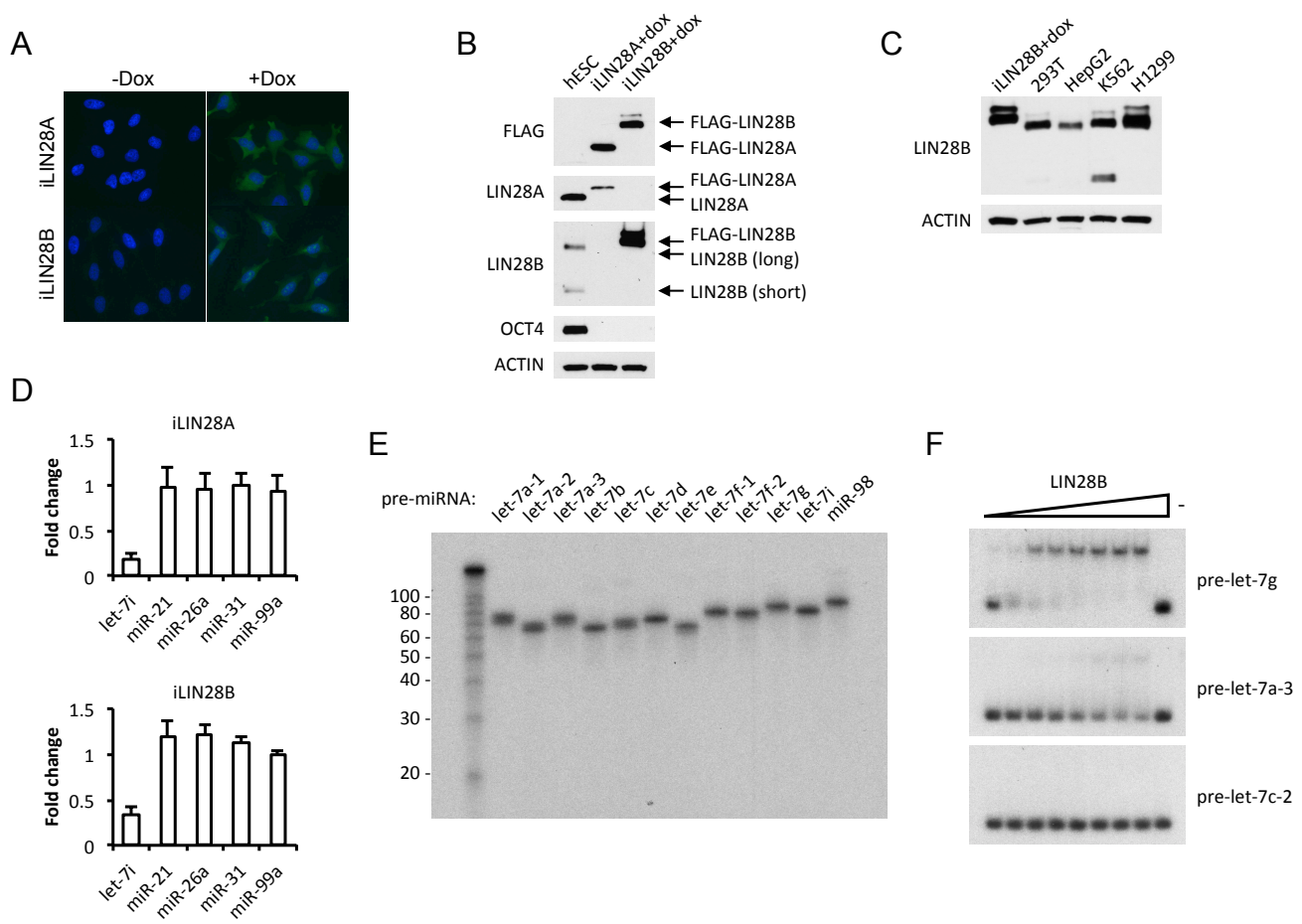


**Figure S1**



**Supplemental Figure 1 (Related to Figure 1): (A)** Immunofluorescence analyses of FLAG-LIN28A and FLAG-LIN28B localization in doxycycline-inducible HeLa cell lines. **(B)** Western blot for comparative analysis of LIN28A and LIN28B expression between human ESCs and doxycycline-treated iLIN28A and iLIN28B HeLa cell lines. **(C)** Western blot for comparative analysis of LIN28B expression between different human cancer cell lines and doxycycline-treated iLIN28B HeLa cell line. **(D)** q.RT-PCR analyses of miRNA expression in iLIN28A (left) and iLIN28B (right) HeLa cells. Results are the average of two biological replicate experiments represented as fold change relative to mock treated cells. Error bars represent SD. **(E)** Denaturing gel analysis for individual in vitro transcribed human let-7 pre-miRNA. **(F)** EMSA showing relative LIN28B binding to pre-let-7g, pre-let-7a-3 and pre-let-7c-2.

**Table S1 (related to experimental procedures):****PCR primers sequences for pri-miRNA subcloning into pCDNA3 plasmid**

Name	Sequence
hsa-pri-let-7a-1 BamHI	AACAAGGATCCTCAACGTAAGTGAATGAAAATGGTCTAATAC
hsa-pri-let-7a-1 NotI	AACAAGCGGCCGCCAGTGTACTTGCTACAGACTTTATC
hsa-pri-let-7a-2 BamHI	AACAAGGATCCTCTCTTCTATCATCAGACTCTTTGATATGTTG
hsa-pri-let-7a-2 NotI	AACAAGCGGCCGCATATGAAAATACAGCATGGGTACAAGG
hsa-pri-let-7a-3 BamHI	AACAAGGATCCTGCCCGCCAGAATCCCT
hsa-pri-let-7a-3 NotI	AACAAGCGGCCGCTCACACAGCAAGTGGCACCTAG
hsa-pri-let-7b BamHI	AACAAGGATCCCAAGACCAGCCAGCAGCAGC
hsa-pri-let-7b NotI	AACAAGCGGCCGCTAGCTCCCAGATGCCACCCAGC
hsa-pri-let-7c BamHI	AACAAGGATCCAGAAACAGTTAGCAGCAAAGTTTACTGG
hsa-pri-let-7c NotI	AACAAGCGGCCGCAGATACAAATGGAATAATCTAATAGGGCTG
hsa-pri-let-7d BamHI	AACAAGGATCCAAGTTCATATGGCCAGATAACTTTTC
hsa-pri-let-7d NotI	AACAAGCGGCCGCAGCACCATGCTGCAGTTTATAATC
hsa-pri-let-7e BamHI	AACAAGGATCCTTCGCCGCACACAAGCTCGTG
hsa-pri-let-7e NotI	AACAAGCGGCCGCTTTCAGGGGAAGGAGGGGATGCAGG
hsa-pri-let-7f-1 BamHI	AACAAGGATCCTGGGGAAACCTTTTGCTTCTTG
hsa-pri-let-7f-1 NotI	AACAAGCGGCCGCTGTTAATCTGTGCCTACTGTACTACTTG
hsa-pri-let-7f-2 BamHI	AACAAGGATCCACTGCATCTAAGTCAAATGACTCCAC
hsa-pri-let-7f-2 NotI	AACAAGCGGCCGCACTTCAAAGATGTACATGAACATGCTG
hsa-pri-let-7g BamHI	AACAAGGATCCGGATTCTCCAAATGTGGTGCAAG
hsa-pri-let-7g NotI	AACAAGCGGCCGCTAGTTATTCTACTGCTAACAATGACTCACC
hsa-pri-let-7i BamHI	AACAAGGATCCGACGTCGGCGGCGAGGAAGG
hsa-pri-let-7i NotI	AACAAGCGGCCGCGAGGCTTCGCGGGCTGTCCC
hsa-pri-miR-98 BamHI	AACAAGGATCCAATGATTGTACCAAAGCCTGCAC
hsa-pri-miR-98 NotI	AACAAGCGGCCGCACTTTATAATGACATCTGCCTCACCC
mmu-pri-let-7b HindIII	AACAACAAGCTTGGCTTCCTCAATCTGTTTGGTAGG
mmu-pri-let-7b XhoI	AACAACCTCGAGTCTCATGTCTCTACGGCCAG
mmu-pri-let-7c-2 HindIII*	AACAACAAGCTTTCCCAGTCTATTAATACCCTCAGC
mmu-pri-let-7c-2 XhoI*	AACAACCTCGAGAGAGGTCAAGGGACTATCCAAGG

\* these primers were also used for genotyping PCR of *let-7c-2<sup>-/-</sup>* clones

**PCR primers sequences for DNA template syntheses for in vitro transcription**

Name	Sequence
T7 hsa-pre-let-7a-1 F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGGTTGTATAGTTTT AGGGT
T7 hsa-pre-let-7a-1 R	GAAAGACAGTAGATTGTATAGTTATCTCCCAG
T7 hsa-pre-let-7a-2 F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGGTTGTATAGTTTA GAATTACATC
T7 hsa-pre-let-7a-2 R	GGAAAGCTAGGAGGCTGTACAGTTATCTCC
T7 hsa-pre-let-7a-3 F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGGTTGTATAGTTTT GG

T7 hsa-pre-let-7a-3 R	GAAAGACAGTAGATTGTATAGTTATCCCATAGC
T7 hsa-pre-let-7c F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGGTTGTATGGTTT GAGTTA
T7 hsa-pre-let-7c R	GGAAAGCTAGAAGGTTGTACAGTTAACTC
T7 hsa-pre-let-7d F	ACGGTTCAGCTAATACGACTCACTATAGGGAGAGGTAGTAGGTTGCATAGTTTT AGG
T7 hsa-pre-let-7d R	AAGAAAGGCAGCAGGTCGTATAG
T7 hsa-pre-let-7e F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGGAGGTTGTATAGTTG AGG
T7 hsa-pre-let-7e R	GGAAAGCTAGGAGGCCGTATAGTG
T7 hsa-pre-let-7f-1 F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGATTGTATAGTTGT GGGG
T7 hsa-pre-let-7f-1 R	GGGAAGGCAATAGATTGTATAGTTATCTCC
T7 hsa-pre-let-7f-2 F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGATTGTATAGTTTT AGGGTCATAC
T7 hsa-pre-let-7f-2 R	GGAAAGACAGTAGACTGTATAGTTATCTCCA
T7 hsa-pre-let-7g GGAG/UGCG F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGTTTGTACAGTTTG AGG
T7 hsa-pre-let-7g GGAG/UGCG R	GCAAGGCAGTGGCCTGTACAGTTATC
T7 hsa-pre-let-7i F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGTTTGTGCTGTTGG
T7 hsa-pre-let-7i R	AGCAAGGCAGTAGCTTGC
T7 hsa-pre-miR-98 F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAAGTTGTATTGTTGT GGG
T7 hsa-pre-miR-98 R	GGGAAAGTAGTAAGTTGTATAGTTATCTTCTAATTGG
T7 mmu-pre-let-7b F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGGTTGTGTGGTTTC
T7 mmu-pre-let-7b R	GGGAAGGCAGTAGGTTGTATAGTTATC
T7 mmu-pre-let-7c-2 UGCG/GGAG F	ACGGTTCAGCTAATACGACTCACTATAGGGTGAGGTAGTAGGTTGTATGGTTTT GG
T7 mmu-pre-let-7c-2 UGCG R	GGAAAGACAGTAGATTGTATAGTTACCGC
T7 mmu-pre-let-7c-2 GGAG R	GGAAAGACAGTAGATTGTATAGTTACCTCC

### PCR primer sequences for subcloning LIN28 cDNA into pCDNA5

Name	Sequence
FLAG-LIN28A EcoRV	AACAAGATATCACCATGGACTACAAAGACGATGAC
FLAG-LIN28A NotI	AACAAGCGGCCGCTTAATTCTGTGCCTCCGGGAG
FLAG-LIN28B EcoRV	AACAAGATATCACCATGGACTACAAAGACGATGACGACAAGGCCGAAGGCCGGG GCTAGC
FLAG-LIN28B NotI	AACAAGCGGCCGCTTATGTCTTTTTCTTTTTTGAAGTGAAGG

### PCR primer sequences for mutagenesis

Name	Sequence
mmu-pre-let-7c-2 GGAG sense	GGGCTCTGCCCGCTCGGAGGTAACATACAATCTA
mmu-pre-let-7c-2 GGAG antisense	TAGATTGTATAGTTACCTCCGAGCGGGGCAGAGCCC
mmu-pre-let-7c-2 UCACA sense	AGGTAGTAGGTTGTATGGTTTTGGGTCACACCCCGCTCTGCGGTAACATACAA T

mmu-pre-let-7c-2 UCACA antisense	ATTGTATAGTTACCGCAGAGCGGGGTGTGACCCAAAACCATACAACCTACTAC CT
mmu-pre-let-7c-2 CCACG sense	GGTTGTATGGTTTTGGGCCACGCCCGCTCTGCGGTAAC
mmu-pre-let-7c-2 CCACG antisense	GTTACCGCAGAGCGGGGCGTGGCCAAAACCATACAACC
mmu-pre-let-7c-2 CUAUG sense	GGTTGTATGGTTTTGGGCTATGCCCGCTC
mmu-pre-let-7c-2 CUAUG antisense	GAGCGGGGCATAGCCAAAACCATACAACC
mmu-pre-let-7c-2 UUCUA sense	GCAGAGCGGGGTAGAACCCAAAACCATACAACC
mmu-pre-let-7c-2 UUCUA antisense	GGTTGTATGGTTTTGGGTTCTACCCCGCTCTGC
mmu-pre-let-7g UGCG sense	ATGATACCACCCGGTACATGCGATAACTGTACAGGCCAC
mmu-pre-let-7g UGCG antisense	GTGGCCTGTACAGTTATCGCATGTACCGGGTGGTATCAT

### Oligonucleotide sequences for gRNA cloning into pX330 for CRISPR/Cas9 genome editing

Name	Sequence
let-7c-2 gRNA#1 sense	CACCGAGTGGATACAACGGCGGCC
let-7c-2 gRNA#1 antisense	AAACGGGCCCGCGTTGTATCCACTC
let-7c-2 gRNA#2 sense	CACCGTGTATAGTTACCGCAGAGCG
let-7c-2 gRNA#2 antisense	AAACCGCTCTGCGGTAACATACAC