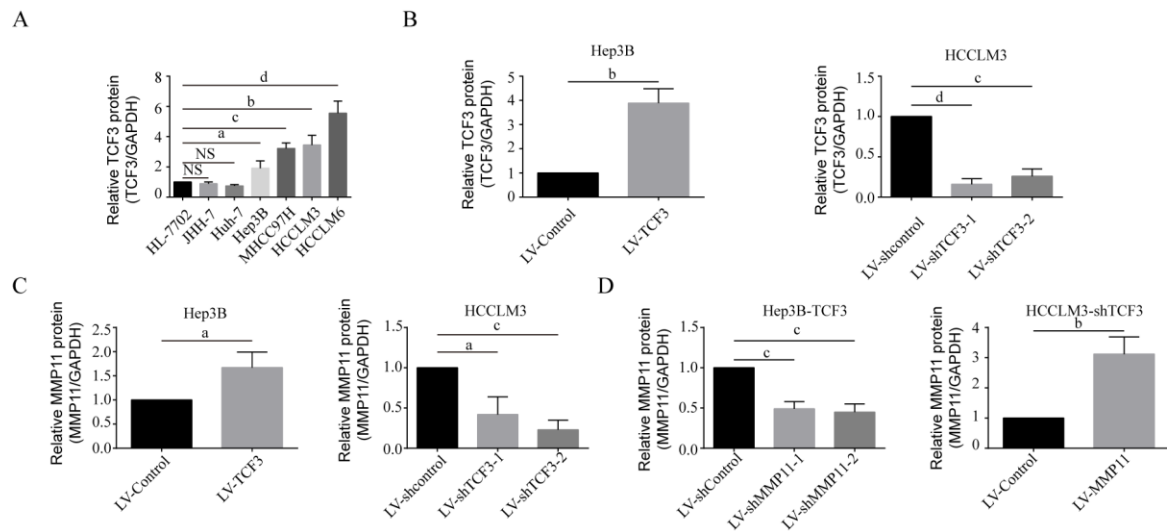




Supplementary Figure 1 Semi-quantitative and statistical analysis of western blotting data was conducted. A: Semi-quantitative analysis of Western blot results for transcription factor 3 (TCF3) expression was performed across various hepatocellular carcinoma (HCC) cell lines and a normal liver hepatocyte cell line; B: The semi-quantitative analysis of western blot results for TCF3 expression was conducted in Hep3B cells with TCF3 overexpression and in HCCLM3 cells with TCF3 knockdown; C: Semi-quantitative analysis of western blot results for matrix metalloproteinase-11 (MMP11) expression was performed in Hep3B cells with TCF3 overexpression and in HCCLM3 cells with TCF3 knockdown; D: The semi-quantitative analysis of western blot results for MMP11 expression was conducted in Hep3B-TCF3 cells transfected with an MMP11 knockdown lentivirus and in HCCLM3-short hairpin TCF3 cells transfected with an MMP11 overexpression lentivirus. ^a $P < 0.05$, ^b $P < 0.01$, ^c $P < 0.001$, ^d $P < 0.0001$, NS: No significance. HCC: Hepatocellular carcinoma; MMP11: Matrix metalloproteinase-11; Sh: Short hairpin; TCF3: Transcription factor 3.

Supplementary Table 1 Primer and short hairpin RNA sequences used in the present study

Primer/shRNA name	Primer sequence	Enzyme
Primers for reverse transcription-quantitative polymerase chain reaction		
TCF3sense	5'-AGGAGAAGGAGGACGAGGAG-3'	
TCF3antisense	5'-AAAGGCCTCGTTGATGTCAC-3'	
MMP11 sense	5'-CTAG ACTAGT CGAGCCTGCCAACACT-3'	
MMP11 antisense	5'-CCCAAGCTTGGGATACAGCAAGGACAC-3'	
GAPDH sense	5'-GCACCGTCAAGGCTGAGAAC-3'	
GAPDH antisense	5'-TGGTGAAGACGCCAGTGGA-3'	
Primers for MMP11 promoter construct		
(-1,918/+189)	5'-TATAGGTACCGACTGAGGCAGGAGAATG-3'	KpnI
MMP11sense		
(-1,655/+189) MMP11 sense	5'-TATAGGTACCGACCACAGCCTCACCTTT-3'	KpnI
(-1,209/+189) MMP11	5'-TATAGGTACCTGCTCTCCAACCTCCTGAC-3'	KpnI

sense

(-1,030/+189) MMP11 5'- TATAGGTACCTACTCCACTCAGACACCTC -3'

KpnI

sense

Antisense 5'- ATATAAGCTTTTAAGAAGCCACTGTAGCA -3'

HindIII

Primers for MMP11

promoter site-directed

mutagenesis

Binding site 1 mutation 5'-CTGTGAAACATTTcCaacatatagtCCCTTTCTTGG -3'

sense

Binding site 1 mutation 5'-CCAAGAAAGGGactatatgttgGAAATGTTTCACAG -3'

antisense

Binding site 2 mutation 5'-AGGTGTGTGCCACcgagttgaattAATTTTGTATTTT -3'

sense

Binding site 2 mutation 5'-AAAAATACAAAAATTaattcaactcgGTGGCACACACCT-3'

antisense

Binding site 3 mutation 5'-GACCTCGTGA taagttgatatCGGCCTCCCAAAG -3'

sense

Binding site 3 mutation 5'-CTTTGGGAGGCCGatatcaacttaTCACGAGGTC -3'

antisense

Primers used for
chromatin
immunoprecipitation in
the MMP11 promoter

Distant region sense 5'- GACACTGAAGCAGGCATT -3'

Distant region antisense 5'- CGGATCACAAGAGACCATC -3'

Binding site 1 sense 5'- CTTGCAGTGAGCCGAGAT -3'

Binding site 1 antisense 5'- AGAAGGAAAGGAGGTGAGAC -3'

Binding site 2, 3 sense 5'- ATGGAGTCTCGCTCTGTC -3'

Binding site 2, 3 antisense 5'- GAAGGATGCTGTCAGAAGG -3'

ShRNA sequences

LV-shTCF3-1 sense 5'-CCGGCAGCCTCTCTTCATCCACATTCTCGAGAATGTGGATGAAGAGAGGCTGTTTTTG-3'

LV-shTCF3-1 antisense 5'-

AATTCCAAAAACAGCCTCTCTTCATCCACATTCTCGAGAATGTGGATGAAGAGAGGCT-3'

LV-shTCF3-2 sense 5'- CCGGCCCCGGATCACTCAAGCAATAACTCGAGTTATTGCTTGAGTGATCCGGGTTTTTG-3'

LV-shTCF3-2 antisense 5'-

AATTCCAAAAACCCGGATCACTCAAGCAATAACTCGAGTTATTGCTTGAGTGATCCGG-3'

LV-shMMP11-1 sense 5'-CCGGGATGTCCACTTCGACTATGATCTCGAGATCATAGTCGAAGTGGACATCTTTTTG-3'

LV-shMMP11-1 antisense 5'-AATTCCAAAAAGATGTCCACTTCGACTATGATCTCGAGATCATAGTCGAAGTGGACAT-3'

LV-shMMP11-2 sense	5'-CCGGCGCCTTCTACACCTTTCGCTACTCGAGTAGCGAAAGGTGTAGAAGGCGTTTTTG-3'
LV-shMMP11-2 antisense	5'-AATTCCAAAAACGCCTTCTACACCTTTCGCTACTCGAGTAGCGAAAGGTGTAGAAGGC-3'
LV-shControlsense	5'-ccggTTCTCCGAACGTGTCACGTCTCGAGACGTGACACGTTCCGAGAATTTTTg-3'
LV-shControlantisense	5'-aattcAAAAATTCTCCGAACGTGTCACGTCTCGAGACGTGACACGTTCCGAGAA-3'

MMP11: Matrix metalloproteinase-11; Sh: Short hairpin; TCF3: Transcription factor 3.

Supplementary Table 2 List of genes differentially expressed in Hep3B-transcription factor 3 *vs* Hep3B-control cells using a human tumor Metastasis RT² Profiler PCR Array

Symbol	Hep3B-transcription factor 3/Hep3B-control	Description
MMP11	5.803169	Matrix metalloproteinase 11 (stromelysin 3)
CTSL	4.929974	Cathepsin L1
ETV4	4.875143	Ets variant 4
KRAS	3.527235	V-Ki-ras2 Kirsten rat sarcoma viral oncogene homolog
HRAS	3.258437	V-Ha-ras Harvey rat sarcoma viral oncogene homolog
B2M	3.210097	Beta-2-microglobulin
TSHR	3.117362	Thyroid stimulating hormone receptor
SYK	3.102342	Spleen tyrosine kinase
IGF1	3.040211	Insulin-like growth factor 1 (somatomedin C)

MMP13	3.038544	Matrix metallopeptidase 13 (collagenase 3)
SET	3.038133	SET nuclear oncogene
SRC	2.952675	V-src sarcoma (Schmidt-Ruppin A-2) viral oncogene homolog (avian)
MMP9	2.603238	Matrix metallopeptidase 9 (gelatinase B, 92 kDa gelatinase, 92 kDa type IV collagenase)
ITGB3	2.122566	Integrin, beta 3 (platelet glycoprotein IIIa, antigen CD61)
RPSA	2.12027	Ribosomal protein SA
MET	2.000382	Met proto-oncogene (hepatocyte growth factor receptor)
MMP2	1.89206	Matrix metallopeptidase 2 (gelatinase A, 72 kDa gelatinase, 72 kDa type IV collagenase)
PLAUR	1.888349	Plasminogen activator, urokinase receptor
CXCR2	1.823738	Chemokine (C-X-C motif) receptor 2
RPLP0	1.736422	Ribosomal protein, large, P0
CST7	1.516215	Cystatin F (leukocystatin)
APC	1.516204	Adenomatous polyposis coli
EPHB2	1.413144	EPH receptor B2
FLT4	1.330847	Fms-related tyrosine kinase 4
RORB	1.267176	RAR-related orphan receptor B
IL18	1.244491	Interleukin 18 (interferon-gamma-inducing factor)

TIMP4	1.238329	TIMP metalloproteinase inhibitor 4
CDH11	1.222958	Cadherin 11, type 2, OB-cadherin (osteoblast)
TP53	1.161222	Tumor protein p53
SMAD4	1.112388	SMAD family member 4
NR4A3	1.111114	Nuclear receptor subfamily 4, group A, member 3
MYC	1.033829	V-myc myelocytomatosis viral oncogene homolog (avian)
CDKN2A(p16)	1.020416	Cyclin-dependent kinase inhibitor 2A (melanoma, p16, inhibits CDK4)
TRPM1	1.005413	Transient receptor potential cation channel, subfamily M, member 1
METAP2	1.004915	Methionyl aminopeptidase 2
FXRD5	1.004866	FXRD domain containing ion transport regulator 5
CTNNA1	1.004558	Catenin (cadherin-associated protein), alpha 1, 102 kDa
TIMP3	1.004087	TIMP metalloproteinase inhibitor 3
PNN	1.003893	Pinin, desmosome associated protein
FAT1	1.003698	FAT tumor suppressor homolog 1 (Drosophila)
KISS1	1.003266	KISS-1 metastasis-suppressor
CDH1(E-cadherin)	1.002999	Cadherin 1, type 1, E-cadherin (epithelial)
KISS1R	1.002721	KISS1 receptor
IL1B	1.002542	Interleukin 1, beta

MTSS1	1.001997	Metastasis suppressor 1
BRMS1	1.000768	Breast cancer metastasis suppressor 1
TIMP2	-1.00969	TIMP metalloproteinase inhibitor 2
NF2	-1.00983	Neurofibromin 2 (merlin)
TNFSF10	-1.00986	Tumor necrosis factor (ligand) superfamily, member 10
CTBP1	-1.12847	C-terminal binding protein 1
HPSE	-1.18492	Heparanase
VEGFA	-1.18649	Vascular endothelial growth factor A
COL4A2	-1.19047	Collagen, type IV, alpha 2
CXCR4	-1.20391	Chemokine (C-X-C motif) receptor 4
CHD4	-1.27096	Chromodomain helicase DNA binding protein 4
MMP7	-1.42336	Matrix metalloproteinase 7 (matrilysin, uterine)
MTA1	-1.4516	Metastasis associated 1
FN1(fibronectin)	-1.55503	Fibronectin 1
CXCL12	-1.58334	Chemokine (C-X-C motif) ligand 12
MMP10	-1.61165	Matrix metalloproteinase 10 (stromelysin 2)
MCAM	-1.62911	Melanoma cell adhesion molecule
TGFB1	-1.67975	Transforming growth factor, beta 1
FGFR4	-1.71472	Fibroblast growth factor receptor 4

CCL7	-1.71564	Chemokine (C-C motif) ligand 7
ITGA7	-1.76771	Integrin, alpha 7
HGF	-1.78205	Hepatocyte growth factor (hepapoietin A; scatter factor)
SMAD2	-1.81952	SMAD family member 2
CTSK	-1.85731	Cathepsin K
NME4	-1.92557	Non-metastatic cells 4, protein expressed in
TCF20	-1.92803	Transcription factor 20 (AR1)
ACTB	-1.92907	Actin, beta
SSTR2	-1.9299	Somatostatin receptor 2
PTEN	-2.32518	Phosphatase and tensin homolog
MGAT5	-2.3345	Mannosyl (alpha-1,6-)-glycoprotein beta-1,6-N-acetyl- glucosaminyltransferase
SERPINE1	-2.36314	Serpin peptidase inhibitor, clade E (nexin, plasminogen activator inhibitor type 1), member 1
GNRH1	-2.64297	Gonadotropin-releasing hormone 1 (luteinizing-releasing hormone)
NME1	-2.77507	Non-metastatic cells 1, protein (NM23A) expressed in
MDM2	-2.80002	Mdm ² p53 binding protein homolog (mouse)
CDH6	-2.87875	Cadherin 6, type 2, K-cadherin (fetal kidney)
CD82	-2.90771	CD82 molecule

HPRT1	-2.9407	Hypoxanthine phosphoribosyltransferase 1
CD44	-2.94449	CD44 molecule (Indian blood group)
HTATIP2	-3.10121	HIV-1 Tat interactive protein 2, 30kDa
GAPDH	-3.13434	Glyceraldehyde-3-phosphate dehydrogenase
MMP3	-3.18465	Matrix metalloproteinase 3 (stromelysin 1, progelatinase)
RB1	-3.27727	Retinoblastoma 1
MYCL	-3.28467	V-myc myelocytomatosis viral oncogene homolog 1, lung carcinoma derived (avian)
EWSR1	-3.35678	Ewing sarcoma breakpoint region 1
DENR	-3.40619	Density-regulated protein
