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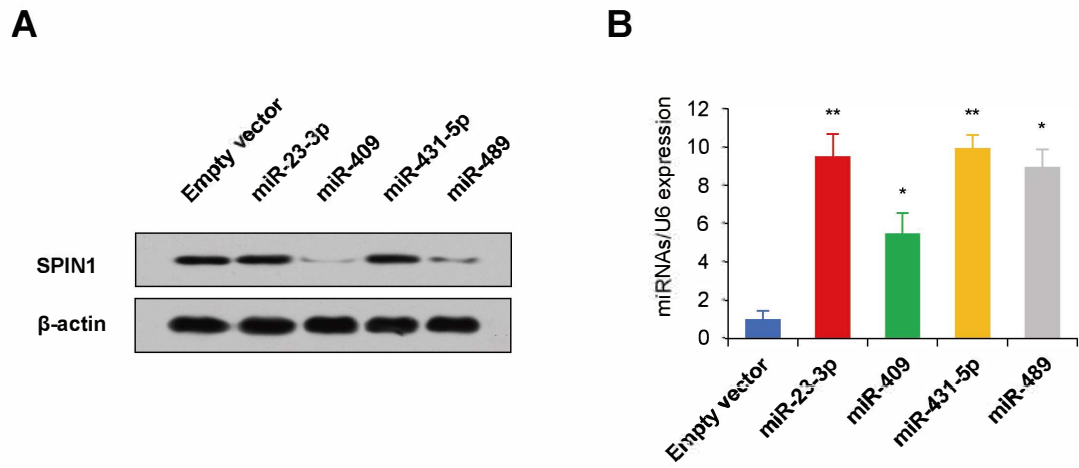
## **Supplemental Information**

**miR-409 Inhibits Human Non-Small-Cell Lung**

**Cancer Progression by Directly Targeting SPIN1**

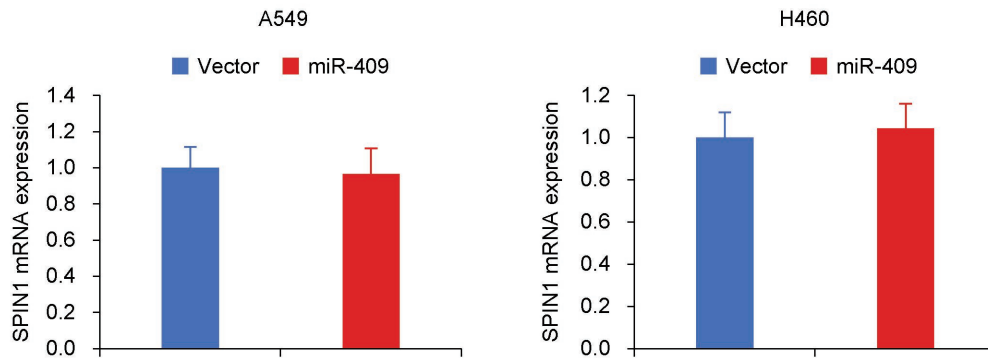
**Qi Song, Quanbo Ji, Jingbo Xiao, Fang Li, Lingxiong Wang, Yin Chen, Yameng Xu, and Shunchang Jiao**

Figure S1



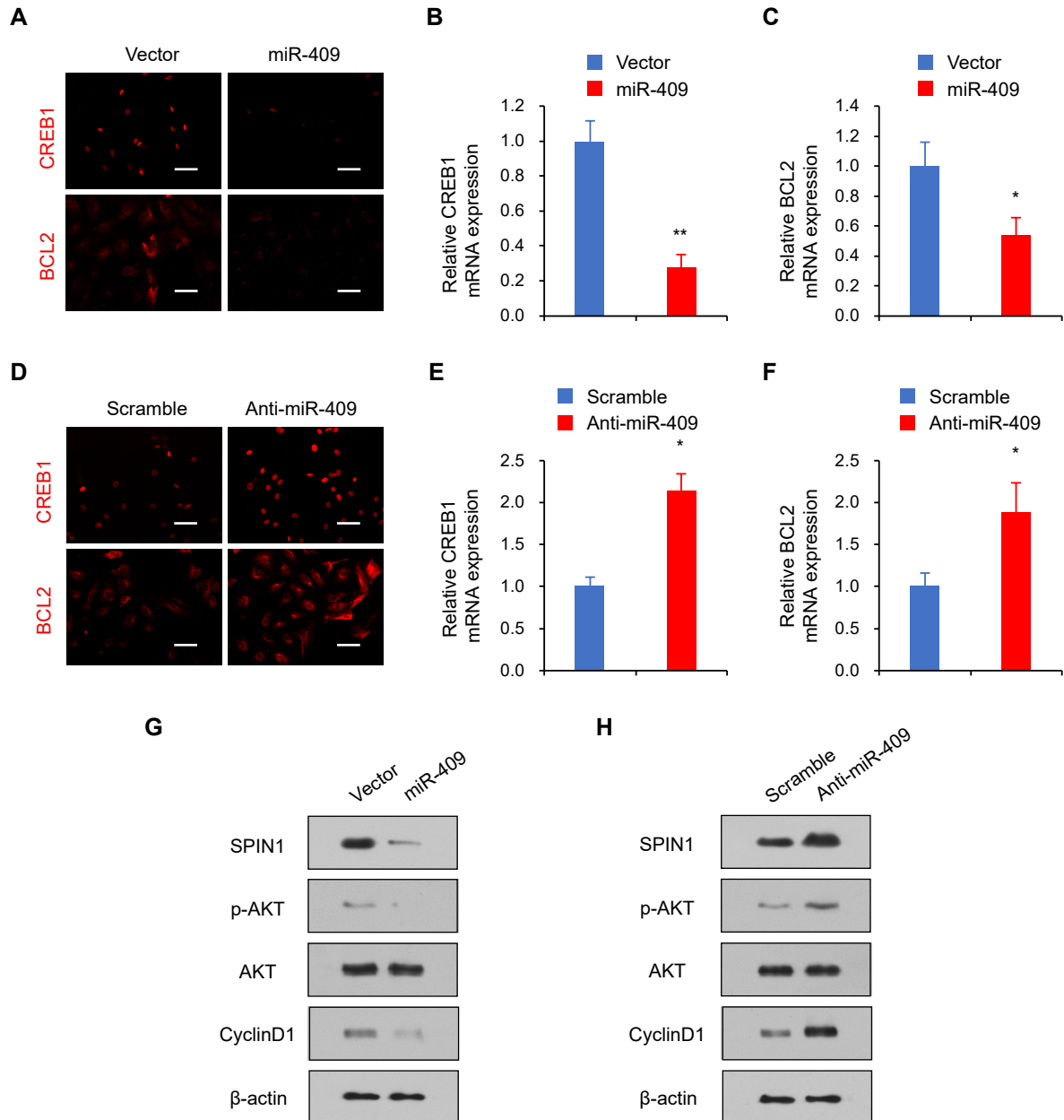
**Figure S1. Screening for potential SPIN1-targeting miRNAs.** Immunoblot analysis of the A549 cells transfected with empty vector, miR-23-3p, miR-409, miR-431-5p, or miR-489. Histograms on the right of the immunoblot graphs show corresponding expression levels of miRNAs by qRT-PCR. \* $p < 0.05$  and \*\* $p < 0.01$ .

## Figure S2



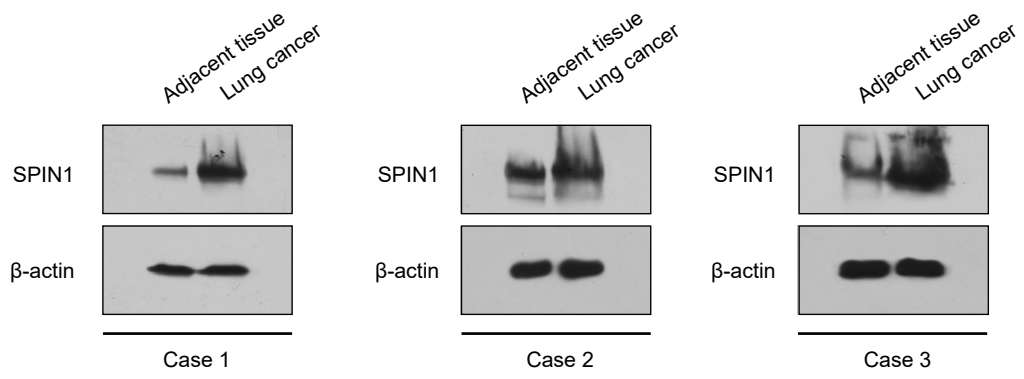
**Figure S2. miR-409 regulates SPIN1 expression in a posttranscriptional way.** Analysis of A549, H460 cells transfected with scramble or miR-409 mimics. The corresponding miRNA expression was determined using qRT-PCR. Each bar represents the mean of at least three independent experiments performed in triplicate  $\pm$  standard deviation. \* $p < 0.05$  and \*\* $p < 0.01$ .

**Figure S3**



**Figure S3. miR-409 regulates the activation of PI3K/AKT pathway.** (A) Immunofluorescent staining were conducted in A549 cells transfected with miR-409. (B) CREB1 expression levels were assessed by qRT-qPCR. (C) BCL2 expression levels were assessed by qRT-qPCR. (D) Immunofluorescent staining were conducted in A549 cells transfected with anti-miR-409. (E) CREB1 expression levels were assessed by qRT-qPCR. (F) BCL2 expression levels were assessed by qRT-qPCR. \* $p < 0.05$ , \*\* $p < 0.01$ . (G) Immunoblot analysis of the A549 transfected with miR-409. (H) Immunoblot analysis of the A549 transfected with anti-miR-409.

**Figure S4**



**Figure S4. SPIN1 is upregulated in NSCLC patients.** SPIN1 expression was analyzed in human lung cancer tissues and adjacent tissues from 3 representative donors by Western blotting.

**Supplementary Table 1** Clinicopathological characteristics of NSCLC patients

<b>Characteristics</b>	<b>n (%)</b>
Age	
$\geq$ 60	47 (55.3%)
$<$ 60	38 (44.7%)
Gender	
Male	62 (72.9%)
Female	23 (27.1%)
Tumor size (cm)	
$>$ 3	34 (40.0%)
$\leq$ 3	51 (60.0%)
TNM Stage	
I	43 (50.6%)
II	31 (36.5%)
III	11 (12.9%)
Pleural invasion	
NO	68 (80.0%)
Yes	17 (20.0%)
Metastasis	
NO	51 (60.0%)
Yes	34 (40.0%)

**Supplementary Table 2** Primer sequences of Oligonucleotides

<b>Name</b>	<b>Forward (5'→3')</b>	<b>Reverse (5'→3')</b>
<b>Primer sequences for real-time quantitative RT-PCR</b>		
<b>SPIN1</b>	GTTCTGGACCAGGTGCCTGTAAA	CTGTCTCAAACATATGTCCACTG
<b>β-actin</b>	ATCACCATTGGCAATGAGCG	TTGAAGGTAGTTTCGTGGAT
<b>Primers sequences for PCR</b>		
<b>SPIN1 3'UTR</b>	ATGTCATCACAACTCTGCCAAA	TTTACAGTCAGGAGTTGCACATAAA
<b>SPIN1 3'UTR Mut</b>	CTTGTTCCAGGAATTACCACGGA	TCCGTGGTAATTCCTGGAACAAG
<b>SPIN1</b>	ATGAAGACCCCATTCGGAAAGAC	CTAGGATGTTTTCACCAAATCGTA