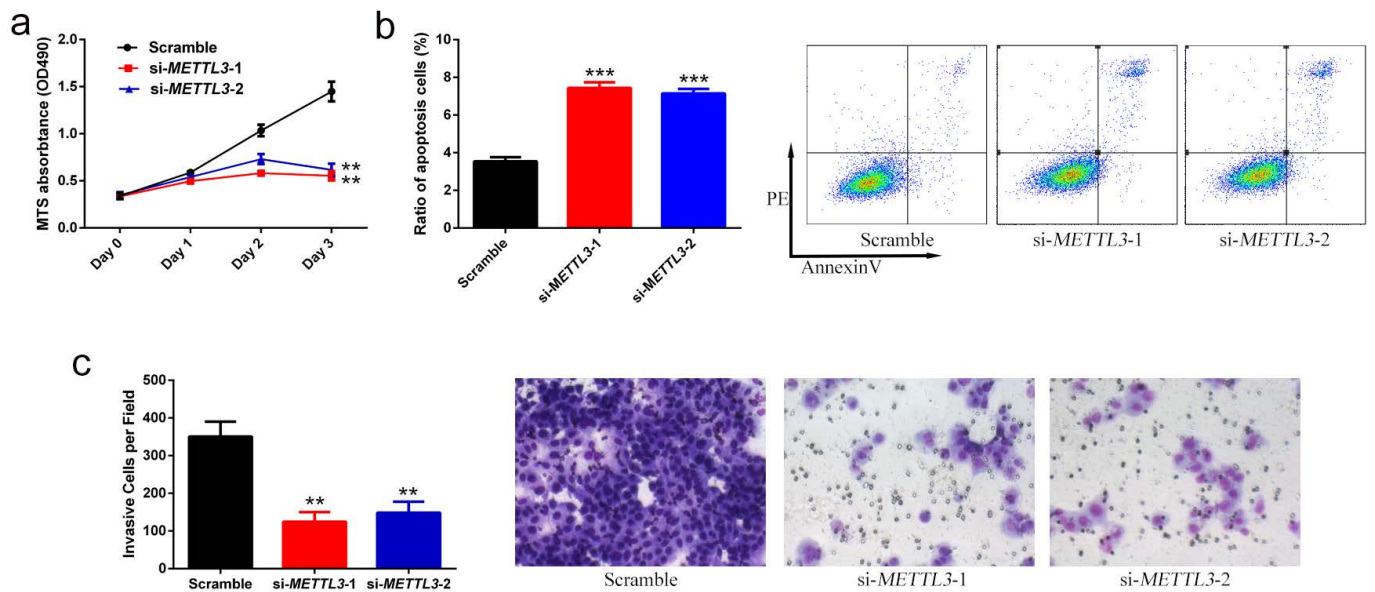
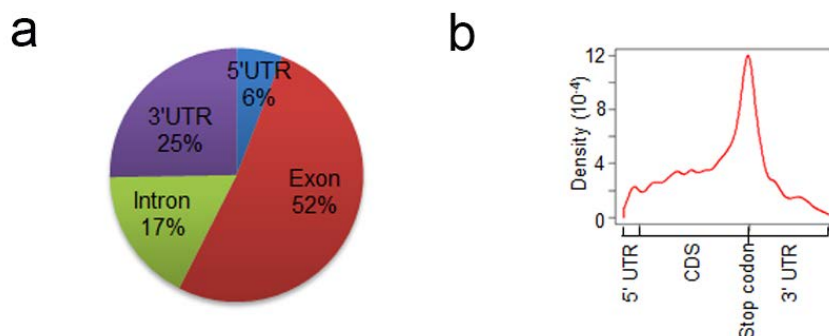


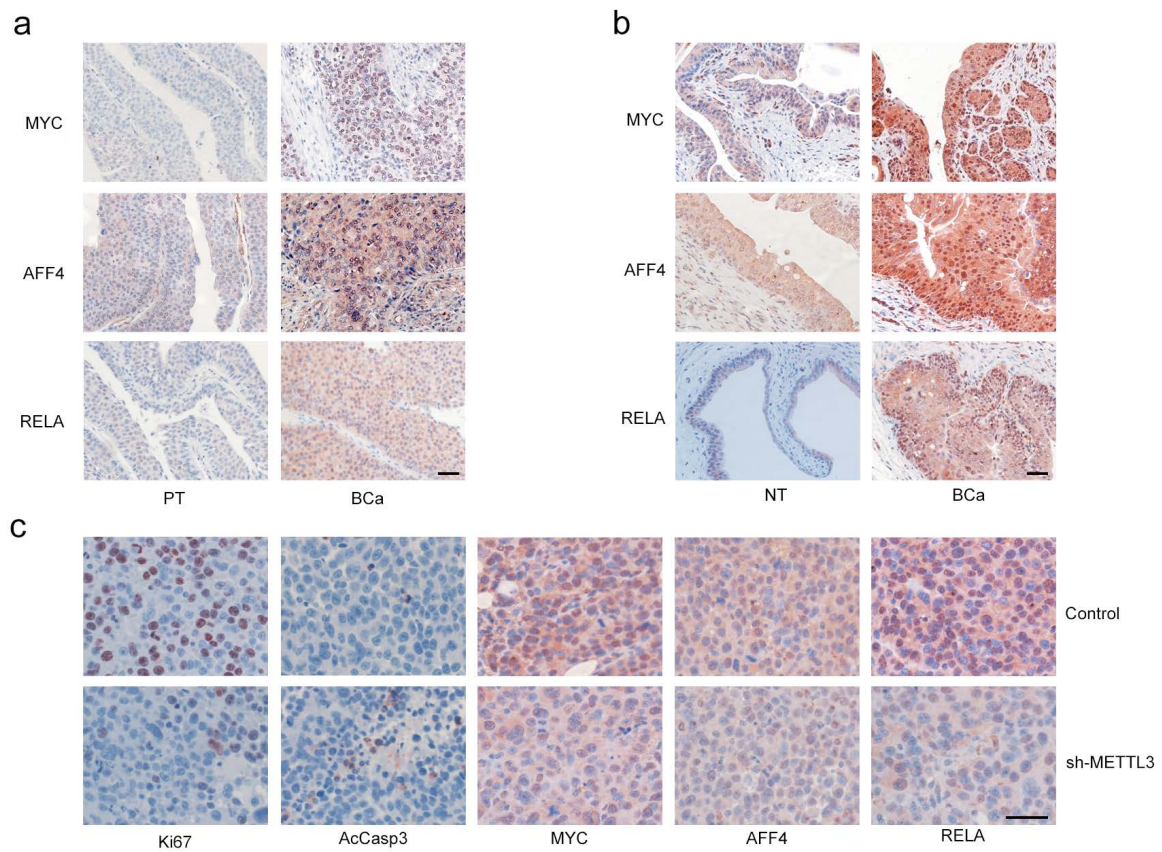
Supplementary Files:



Supplementary Figure S1 Knockdown of METTL3 suppressed UM-UC-3 cell proliferation, invasion and promote its apoptosis. a, MTS assay of cellular proliferation in UM-UC-3 cells (**, $P < 0.01$ Two Way ANOVA). b, Annexin V/PI staining of METTL3 knockdown and control UM-UC-3 cells analyzed by FACS. Quantification of apoptotic cells were plotted, numbers represent the sum of early and late apoptotic cells (***, $P < 0.001$ relative to scramble, One Way ANOVA). f, *In vitro* cell invasion assay, Quantification of invasive UM-UC-3 cells (**, $P < 0.01$ relative to scramble, One-way ANOVA). Bar graphs: mean $\pm$ S.D., $n = 3$.



Supplementary Figure S2. m6A features in 5637 cells. a, Pie chart representing m6A peak distribution. b, Metagene analyses revealed that the m6A sites were predominantly localized near the translation stop codons.



Supplementary Figure S3. The tumor tissues from METTL3 stable knockdown xenografts (a, related to Figure 6f), human BCa samples (b, related to Figure 6g) and mouse BCa samples (c, related to Figure 6h) were fixed and immunostained with the indicated antibodies. Scale bar. 50µm.

Supplementary Table S1. Pathways affected by METTL3 knockdown in BCa cells (Indicated by GSEA).

Pathways downregulated after METTL3 knockdown			
NAME	SIZE	ES	NOM p-val
HALLMARK_MYC_TARGETS_V1	198	-0.3586	0
HALLMARK_OXIDATIVE_PHOSPHORYLATION	197	-0.22598	0
HALLMARK_TNFA_SIGNALING_VIA_NFKB	189	-0.21794	0
HALLMARK_MYC_TARGETS_V2	58	-0.39205	0
HALLMARK_ALLOGRAFT_REJECTION	135	-0.2169	0
HALLMARK_KRAS_SIGNALING_UP	153	-0.19672	0
HALLMARK_MTORC1_SIGNALING	196	-0.17052	0
HALLMARK_E2F_TARGETS	198	-0.16644	0

HALLMARK_INFLAMMATORY_RESPONSE	156	-0.14065	0.004166667
HALLMARK_INTERFERON_ALPHA_RESPONSE	94	-0.1611	0.015810277
HALLMARK_DNA_REPAIR	140	-0.13468	0.006
HALLMARK_PROTEIN_SECRETION	95	-0.15089	0.021645023
HALLMARK_INTERFERON_GAMMA_RESPONSE	179	-0.10692	0.037037037
HALLMARK_UNFOLDED_PROTEIN_RESPONSE	109	-0.13277	0.043737575
HALLMARK_IL6_JAK_STAT3_SIGNALING	73	-0.15898	0.044989776
Pathways upregulated after METTL3 knockdown			
NAME	SIZE	ES	NOM p-val
HALLMARK_KRAS_SIGNALING_DN	116	0.192376	0
HALLMARK_MYOGENESIS	152	0.166367	0
HALLMARK_G2M_CHECKPOINT	197	0.140024	0.002061856
HALLMARK_MITOTIC_SPINDLE	195	0.126596	0.003960396
HALLMARK_PI3K_AKT_MTOR_SIGNALING	97	0.168131	0.008403362
HALLMARK_ESTROGEN_RESPONSE_LATE	173	0.126332	0.004291846
HALLMARK_EPITHELIAL_MESENCHYMAL_TRANSITION	185	0.12368	0.005988024
HALLMARK_GLYCOLYSIS	184	0.120416	0.015533981
HALLMARK_APICAL_JUNCTION	173	0.119838	0.020283977
HALLMARK_NOTCH_SIGNALING	28	0.277111	0.019193858
HALLMARK_COAGULATION	98	0.148076	0.041152265
HALLMARK_ADIPOGENESIS	181	0.109702	0.036538463
HALLMARK_HYPOXIA	179	0.106614	0.037698414
HALLMARK_HEDGEHOG_SIGNALING	32	0.24372	0.03954802
HALLMARK_APICAL_SURFACE	35	0.230663	0.036398467
HALLMARK_UV_RESPONSE_UP	142	0.114397	0.046464648
HALLMARK_PEROXISOME	92	0.135703	0.049462367

Supplementary Table S2. Patient characteristics used for xenograft generating.

Case	NO.1	NO.2
Gender	Male	Male
Age	55	70
Stage	TaNOm0	T3aNOm0
Grade	Non-invasive low grade urothelial carcinoma	High grade muscle invasive urothelial carcinoma

Supplementary Table S3 Information of Primers

	Primer Names	Sequences
For qRT PCR	hMETTL3 F	5'-CAAGCTGCACTTCAGACGAA-3'
	hMETTL3 R	5'-GCTTGGCGTGTGGTCTTT-3'
	hMYC F	5'-TTCGGGTAGTGGAAAACCAG-3'
	hMYC R	5'-AGTAGAAATACGGCTGCACC-3'
	hAFF4 F	5'-AAAGGCCAGCATGGATCAGAA-3'
	hAFF4 R	5'-GTGATTTGGAGCGTTGATGTTC-3'
	hIKBKB F	5'-CTGGCCTTTGAGTGCATCAC-3'
	hIKBKB R	5'-CGCTAACAACAATGTCCACCT-3'
	hRELA F	5'-GTGGGGACTACGACCTGAATG-3'
	hRELA R	5'-GGGGCACGATTGTCAAAGATG-3'
	hBCL2A1 F	5'-TACAGGCTGGCTCAGGACTAT-3'
	hBCL2A1 R	5'-CGCAACATTTTGTAGCACTCTG-3'
	hBIRC3 F	5'-TTTCCGTGGCTCTTATTCAAAC-3'
	hBIRC3 R	5'-GCACAGTGGTAGGAACCTCTCAT-3'
	hKITLG F	5'-AATCCTCTCGTCAAACTGAAGG-3'
	hKITLG R	5'-CCATCTCGCTTATCCAACAATGA-3'
	hMMP9 F	5'-TGTACCGCTATGGTTACACTCG-3'
	hMMP9 R	5'-GGCAGGGACAGTTGCTTCT-3'
	hPLAU F	5'-CCATCTACAGGAGGCACCG-3'
	hPLAU R	5'-GCGACCCAGGTAGACGATG-3'
	hGAPDH F	5'-AGGTCGGTGTGAACGGATTG-3'
	hGAPDH R	5'-GGGGTCGTTGATGGCAACA-3'
m6A-qPCR	MYC-m6A-F	5'-GCATACATCCTGTCCGTCCA-3'
	MYC-m6A-R	5'-GTCGTTTCCGCAACAAGTCC-3'
	AFF4-m6A-F	5'-GGAATAGTTTTCTCCTCGTTGGTTC-3'
	AFF4-m6A-R	5'-ATCAATGTGCTGCAGATTTAAAAGC-3'
	IKBKB-m6A-F	5'-CCAGGGGTCTCTGGTATCCAGATGG-3'
	IKBKB-m6A-R	5'-GAGGACCTCCAGTGTGCAGACAAGG-3'
	RELA-m6A-F	5'-GGGAGCCATATTTTATTCTTTTATTG-3'
	RELA-m6A-R	5'-TCAGGACAGGGGAAAAGTTTGAG-3'
CHIP-qPCR	MYC promoter F	5'-GCAATGCGTTGCTGGGTATT-3'
	MYC promoter R	5'-GCGTTCAGAGCGTGGGATGTT-3'
	MYC 10KB F	5'-AATTGCCTGCATTCACAGAGC-3'
	MYC 10KB R	5'-TCCATCCCATTATCTTGGTATCT-3'

Supplementary Table S4 Information of Antibodies

Protein target	Company	Catalog Number	Host species	Clonality
METTL3	Protein Tech	15073-1-AP	Rabbit	polyclonal
MYC	Protein Tech	10828-1-AP	Rabbit	polyclonal
AFF4	Protein Tech	14662-1-AP	Rabbit	polyclonal
RELA	Protein Tech	10745-1-AP	Rabbit	polyclonal

IKBKB	Protein Tech	15649-1-AP	Rabbit	polyclonal
α -Tubulin	Protein Tech	11224-1-AP	Rabbit	polyclonal
Ki67	Thermo Fisher	PA5-19462	Rabbit	polyclonal
Active Caspase-3	Abcam	ab2302	Rabbit	polyclonal
