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## **Supplemental Information**

### **YTHDF1 Facilitates the Progression of Hepatocellular Carcinoma by Promoting FZD5 mRNA Translation in an m6A-Dependent Manner**

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Fig. S1 YTHDF1 is upregulated in HCC and associated with prognosis of patients, related to Figure 1. **a** H-score of YTHDF1 in HCC tissues and ANTs. **b** Detection of YTHDF1 protein levels in HCC cell lines and LO2 cells by western blot. **c** Kaplan-Meier survival curves of OS, RFS, PFS, and DSS of HCC patients based on YTHDF1 expression in Kaplan-Meier Plotter database.

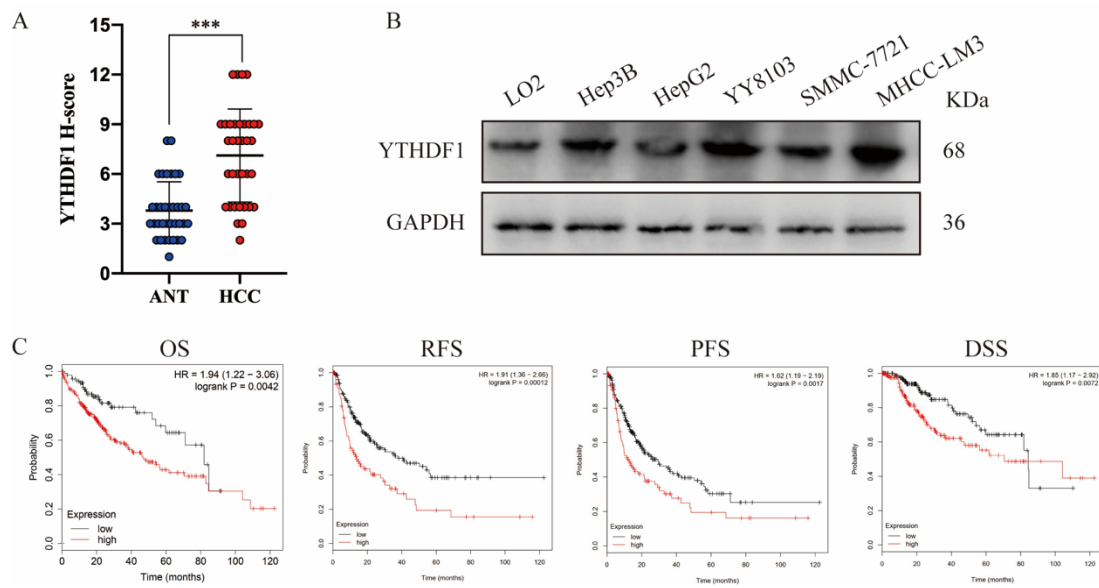
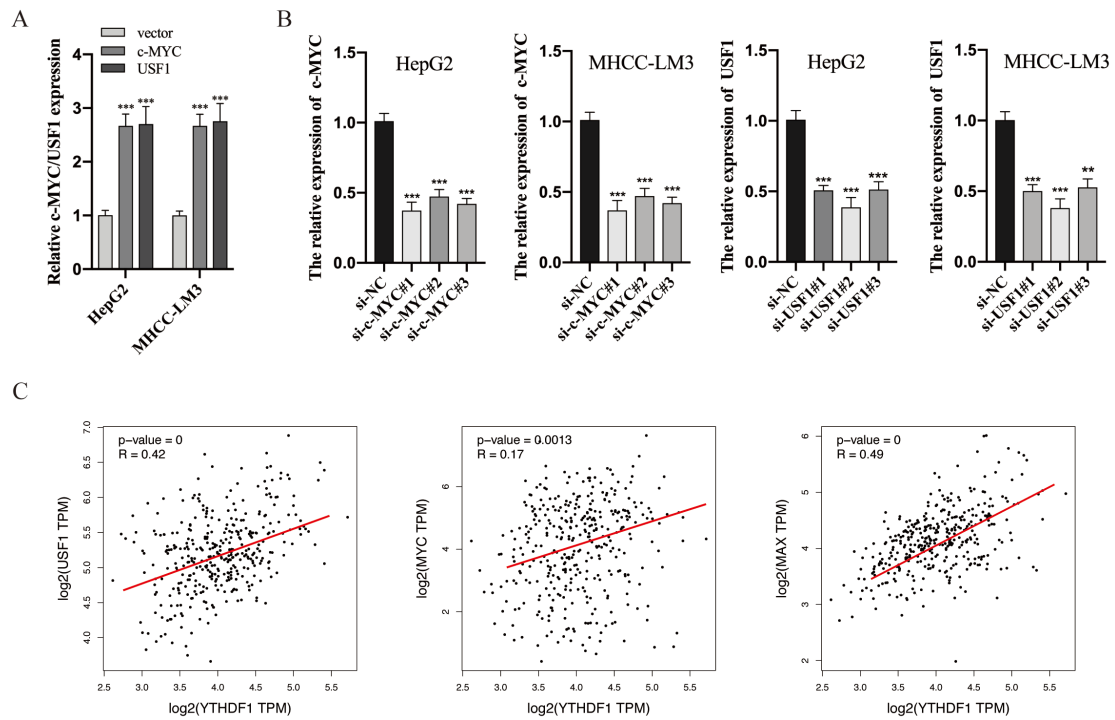


Fig. S2 c-MYC and USF1 promote YTHDF1 expression, related to Figure 2. **a** qRT-PCR analysis of the effect of transfecting c-MYC and USF1 overexpression vectors in HepG2 and MHCC-LM3 cells. **b** qRT-PCR analysis of inhibitory effect of si-c-MYC and si-USF1 in HepG2 and MHCC-LM3 cells. **c** c-MYC/MAX and USF1 expression was positively correlated with YTHDF1 expression in HCC based on TCGA database.



**Table S1:** The correlations between YTHDF1 expression and clinical characteristics of HCC patients ( $n = 84$ ).

Clinicopathologic parameters	Number	YTHDF1 expression		P value
		High	Low	
Gender				
Male	53	28	25	
Female	31	14	17	0.65
Age (years)				
≥60	47	24	23	
<60	37	18	19	0.91
HBV surface antigen				
Negative	27	12	15	
Positive	57	30	27	0.61
Cirrhosis				
Absent	27	11	16	
Present	57	31	26	0.36
Tumor size (cm)				
≥5	45	28	17	
<5	39	14	25	0.028
Differentiation				
High + moderate	41	19	22	
Low	43	23	20	0.67
TNM stage				
I+II	65	27	36	
III+IV	19	15	6	0.042
Barcelona stage				
A+B	60	26	34	
C+D	24	16	8	0.09
Microvascular invasion				
Absent	40	14	26	
Present	44	28	16	0.016

**Table S2:** RNA sequences used in this study.

<b>Name</b>	<b>Sequence (5' - 3')</b>	<b>Application</b>
si-myc#1	GCGAGGATATCTGGAAGAA	siRNA target site
si-myc#2	CGATGTTGTTTCTGTGGAA	siRNA target site
si-myc#3	GGCGAACACACAACGUCUU	siRNA target site
si-USF1#1	GCTGGACAATGACGTGCTT	siRNA target site
si-USF1#2	GACGACTCGGGATGAGAAA	siRNA target site
si-USF1#3	CGCCGAGACAAGATCAACA	siRNA target site
si-METTTL3#1	GCACTTGGATCTACGGAAT	siRNA target site
si-METTTL3#2	CGACTACAGTAGCTGCCTT	siRNA target site
si-METTTL3#3	CAGTGGATCTGTTGTGATA	siRNA target site
sh-YTHDF1	CGAAAGAGTTTGAGTGGAA	shRNA target site
sg-YTHDF1	CACGCCCCACGTGACTTCCC	CRISPR/dcas9 target site

**Table S3:** Primer sequences used in this study.

<b>Name</b>	<b>Sequence (5'-3')</b>	<b>Application</b>
YTHDF1- forward	GACATTGGCACCTGGGATA	qRT-PCR
YTHDF1-Reverse	GCTCTGATACTGCGGTTGAG	qRT-PCR
FZD5- forward	GTCACACCCGCTCTACAACA	qRT-PCR
FZD5-Reverse	GGACGTGGAGATGAAGCACA	qRT-PCR
USF1- forward	GTGATCCAGGGTGCTTTCAC	qRT-PCR
USF1-Reverse	CCTCTGAGCCCTGGGTAGTA	qRT-PCR
c-MYC- forward	CCACGAAACTTTGCCCATAG	qRT-PCR
c-MYC-Reverse	TGCAAGGAGAGCCTTTCAGAG	qRT-PCR
METTL3- forward	CTATCTCCTGGCACTCGCAAGA	qRT-PCR
METTL3-Reverse	GCTTGAACCGTGCAACCACATC	qRT-PCR
GAPDH- forward	GAACGGGAAGCTCACTGG	qRT-PCR
GAPDH-Reverse	GCCTGCTTCACCACCTTCT	qRT-PCR
FZD5 m6A- forward	GGCGCTGAGCTCCGTGGAC	MeRIP-qRT-PCR
FZD5 m6A -Reverse	CGGCGCGTCGCTCAATCC	MeRIP-qRT-PCR
YTHDF1 promoter- forward 1	CGCGACCTGGAACACGC	ChIP-qRT-PCR
YTHDF1 promoter- Reverse 1	AGGTCCGAAGACTAAACGC	ChIP-qRT-PCR
YTHDF1 promoter- forward 2	TGTGTGGGCGGCTGTA	ChIP-qRT-PCR
YTHDF1 promoter- Reverse 2	TCCCAGTCTCGTGCGG	ChIP-qRT-PCR

**Table S4:** Antibodies used in this study.

<b>Antibody</b>	<b>Company</b>	<b>Application</b>	<b>Concentration</b>
YTHDF1	Proteintech (17479-1-AP)	WB	1:1000
		IHC	1:100
		RIP	2.0 µg
FZD5	Abcam (ab75234)	WB	1:500
		IHC	4.0 µg
c-MYC	Proteintech (10828-1-AP)	WB	1:2000
		ChIP	2.0 µg
	Abcam (ab32072)	IHC	1:100
USF1	SANTA CRUZ (sc-390027)	IHC	1:50
		ChIP	2.0
β-catenin	Proteintech (51067-2-AP)	WB	1:2000
		IHC	1:1000
GAPDH	Proteintech (60004-1-Ig)	WB	1:5000
β-actin	Proteintech (60008-1-Ig)	WB	1:5000
HRP-conjugated Affinipure Goat Anti-Mouse IgG(H+L)	Proteintech (SA00001-1)	WB	1:10000
HRP-conjugated Affinipure Goat Anti-Rabbit IgG(H+L)	Proteintech (SA00001-2)	WB	1:10000