miR-99a-5p wild-type vector

gatatttccaggcttacgaccctgggctcacgggtacctatttatatg

miR-99a-5p deletion-type vector

gatatttccaggcttacgaccctgggctc------acctatttatatgctcagtg

miR-99a-3p wild-type vector

ttctgctttcctaggggactcttgagcttagaaactcatcgtacacttg

miR-99a-3p deletion-type vector

ttctgctttcctaggggactctt------agaaactcatcgtacacttgacct

Figure S1. Vector inserted sequences.

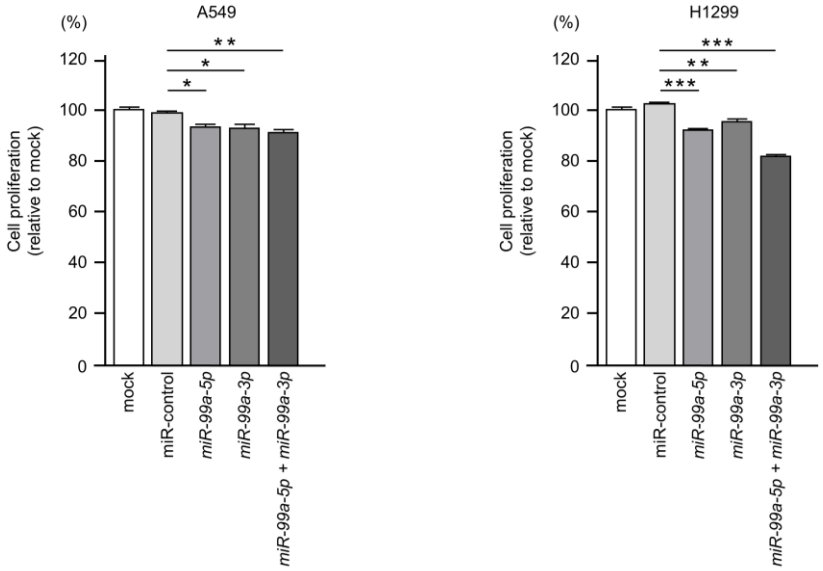

Figure 2. Effects of ectopic expression of *miR-99a-5p* and *miR-99a-3p* on LUAD cells. Cell proliferation was determined by XTT assays 72 h following transfection with *miR-99a-5p* (5nM), *miR-99a-3p* (5nM) or *miR-99a-5p* (5nM) + *miR-99a-3p* (5nM) (*$p < 0.05$, **$p < 0.01$, ***$p < 0.0001$).

Table S1. Reagent used in this study.

Antibody	Dilution	Catalog number	Company
FAM64A	IHC 1:100	ab251896	Abcam, Cambridge, UK

miRNA species	Concentration	Assay ID	Company
miR-99a-5p	10 nM	PM 10719	Applied Biosystems, Foster City, CA, USA
miR-99a-3p	10 nM	PM 12983	Applied Biosystems, Foster City, CA, USA
anti-miR Negative Control #1	10 nM	AM 17010	Applied Biosystems, Foster City, CA, USA

siRNA	Concentration	Catalog number	Company
si*FAM64A*	10 nM	HSS147670 and HSS147671	Invitrogen, Carlsbad, CA, USA

primer and probe	Assay ID	Company
FAM64A	Hs00760463_m1	Applied Biosystems, Foster City, CA, USA
GUSB	Hs00939627_m1	Applied Biosystems, Foster City, CA, USA

Plasmid vector	Catalog number	Company
psiCHECK-2 vector	C8021	Promega, Madison, WI, USA

Table S2. Characteristics of the patients used in immunostaining.

Patients	Age	Sex	T	N	M	Pathological stage	Type
A	47	M	2	0	0	IB	LUAD
B	56	F	2	1	0	IIA	LUAD
C	67	M	2	0	0	IB	LUAD
D	16	F	-	-	-	-	Normal lung tissue

LUAD: lung adenocarcinoma

Table S3. 23 target genes analyzed by Benjamini-Hochberg method.

Entrez GeneID	Gene symbol	*p*-value	i	qi=pi*N/i	FDR threshold	Judgement
983	*CDK1*	0.0003	3	0.0023	0.05	Significant
4288	*MKI67*	0.0005	4	0.0029	0.05	Significant
57082	*CASC5*	0.0003	2	0.0035	0.05	Significant
4751	*NEK2*	0.0002	1	0.0046	0.05	Significant
6241	*RRM2*	0.002	6	0.0077	0.05	Significant
1033	*CDKN3*	0.0017	5	0.0078	0.05	Significant
3832	*KIF11*	0.0034	8	0.0098	0.05	Significant
4173	*MCM4*	0.0032	7	0.0105	0.05	Significant
1063	*CENPF*	0.0059	9	0.0151	0.05	Significant
1163	*CKS1B*	0.0073	10	0.0168	0.05	Significant
11130	*ZWINT*	0.0094	12	0.0180	0.05	Significant
84908	*FAM136A*	0.0087	11	0.0182	0.05	Significant
55215	*FANCI*	0.0108	13	0.0191	0.05	Significant
56888	*KCMF1*	0.0125	14	0.0205	0.05	Significant
417685	*FAM64A*	0.0175	15	0.0268	0.05	Significant
64151	*NCAPG*	0.0208	16	0.0299	0.05	Significant
151648	*SGOL1*	0.0235	18	0.0300	0.05	Significant
157570	*ESCO2*	0.0235	17	0.0318	0.05	Significant
10797	*MTHFD2*	0.0321	19	0.0389	0.05	Significant
11260	*XPOT*	0.0669	20	0.0769	0.05	NS
55854	*ZC3H15*	0.0942	22	0.0985	0.05	NS
51290	*ERGIC2*	0.0929	21	0.1017	0.05	NS
993	*CDC25A*	0.1585	23	0.1585	0.05	NS

NS: not significant

Table S4A. Significantly enriched annotations regulated by *miR-99a-5p* in LUAD cells.

Genes	*p*-value	Annotations
CKS1B *CENPF* *CDC25A* *FAM64A*	4.14×10^{-5}	(GO) 0051301: cell division
CKS1B *CENPF* *CDC25A* *FAM64A*	0.000137009	(GO) 0007049: cell cycle
CKS1B *CDC25A*	0.00127197	(GO) 0000079: regulation of cyclin-dependent protein serine/threonine kinase activity
CDC25A	0.00529465	(GO) 0110032: positive regulation of G2/MI transition of meiotic cell cycle
CKS1B *CDC25A*	0.00547439	(GO) 0008283: cell population proliferation

Table S4B. Significantly enriched annotations regulated by *miR-99a-3p* in LUAD cells.

Genes	*p*-value	Annotations
ZWINT *CDKN3* *CDK1* *NEK2* *MKI67* *MCM4* *KIF11* *ESCO2* *SGOL1* *NCAPG* *CASC5* *FANCI* *FAM64A*	9.15×10^{-14}	(GO) 0007049: cell cycle
ZWINT *CDK1* *NEK2* *KIF11* *SGOL1* *NCAPG* *CASC5* *FAM64A*	1.10×10^{-7}	(GO) 0051301: cell division
NEK2 *ESCO2* *SGOL1* *CASC5*	7.25×10^{-5}	(GO) 0007059: chromosome segregation
ZWINT *NEK2* *SGOL1*	0.000165787	(GO) 0000070: mitotic sister chromatid segregation
NEK2 *KIF11*	0.000243324	(GO) 0046602: regulation of mitotic centrosome separation

Table S4C. Pathways regulated by *miR-99a-5p* in LUAD cells.

Cell division pathway

Gene symbol	Gene name
CKS1B	CDC28 protein kinase regulatory subunit 1B
CENPF	Centromere protein F
CDC25A	Cell division cycle 25A
FAM64A	Family with sequence similarity 64 member A

Cell cycle pathway

Gene symbol	Gene name
CKS1B	CDC28 protein kinase regulatory subunit 1B
CENPF	Centromere protein F
CDC25A	Cell division cycle 25A
FAM64A	Family with sequence similarity 64 member A

Regulation of cyclin-dependent protein serine/threonine kinase activity pathway

Gene symbol	Gene name
CKS1B	CDC28 protein kinase regulatory subunit 1B
CDC25A	Cell division cycle 25A

Positive regulation of G2/MI transition of meiotic cell cycle pathway

Gene symbol	Gene name
CDC25A	Cell division cycle 25A

Cell population proliferation pathway

Gene symbol	Gene name
CKS1B	CDC28 protein kinase regulatory subunit 1B
CDC25A	Cell division cycle 25A

Table S4D. Pathways regulated by *miR-99a-3p* in LUAD cells.

Cell cycle pathway

Gene symbol	Gene name
ZWINT	ZW10 interacting kinetochore protein
CDKN3	Cyclin dependent kinase inhibitor 3
CDK1	Cyclin dependent kinase 1
NEK2	NIMA related kinase 2
MKI67	Marker of proliferation Ki-67
MCM4	Minichromosome maintenance complex component 4
KIF11	Kinesin family member 11
ESCO2	Establishment of sister chromatid cohesion N-acetyltransferase 2
SGOL1	Shugoshin 1
NCAPG	Non-SMC condensin I complex subunit G
CASC5	Cancer susceptibility candidate 5
FANCI	FA complementation group I
FAM64A	Family with sequence similarity 64 member A

Cell division pathway

Gene symbol	Gene name
ZWINT	ZW10 interacting kinetochore protein
CDK1	Cyclin dependent kinase 1
NEK2	NIMA related kinase 2
KIF11	Kinesin family member 11
SGOL1	Shugoshin 1
NCAPG	Non-SMC condensin I complex subunit G
CASC5	Cancer susceptibility candidate 5
FAM64A	Family with sequence similarity 64 member A

Chromosome segregation pathway

Gene symbol	Gene name
NEK2	NIMA related kinase 2
ESCO2	Establishment of sister chromatid cohesion N-acetyltransferase 2
SGOL1	Shugoshin 1
CASC5	Cancer susceptibility candidate 5

Mitotic sister chromatid segregation pathway

Gene symbol	Gene name
ZWINT	ZW10 interacting kinetochore protein
NEK2	NIMA related kinase 2
SGOL1	Shugoshin 1

Regulation of mitotic centrosome separation pathway

Gene symbol	Gene name
NEK2	NIMA related kinase 2
KIF11	Kinesin family member 11