Supplementary Figures

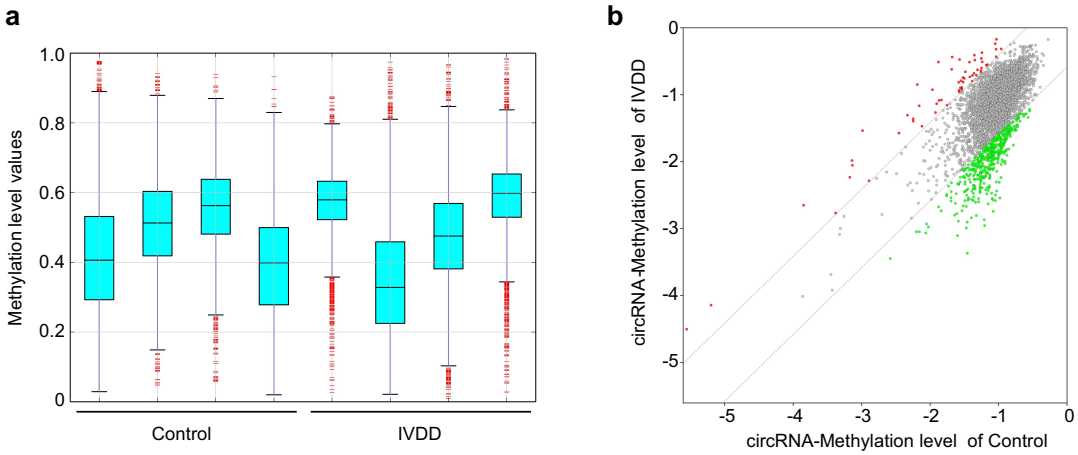


Fig. S1 (a) The expression levels of circRNAs methylation level were recorded by a box plot as the mean ± standard deviation in the Control and IVDD groups. (b) A scatter plot was used to evaluate the variation of circRNAs methylation level. The dotted lines are fold-change lines, the methylation level of circRNAs above the top line and below the bottom line indicate greater than 1.5-fold changes between the two groups.

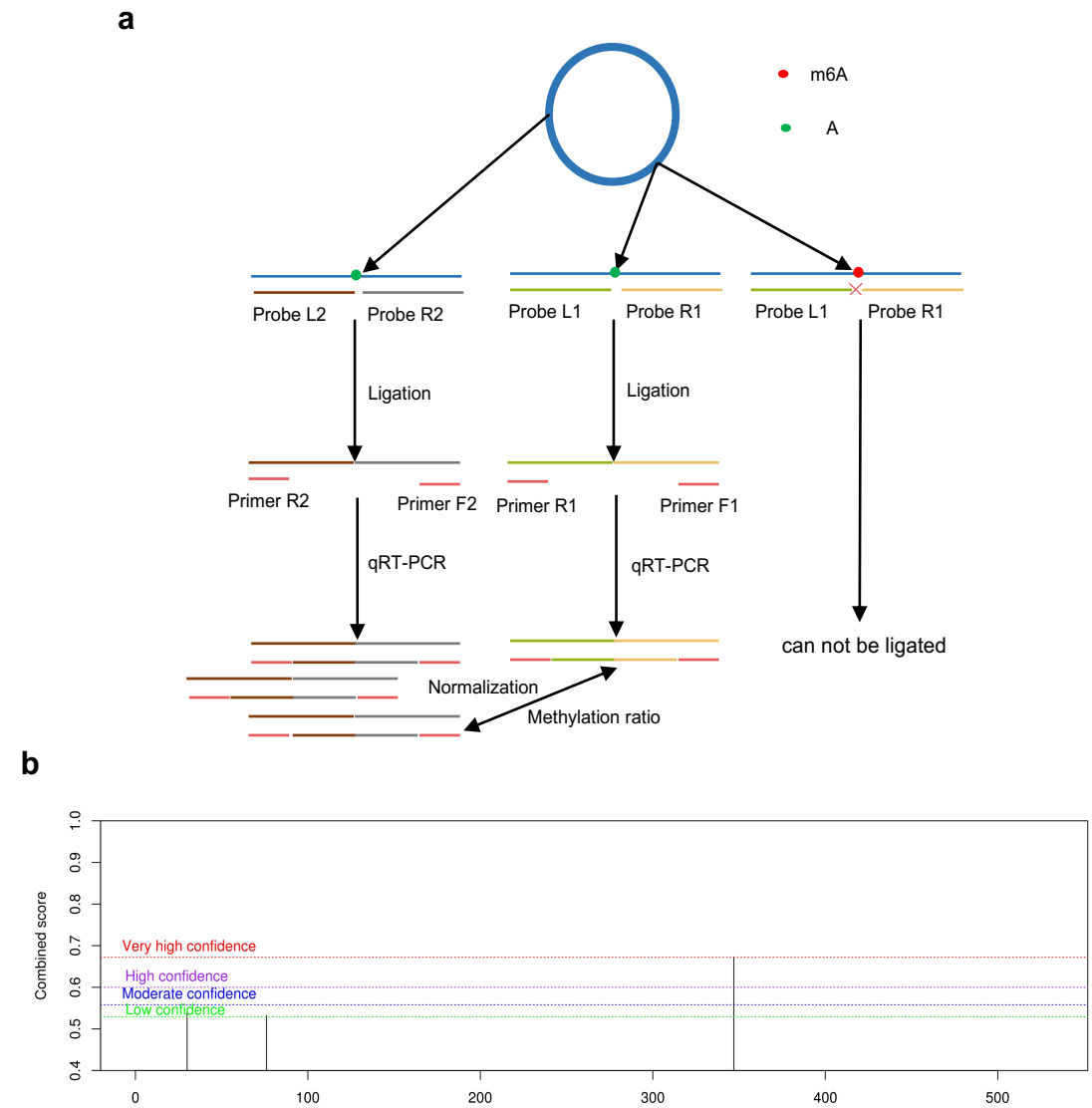


Fig. S2 (a) A schematic procedure of T3 DNA ligase assay for N6-methyladenosine (m6A) detection. (b) Prediction score of m6A site along the circGPATCH2L sequence by SRAMP prediction server.

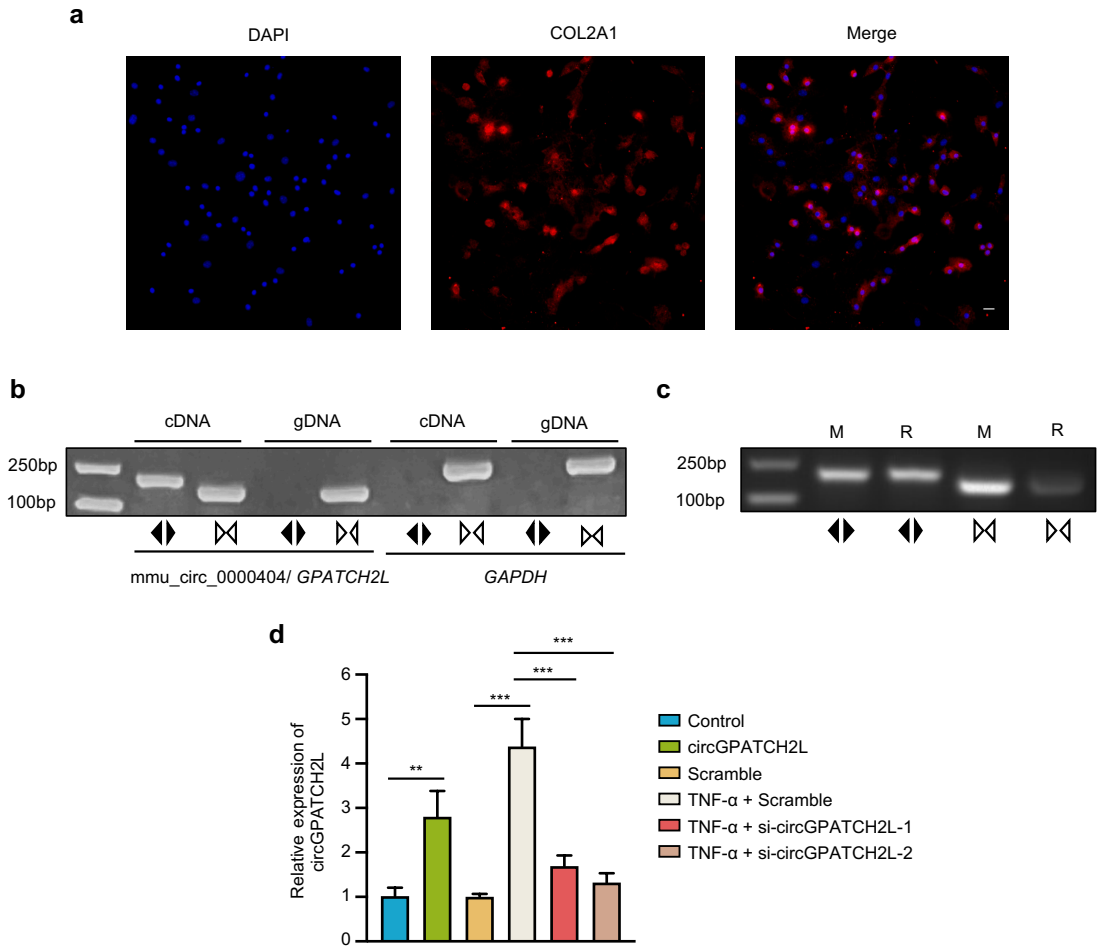
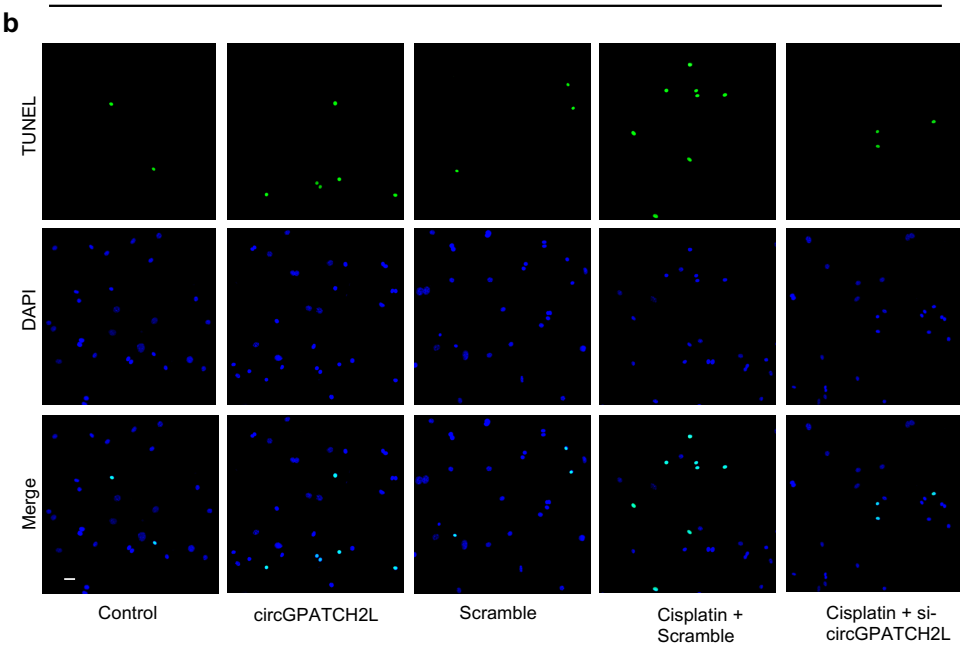


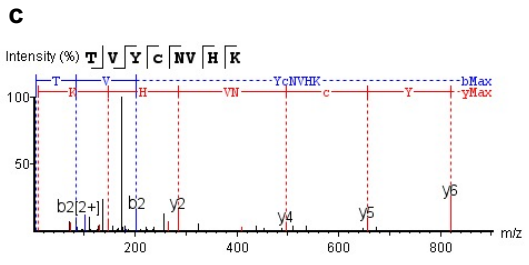
Fig. S3 (a) Identification of primary nucleus pulposus cells (NPCs). COL2A1 expression was detected by Immunofluorescence staining in NPCs. Scale bar = 50 μm. (b) PCR analysis of mmu_circ_0000404 (circGPATCH2L in mouse) and Gapdh using divergent and convergent primers in cDNA and gDNA from mouse nucleus pulposus tissue. (c) PCR analysis for mmu_circ_0000404 using divergent and convergent primers after RNase R treatment in mouse nucleus pulposus tissue. (d) qRT–PCR analysis of circGPATCH2L expression in different groups. The control group was transfected with a blank vector with flanking introns containing complementary Alu elements. NPCs transfected with sequence-scrambled DNA oligos were named Scramble. TNF-α (10 ng/ml) was used to induce an *in vitro* IVDD model. The circGPATCH2L overexpression plasmid successfully upregulated circGPATCH2L expression. Both siRNAs for circGPATCH2L downregulated circGPATCH2L expression. n=3. NPCs, nucleus pulposus cells; DAPI, 4,6-diamidino-2-phenylindole; COL2A1, collagen type II alpha 1 chain; M, mock; R, RNase R; PCR, polymerase chain reaction; cDNA, complementary DNA; gDNA, genomic DNA; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; qRT–PCR, quantitative real-time PCR;

TNF-α, tumour necrosis factor-α. Data are shown as the mean ± S.D. *** $p < 0.001$; ** $p < 0.01$.

a

Accession	Protein name	-10lgP
P49327	FASN	377.19
Q14204	DYNC1H1	343.43
P68371	TUBB4B	340.13
P04350	TUBB4A	330.01
P78527	PRKDC	326.17
P10809	HSPD1	324.66
Q9BQE3	TUBA1C	319.85
Q00610	CLTC	317.99
Q71U36	TUBA1A	314.62
P08238	HSP90AB1	310.23
Q9BVA1	TUBB2B	307.48
Q13885	TUBB2A	307.35
P07900	HSP90AA1	304.08
P49327	TRIM28	299.5
Q14697	GANAB	296.8





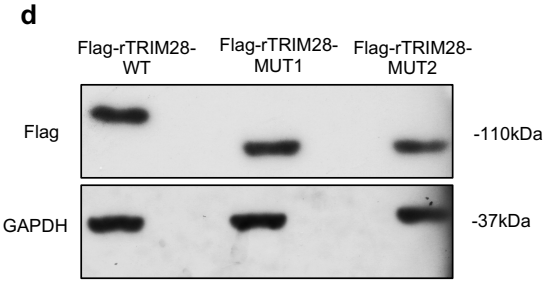


Fig. S4 (a) Identification of TRIM28 as circGPATCH2L-associated protein by Mass Spectrogram analysis in NPCs. (b) Representative images of TUNEL

immunofluorescence staining showing that circGPATCH2L induces DNA damage and apoptosis, while inhibiting circGPATCH2L rescues these phenotypes. Scale bar = 50 μm. (c) Mass spectrum of TRIM28. (d) Western blot confirming the overexpression efficiency of different mutant TRIM28 at the protein level in RIP assay. TRIM28, tripartite motif containing 28; GAPDH, glyceraldehyde 3-phosphate dehydrogenase; WT, wild type; MUT, mutant.

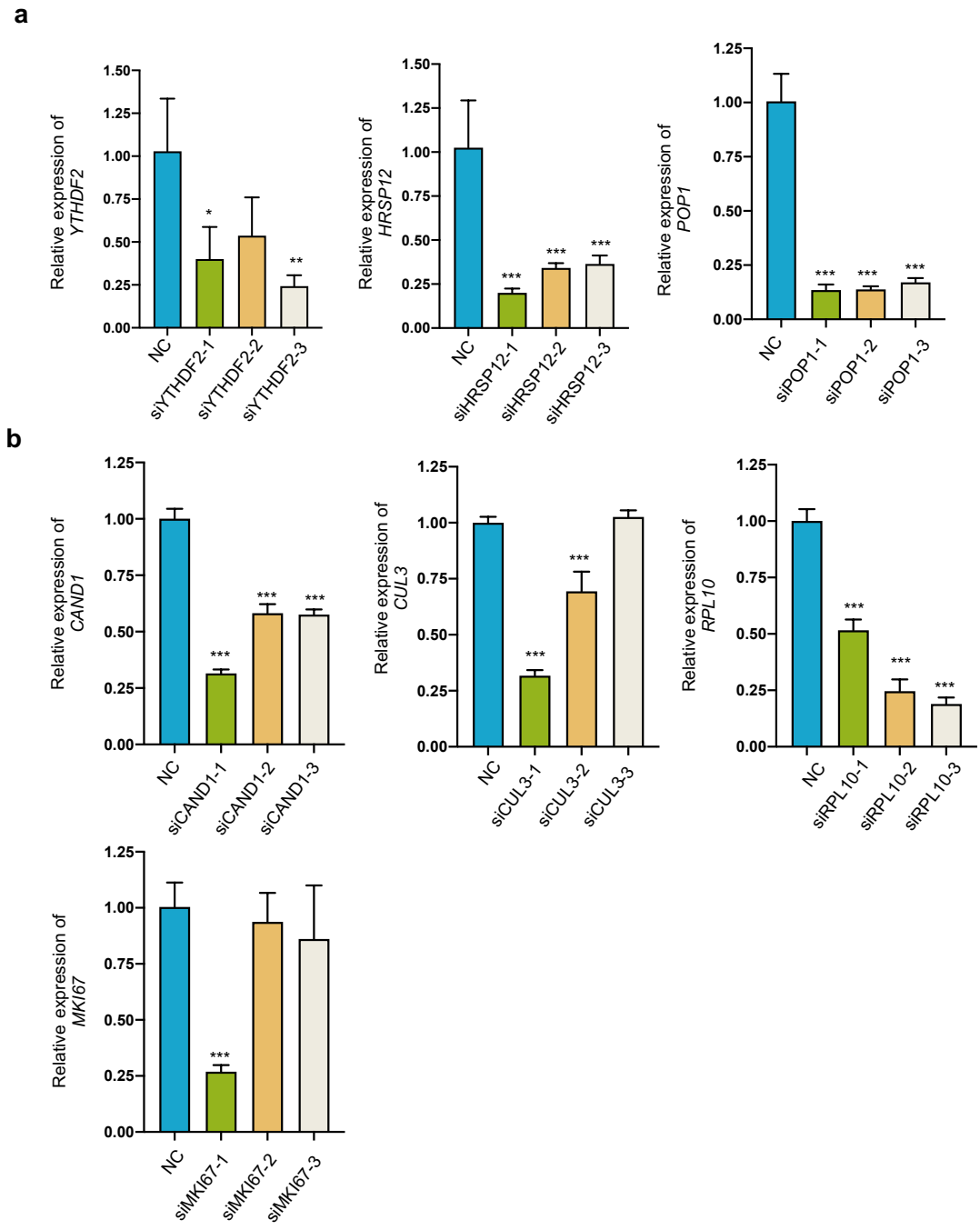


Fig. S5 (a) qRT–PCR analysis of YTHDF2, HRSP12, and POP1 in NPCs transfected

by relevant siRNAs. siYTHDF2-3, siHRSP12-1, and siPOP1-1 were selected for further experiments. NC group was treated with transfection reagent only. n = 3. (b) qRT–PCR analysis of CAND1, CUL3, RPL10, and MKI67 in NPCs transfected by relevant siRNAs. siCAND1-1, siCUL3-1, siRPL10-3, and siMKI67-1 were selected for further experiments. NC group was treated with transfection reagent only. n = 3. YTHDF2, YTH N6-Methyladenosine RNA binding protein 2; qRT–PCR, quantitative real-time polymerase chain reaction; HRSP12, 2-iminopropanoate deaminase; POP1, Ribonucleases P/MRP protein subunit POP1; CAND1, cullin-associated and neddylation-dissociated 1; RPL10, ribosomal protein L10; CUL3, cullin 3; MKI67, marker of proliferation Ki-67. Data are shown as the mean ± S.D. *** $p<0.001$; ** $p<0.01$; * $p<0.05$.