

Supplementary material

QKI-5 regulates the alternative splicing of cytoskeletal gene ADD3 in lung cancer

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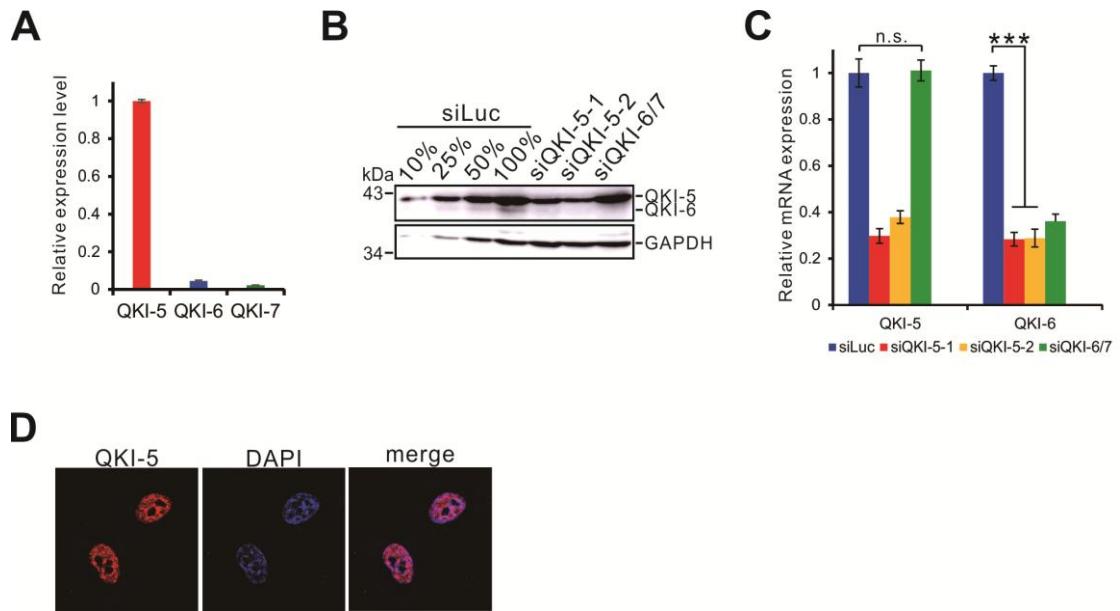
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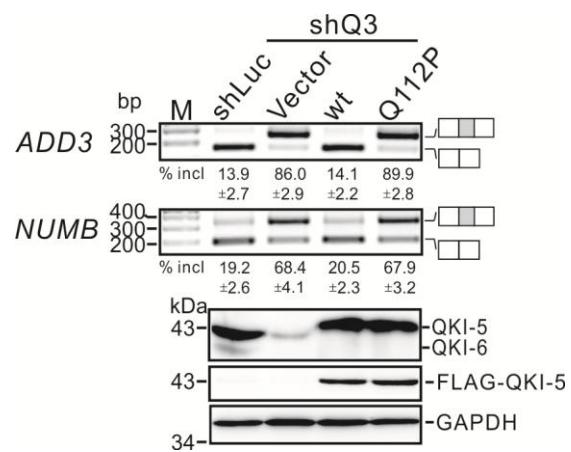
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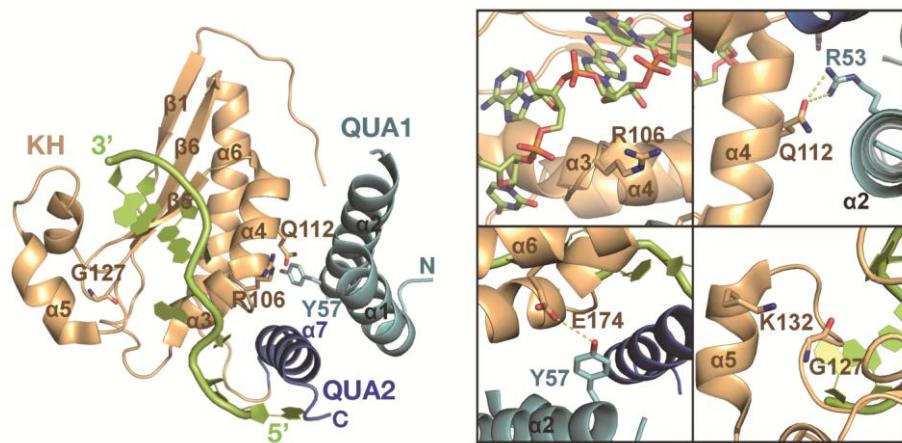
1 Supplementary Table



Supplementary Figure S1 QKI-5 is the dominant form of QKI proteins in lung cancer cells and required for QKI-6 expression. **(A)** RT-qPCR analysis of QKI-5, QKI-6, and QKI-7 mRNAs in BEAS2B cells. **(B)** Western blot analysis of QKI expression in BEAS2B cells transfected with control-, QKI-5, QKI-6/7 siRNAs. **(C)** RT-qPCR analysis of QKI-5 and QKI-6 mRNAs in cells treated as in **(B)**. **(D)** Immunofluorescent staining of QKI-5 (red) and DAPI (blue) in A549 cells.



Supplementary Figure S2 Q112P mutation in QKI does not affect the splicing of ADD3 and NUMB. RT-PCR analysis of the splicing patterns of ADD3 and NUMB in control- or QKI-specific knockdown BEAS2B cells or QKI-knockdown cells expressing wildtype or Q112P mutant QKI-5. The average percentages of exon inclusion with standard deviations are shown below (n=3).



Supplementary Figure S3 Cartoon representation of the structure of QKI-5-RNA complex (PDB 4Jvh). Key residues Y57, R106, Q112, and G127 are shown in stick.

Supplementary Table S1

Oligonucleotide name	Oligonucleotide sequences (5'-3')
ADD3-HindIII-for	TTAAAAAGCTTGAAGATGATGATCATGGCCC
ADD3-Xhol-rev	TTAAACTCGAGTCGCTTAGCAAGCTCATCTTC
ligation primer1	TAGCTACTCTGGTTTCTAATGCTAGCAAGA
ligation primer2	TTAGAAAACCAGAGTAGCTAGGACTAC
ligation primer 3	GCTGAGGCAGGAATTAAACATCTCCCAATCAG
ligation primer 4	TGTTTAATTCCCTGCCTCAGCCTCCAAA
ADD3 Mut1 for	GGCTTTGACTGAACCTTATCCAACAGATGCT
ADD3 Mut1 rev	AAGAGTTCAGTCAAAAGCCATGCAAAACAAAAC
ADD3 Mut2 for	TGCGCTGACCTCCCTGAAATCATATGCTGCTTGTTT
ADD3 Mut2 rev	TTTCAGGGAGGTCA CGCATA CATCAAATGCCTGTAT
ADD3 Mut4 for	AATGTTGACACATTGACCTCCTCTTGATGTATGATTATGGATATGGAT GACTGTTAGCA
ADD3 Mut4 rev	AGGTCAATGTGTCAACATTCTGGCTGCTAATGGTCATTCACGTACAG GTCACAAAAGATG
ADD3 Mut5 for	GCCAGAAATGTTGACACATTGACCTCCTTAATGTATGATTATGGAT
ADD3 Mut5 rev	GCATGCTAACAGTCATCCCATAATCCATAATCATAACATTAAGAGG
ADD3 Mut6 for	TTTGTGACCTGTACGTGAAATGACCATT
ADD3 Mut6 rev	GCATGCTAACAGTCATCCCATAATCCATAATCATAACATTAAGAGG
QKI5-FLAG-rev	TTAAATCTAGATTACTTGTGTCATCGTCTTGATCGTTGCCGGTGGC GGCTC
QKI5-for	TTAAAGGATCCATGGTCGGGGAAATGGAAAC
QKI5 Mut1 for	AAGACATGCCACAATGACACATTAATGGCA
QKI5 Mut1 rev	TGTGTCATTGTGCATGTCTTCCGTACTCTGC
QKI5 Mut2 for	GACCTACAGGACTTACAGCCAAACA
QKI5 Mut2 rev	GGCTGTAAGTCCTGTAGGTCCAAGGATTCTCCC
QKI5 Mut3 for	AAACCACTTGAAGCAGAAACCGGATG
QKI5 Mut3 rev	GTTTCTGCTTCAAGTGGTTGGCTGTAAGTCCTCTA
QKI5 Mut4 for	ATTATTGGTACGTGCAGCAGAAGGAGAAGACA
QKI5 Mut4 rev	CTGCTGCACGTACCAATAATTCTTCACTTCT
QKI5 Mut5 for	GAGGCAAAGTCTCAATGAGGGATAAAAAAAA
QKI5 Mut5 rev	CCTCATTGAGACTTGCCTCGGACCATGAT
QKI5 Mut6 for	GCAGTTGAAAAAGTGAAGAAATTATTGGTA

QKI5 Mut6 rev	TTCTTCACTTTCAACTGCTCTTCATTTA
QKI5 Mut7 for	AAGCAGAAACCAGATGTAAAATCATGGTCCGA
QKI5 Mut7 rev	TTTACATCTGGTTCTGCTCAAGTTGTTT
QKI5 Mut8 for	TGATAGAGCTTGCGATTCTGAATGG
QKI5 Mut8 rev	CAGAACGCAAGCTCTATCAGCTGCATCTTCTTC
QKI5 Mut9 rev	TTAAATCTAGATTACTTGTGTCATCGTCTTGAGTCTCCTCTGCTGCA GGTAC
ADD3 for	ACCAGCTCCTCCTAACCCA
ADD3 rev	CATCCTGCCATTTACTACC
NUMB for	AGGGGAGGCAGAGAGCAT
NUMB rev	TCTATGACCGGCCTGGAA
ctrl ASO	CCUAUAGGACUAUCCAGGAA (each base with 2'-O-methoxyethyl-phosphorothioate modifications)
ADD3 ASO-1	ACAUUGAGUACCUUCUAAUUUU (each base with 2'-O-methoxyethyl-phosphorothioate modifications)
ADD3 ASO-2	CUGCUCAGCAUCUGUUGGAUAA (each base with 2'-O-methoxyethyl-phosphorothioate modifications)
shLuc target sequence	CGTACGCGGAATACTTCGA
shQ3 target sequence	GAAGAGAGCAGTTGAAGAA
siRNA-QKI5-1	CUAUGACCUUCUGACCUCUGAAUU
siRNA-QKI5-3	AGUAGAUACUUUUACUAUACAAGGUU
siRNA-QKI6/7	GAAUUCAGAACGGUCUUAAUU