

Figure S1. 18S rRNA m⁶A level in indicated cells. (A and B) The 18S rRNA of indicated cells were isolated and quantified the m⁶A level by LC-MS/MS. Consistent data were obtained from three independent experiments. m⁶A, N⁶ methyladenosine; METTL5, methyltransferase N⁶-adenosine; sh, short hairpin RNA; OE, overexpression vector.

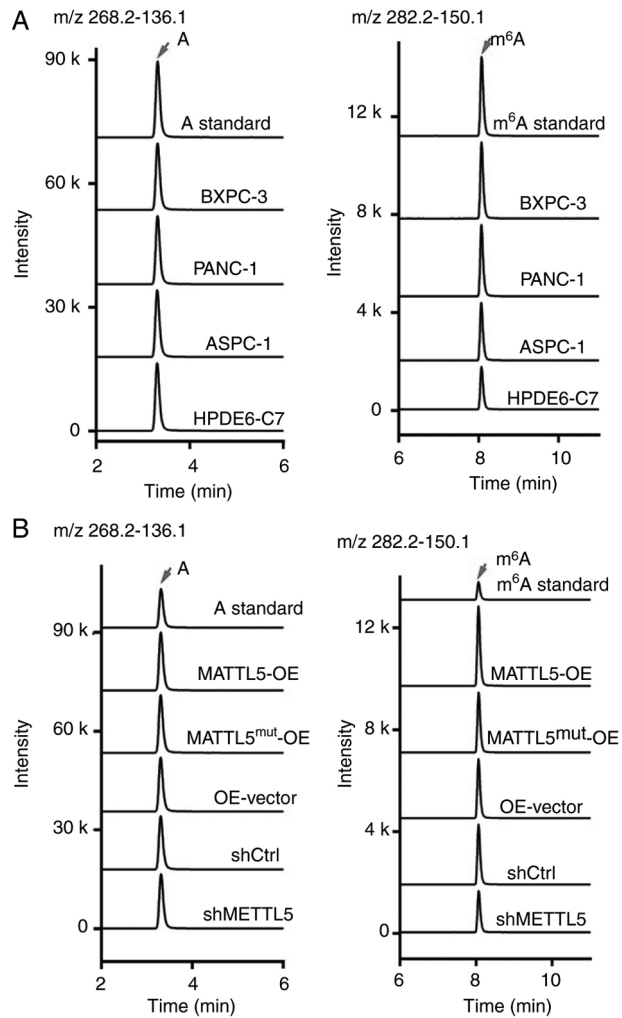


Figure S2. Oncogenic role of METTL5 and c-Myc in pancreatic cancer cells. (A) Transwell assays of BXPC-3 cells overexpressing vector and METTL5. (B) Proliferation of BXPC-3 cells overexpressing vector and METTL5. (C) Transwell assays of ASPC-1 cells overexpressing vector and METTL5. (D) Proliferation of ASPC-1 cells overexpressing vector and METTL5. (E) and (F) c-Myc knockdown abolished the increase in (E) invasion and (F) cell proliferation mediated by overexpression of METTL5 in PANC-1 cells. Data are shown as the mean $\pm$ SD (n=3). *P<0.05, **P<0.01 and ***P<0.001. METTL5, methyltransferase N6-adenosine; sh, short hairpin RNA; OE, overexpression vector.

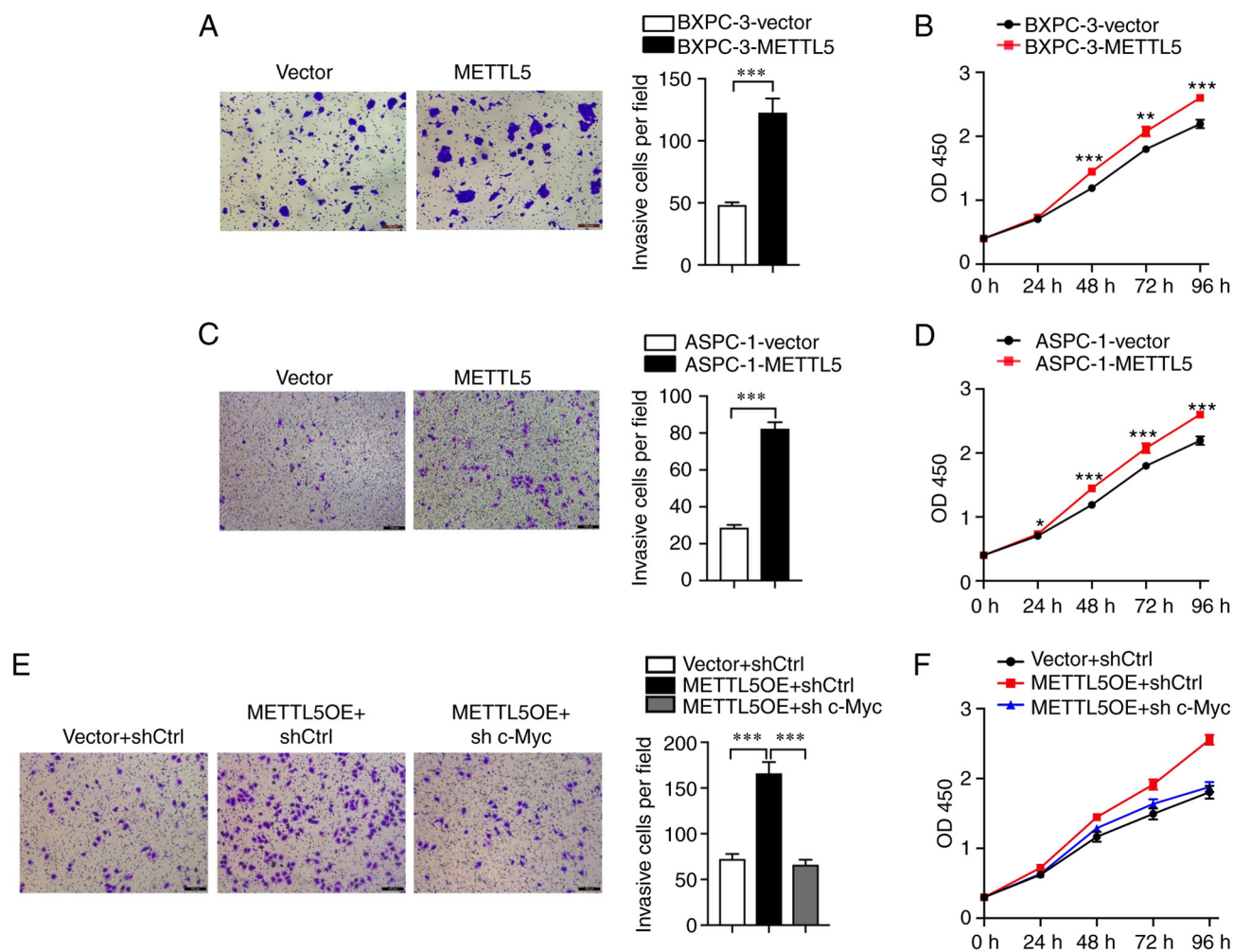


Figure S3. Protein levels of p53, Oct4, E-cadherin, N-cadherin, vimentin, snail and slug in METTL5-overexpressing PANC-1 cells and control cells. METTL5, methyltransferase N6-adenosine.

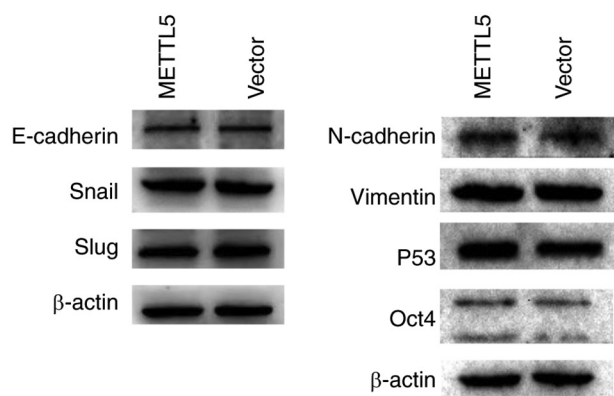


Figure S4. METTL5 and TRMT112 may function together in pancreatic cancer. The combination of TRMT112 and METTL5 overexpression further enhanced the increase in (A) cell proliferation, (B) colony formation capacity, (C) migration and (D) invasion capacity compared with TRMT112 overexpression alone in PANC-1 cells. Data are shown as the mean $\pm$ SD (n=3). *P<0.05, **P<0.01, ***P<0.001 and ****P<0.0001. METTL5, methyltransferase N6-adenosine; TRMT112, multifunctional methyltransferase subunit TRMT112-like protein; OE, overexpression vector.

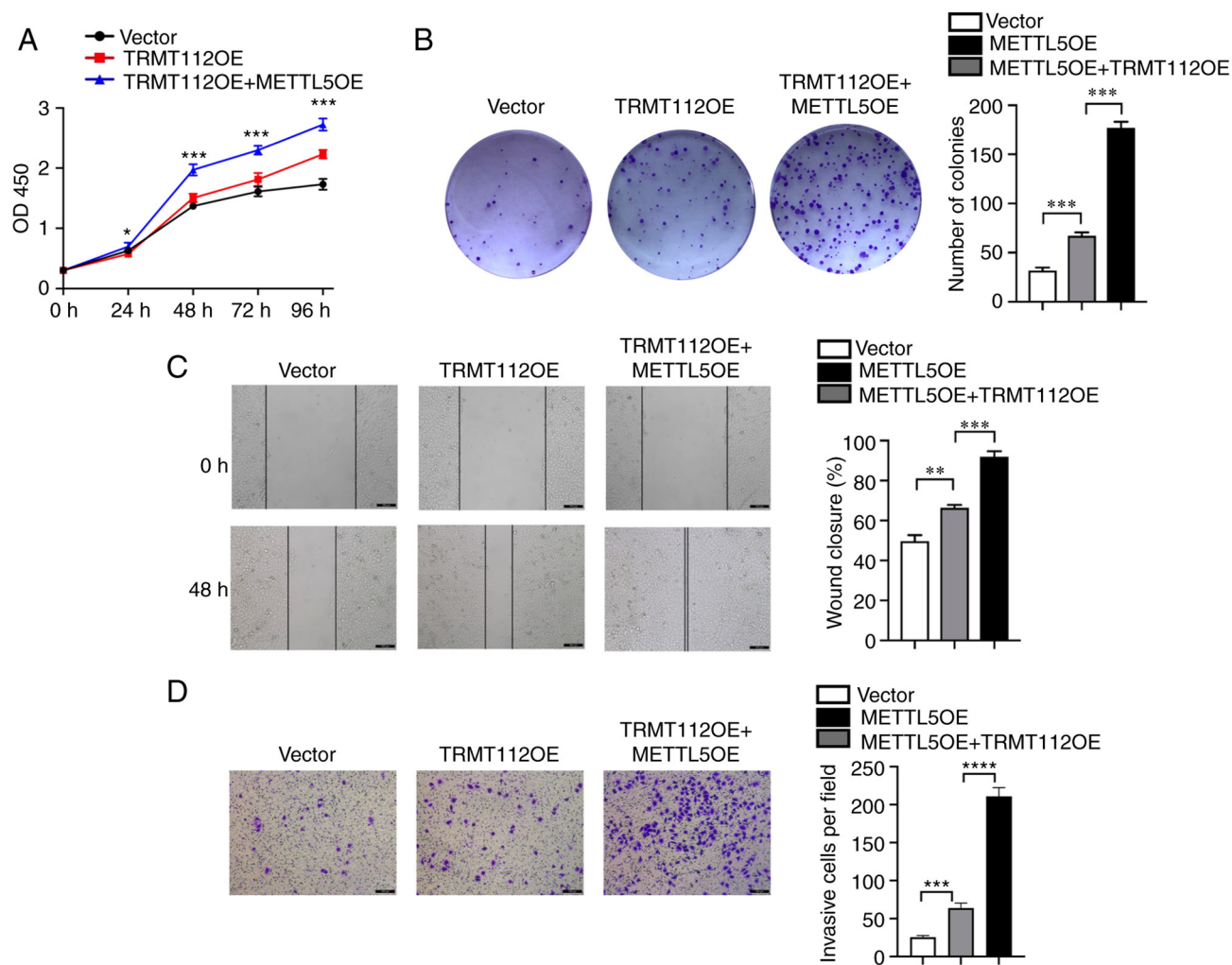


Figure S5. METTL5 knockdown abolishes the TRMT112 function in pancreatic cancer. METTL5 knockdown abolished the increase in (A) cell proliferation, (B) colony formation capacity, (C) migration and (D) invasion induced by overexpression of TRMT112 in PANC-1 cells. Data are shown as the mean $\pm$ SD (n=3). *P<0.05, **P<0.01 and ***P<0.001. METTL5, methyltransferase N6-adenosine; TRMT112, multifunctional methyltransferase subunit TRMT112-like protein; sh, short hairpin RNA; OE, overexpression vector.

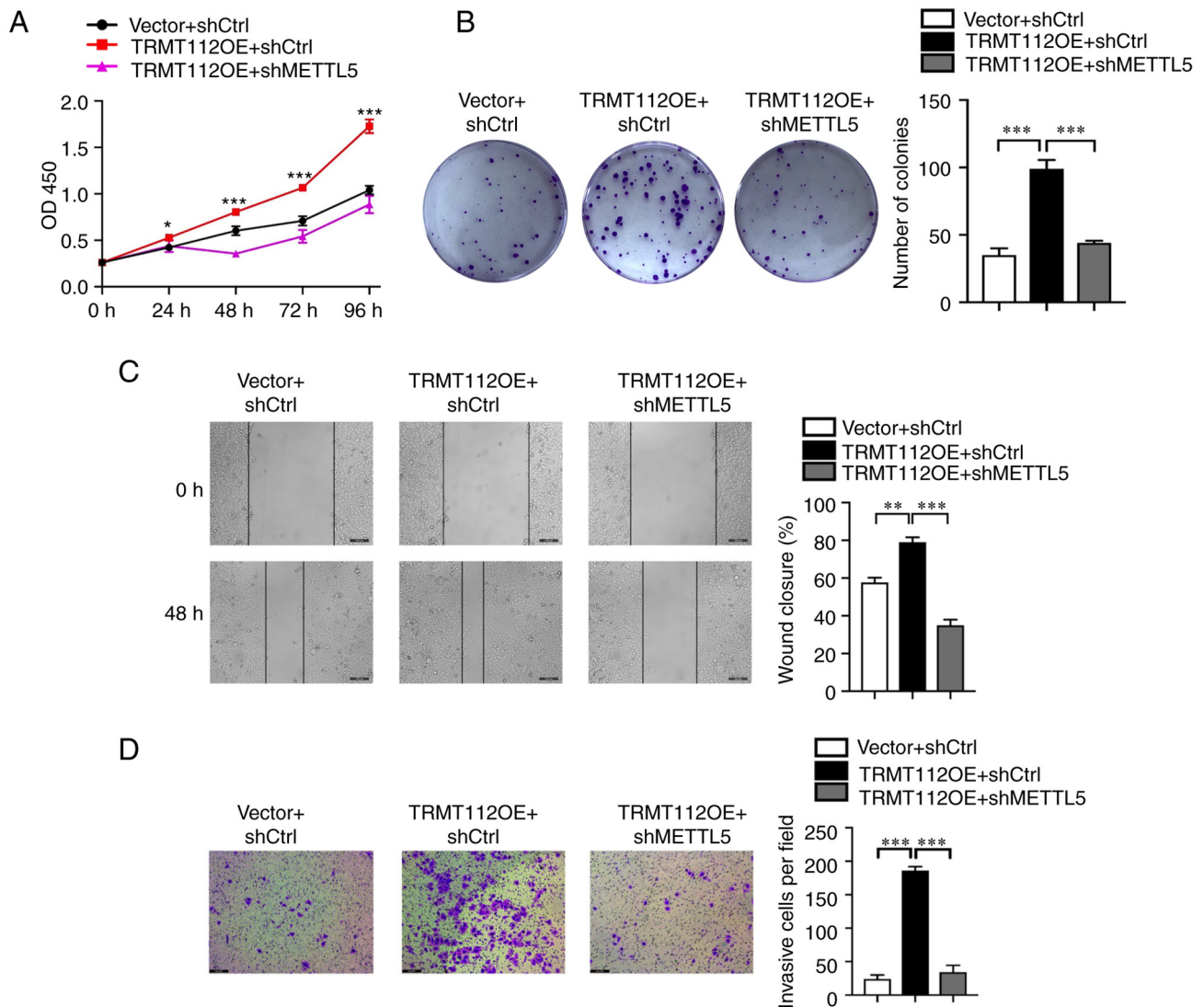


Table SI. shRNA sequences for target gene knockdown.

shRNA name	shRNA sequence (5'→3')
shMETTL5-1	GTCATTTCGATACAGTAATTAT
shMETTL5-2	TTGCAGCATGTATGCTCTATA
shMETTL5-3	ATGTGATGTGTGCTTATTATC
shTRMT112-1	AACTTCGTGGCGCGTATGATA
shTRMT112-2	CCCAATGACACCAAACACAGT
Scramble shRNA	CCTAAGGTTAAGTCGCCCTCG

shRNA/sh, short hairpin RNA; METTL5, methyltransferase N6-adenosine; TRMT112, multifunctional methyltransferase subunit TRMT112-like protein.

Table SII. Primer sequences for quantitative PCR.

Primer name	Primer sequence (5'→3')
β-actin	F: CATGTACGTTGCTATCCAGGC R: CTCCTTAATGTCACGCACGAT
METTL5	F: AAGGAACTAGAGAGTCGCCTG R: GCGGCCTGGTAGGATACTG
F, forward; R, reverse; METTL5, methyltransferase N6-adenosine.	

Table SIII. Characteristics of antibodies used in the present study.

Antigen or description	Cat. no.	Cat. no. and supplier	Dilution
Primary antibodies			
METTL5	16791-1-AP	ProteinTech Group, Inc.	1:1,000
c-Myc	18583S	Cell Signaling Technology, Inc.	1:1,000
Snail	3879S	Cell Signaling Technology, Inc.	1:1,000
OCT4	2750S	Cell Signaling Technology, Inc.	1:1,000
TP53	2527S	Cell Signaling Technology, Inc.	1:1,000
E-cadherin	3195S	Cell Signaling Technology, Inc.	1:1,000
N-cadherin	4061S	Cell Signaling Technology, Inc.	1:1,000
Vimentin	5741S	Cell Signaling Technology, Inc.	1:1,000
Slug	9585S	Cell Signaling Technology, Inc.	1:1,000
TRMT112	Sc-398481	Santa Cruz Biotechnology, Inc.	1:5,00
β -actin	4970S	Cell Signaling Technology, Inc.	1:1,000
Secondary antibodies			
Goat anti-rabbit IgG antibody (HRP)	orb43514	Biorbyt	1:5,000
Goat anti-mouse IgG (H + L) antibody (HRP)	orb688922	Biorbyt	1:5,000

HRP, horseradish peroxidase; METTL5, rRNA N6-adenosine-methyltransferase METTL5; TRMT112, multifunctional methyltransferase subunit TRMT112-like protein.

Table SIV. dRfxCas13d gRNA sequences for targeting c-Myc mRNA.

Name	gRNA sequence (5'→3')
gRNA-1	GGGTGTTGTAAGTTCCAGTGCAAAGT GCCCCG
gRNA-2	GGCCGAGAAGCCGCTCCACATACAGT CCTGG
gRNA-3	TACTTTTCCTTACGCACAAGAGTTCCG TAGC
gRNA-4	TATGCCCAAAGTCCAATTTGAGGCAGT TTAC
gRNA, guide RNA.	