



**Supplementary Table S1.** miR-133b expression and clinicopathological features in 40 patients with breast cancer.

Characteristics	Expression of miR-133b		p value*
	Low	High	
<b>Sex</b>			
male	20	20	
female	0	0	
<b>Age</b>			0.185
≤60	11	15	
>60	9	5	
<b>Grade</b>			0.125
I /I-II, well-differentiated	4	8	
II /II-III, moderately differentiated	9	10	
III, poorly differentiated	7	2	
<b>Tumor histological</b>			0.001*
Ductal carcinoma in situ	1	10	
Invasive ductal carcinoma	19	10	
<b>T Classification</b>			0.062
T1	12	5	
T2	7	11	
T3	1	4	
<b>N Classification</b>			0.688
N0	7	6	
N1	3	1	
N2	6	8	
N3	4	5	
<b>ER status</b>			0.206
Negative	12	8	
Positive	8	12	
<b>PR status</b>			0.288
Negative	16	13	
Positive	4	7	
<b>HER2 status</b>			0.736
Negative	6	7	
Positive	14	13	
<b>Tumor size(cm<sup>3</sup>)</b>			0.109
≤ 6	9	14	
> 6	11	6	
<b>Lymph node metastasis</b>			0.008*
Negative	9	17	

Positive

11

3

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Median expression level was used as a cutoff to divide the 40 patients into miR-133b low group ( $n = 20$ ) and miR-133b high group ( $n = 20$ ). Two-sided  $\chi^2$  test. \*  $p < 0.05$ .

**Supplementary Table S2.** Sequences of primers used for RT-qPCR and plasmid construction.

<b>Primer Names</b>	<b>Sequences</b>
<b>Sequences of primers used for RT-qPCR</b>	
hsa-miR-133b forward	5'-CTCAGCTTTGGTCCCCTTCAAC-3'
hsa-miR-133b reverse	5'-GTGCAGGGTCCGAGGT-3'
U6 forward	5'-ATTGGAACGATACAGAGAAGATT-3'
U6 reverse	5'-GGAACGCTTCACGAATTTG-3'
TIMM17A forward	5'-TTGTGGATGACTGTGGTG-3'
TIMM17A reverse	5'-CCAAAAGGTGAGGAAGGT -3'
GAPDH forward	5'-GAGTCAACGGATTTGGTCGT-3'
GAPDH reverse	5'-TTGATTTTGGAGGGATCTCG-3'
NEAT1 forward	5'-TGGCTAGCTCAGGGCTTCAG-3'
NEAT11 reverse	5'-TCTCCTTGCCAAGCTTCCTTC- 3'
<b>Sequences of primers used for plasmid construction</b>	
pcDNA3.1-TIMM17A forward	5'-CAGGAATTCTGGAGGAGTACGCGCGAG- 3'
pcDNA3.1-TIMM17A reverse	5'-GACCTCGAGCTACTGATATTGTCGATA- 3'
pcDNA3.1-NEAT1 forward	5'-CGCGGAGAGTTAGCGACAGGGAGGGAT-3'
pcDNA3.1-NEAT1 reverse	5'-TGCTCTCTAATGAGTTTAGAACTCAAAC-3'

**Supplementary Table S3.** Sequences of miR-133b mimics and inhibitor, and siRNAs.

<b>RNA Names</b>	<b>Sequences</b>
miR-133b mimics	5'-UUUGGUCCCCUUCAACCAGCUA-3'
mimics NC	5'- UUCUCCGAACGUGUCACGUTT-3'
miR-133b inhibitor	5'-UAGCUGGUUGAAGGGGACCAAA-3'
inhibitor NC	5'-CAGUACUUUUGUGUAGUACAA-3'
NEAT1-siRNA-1	5'-GGTCTGTGTGGAAGGAGGAAGGCAG- 3'
NEAT1-siRNA-2	5'-GCCAUCAGCUUUGAAUAAAUU-3'
NEAT1-siRNA-3	5'-GGUGUUAUCAAGUGAAUUAUU-3'
TIMM17A-siRNA-1	5'-GCAUGAUGUUAGUAAUUACA-3'
TIMM17A-siRNA-2	5'-GGCUAUAAGAGACAUUUAGC-3'
TIMM17A-siRNA-3	5'-GAUGUUUCAUGCUCAUGUACU-3'

**Supplementary Table S4.** Antibodies used for western blotting (WB), RNA-binding protein immunoprecipitation (RIP) and flow cytometry (FC).

<b>Protein</b>	<b>Applications</b>	<b>Antibody</b>	<b>Origin</b>	<b>Dilution</b>	<b>Molecular Weight</b>
GAPDH	WB	D16H11, Cell Signaling Technology	Rabbit	1:1000	36 KD
TIMM17A	WB, CHIP	ab126044, Abcam	Rabbit	1:500	18KD
Ago2	RIP	03-110, Merck Millipore	Mouse	1:10	100KD
GFP	RIP	ab290, Abcam	Rabbit	1:20	28KD

Supplementary Table S5. Screening of 111 predicted targets of miR-133b.

No.	Gene Name	Breast Cancer Related	Migration or Invasion Related	Reported 133b-target	Expression in Tumor	Fold Change	p-Value	Survival Related	p-Value
1	TIMM17A	√	√	×	high	2.20	7.80E-79	√	2.60E-15
2	ELAVL1	√	√	×	high	1.53	9.60E-67	√	0.007
3	NDRG1	√	√	×	high	1.06	1.40E-05	√	2.20E-09
4	SOX4	√	√	×	high	1.75	5.90E-24	×	0.46
5	PFN2	√	√	×	high	1.61	0.00061	×	0.12
6	CTBP2	√	√	×	high	1.38	9.10E-10	×	0.48
7	RAPH1	√	√	×	low	0.86	0.00054		
8	SGMS2	√	√	×	low	0.76	2.00E-08		
9	CRK	√	√	×	low	0.75	2.10E-26		
10	RBPJ	√	√	×	low	0.74	4.70E-27		
11	TCF7	√	√	×	low	0.59	4.90E-16		
12	MEIS1	√	√	×	low	0.53	7.20E-34		
13	GABARAPL1	√	√	×	low	0.37	5.00E-86		
14	SGK1	√	√	×	low	0.37	1.40E-47		
15	USP6	√	√	×	low	0.36	1.10E-30		
16	PFKFB3	√	√	×	low	0.29	3.70		
17	AKAP9	√	√	×		1.13	0.99		
18	NUP153	√	√	×		1.02	0.30		
19	SP3	√	√	×		1.00	0.58		
20	YES1	√	√	×		0.99	0.014		
21	MAML1	√	√	√					
22	FGFR1	√	√	√					
23	MCL1	√	√	√					
24	LASP1	√	√	√					
25	BCL2L2	√	√	√					
26	DUSP1	√	√	√					
27	MEIS2	√	×						
28	UBA2	√	×						
29	BNIP3L	√	×						
30	PREX1	√	×						
31	PTPRD	√	×						
32	POU4F1	√	×						

33	ATP6AP2	√	×
34	FTL	√	×
35	RARB	√	×
36	TRHDE	√	×
37	MAP3K3	√	×
38	PPP2CA	√	×
39	SH3GL2	√	×
40	AFAP1	√	×
41	USP32	√	×
42	SMARCD1	√	×
43	MECOM	√	×
44	RB1CC1	√	×
45	SV2A	√	×
46	SLC7A8	√	×
47	SUMO1	√	×
48	QKI	√	×
49	VAPB	√	×
50	FOXL2	√	×
51	SEPHS2	√	×
52	TBPL1	√	×
53	PPP2CB	√	×
54	CMPK1	√	×
55	TFE3	√	×
56	SESN1	√	×
57	EPHA7	√	×
58	UBE2Q1	√	×
59	PEX5L	×	
60	AFTPH	×	
61	BTBD3	×	
62	PAN3	×	
63	ARFIP2	×	
64	VPS54	×	
65	ZC3H14	×	
66	ADCYAP1	×	
67	ARHGDI1	×	
68	PRRT2	×	
69	JAZF1	×	

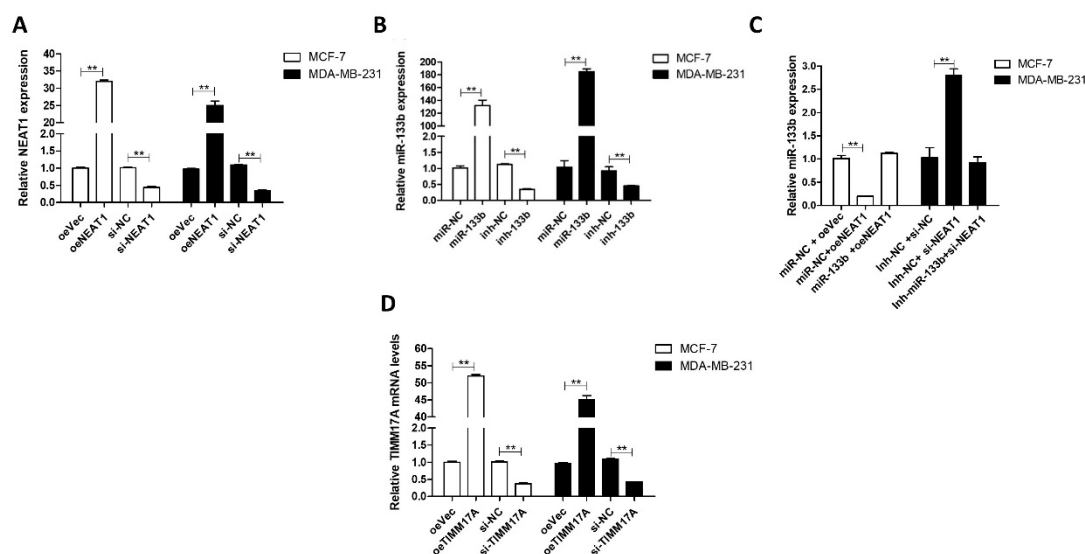
70	DOLPP1	×
71	ZNF436	×
72	CCDC117	×
73	FAM117B	×
74	GARNL3	×
75	GPM6A	×
76	EXD2	×
77	GABPB2	×
78	SUPT16H	×
79	STX5	×
80	ANKRD12	×
81	CLTA	×
82	SOBP	×
83	SACM1L	×
84	FBXL2	×
85	MED12L	×
86	TMOD3	×
87	CDK13	×
88	XPO4	×
89	PPFIA3	×
90	ARHGAP12	×
91	ZNF362	×
92	GRM5	×
93	TRAM2	×
94	TMEM167A	×
95	RAP2C	×
96	PTBP2	×
97	SLC6A1	×
98	FBXW11	×
99	GDI2	×
100	RAVER1	×
101	NRIP3	×
102	SEC61B	×
103	SYT1	×
104	MLLT3	×
105	CRTAM	×
106	RBMXL1	×



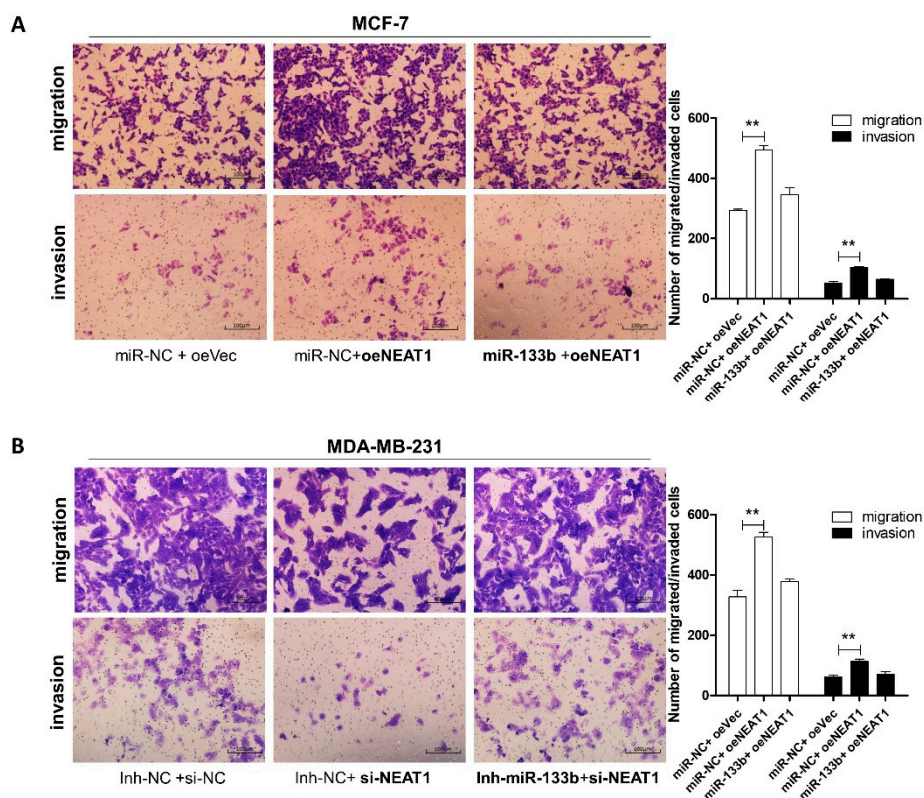
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107	LRRC7	×
108	TFAP2D	×
109	SHISA5	×
110	SNRK	×
111	MTMR4	×

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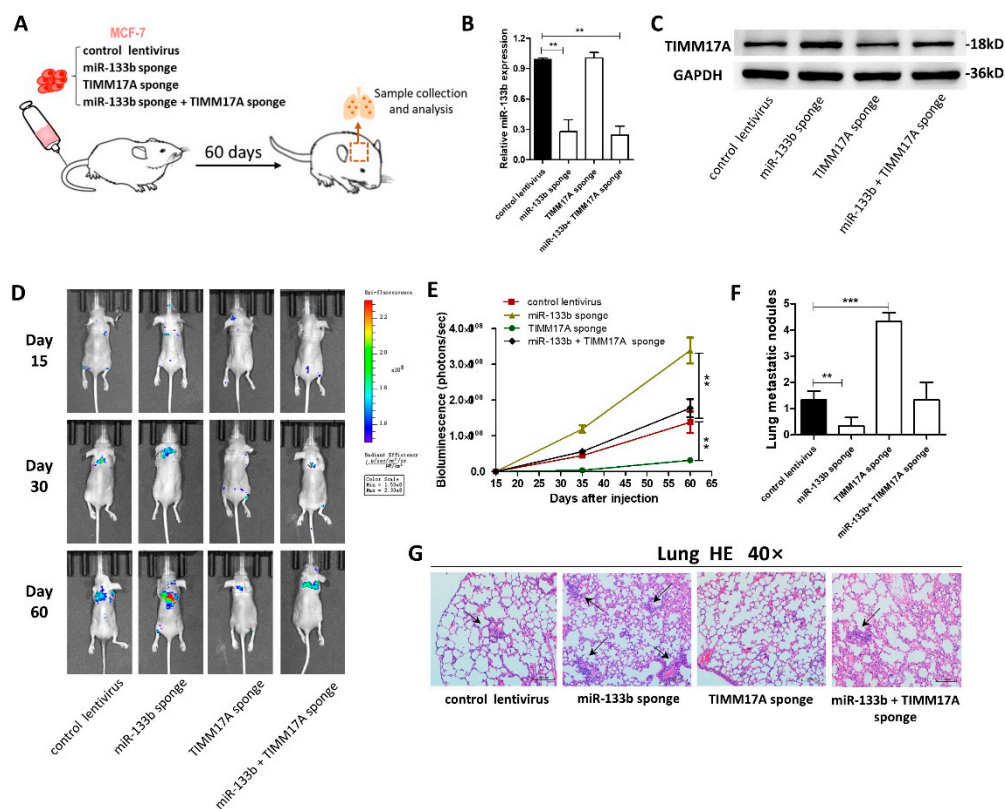


**Figure S1.** Verification of miR-133b, NEAT1 and TIMM17A overexpression or knockdown efficiencies in breast cancer cells. (A) NEAT1 levels in MCF-7 cells and MDA-MB-231 cells transfected with oeVec, oeNEAT1, si-NC or si-NEAT1. (B) miR-133b levels in MCF-7 cells and MDA-MB-231 cells transfected with miR-NC, miR-133b, inh-NC or inh-miR-133b. (C) miR-133b levels in MCF-7 cells transfected with miR-NC plus oeVec, miR-NC plus oeNEAT1, or miR-133b plus oeNEAT1, and MDA-MB-231 cells transfected with inh-NC plus si-NC, inh-NC plus si-NEAT1, or inh-miR-133b plus si-NEAT1. (D) TIMM17A mRNA levels in MCF-7 cells and MDA-MB-231 cells transfected with oeVec, oeTIMM17A, si-NC or si-TIMM17A. \*\* $p < 0.01$ ; \*\*\* $p < 0.001$ .



**Figure S2.** NEAT1 promotes breast cancer cells migration and invasion via silencing miR-133b. (A,B) Migration and invasion of MCF-7 cells transfected with miR-NC plus oeVec, miR-NC plus oeNEAT1,

or miR-133b plus oeNEAT1 (A), and MDA-MB-231 cells transfected with inh-NC plus si-NC, inh-NC plus si-NEAT1, or inh-miR-133b plus si-NEAT1 (B) detected by transwell assay. Scale bar, 100  $\mu$ m.



**Figure S3.** Effects of TIMM17A-targeted miR-133b on the lung colonization of MCF-7 cells xenografts in mice. (A) Experimental design: immunocompromised mice were injected through tail vein with MCF-7 cells transfected with either the control lentivirus, miR-133b sponge, TIMM17A sponge, miR-133b sponge plus TIMM17A sponge. (B,C) miR-133b levels (B) and TIMM17A protein levels (C) in MCF-7 cells transfected with either the control lentivirus, miR-133b sponge, TIMM17A sponge, miR-133b sponge plus TIMM17A sponge. (D,E) Representative BLI images (D) and quantitative analysis of the fluorescence intensities (E) of mice of five groups. The BLI was performed on days 15, 35, and 60 after injection. The intensity of BLI is represented by the color. (F,G) Numbers of metastatic nodules (F) and representative H&E-stained sections of lung tissues isolated from the intravenously injected mice. Black arrows indicate metastatic nodules. Scale bar, 200  $\mu$ m. \*\* $p < 0.01$ ; \*\*\* $p < 0.001$ .