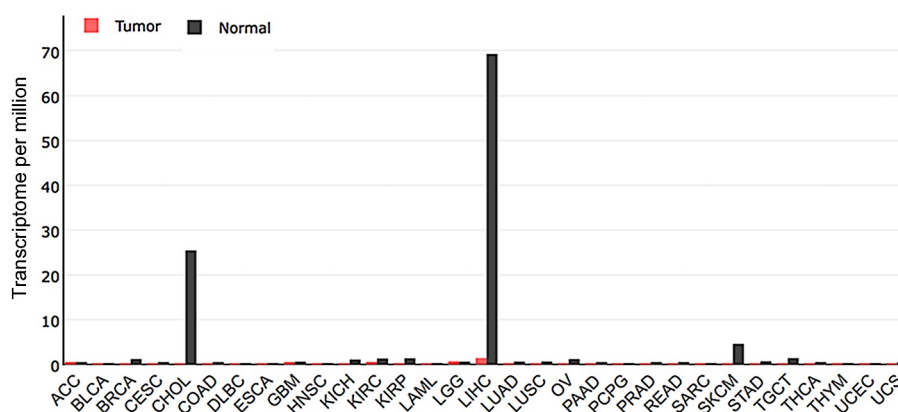


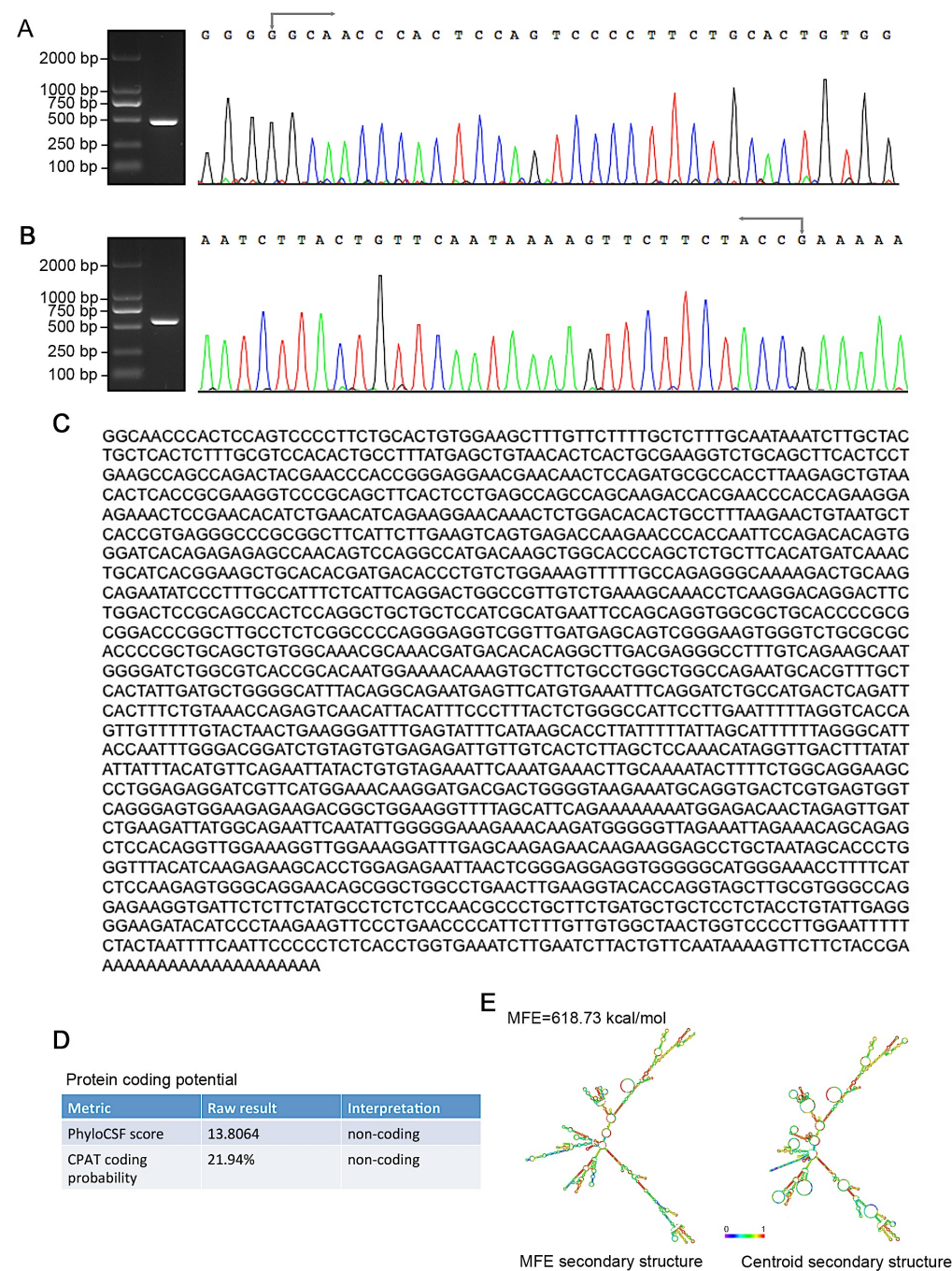
Supplementary data



Zheng *et al.* Figure S1

Figure S1. Relative expression of *LINC01554* in tumor tissues and corresponding nontumor tissues of different cancer types provided by GEPIA (<http://gepia.cancer-pku.cn>). ACC, adrenocortical carcinoma; BLCA, bladder urothelial carcinoma; BRCA, breast invasive carcinoma; CESC, cervical squamous cell carcinoma and endocervical adenocarcinoma; CHOL, cholangio carcinoma; COAD, colon adenocarcinoma; DLBC, diffuse large B-cell lymphoma; ESCA, esophageal carcinoma; GBM, glioblastoma multiforme; HNSC, head and neck squamous cell carcinoma; KICH, kidney chromophobe; KIRC, kidney renal clear cell carcinoma; KIRP, kidney renal papillary cell carcinoma; LAML, acute myeloid leukemia; LGG, brain lower grade glioma; LIHC, liver hepatocellular carcinoma; LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma; MESO, mesothelioma; OV, ovarian serous cystadenocarcinoma; PAAD, pancreatic adenocarcinoma; READ, rectum adenocarcinoma; SARC, sarcoma; SKCM, skin cutaneous melanoma; STAD, stomach adenocarcinoma; TGCT, testicular germ cell tumors; THCA, thyroid carcinoma; THYM, thymoma; UCEC, uterine corpus

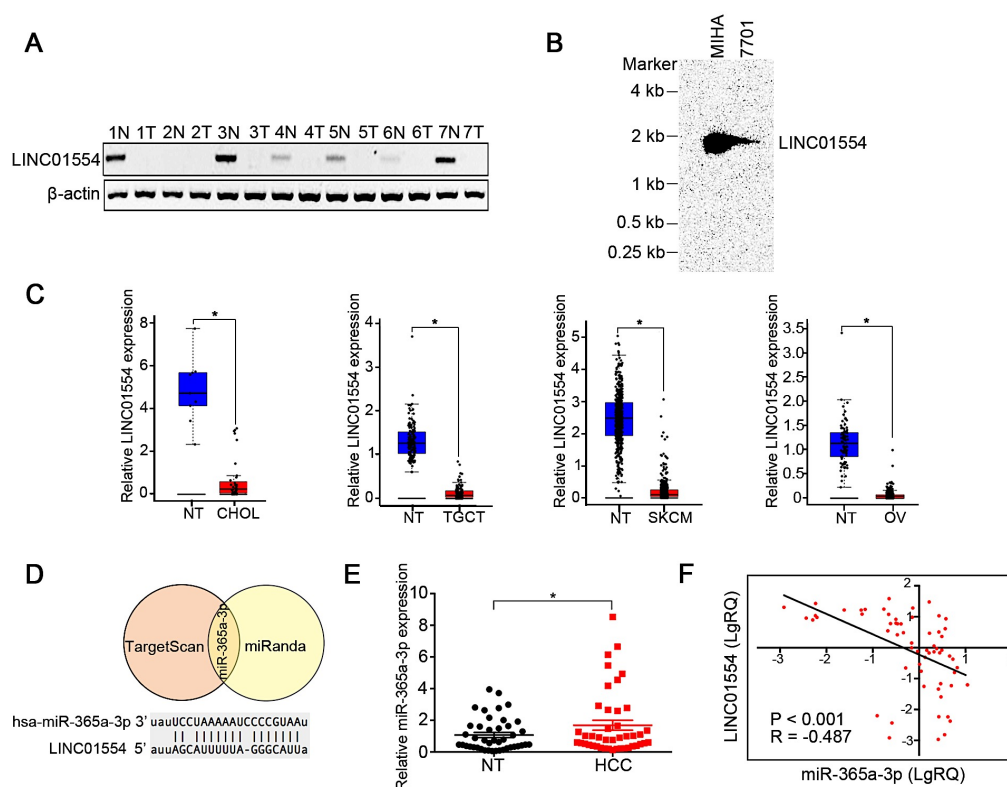
endometrial carcinoma; UCS, uterine carcinosarcoma.



Zheng *et al.* Figure S2

Figure S2. Characterization of full-length human *LINC01554* in MIHA cells and protein coding potential as well as secondary structure of *LINC01554*. (A,B) Representative images of the PCR products from 5' RACE (A) and 3' RACE (B) (left). Sequencing of PCR products determined the boundary between the universal anchor

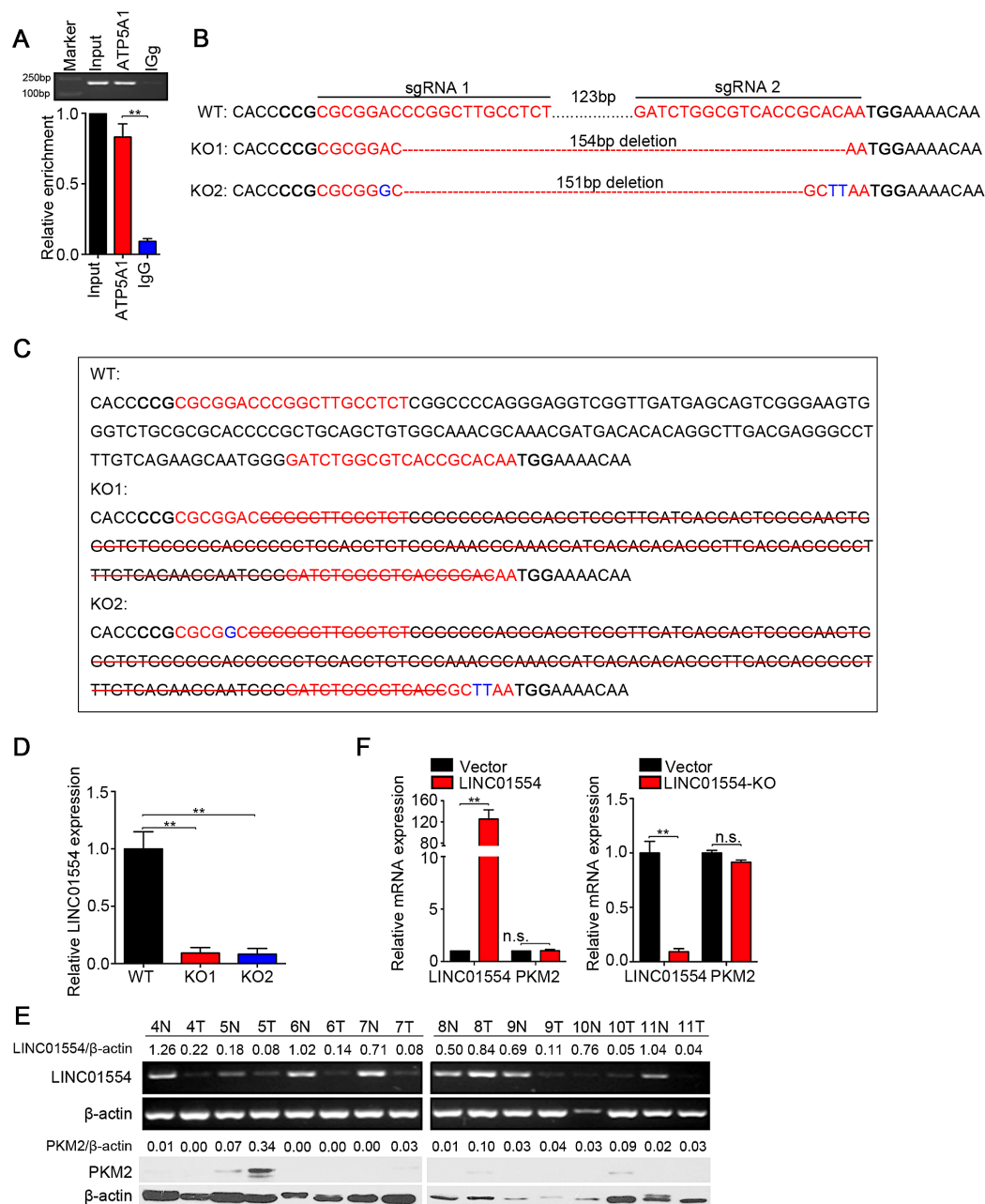
primer and *LINC01554* sequences (right). (C) The nucleotide sequence of full-length human *LINC01554*. (D) Protein coding potential of *LINC01554* predicted using LNCipedia (<https://lncipedia.org>). (E) The RNA secondary structure of *LINC01554* predicted using RNAfold Webserver (<http://rna.tbi.univie.ac.at/>) based on minimum free energy (MFE) and partition function. Color scale indicates the predictive confidence from weak to strong in line with the changes of color from purple to red.



Zheng *et al.* Figure S3

Figure S3. *LINC01554* is downregulated in human cancers and negatively regulated by miR-365a. (A) Relative expression of *LINC01554* in 7 pairs of HCC tissues and adjacent non-tumor tissues by RT-PCR. N, non-tumor tissue; T, tumor tissue. (B) Northern blot analysis of *LINC01554* isolated from MIHA and QGY7701, showing its expression level and transcript size in cell lines. (C) Downregulation of *LINC01554* in cholangiocarcinoma (CHOL), testicular germ cell tumors (TGCT),

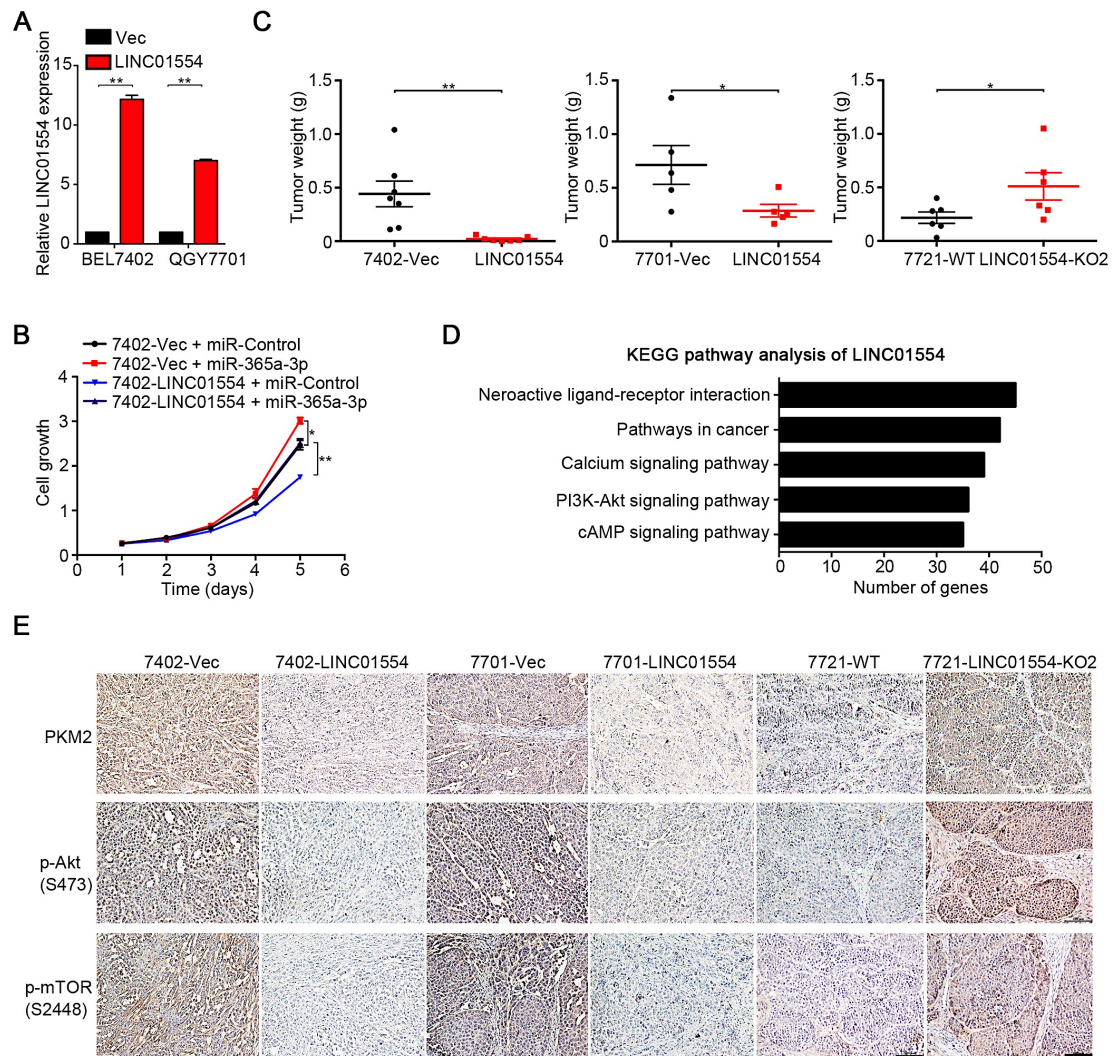
skin cutaneous melanoma (SKCM) and ovarian serous cystadenocarcinoma (OV), compared with their corresponding normal tissues, analyzed from TCGA database. (D) miR-365a-3p and the complementary sequences between miR-365a-3p and *LINC01554* predicted by TargetScan and miRanda. (E) Higher expression levels of miR-365a-3p detected by qRT-PCR in 43 HCC tissues compared with their adjacent non-tumor tissues. *, $P < 0.05$. (F) Correlation between *LINC01554* and miR-365a-3p in 62 HCC cases determined by qRT-PCR ($P < 0.001$).



Zheng *et al.* Figure S4

Figure S4. LINC01554 interacts with PKM2 and ATP5A1, and verification of LINC01554-knockout 7721 cells with the CRIPSR/Cas9 system by sequencing and qRT-PCR. **(A)** RIP assay of *LINC01554* in BEL7402 cells using anti-ATP5A1 and IgG antibodies. The percentage of RIP-enriched RNA relative to input was calculated by RT-PCR. Error bars represent SD in triplicate experiments. **, $P < 0.01$. **(B and C)** DNA sequences for each of the PCR products from WT, KO1 and KO2. The sgRNA

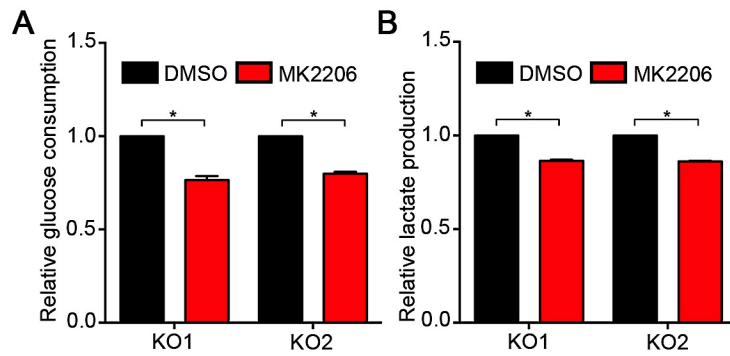
target sites are highlighted in red. PAM regions are highlighted in bold. Mutant nucleotides are labeled in blue. Nucleotide deletions are shown in red dashed line, with detailed editing length labeled above. The length between two sgRNAs is labeled above the black dotted lines. Detailed sequences information is shown in Figure (C) and cross-cut indicates a deletion. (D) Detection of *LINC01554* level by qRT-PCR in SMMC7721-WT and -KO clones. **, $P < 0.01$. (E) *LINC01554* expression level detected by RT-PCR and PKM2 protein level analyzed by Western blotting in other 8 pairs of HCC tissues and corresponding non-tumor tissues. N, non-tumor tissue; T, tumor tissue. (F) Detection of PKM2 mRNA level in *LINC01554*-transfected QGY7701 cells and *LINC01554*-KO SMMC7721 cells. WT, wild-type cells; KO, *LINC01554*-knockout cells.



Zheng *et al* Figure S5

Figure S5. Overexpression of *LINC01554* in BEL7402 and QGY7701 cells (A), Cell growth assay in *LINC01554*-transfected and vector BEL7402 cells after introduction of miR-365a-3p mimics or control. (B), tumor weight of xenograft tumors derived from *LINC01554*-transfected cells, knockout cells and their corresponding vectors (C), KEGG pathway analysis of *LINC01554* co-expressed genes using COEXPREDdb (<http://coxpresdb.jp/>), which was based on microarray and RNA-Seq data (D) and representative IHC images of PKM2, p-Akt (S473) and p-mTOR (S2448) expression in xenograft tumors derived from *LINC01554*-transfected cells, knockout cells and

their corresponding vectors, scale bar = 100 μ m (E). Vec, empty vector-transfected cells; LINC01554, *LINC01554*-transfected cells; WT, wild-type cells; KO, knockout cells. *, $P < 0.05$; **, $P < 0.01$ in independent Student's t test.



Zheng *et al.* Figure S6

Figure S6. Detection of glucose consumption (A) and lactate production (B) in SMMC7721 *LINC01554*-KO cells after treatment with 5 μ M MK2206 for 24 h. KO, *LINC01554*-knockout cells. *, $P < 0.05$ in independent Student's t test.

Table S1. Sequences of primers, probes and sgRNAs used in the study

Sequences of gene-specific PCR primers for RACE
5' RACE: 5'-CCATGACAAGCTGGCACCCA-3'
3' RACE: 5'-ACGGCTGGAAGGTTTATAGCA-3'
Sequences of primers used for pcDNA3.1-<i>LINC01554</i> construction
5'-CGGGATCCGGCAACCCACTCCAGTCCCCTTCT-3' (forward)
5'-CCGCTCGAGTTTCGGTAGAAGAAGTCTTTATTGAA-3' (reverse)
Sequences of <i>in vitro</i> transcription primers of <i>LINC01554</i>
Sense
5'-TAATACGACTCACTATAGGGAGAGGCAACCCACTCCAGTCCCCTTCT-3' (forward)
5'-TTTCGGTAGAAGAAGTCTTTATTGAA-3' (reverse)
Antisense
5'-GGCAACCCACTCCAGTCCCCTTCT-3' (forward)
5'-TAATACGACTCACTATAGGGAGATTTCGGTAGAAGAAGTCTTTATTGAA-3' (reverse)
Sequences of probes for <i>LINC01554</i> RNA FISH
5'-GGACTGGCCGTTGTCTGAAAGCAAACCTCAAGGACAGGACTTCTGGAC TCCGCAGCCACTTCAGGCTGCTGCTCCATCGCATGAATTCCAGCAGGTGG CGCTGCACCCCGCGCGGACCCGGCTTGCCTCTCGGCCCCAGGGAGGTCGG TTGATGAGCAGTCGGGA-3'
5'-GGAACAGCAGCTGGCCTGAACTTGAAGGTACACCAGGTAGCTTGCGTG GGCCAGGAGAAGGTGATTCTTCTATGCCTCTCTCCAACGCCCTGCTTCT GATGCTGCTCC-3'
Sequences of primers and probes for qPCR
<i>LINC01554</i>
5'-GAGGGCAAAAGACTGCAAGC-3' (forward)
5'-CTCATCAACCGACCTCCCTG-3' (reverse)
U6
5'-CTCGCTTCGGCAGCACA-3' (forward)
5'-AACGCTTCACGAATTTGCGT-3' (reverse)
β-actin
5'-CATGTACGTTGCTATCCAGGC-3' (forward)
5'-CTCCTTAATGTCACGCACGAT-3' (reverse)
Probes for Taqman Assay
U6
5'-CGCAAGGATGACACGCAAATTCGTGAAGCGTTCCATATTTTT-3'
miR-365a-3p
5'-UAAUGCCCCUAAAAAUCCUUAU-3'
Sequences of SgRNAs for CRISPR/Cas9
1: 5'-AGAGGCAAGCCGGGTCCGCG-3'
2: 5'-GATCTGGCGTCACCGCACAA-3'

Table S2. List of antibodies used in western blot analysis and IHC

Antibody	Vendor	Catalog No.	Application
Rabbit anti-human PKM2	Cell Signaling	4053	WB, 1:1000; IHC, 1:600
Rabbit anti-human Akt	Cell Signaling	4691	WB, 1:1000
Rabbit anti-human Phospho-Akt (Ser473)	Cell Signaling	4060	WB, 1:1000; IHC, 1:100
Rabbit anti-human mTOR	Cell Signaling	2983	WB, 1:1000
Rabbit anti-human Phospho-mTOR (Ser2448)	Cell Signaling	5536	WB, 1:1000;
Rabbit anti-human Phospho-mTOR (Ser2448)	Cell Signaling	2976	IHC, 1:100
Rabbit anti-human Phospho-p70 S6K (Ser371)	Cell Signaling	9208	WB, 1:1000
Mouse anti-human Ubiquitin	Cell Signaling	3936	WB, 1:1000
Rabbit anti-human β -actin	Cell Signaling	4970	WB, 1:1000
Rabbit anti-human ki67	Abcam	16667	IHC, 1:600

Table S3. Top 10 highest score of glycolytic proteins interacting with LINC01554 identified by mass spectrometry

No.	Protein Mass	Name	NCBI-GI	Score
1	44615	Phosphoglycerate kinase 1	4505763	237.34
2	59751	ATP synthase subunit alpha	4757810	213.03
3	57000	Phospholipase C-alpha	303618	164.48
4	70906	Thioredoxin reductase	7296	160.67
5	61830	Pyruvate kinase M2	35505	146.03
6	36053	Glyceraldehyde-3-phosphate dehydrogenase	31645	130.19
7	61449	Phosphoglucomutase 1	189926	122.59
8	56651	Phosphoglycerate dehydrogenase	5620462	96.14
9	53140	Phosphogluconate dehydrogenase	984325	85.53
10	59257	Glucose-6-phosphate dehydrogenase	111052653	78.32

Table S4. List of LINC01554-interacting proteins identified by mass spectrometry

Accession No.	Protein Name
gi 4503571	alpha-enolase [Homo sapiens]
gi 37492	alpha-tubulin [Homo sapiens]
gi 194385120	unnamed protein product [Homo sapiens]
gi 18088719	Tubulin, beta [Homo sapiens]
gi 5174735	tubulin beta-2C chain [Homo sapiens]
gi 5453603	T-complex protein 1 subunit beta isoform 1 [Homo sapiens]
gi 4507729	tubulin beta-2A chain [Homo sapiens]
gi 21361322	tubulin beta-4 chain [Homo sapiens]
gi 14488680	Chain A, Crystal Structure Of Human Phosphoglucose Isomerase neuroleukinautocrine motility factormaturation Factor [Homo sapiens]
gi 4757810	ATP synthase subunit alpha, mitochondrial precursor [Homo sapiens]
gi 12804225	Unknown (protein for IMAGE:3543711) [Homo sapiens]
gi 4505763	phosphoglycerate kinase 1 [Homo sapiens]
gi 35959	tubulin 5-beta [Homo sapiens]
gi 73535278	Chain A, Human Pyruvate Kinase M2 [Homo sapiens]
gi 178685	lymphocyte activation antigen [Homo sapiens]
gi 177207	4F2 antigen heavy chain [Homo sapiens]
gi 178045	gamma-actin [Homo sapiens]
gi 32709	IFP53 [Homo sapiens]
gi 5729877	heat shock cognate 71 kDa protein isoform 1 [Homo sapiens]
gi 31543	unnamed protein product [Homo sapiens]
gi 21361621	phosphoglucomutase-1 isoform 1 [Homo sapiens]
gi 34535921	unnamed protein product [Homo sapiens]
gi 5107666	Chain A, Structure Of Importin Beta Bound To The Ibb Domain Of Importin Alpha
gi 1184537	thioredoxin reductase [Homo sapiens]
gi 306890	chaperonin (HSP60) [Homo sapiens]
gi 31645	glyceraldehyde-3-phosphate dehydrogenase [Homo sapiens]
gi 4503483	elongation factor 2 [Homo sapiens]
gi 386785	heat shock protein [Homo sapiens]
gi 28931	beta-subunit (AA 1-312) [Homo sapiens]
gi 30582781	tubulin, beta, 4 [Homo sapiens]
gi 386758	GRP78 precursor [Homo sapiens]
gi 14210536	tubulin beta-6 chain [Homo sapiens]
gi 4507877	vinculin isoform VCL [Homo sapiens]
gi 984325	phosphogluconate dehydrogenase [Homo sapiens]
gi 12653415	Heat shock 70kDa protein 9 (mortalin) [Homo sapiens]
gi 1297274	beta-tubulin [Homo sapiens]
gi 9507215	tubulin alpha-8 chain isoform 1 [Homo sapiens]
gi 28336	mutant beta-actin (beta'-actin) [Homo sapiens]
gi 303618	phospholipase C-alpha [Homo sapiens]
gi 4506243	polypyrimidine tract-binding protein 1 isoform a [Homo sapiens]

gi 31170	unnamed protein product [Homo sapiens]
gi 2851393	RecName: Full=NADPH--cytochrome P450 reductase; Short=CPR; Short=P450R
gi 825671	B23 nucleophosmin (280 AA) [Homo sapiens]
gi 8569616	Chain A, Crystal Structure Of The Moesin Ferm DomainTAIL DOMAIN Complex [Homo sapiens]
gi 181400	cytokeratin 8 [Homo sapiens]
gi 2580550	dead box, X isoform [Homo sapiens]
gi 30293	unnamed protein product [Homo sapiens]
gi 1200072	keratin [Homo sapiens]
gi 30311	cytokeratin 18 (424 AA) [Homo sapiens]
gi 4506467	radixin [Homo sapiens]
gi 189054795	unnamed protein product [Homo sapiens]
gi 5453998	importin-7 [Homo sapiens]
gi 4503481	elongation factor 1-gamma [Homo sapiens]
gi 119607256	hCG1988300, isoform CRA_a [Homo sapiens]
gi 193785329	unnamed protein product [Homo sapiens]
gi 28334	unnamed protein product [Homo sapiens]
gi 5771519	3-phosphoglycerate dehydrogenase [Homo sapiens]
gi 180631	calnexin [Homo sapiens]
gi 24485	unnamed protein product [Homo sapiens]
gi 1419564	cytokeratin [Homo sapiens]
gi 4826760	heterogeneous nuclear ribonucleoprotein F [Homo sapiens]
gi 704416	elongation factor Tu [Homo sapiens]
gi 6005942	transitional endoplasmic reticulum ATPase [Homo sapiens]
gi 4507943	exportin-1 [Homo sapiens]
gi 4758756	nucleosome assembly protein 1-like 1 [Homo sapiens]
gi 9802306	DNA-binding protein TAXREB107 [Homo sapiens]
gi 306891	90kDa heat shock protein [Homo sapiens]
gi 348239	unnamed protein product [Homo sapiens]
gi 1263196	AICAR formyltransferase/IMP cyclohydrolase bifunctional enzyme [Homo sapiens]
gi 63102283	CKAP4 protein [Homo sapiens]
gi 8546856	ENO3, muscle enolase 3 beta [Homo sapiens]
gi 30130	colligin [Homo sapiens]
gi 37267	transketolase [Homo sapiens]
gi 1184951	phosphotyrosine independent ligand p62B for the Lck SH2 domain B-cell isoform [Homo sapiens]
gi 1145799	p62/SQSTM1 [Homo sapiens]
gi 10439989	unnamed protein product [Homo sapiens]
gi 12654329	HSP90AA1 protein [Homo sapiens]
gi 1060888	26S proteasome subunit p97 [Homo sapiens]
gi 493066	glycyl-tRNA synthetase [Homo sapiens]
gi 1870688	Bruton's tyrosine kinase-associated protein-135 [Homo sapiens]
gi 434755	KIAA0123 [Homo sapiens]
gi 119581848	nucleophosmin (nucleolar phosphoprotein B23, numatrin), isoform CRA_b [Homo sapiens]
gi 226021	growth regulated nuclear 68 protein [Homo sapiens]
gi 189306	nucleolin [Homo sapiens]

gi 4503509	eukaryotic translation initiation factor 3 subunit A [Homo sapiens]
gi 577295	KIAA0088 [Homo sapiens]
gi 4757756	annexin A2 isoform 2 [Homo sapiens]
gi 36138	ribosomal protein L6 [Homo sapiens]
gi 111052653	glucose-6-phosphate dehydrogenase [Homo sapiens]
gi 3273228	very-long-chain acyl-CoA dehydrogenase [Homo sapiens]
gi 288100	initiation factor 4B [Homo sapiens]
gi 49456715	ALDOA [Homo sapiens]
gi 4502643	T-complex protein 1 subunit zeta isoform a [Homo sapiens]
gi 14141166	poly(rC)-binding protein 2 isoform b [Homo sapiens]
gi 33636742	procollagen-lysine,2-oxoglutarate 5-dioxygenase 2 isoform 1 precursor [Homo sapiens]
gi 4505343	nuclear cap-binding protein subunit 1 [Homo sapiens]
gi 194391318	unnamed protein product [Homo sapiens]
gi 23712	myoblast antigen 24.1D5 [Homo sapiens]
gi 14043666	ATAD3A protein [Homo sapiens]
gi 119594944	hCG2016942, isoform CRA_a [Homo sapiens]
gi 5031573	actin-related protein 3 [Homo sapiens]
gi 7022978	unnamed protein product [Homo sapiens]
gi 288563106	Chain A, Crystal Structure Of The Mlle Domain Of Poly(A)-Binding Protein
gi 1200184	stimulator of TAR RNA binding [Homo sapiens]
gi 38202255	threonyl-tRNA synthetase, cytoplasmic [Homo sapiens]
gi 122921194	Chain A, Crystal Structure Of Protein Phosphatase 2a (Pp2a) Holoenzyme With The Catalytic Subunit Carboxyl Terminus Truncated [Homo sapiens]
gi 34234	laminin-binding protein [Homo sapiens]
gi 3288815	citrate synthase [Homo sapiens]
gi 347134	succinate dehydrogenase flavoprotein subunit [Homo sapiens]
gi 67464147	Chain A, X-Ray Structure Of The Human Alpha-Actinin Isoform 3 At 2.2a Resolution
gi 285975	human rab GDI [Homo sapiens]
gi 1710248	protein disulfide isomerase-related protein 5 [Homo sapiens]
gi 4503445	thymidine phosphorylase precursor [Homo sapiens]
gi 119590453	EDAR-associated death domain, isoform CRA_a [Homo sapiens]
gi 5531851	homologous yeast-44.2 protein [Homo sapiens]
gi 119602169	dynein, cytoplasmic 1, heavy chain 1, isoform CRA_e [Homo sapiens]
gi 755746	p85Mcm protein [Homo sapiens]
gi 7688673	putative 55 kDa protein [Homo sapiens]
gi 4557817	succinyl-CoA:3-ketoacid-coenzyme A transferase 1, mitochondrial precursor [Homo sapiens]
gi 505108	KIAA0070 [Homo sapiens]
gi 119620305	hCG21219 [Homo sapiens]
gi 5821140	ASY [Homo sapiens]
gi 4507677	endoplasmin precursor [Homo sapiens]
gi 971270	proteasome subunit p40 / Mov34 protein [Homo sapiens]
gi 450469	type 2 inositol 1,4,5-trisphosphate receptor [Homo sapiens] ITRP2 high expression in AML
gi 119602279	hCG2029577, isoform CRA_c [Homo sapiens]
gi 55959084	seryl-tRNA synthetase [Homo sapiens]

gi 7513059	hypothetical protein KIAA0670 - human (fragment)
gi 6807655	hypothetical protein [Homo sapiens]
gi 14575679	hemicentin [Homo sapiens]
gi 194374631	unnamed protein product [Homo sapiens]
gi 6272557	ERO1L [Homo sapiens]
gi 5106557	MLL septin-like fusion protein [Homo sapiens]
gi 28872	unnamed protein product [Homo sapiens]
gi 1015321	alanyl-tRNA synthetase [Homo sapiens]
gi 340219	vimentin [Homo sapiens]
gi 5453607	T-complex protein 1 subunit eta isoform a [Homo sapiens]
gi 4507491	thimet oligopeptidase [Homo sapiens]
gi 178743	apurinic endonuclease [Homo sapiens]
gi 15825665	Chain B, Crystal Structure Of The Ku Heterodimer
gi 5031703	ras GTPase-activating protein-binding protein 1 [Homo sapiens]
gi 801893	leucine-rich PPR-motif containing protein [Homo sapiens]
gi 33878473	DHX9 protein [Homo sapiens]
gi 55960095	dystonin [Homo sapiens]
gi 5410326	RNA helicase [Homo sapiens]
gi 2996636	BIGH3 [Homo sapiens]
gi 14042277	unnamed protein product [Homo sapiens]
gi 2224663	KIAA0361 [Homo sapiens]
gi 5174613	nucleosome assembly protein 1-like 4 [Homo sapiens]
gi 71891729	KIAA1503 protein [Homo sapiens]
gi 403456	26S protease (S4) regulatory subunit [Homo sapiens]
gi 7661744	basic leucine zipper and W2 domain-containing protein 2 [Homo sapiens]
gi 409027	CDC42 GTPase-activating protein [Homo sapiens]
gi 5733122	Iodestar protein [Homo sapiens]
gi 485388	eukaryotic initiation factor 4AII [Homo sapiens]
gi 16075820	immunoglobulin heavy chain variable region [Homo sapiens]
gi 10439926	unnamed protein product [Homo sapiens]
gi 38260	MBP-2 (MHC Binding Protein-2) [Homo sapiens]
gi 31958	glutaminyl-tRNA synthetase [Homo sapiens]
gi 221042282	unnamed protein product [Homo sapiens]
gi 194389714	unnamed protein product [Homo sapiens]
gi 40788974	KIAA0886 protein [Homo sapiens]
gi 119585350	hCG2002013, isoform CRA_a [Homo sapiens]
gi 219588	DnaJ protein homolog [Homo sapiens]
gi 157833780	Chain A, Human Annexin V With Proline Substitution By Thioproline
gi 2695641	mammary tumor-associated protein INT6 [Homo sapiens]
gi 24660442	MYO18A protein [Homo sapiens]
gi 913393	nucleoprotein interactor 1 [Homo sapiens]
gi 560623	CDC10 homolog [Homo sapiens]
