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Supplemental information

A diagnostic and predictive lncRNA *lnc-MPEG1-1* promotes the proliferation and metastasis of papillary thyroid cancer cells by occupying miR-766-5p

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Supplementary Table 1 Differentially expressed lncRNAs in the discovery stage

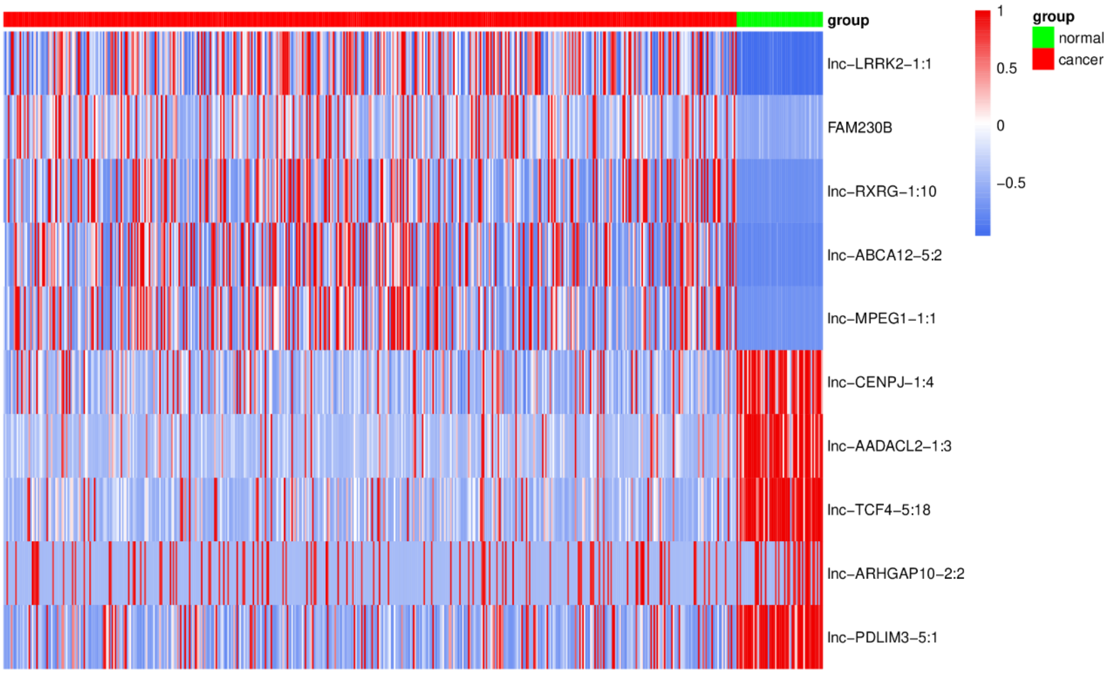
Supplementary table 2. Univariate and multivariate logistic regression analysis of the hub lncRNA and other clinicopathological factors for LN metastasis in TCGA cohort

Risk factors	Univariate analysis		Multivariate analysis	
	OR(95%CI)	P value	OR(95%CI)	P value
Age	0.99(0.98–1.00)	0.04	0.98(0.96–0.99)	2.60×10^{-3}
T stage				
T1				
T2	1.71(1.05–2.81)	0.03	1.96（1.14–3.40)	0.02
T3	3.03(1.87–4.97)	8.07×10^{-6}	3.37(1.96–5.87)	1.33×10^{-5}
T4	8.90(3.10–32.24)	1.77×10^{-4}	10.43(3.35–40.23)	1.60×10^{-4}
Histological type				
Papillary adenocarcinoma				
Papillary carcinoma, columnar cell	1.26(0.63–2.60)	0.52	0.87(0.40–1.93)	0.72
Papillary carcinoma, follicular variant	0.17(0.09–0.31)	2.47×10^{-8}	0.38(0.18–0.78)	9.72×10^{-3}
lnc-LRRK2-1:1				
Low				
High	4.49(2.47–8.67)	2.30×10^{-6}		
FAM230B				
Low				
High	3.95(2.28–7.13)	2.01×10^{-6}		
lnc-RXRG-1:10				
Low				
High	4.44(2.78–7.27)	1.01×10^{-9}		
lnc-MPEG1-1:1				
Low				
High	4.02(2.71–6.03)	8.65×10^{-12}	1.79(1.04–3.09)	3.61×10^{-2}
lnc-ABCA12-5:2				
Low				
High	6.26(3.94–10.23)	3.80×10^{-14}	2.84(1.51–5.40)	1.28×10^{-3}

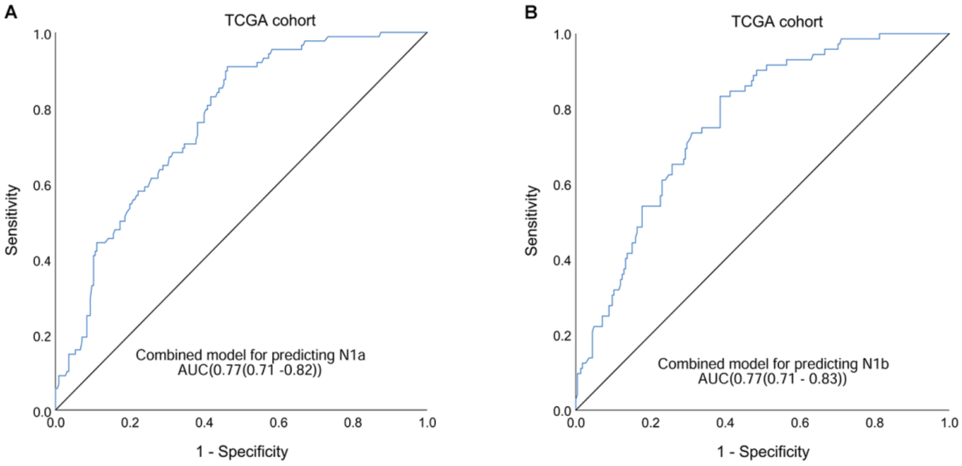
Supplementary table 3. The sequences of all primers in RT-qPCR assay

ID	Sequence (5’- 3’)
GAPDH (Forward)	TGTTCGTCATGGGTGTGAAC
GAPDH (Reverse)	ATGGCATGGACTGTGGTCAT
U6 F	CTCGCTTCGGCAGCACA
U6 R	AACGCTTCACGAATTTGCGT
Lnc-MPEG1-1 F	CTCTTCCACTTAATGCCTGTT
Lnc-MPEG1-1 R	GTTTGAATCCCTGCTCCAC
FAM230B.F	GCACACCAACCAGTGTAGGA
FAM230B.R	AGAGTGGGCTGAAGCTTGTC
lnc-RXRG-1:10.F	AATCCTGGCACTGTTACTT
lnc-RXRG-1:10.R	CCTTCCCTATGTCTTTCG
LNC-MPEG1-1:1.F	TCTGAATTTCGCTTGCTC
LNC-MPEG1-1:1.R	AGGTTTGGTGGATGTCTT
lnc-ABCA12-5:2.F	AGAGCCCGTGTCATTCCT
lnc-ABCA12-5:2.R	TGCCTCTGCTGGTCTTTC
lnc-CENPJ-1:4.F	CTCCCGGTTCAAGTGATT
lnc-CENPJ-1:4.R	TCTCCGAGTGCGAGTTTT
LNC-AADACL2-1:3.F	CTCATAGAAGACCCAGAT
LNC-AADACL2-1:3.R	GTATTCCCTACAGAACCT
lnc-TCF4-5:18.F	TCCTCGCCTACTCCACAT
lnc-TCF4-5:18.R	GTTCACGCCATTCTCCTG
lnc-ARHGAP10-2:2.F	TGCTCACCGATCTCTCAATCTG
lnc-ARHGAP10-2:2.R	CTGGCCTTGGAACGTCACTA
lnc-PDLIM3-5:1.F	CAAAAGCCACTCCGACAACA
lnc-PDLIM3-5:1.R	TCCTTGGCTCAGACCGAAAT
Fibronectin F	CGGTGGCTGTCAGTCAAAG
Fibronectin R	AAACCTCGGCTTCCTCCATAA
ITGB1 F	CCTACTTCTGCACGATGTGATG

ITGB1 R	CCTTTGCTACGGTTGGTTACATT
E-cadherin F	ATTTTTCCCTCGACACCCGAT
E-cadherin R	TCCCAGGCGTAGACCAAGA
Vimentin F	AGTCCACTGAGTACCGGAGAC
Vimentin R	CATTTCACGCATCTGGCGTTC
N-cadherin F	TGCGGTACAGTGTAACTGGG
N-cadherin R	GAAACCGGGCTATCTGCTCG
β-catenin F	GCAGTTCGCCTTCACTATGG
β-catenin R	ATGTGCACGAACAAGCAACT
miR-766-5p RT	CTCAACTGGTGTCGTGGAGTCGGCAATTCAGTTGAGAAGACCAG
miR-766-5p F	ACACTCCAGCTGGGAGGAGGAATTGGTGCTGG
miR-670-5p RT	CTCAACTGGTGTCGTGGAGTCGGCAATTCAGTTGAGCACCACAT
miR-670-5p F	ACACTCCAGCTGGGGTCCCTGAGTGTATGT
miR-579-3p RT	CTCAACTGGTGTCGTGGAGTCGGCAATTCAGTTGAGAATCGCGG
miR-579-3p F	ACACTCCAGCTGGGTTCATTTGGTATAAACCGC
miR-644-3p RT	CTCAACTGGTGTCGTGGAGTCGGCAATTCAGTTGAGTGTAGGCT
miR-644-3p F	ACACTCCAGCTGGGTTCATTTGCCTCCCAGCC

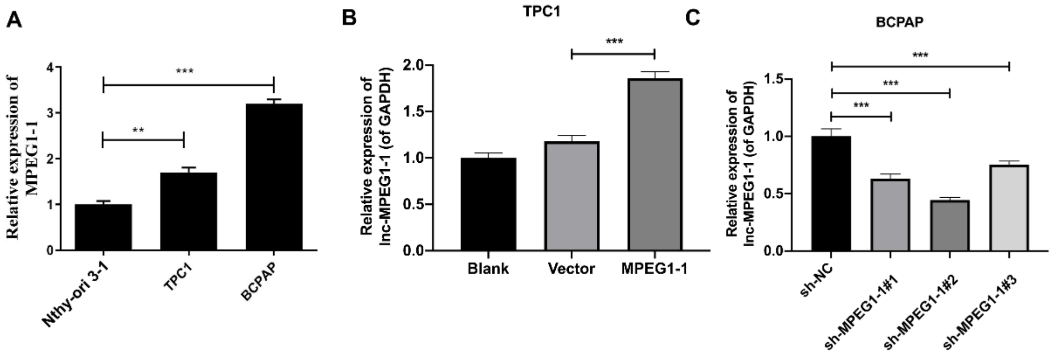


Supplementary Figure 1 Verification of the ten differential expressed lncRNA in TCGA data

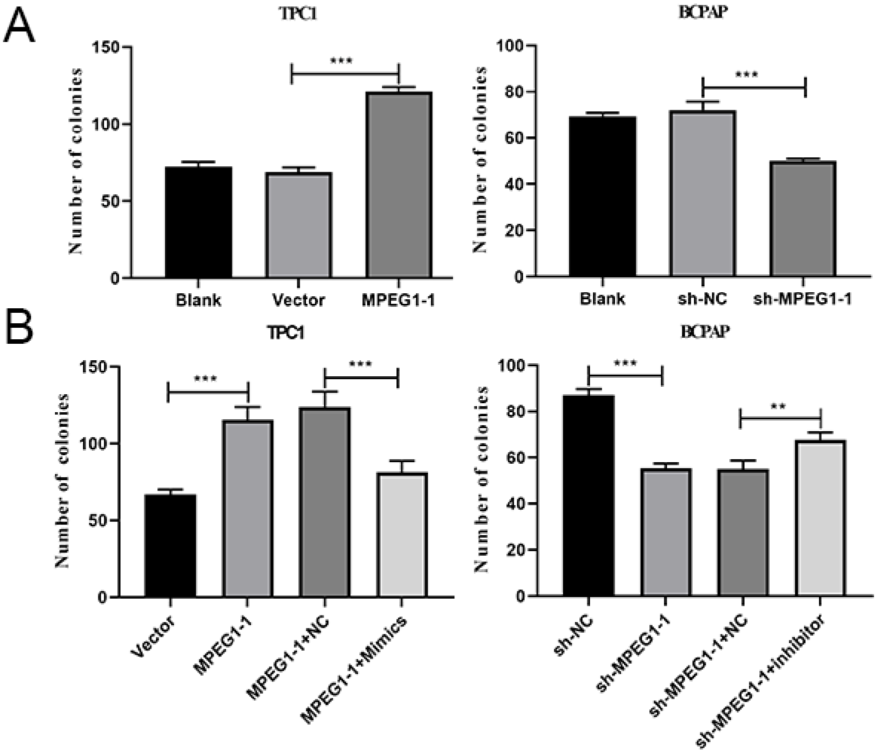


Supplementary Figure 2 Predictive performance of combined model for lymph node metastasis (N1a and N1b) of papillary thyroid cancer in TCGA data. (A) for N1a; (B) for N1b.

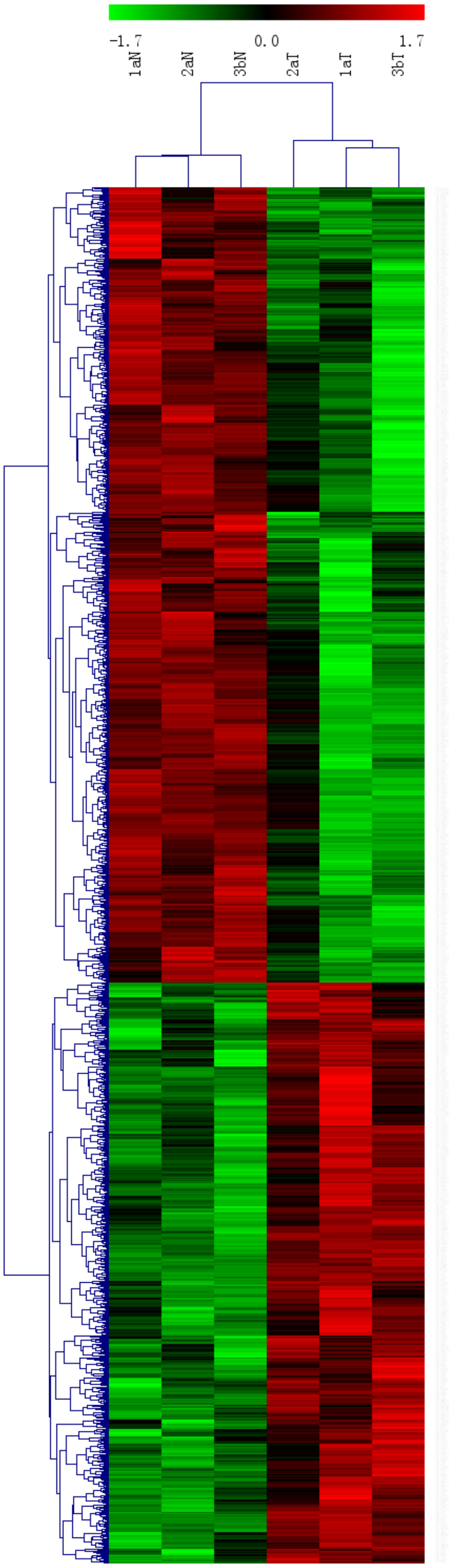
Supplementary Figure 3 lnc-MPEG1-1:1 is located in the intergenic region of the genome, and there is no coding protein gene upstream and downstream (retrieved from http://asia.ensembl.org/； lnc-MPEG1-1:1 (also named ap002358.1-201)



Supplementary Figure 4 MPEG1-1 was successfully overexpressed and silenced in PTC cells by transfection. (A) Expression of MPEG1-1 was verified with RT-qPCR analysis in Nthy-ori 3-1 and 2 PTC cell lines (TPC1 and BCPAP). (B) TPC1 cells were transfected with MPEG1-1-overexpressed plasmid or vector (control), and the overexpression effect of MPEG1-1 was determined with RT-qPCR. (C) After transfection with MPEG1-1 shRNAs (sh-MPEG1-1#1, sh-MPEG1-1#2, and sh-MPEG1-1#3), the respective transfection effects were verified through RT-qPCR analysis in BCPAP cells. ****P* < 0.001.



Supplementary Figure 5 Cell vitality in each group was determined by colony number analysis. (A) Colony number analysis was utilized to evaluate the colony-forming capacity of MPEG1-1-overexpressed on TPC1 cells and MPEG1-1-silenced on BCPAP cells. (B) The effect of MPEG1-1+mimics on TPC1 cell and sh-MPEG1-1+ inhibitor on BCPAP cells were analyzed by clone number analysis. All experiments were performed in triplicate and data are presented as the mean ± SD. ***p* < 0.01; ****p* < 0.001.



Supplementary Figure 6 Hierarchical clustering revealed distinct lncRNA expression profiles between the samples detected by microarray in the discovery stage