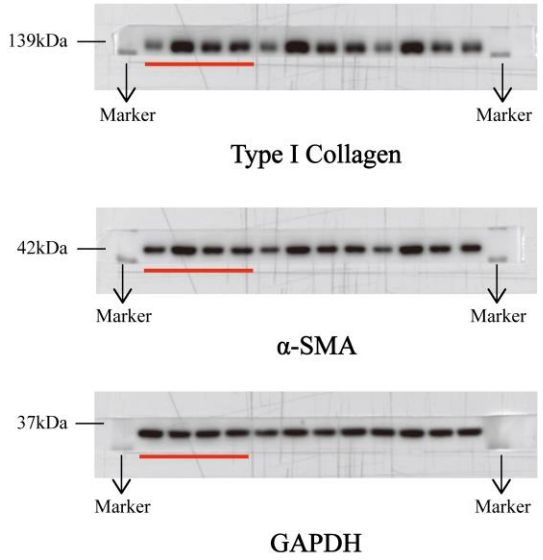
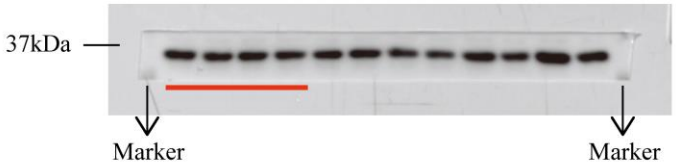


Fig.S3e

Vimentin



GAPDH

330
331

Fig.S6c

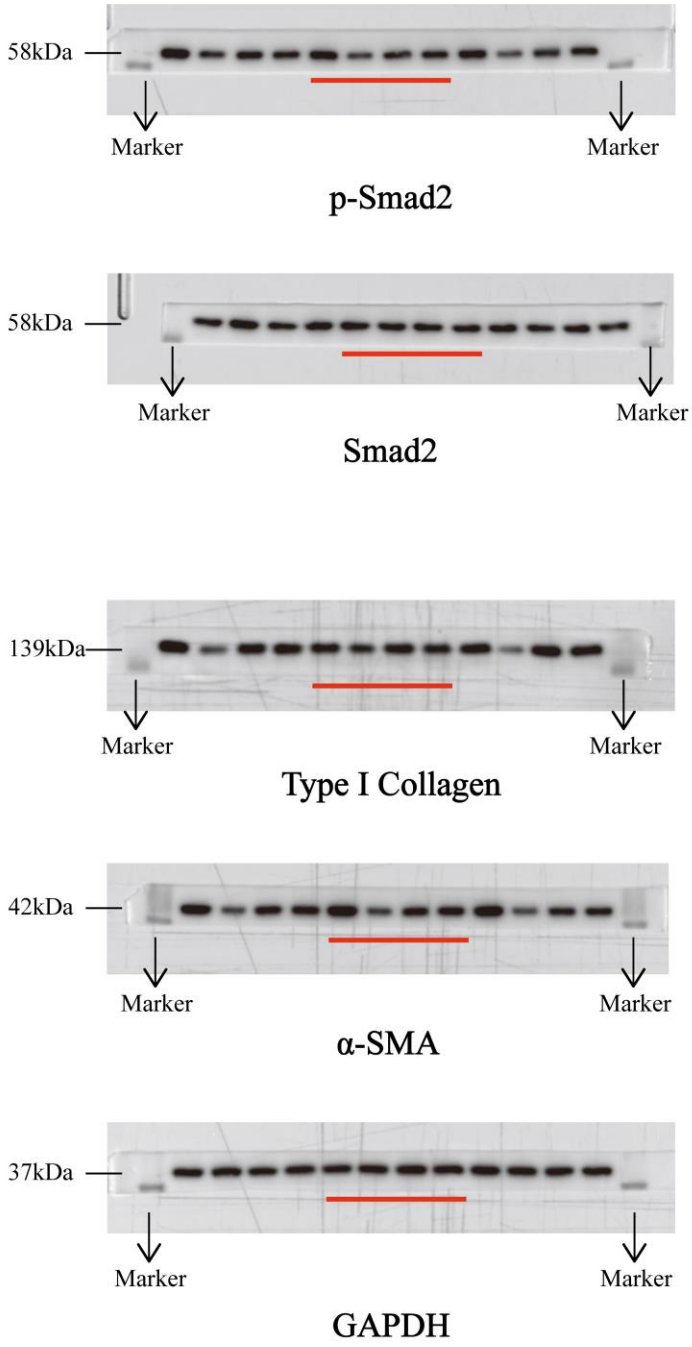
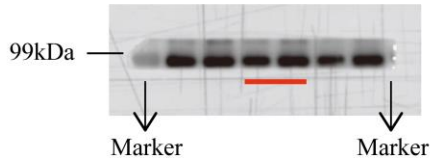
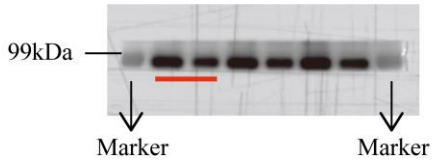

58kDa
Marker
Marker
p-Smad2
58kDa
Marker
Marker
Smad2
139kDa
Marker
Marker
Type I Collagen
42kDa
Marker
Marker
α-SMA
37kDa
Marker
Marker
GAPDH

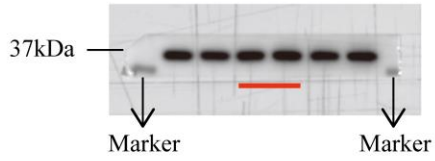
Fig.S7b



Sp1 (Ad-cVIM Group)



Sp1 (Ad-shcVIM Group)



GAPDH (Ad-cVIM Group)

GAPDH (Ad-shcVIM Group)

337
338
339

Fig.S8b

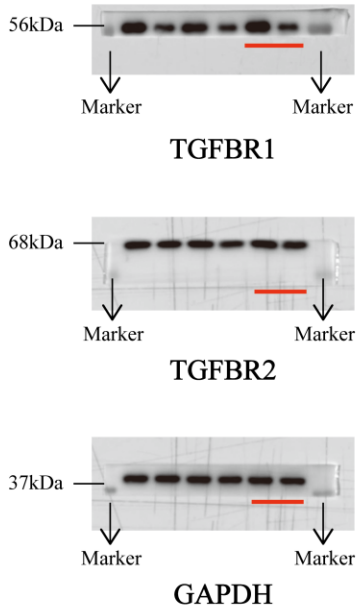
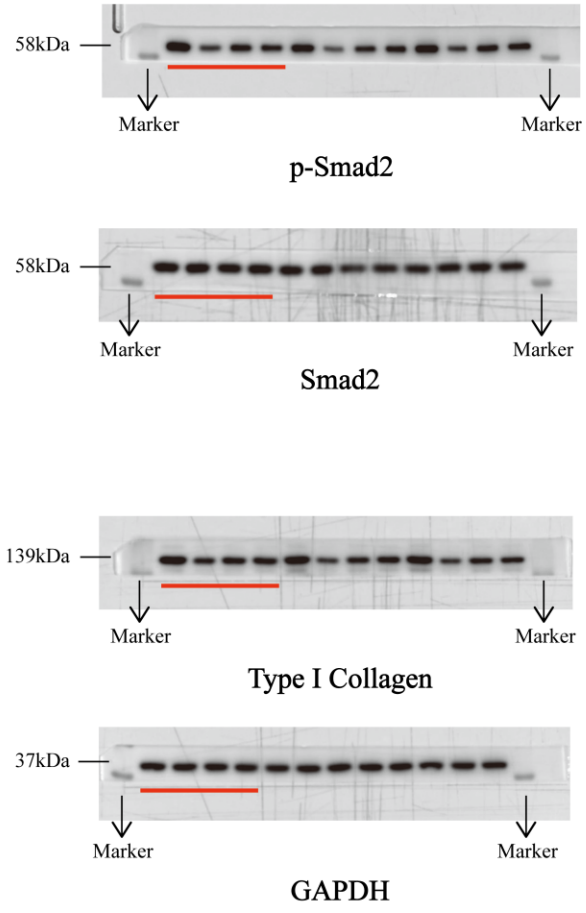


Fig.S8e



Supplementary References

1 Yu, F. *et al.* Epigenetically-Regulated MicroRNA-9-5p Suppresses the Activation of Hepatic Stellate Cells via TGFBR1 and TGFBR2. *Cell Physiol Biochem* **43**, 2242-2252, doi:10.1159/000484303 (2017).

2 Miguel, V., Busnadiego, O., Fierro-Fernandez, M. & Lamas, S. Protective role for miR-9-5p in the fibrogenic transformation of human dermal fibroblasts. *Fibrogenesis & tissue repair* **9**, 7, doi:10.1186/s13069-016-0044-2 (2016).

3 Fierro-Fernandez, M. *et al.* miR-9-5p suppresses pro-fibrogenic transformation of fibroblasts and prevents organ fibrosis by targeting NOX4 and TGFBR2. *EMBO Rep* **16**, 1358-1377, doi:10.15252/embr.201540750 (2015).

4 Yu, F., Jiang, Z., Chen, B., Dong, P. & Zheng, J. NEAT1 accelerates the progression of liver fibrosis via regulation of microRNA-122 and Kruppel-like factor 6. *J Mol Med (Berl)* **95**, 1191-1202, doi:10.1007/s00109-017-1586-5 (2017).

5 Beaumont, J. *et al.* microRNA-122 down-regulation may play a role in severe myocardial fibrosis in human aortic stenosis through TGF-beta1 up-regulation. *Clin Sci (Lond)* **126**, 497-506, doi:10.1042/CS20130538 (2014).

6 Sun, Y. *et al.* miR-24 and miR-122 Negatively Regulate the Transforming Growth Factor-beta/Smad Signaling Pathway in Skeletal Muscle Fibrosis. *Mol Ther Nucleic Acids* **11**, 528-537, doi:10.1016/j.omtn.2018.04.005 (2018).

7 Zeng, C. *et al.* Identification of a novel TGF-beta-miR-122-fibronectin 1/serum response factor signaling cascade and its implication in hepatic fibrogenesis. *Oncotarget* **6**, 12224-12233, doi:10.18632/oncotarget.3652 (2015).