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Supplemental Information

**Cryptotanshinone Protects Cartilage
against Developing Osteoarthritis
through the miR-106a-5p/GLIS3 Axis**

Quanbo Ji, Dengbin Qi, Xiaojie Xu, Yameng Xu, Stuart B. Goodman, Lei Kang, Qi Song, Zhongyi Fan, William J. Maloney, and Yan Wang

Figure S1

A

Target gene	ResfSeq Id
GLIS3	NM_001042413
HMGA2	NM_003483
SMAD7	NM_005904
CCND1	NM_053056
CDK6	NM_001259

B

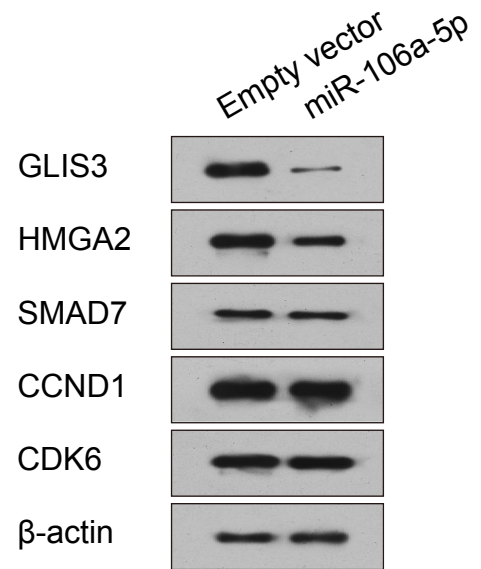


Figure S1. Potential target genes of miR-106a-5p screened. (A) Candidate target genes of miR-106a-5p were found using publicly available databases (TargetScan and miRanda). (B) Immunoblot analysis showing the expression of the candidate target genes in chondrocytes infected with miR-106a-5p.

Figure S2

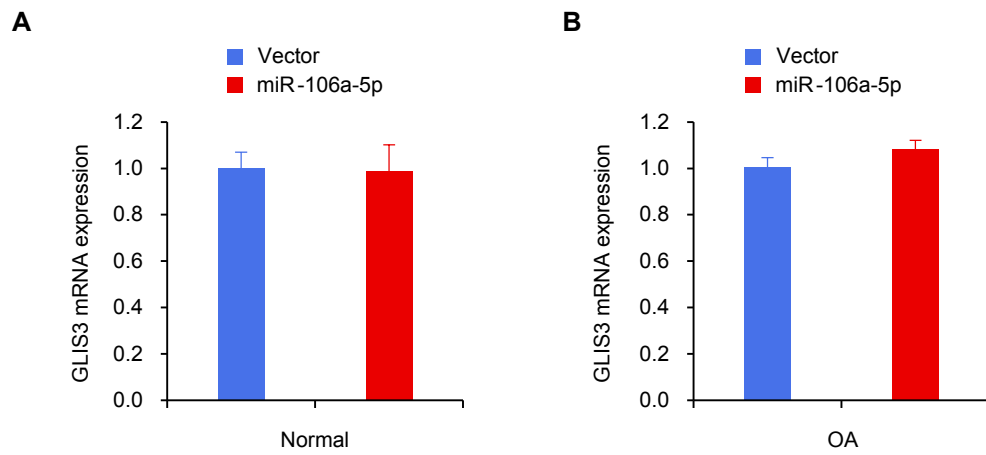


Figure S2. miR-106a-5p regulates GLIS3 expression in a posttranscriptional way. (A and B) Analysis of normal (A) and OA (B) chondrocytes transfected with scramble or miR-106a-5p mimics. The corresponding miRNA expression was determined using qRT-PCR. Each bar represents the mean of at least three independent experiments performed in triplicate \pm standard deviation. * $p < 0.05$, ** $p < 0.01$.

Table S1 Clinical and demographic characteristics of the study population*

Clinical data

Age, years	61.2 ± 3.8
Male/ female (n)	16/24
Disease duration, years	10.2 ± 3.6
Swollen joints (n)	1.3 ± 0.2
Tender joints (n)	1.2 ± 0.6
HSS score ^a	46.7 ± 6.2
CRP, mg/dl	0.5 ± 0.3
ESR, mm/h	14.2 ± 7.1

* Data are shown as the mean ± standard deviation or absolute numbers.

^a The Hospital for Special Surgery Knee Score (HSS) was used. Scores can change from 0 to 100, with lower scores suggesting greater disease activity.

CRP, C-reactive protein; ESR, erythrocyte sedimentation rate.

Table S2 Sequences of DNA and RNA Oligonucleotides

Name	Forward (5'→3')	Reverse (5'→3')
Primer sequences for real-time qRT-PCR		
GLIS3	AAGGCAGCTGCAACAATCTAGTGG	CCGTCAGACTCAAGGTCGTGGA
β-actin	ATCACCATTGGCAATGAGCG	TTGAAGGTAGTTTCGTGGAT
Primers sequences for PCR		
GLIS3 3'UTR	AAGCTCTCTTGGCCACTCCTGC	TTTTTTAACATGTATAAAAAGCTTTAATT
GLIS3 3'UTR Mut	TGTTGCTGACATAAATACAGGGG	ACAACGACTGTATTTATGTCCCC
P1 (-186 bp)	AAACCTAGCTTAGACTCTGTAAG	CCTATTCTGTAGCAAAAATTTTAA
P2 (-439 bp)	GAAGACTCGAATACTCATCCTGG	TGTGGTTTCAACCAAATCCTGAGA
P3 (-761 bp)	CITAGGTAAAATAAGCACTTGAAAAT	AGATCATTCAGAGCTTTTGGGC
P4 (-1000 bp)	ATTGATGTATAAGTTTATAAATCAG	AACATACAGCAAGGAAGTGAGC
P5 (-761 bp, Mut)	ATTAATGGGGTGAAATCAAGAATATTCATTAA	TTAAATGAATATTCCTTGATTTCCACCCCATTAAT
GLIS3	ATGAATGGAAGATCATGCAGCATGA	TAGCCTTCGGTGTAGACAGAGGA
PAX5	ATGGATTTAGAGAAAAATTATCCG	TCAGTGACGGTCATAGGCAGTGGC
IRF2	ATGCCGGTGAAAGGATGCGCA	TTAACAGCTCTTGACGCGGGCCT
STAT4	ATGTCTCAGTGGAATCAAGTCCAAC	TCATTACAGCAGAATAAGGAGACTT
