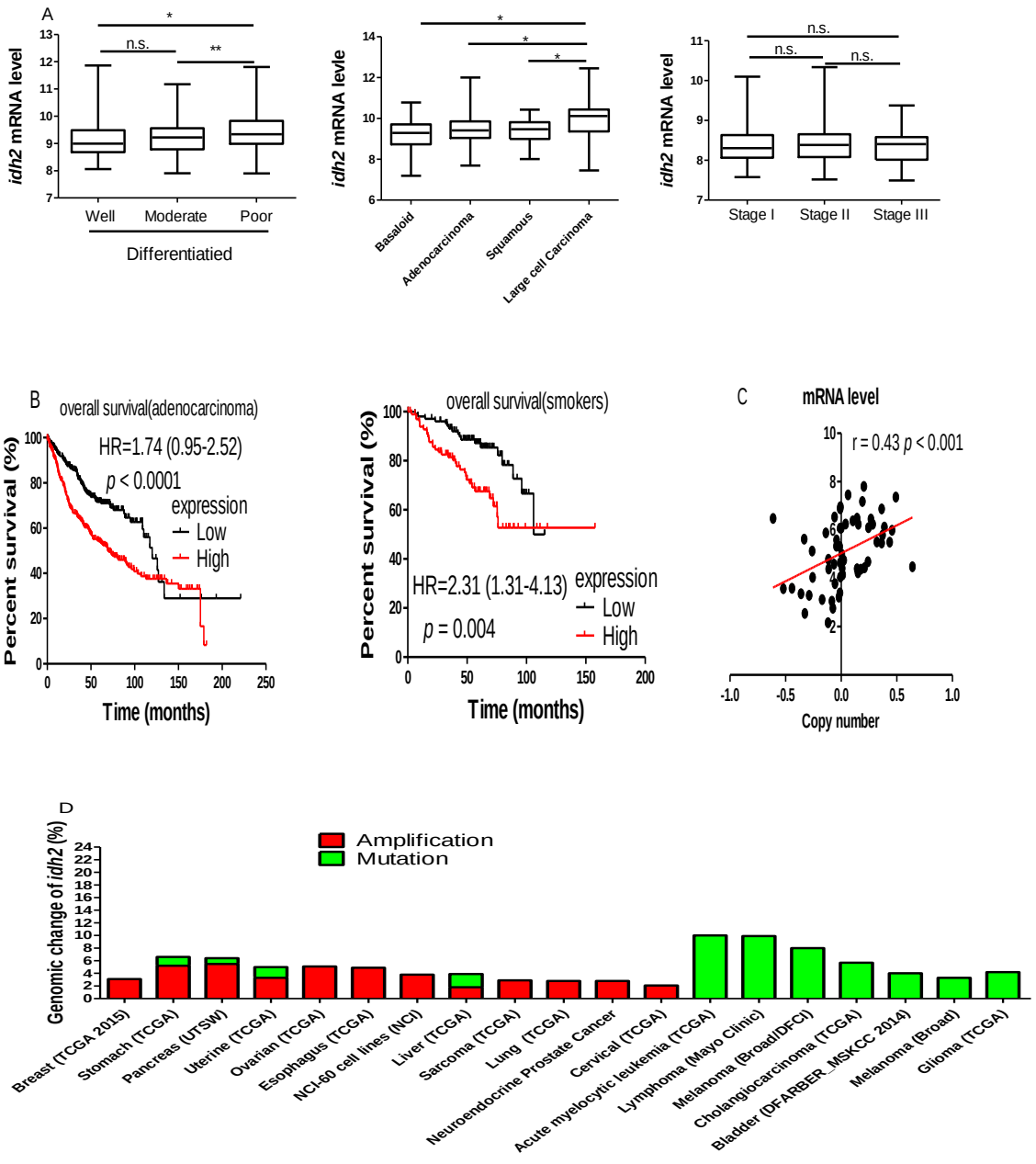

A
idh2 mRNA level
n.s.
**
Well
Moderate
Poor
Differentiatied
idh2 mRNA levle
Basaloid
Adenocarcinoma
Squamous
Large cell Carcinoma
idh2 mRNA level
n.s.
Stage I
Stage II
Stage III
B
overall survival(adenocarcinoma)
HR=1.74 (0.95-2.52)
p < 0.0001
expression
Low
High
Percent survival (%)
Time (months)
overall survival(smokers)
HR=2.31 (1.31-4.13)
p = 0.004
C
mRNA level
r = 0.43 p < 0.001
Copy number
D
Genomic change of idh2 (%)
Amplification
Mutation
Breast (TCGA 2015)
Stomach (TCGA)
Pancreas (UTSW)
Uterine (TCGA)
Ovarian (TCGA)
Esophagus (TCGA)
NCI-60 cell lines (NCI)
Liver (TCGA)
Sarcoma (TCGA)
Lung (TCGA)
Neuroendocrine Prostate Cancer
Cervical (TCGA)
Acute myelocytic leukemia (TCGA)
Lymphoma (Mayo Clinic)
Melanoma (Broad/DFCI)
Cholangiocarcinoma (TCGA)
Bladder (DFARBER_MSKCC 2014)
Melanoma (Broad)
Glioma (TCGA)

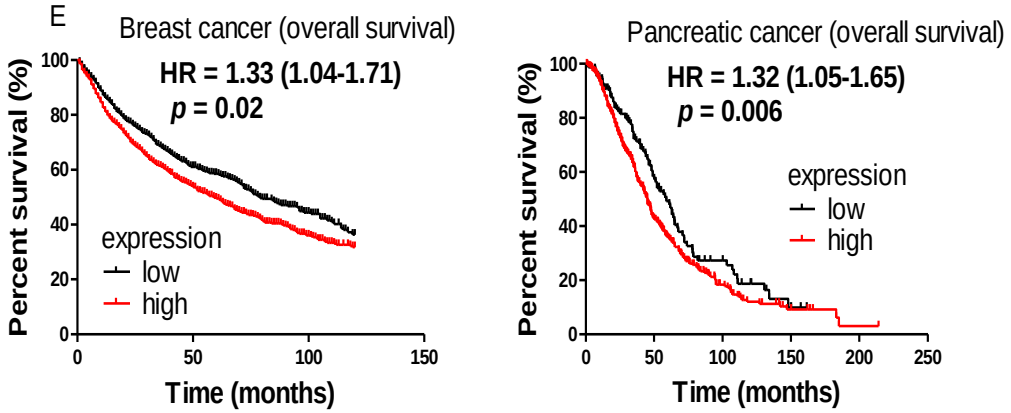


Figure S1–Additional IDH2 clinical information related to Figure 1.
(A) IDH2 expression profiles in different differential and histopathology lung cancer tissues (SurvExpress).
(B) Genomic analysis of cancers according to TCGA database (Cbioportal).
(C) Copy number and corresponding mRNA level in lung cancer cells(CCLE).
(D) Kaplan-Meier survival curves of censored Cox analysis lung cancer stratified by IDH2 expression risk group(The total number of sample analyzed was 720 and 205.) , the median value was used as the cut-off (Kmplot).
(E) Kaplan-Meier survival curves of censored Cox analysis for breast cancer and pancreatic cancer stratified by IDH2 expression risk group (Kmplot and SurvExpress).

Table S1 IDH2 mRNA expression of lung cancer in different datasets[1-8].

Experiments	IDH2 mRNA(cancer vs. normal) in datasets	P value
Hou Lung	1.92(Adenocarcinoma)	2.59E-14
	2.02(Squamous Cell Lung Carcinoma)	8.91E-13
	2.68(Large Cell Lung Carcinoma)	6.74E-7
Su Lung	2.08(Adenocarcinoma)	1.47E-7
Stearman Lung	1.96(Adenocarcinoma)	3.38E-7
Landi Lung	1.96(Adenocarcinoma)	1.90E-16
Beer Lung	1.59(Adenocarcinoma)	9.04E-6
Bhattacharjee lung	4.08(Squamous Cell Lung Carcinoma)	1.69E-4
	2.03(Small Cell Lung Carcinoma)	0.03
Selamat Lung	2.07(Adenocarcinoma)	1.61E-16
Okayama Lung	2.02(Adenocarcinoma)	2.86E-13

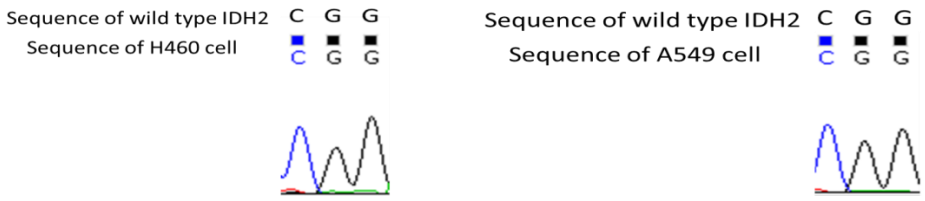
Table S2 The positive correlation of IDH2 mRNA with 43 HIF1α target gene (a total of 74 HIF1α target gene was analyzed)[9].

IDH2	p	IDH2	p	IDH2	p	IDH2	p
RNF24	*	DPM2	**	XPO5	***	RUVBL2	***
CA9	*	SLCO1B3	**	GSS	***	PLAU	***

PSMA7	*	PGK1	**	BNIP3	***	RNPS1	***
PSMB7	*	SLC6A8	**	MIF	***	TPBG	***
VEZT	*	TNS4	**	CDCA4	***	RAN	***
IL8	*	PFKFB4	***	ECE2	***	ANGPTL4	***
TUBB2C	**	PGF	***	GAPDH	***	ADORA2B	***
ANKRD9	**	MNAT1	***	MTX1	***	P4HA1	***
TFAP2C	**	TRMT5	***	ANLN	***	LDHA	***
TANC2	**	PDZD11	***	NME1	***	ALDOA	***
PPARD	**	B4GALT2	***	TPD52L2	***	PDK1	***

The asterisks (*, **, ***) indicate a significant ($p < 0.05$, $p < 0.01$, $p < 0.001$, respectively) difference.

A



B

Score	Expect	Identities	Gaps
854 bits(462)	0.0	462/462(100%)	0/462(0%)

```
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Sbjct  947  ACGCCCATCGTAGGCTTTCAGTATGGTGTTCTTGGTGCTCATGTACAGCGGCCATTTCTT  888

Query  79   CTGGATGGCATACTGGAAGCAGCTGTGCGCAAAACCTGAGATGGACTCGTCGGTGTTGTA  138
Sbjct  887  CTGGATGGCATACTGGAAGCAGCTGTGCGCAAAACCTGAGATGGACTCGTCGGTGTTGTA  828

Query  139  CATGCCCATGCCCACGCCGCCTGCGGGGAAGTTGTACACTTCCCACTCCTTGACACCACT  198
Sbjct  827  CATGCCCATGCCCACGCCGCCTGCGGGGAAGTTGTACACTTCCCACTCCTTGACACCACT  768

Query  199  GCCATCTTTTGGGGTGAAGACCATTTTGAAAGTGCCGGCCCGGTCTGCCACAAAGTCTGT  258
Sbjct  767  GCCATCTTTTGGGGTGAAGACCATTTTGAAAGTGCCGGCCCGGTCTGCCACAAAGTCTGT  708
```
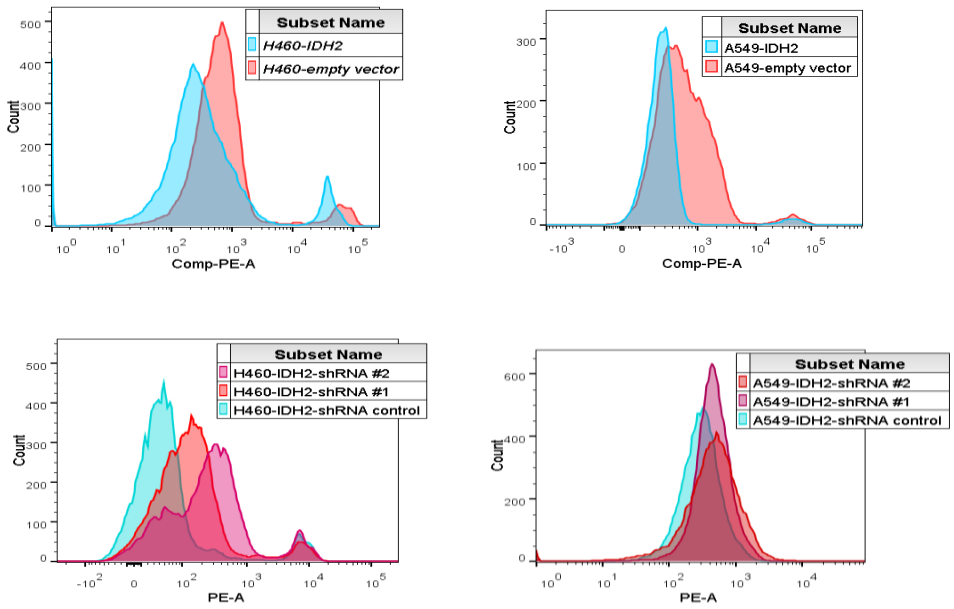

Figure S2 – Sequences analysis of IDH2 in H460 and A549 lung cancer cell lines related to Figure 2.

(A) Hotspot site of IDH2 mRNA sequences in H460 and A549 lung cancer cell lines. The cDNA region contains Hotspot site of IDH2 was amplified and sequencing by Sanger sequencing method.

(B) The sequences of IDH2 cDNA region contain Hotspot site was blasted in NCBI site. A 100% of identities of H460 and A549 cell IDH2 cDNA and IDH2 wild type sequences was found.

A



B

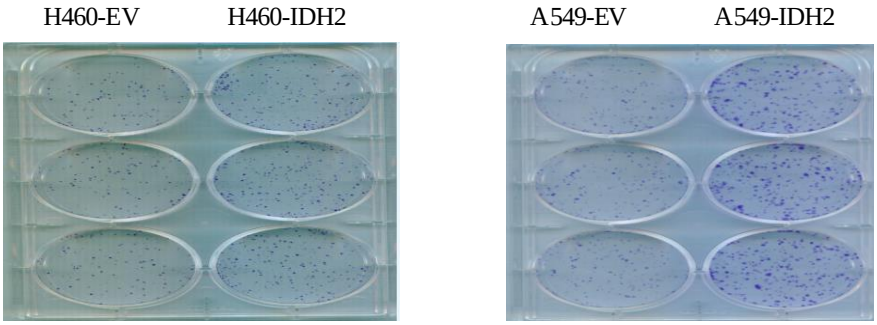
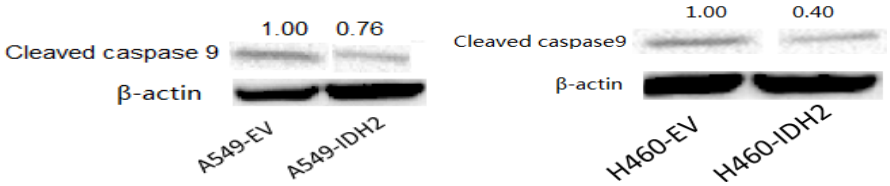


Figure S3 –Additional cell death, ROS and colony number related to Figure 4. (A) Cell death of H460 and A549 was determined by flow cytometer in IDH2 overexpression or knockdown lung cancer cell with its empty vector or controls. (B) Colony number of H460 and A549 were determined by crystal violet staining in IDH2 overexpression lung cancer cell with its empty vector.

A



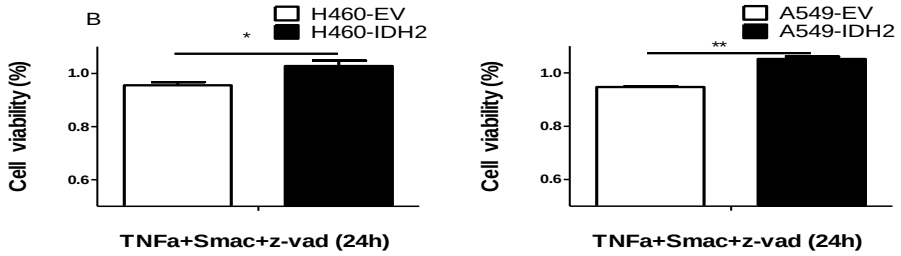


Figure S4 - Additional cell death and cell viability data related to Figure 4.
(A) Cleaved caspase 9 protein levels were determined by western blot in H460 and A549 with IDH2 overexpression lung cancer cell lines with its empty vector. β-Actin served as loading control.
(B) Induced cell necrosis were determined by MTS method in H460 and A549 with IDH2 overexpression lung cancer cell lines treated with TNFα,smac and z-vad.

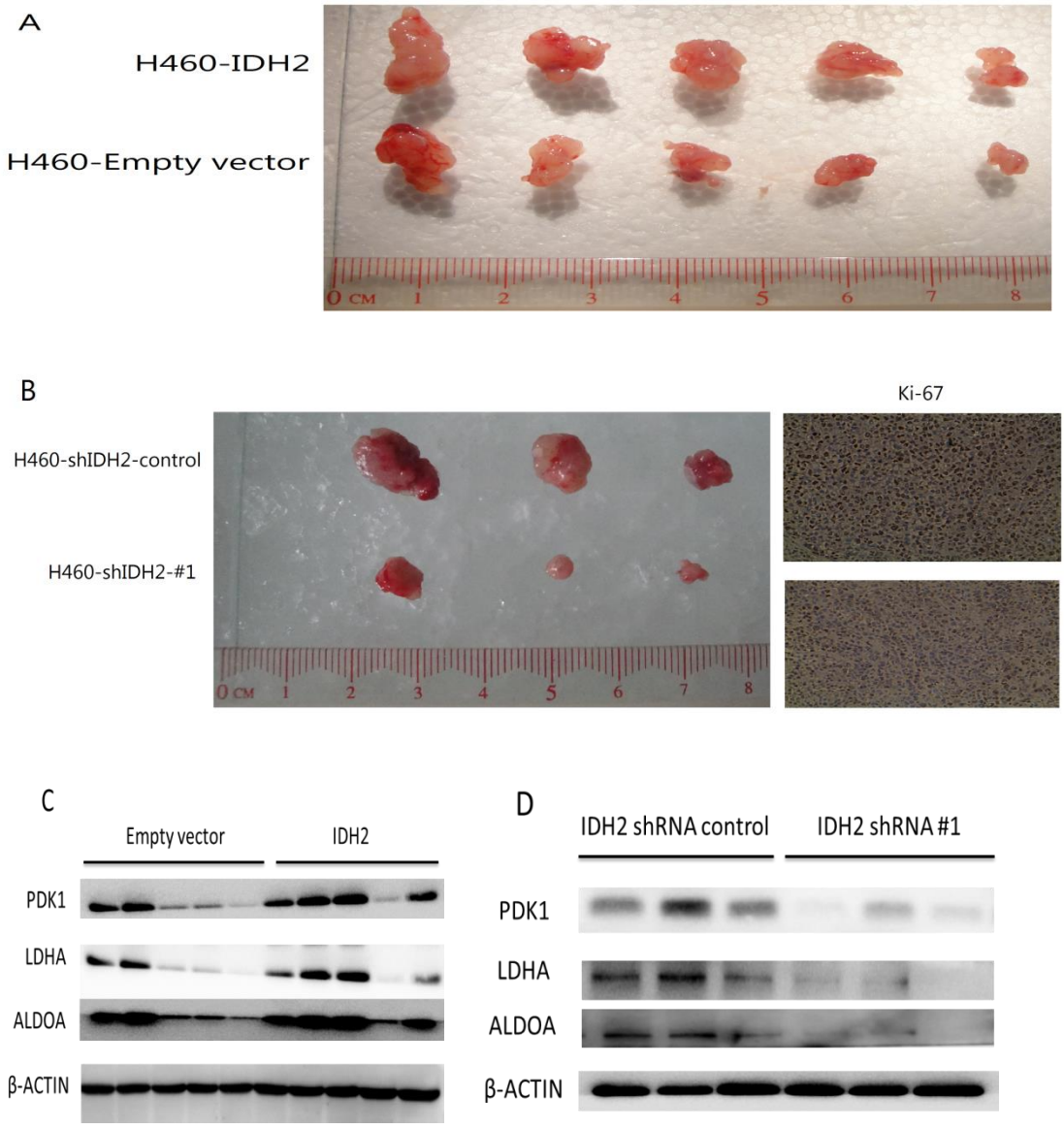


Figure S5 –Wild type IDH2 promote tumor growth related to Figure 4.
(A) H460 with IDH2 overexpression lung cancer cell xenograft tumors with its empty

vector.
(B) H460 with IDH2 knockdown lung cancer cell xenograft tumors with its control. Ki-67 immunohistochemistry staining was shown at the right (X100).
(C, D) Western blot showing HIF1 α targeted glycolysis genes in xenografts tissues from mice implanted with H460 overexpressing (C) or knockdown (D) IDH2 lung cancer cells. β-Actin was used as a loading control.

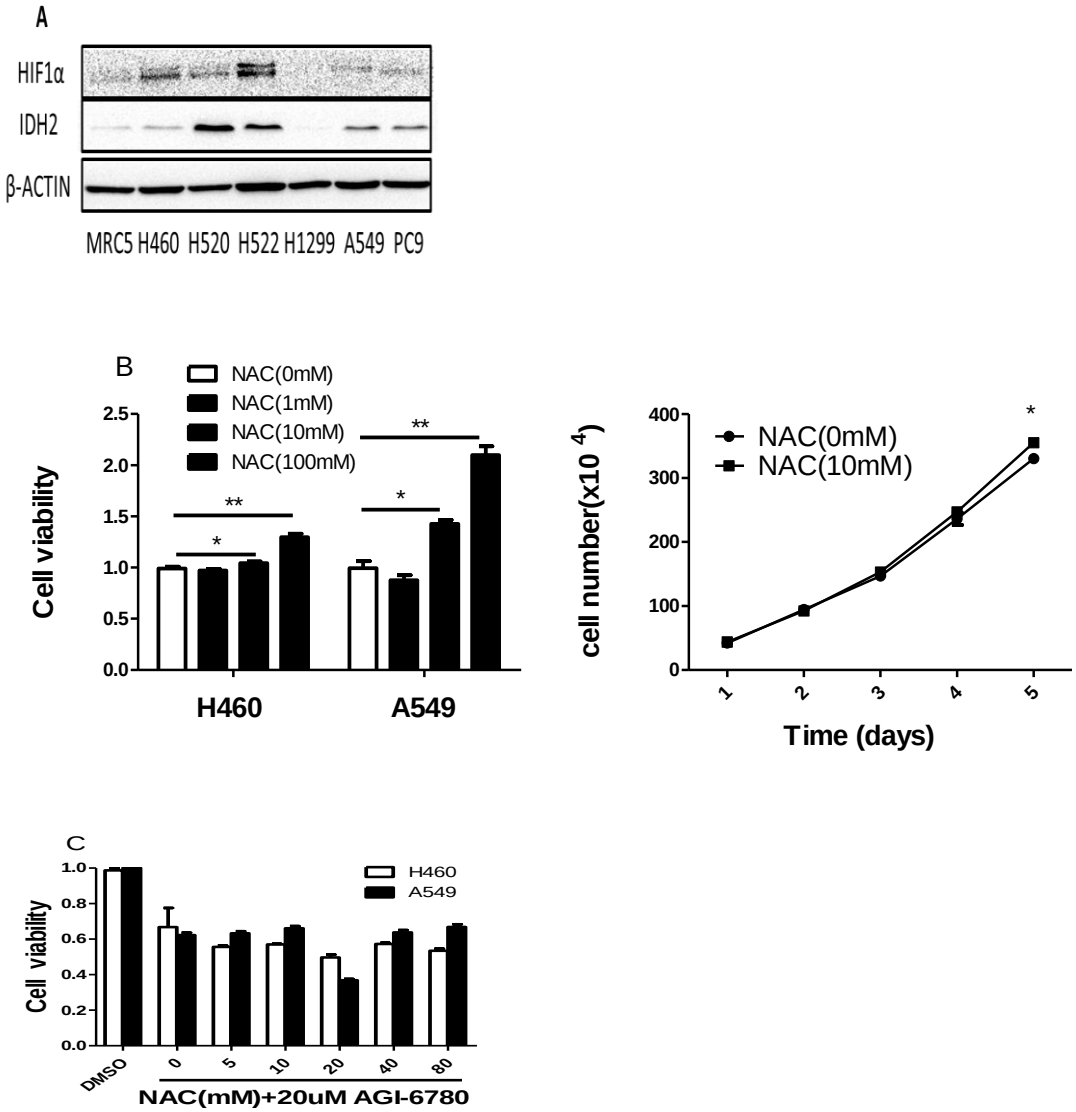


Figure S6 - Additional cell viability data related to Figure 5.

(A) Western blot showing IDH2, HIF1 α levels in lung cancer cells and normal lung fibroblast cell MRC5. β-Actin was used as a loading control.

(B) Cell viability and number were determined in H460 and A549 cells treated with the ROS scavenger, N-acetyl-cysteine.

(C) Cell viability of H460 and A549 lung cancer cell lines treatment with IDH2 inhibitor AGI-6780(20uM)and different concentration of NAC(0-80mM) was measured by MTS method.

Supplemental Experimental Procedures

Bioinformatic analyses

Clinical data were from publically available databases, which were used to explore the genomic change and mRNA expression of IDH2 in cancers including cBioprotal (TCGA) and Oncomine [10-12]. Kmplot was used to compare survival profiles of cancer patients divided with high and low IDH2 expression [13]. Survexpress was utilized to analyze the mRNA of IDH2 in different differential, pathological type and clinical stage lung cancer tissues [14].

Reagents

TNFα was from peprotech. Smac and z-vad were from selleck. Ki-67 antibody and immunohistochemistry reagents were from ZSGB-BIO.

Colony number analysis

$3x10^2$ cells was seeded in 6-well plate for 2 weeks and the culture medium was changed every 3 days. Crystal violet solution was used to stain the cells and cell number was counted by Image J software.

Sequences of Primers Used[15]

β-actin

Forward AGAGCTACGAGCTGCCTGAC

Reverse AGCACTGTGTTGGCGTACAG

aldoa

Forward GCCCGTTATGCCAGTATCT

Reverse AGCCAAGACCTTCTCTGTAA

ldha

Forward GGTTGAGAGTGCTTATGA

Reverse AACACTAAGGAAGACATCA

pdk1

Forward CGGATCAGAAACCGACACA

Reverse ACTGAACATTCTGGCTGGTGA

Supplemental References

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