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Supplemental Information

RPL32 Promotes Lung Cancer Progression by Facilitating p53 Degradation

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Figure S1

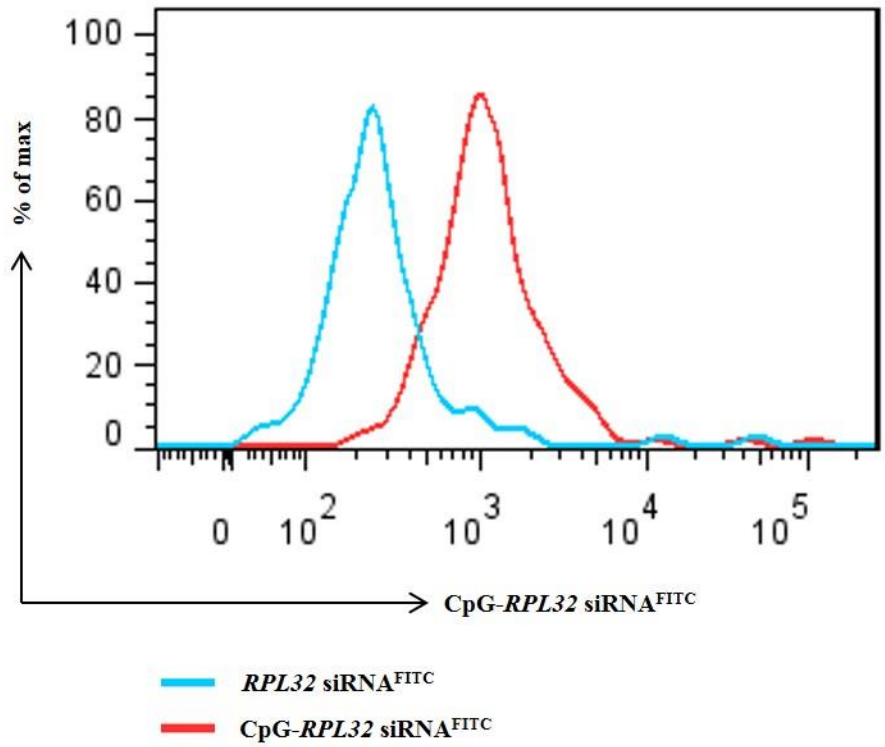

Figure S1. Internalization of CpG-siRNA conjugates by A549 lung cancer cells in vitro. A549 cells were incubated with fluorescently labeled CpG-*RPL32* siRNA^{FITC} or unconjugated *RPL32* siRNA^{FITC} (500 nM) for 1 hour, and the level of uptake was analyzed using flow cytometry.

Figure S2

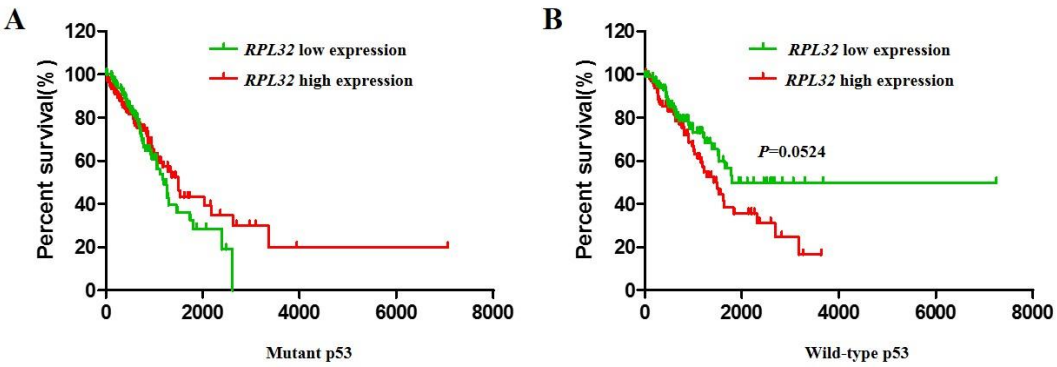

Figure S2. Survival analysis of the relationship between the expression of the *RPL32* gene and prognosis of patients from the TCGA lung adenocarcinoma dataset. (A) Kaplan-Meier survival curves of lung cancer patients with mutant p53 based on *RPL32* expression. (B) Kaplan-Meier survival curves of lung cancer patients with wild-type p53 based on *RPL32* expression.

Figure S3

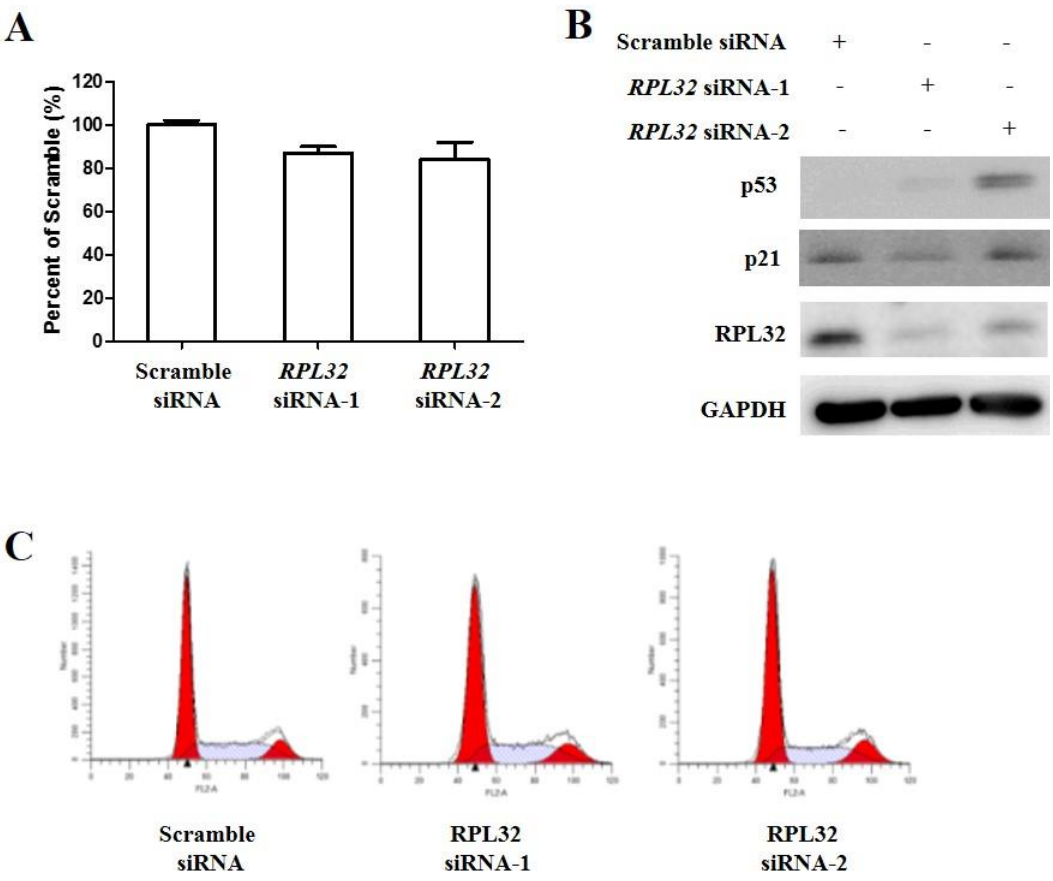

Figure S3. *RPL32* silencing has little effect on the proliferation of the normal lung epithelial cell line BEAS-2B. (A) BEAS-2B cells were counted 96 hours after transfection with scramble siRNA or *RPL32* siRNA (n=3; mean±s.d.) (B) Immunoblot analysis of RPL32, p53 and p21 in BEAS-2B cells transfected with scramble siRNA or *RPL32* siRNA. (C) Representative histograms of cell cycle analysis of BEAS-2B cells transfected with scramble siRNA or *RPL32* siRNA.

Figure S4

A

No. of Positive clone	NCBI Accession	Gene Name
1、2	XM_009248038.1	NAP1L1
3	NM_015014.3	RBM34
4	XM_001156825.5	RPL32
5	NM_000117.2	EMD
6	NM_017779.5	DEPDC1
7、10、12、13、15、23	NM_018407.4	LAPTM4B
8、22	NM_001212.3	C1QBP
9	NM_001330231.1	NAP1L1
11	NM_001190764.1	TMEM238
14	NM_053045.1	TMEM203
16	AY129295.1	NUB1
17、18	NM_005216.4	DDOST
19	NM_006579.2	EBP
20	XM_017000767.1	RPRD2
21	NM_003217.2	TMBIM6
A total of 23 positive clones, encoding 15 proteins		

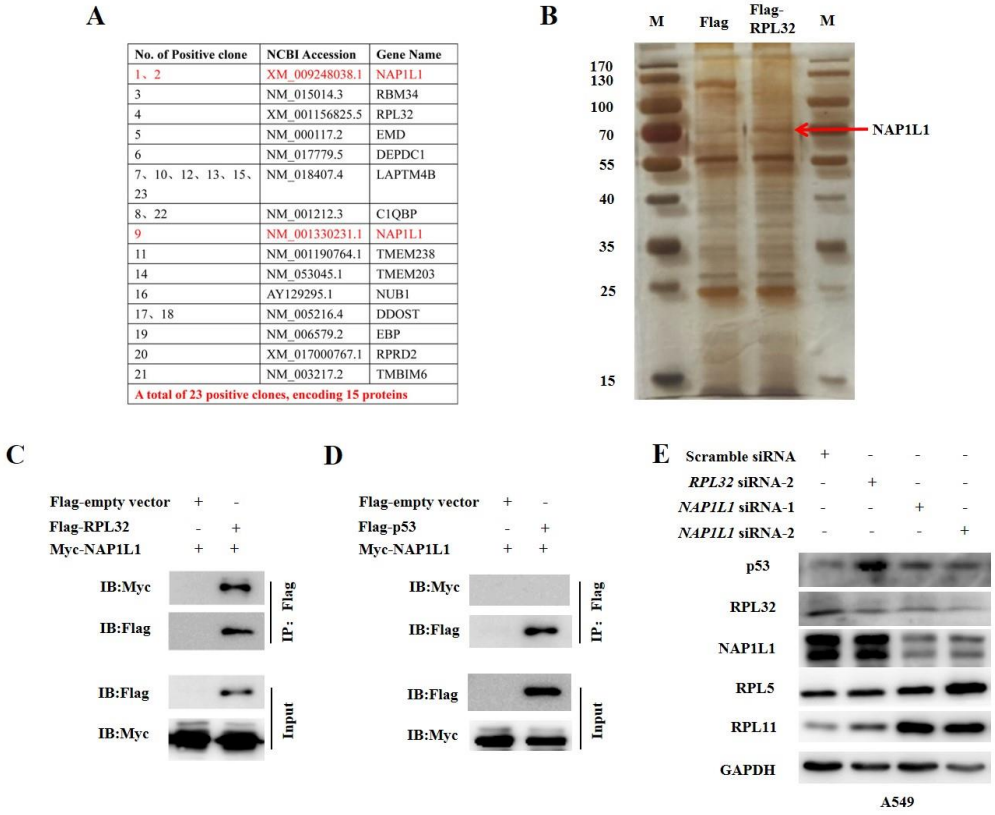

Figure S4. NAP1L1 was screened and validated to interact with RPL32. (A) List of positive cDNA clones screened in the A549 cDNA library using the yeast two-hybrid system and RPL32 as bait. (B) After transfection of pcDNA3.1-Flag or pcDNA3.1-Flag-RPL32, HEK293 cells were lysated and then immunoprecipitated with anti-Flag antibody, resolved on SDS-PAGE gel and silver-stained. Differential strips are indicated with arrows. (C) Validation of the interaction between RPL32 and NAP1L1 by immunoprecipitation. (D) Detection of the interaction between p53 and NAP1L1 by immunoprecipitation. (E) Immunoblot analysis of RPL32, NAP1L1, p53, RPL5 and RPL11 in A549 cells transfected with *RPL32* siRNA and *NAP1L1* siRNA alone or together.

Table S1. Clinical features of lung cancer patients