Ageing and Cancer
Research & Treatment

https://doi.org/10.70401/acrt.2026.0009

Supplementary information

Ythdf1 deficiency exacerbates colitis but prevents colitis-associated cancer in mice

Zhen Zhang, Jie Zhang, Chenzhong Xu, Yuping Ning, Baohua Liu

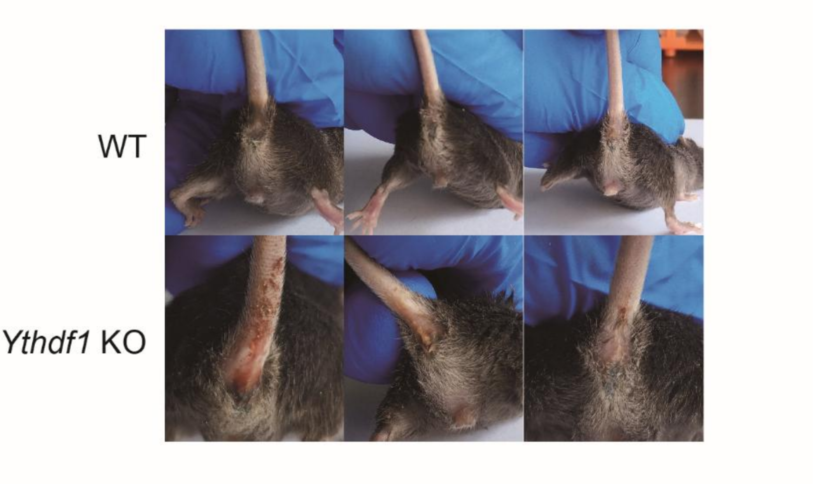

Figure S1. Images showing representative diarrhoea and bloody stools in *Ythdf1* KO mice at day 7 following DSS challenge. Three mice per genotype were analysed.

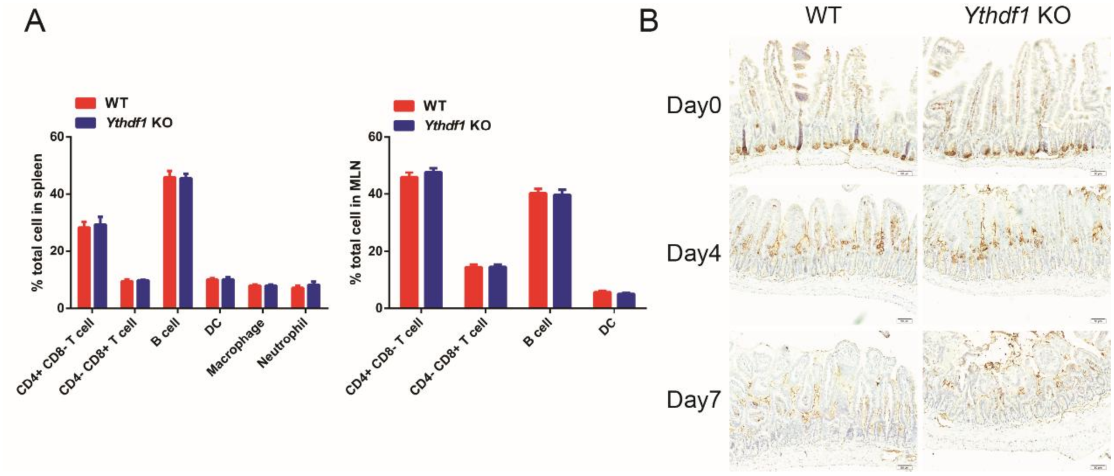

Figure S2. (A) Flow cytometry analysis of immune cell frequency in spleen (left) and MLN (right) after the induction of colitis in WT (*n* = 5) or KO (*n* = 4) mice; (B) Lysozyme expression in ileum sections at different time points after exposure to DSS, as determined with IHC (scale bar, 50 μm).

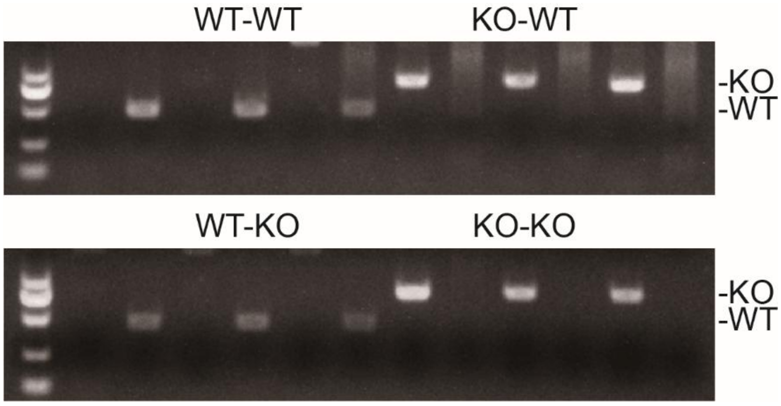

Figure S3. The results of genotyping peripheral blood cells isolated from the indicated 4 groups of chimeric mice with bone marrow reconstitution. WT-WT: WT mice reconstituted with WT bone marrow cells; KO-WT: WT mice reconstituted with *Ythdf1* KO bone marrow cells; WT-KO: *Ythdf1* KO mice reconstituted with WT bone marrow cells; KO-KO: *Ythdf1* KO mice reconstituted with *Ythdf1* KO bone marrow cells. Three mice per group were analysed.

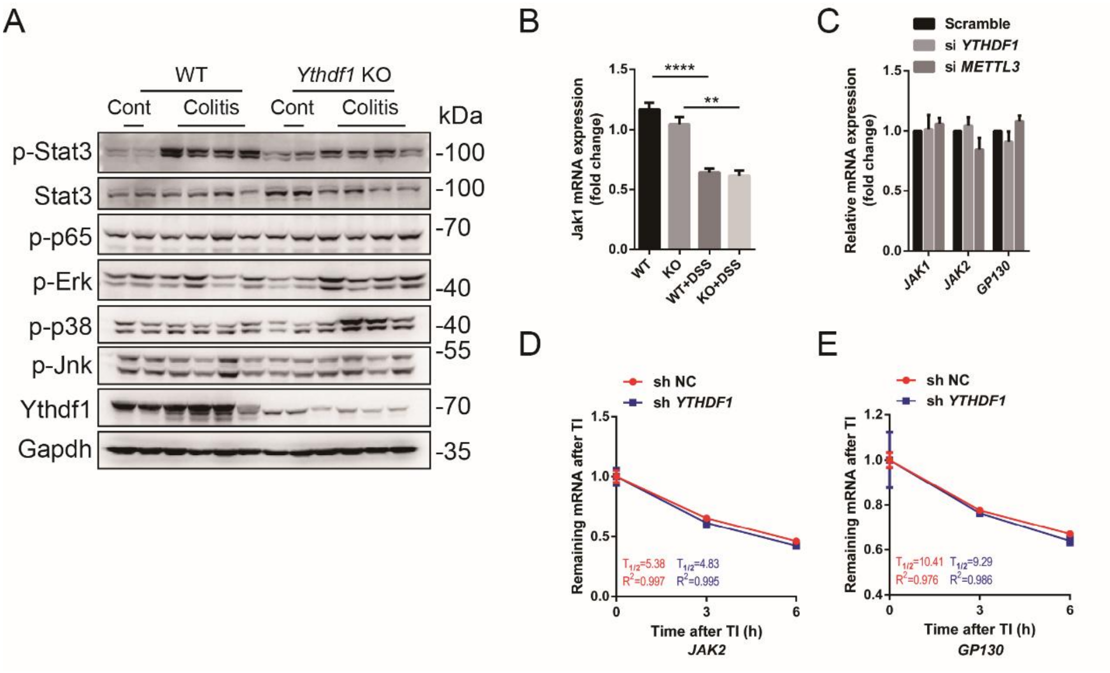

Figure S4. (A) Western blot analysis of Stat3, NF-κB, and MAPK pathways in colon epithelial cells collected from control and DSS-induced colitis mouse models. One lane represents one sample from an individual mouse; (B) qRT-PCR analysis of *Jak1* mRNA level in colon epithelial cells before and after the induction of colitis (*n* = 5 for WT and WT+DSS, *n* = 3 for KO and KO+DSS); (C) qRT-PCR analysis of *JAK1*, *JAK2* and *GP130* expression in HCT116 cells treated with scramble or si*YTHDF1* siRNA; (D-E) RNA lifetime for *JAK2* and *GP130* in shNC and sh*YTHDF1*-transfected HCT116 cells. TI, transcription inhibition; $t_{1/2}$ means RNA lifetime. Data are presented as the means ± s.e.m.

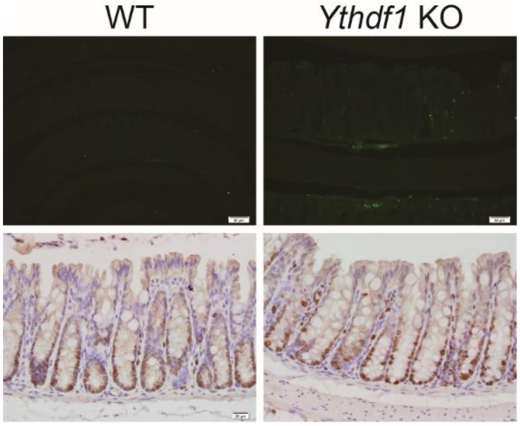

Figure S5. Representative TUNEL-stained apoptotic cells(upper; scale bar, 50 μm) and IHC analysis of Ki-67(lower; scale bar, 20 μm) in WT and *Ythdf1* KO colon sections prior to colitis.

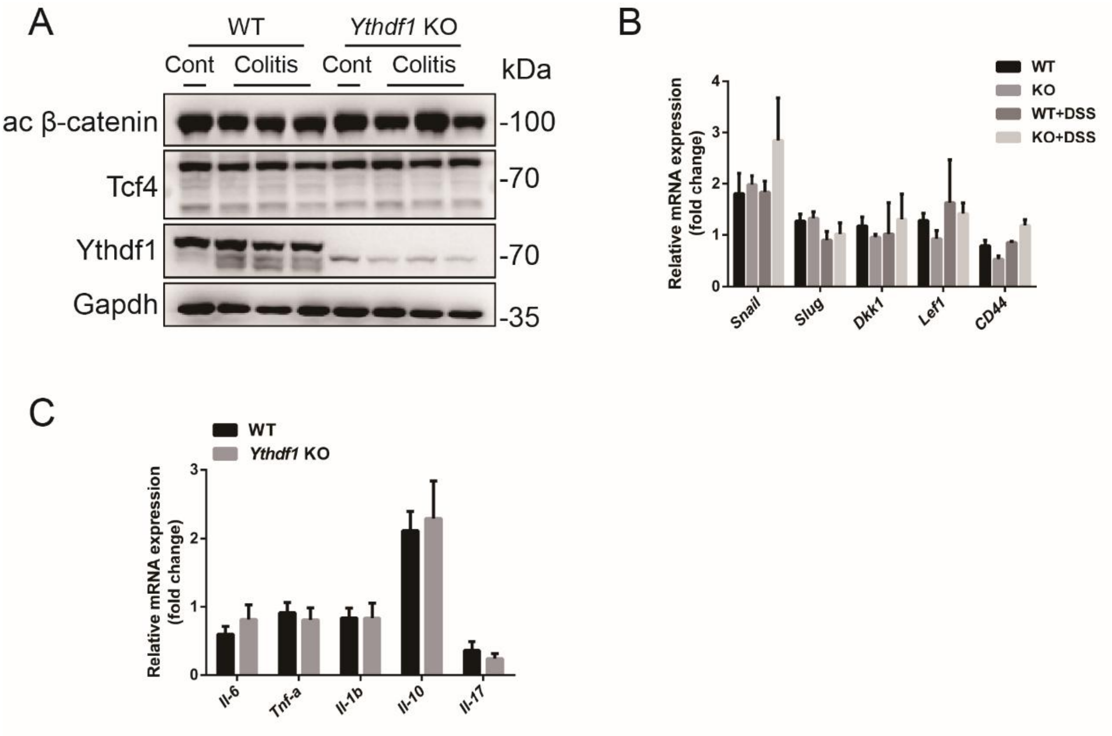

Figure S6. (A) Western blot analysis of activated β-catenin and Tcf4 in control and DSS-induced colitis samples. One lane represents one sample from an individual mouse. The protein lysates are the same as those used in Figure 4A, and the Ythdf1 blot is identical to that shown in Figure 4A; (B) qRT-PCR analysis of the expression of Wnt/β-catenin target genes before and after the induction of colitis in colon epithelial cells (n = 3 for each group); (C) qRT-PCR analysis of genes encoding inflammatory cytokines in WT and *Ythdf1* KO colon tumour tissues (n = 6 for each group). Data are presented as the means ± s.e.m.

Table S1. Primer sequences used for qRT-PCR and siRNA oligos.

Gene name (mouse)	Sense (5’-3’)	Anti-sense (5’-3’)
Il-6	CTGCAAGAGACTTCCATCCAGTT	GGGAAGGCCGTGGTTGTC
Tnf-α	TCAAGGACTCAAATGGGCTTTC	TGCAGAACTCAGGAATGGACAT
Cxcl1	ACTGCACCCAAACCGAAGTC	TGGGGACACCTTTTAGCATCTT
Ccl2	TAAAAACCTGGATCGGAACCAAA	GCATTAGCTTCAGATTTACGGGT
Il-10	CTTACTGACTGGCATGAGGATCA	GCAGCTCTAGGAGCATGTGG
Il-17	GCTCCAGAAGGCCCTCAGA	CTTTCCCTCCGCATTGACA
Bclxl	GCTGCATTGTTCCCGTAGAG	GTTGGATGGCCACCTATCTG
Survivin	CAGGGGAGTGCTTTCTATGC	TACCGAGAACGAGCCTGATT
Ccnd1	TGACTGCCGAGAAGTTGTGC	CTCATCCGCCTCTGGCATT
c-Myc	CCCTATTTCATCTGCGACGAG	GAGAAGGACGTAGCGACCG
Jak1	AGTGCAGTATCTCTCCTCTCTG	GATTCGGTTCGGAGCGTACC
Jak2	GGAATGGCCTGCCTTACAATG	TGGCTCTATCTGCTTCACAGAAT
Bcl2	ATGCCTTTGTGGAACTATATGGC	GGTATGCACCCAGAGTGATGC
Snail	CACACGCTGCCTTGTGTCT	GGTCAGCAAAAGCACGGTT
Slug	CAGCGAACTGGACACACACA	ATAGGGCTGTATGCTCCCGAG
CD44	AGCAAGAAGGGCGAGTATAG	CTGTTCAGTAGGAAGGTAGGTG
Lef1	GCCACCGATGAGATGATCCC	TTGATGTCGGCTAAGTCGCC
Dkk1	CAGTGCCACCTTGAACTCAGT	CCGCCCTCATAGAGAACTCC
Gapdh	TGGCCTTCCGTGTTCCTAC	GAGTTGCTGTTGAAGTCGCA

Gene name (human)	Sense (5’-3’)	Anti-sense (5’-3’)
JAK1	CTTTGCCCTGTATGACGAGAAC	ACCTCATCCGGTAGTGGAGC
JAK2	TCTGGGGAGTATGTTGCAGAA	AGACATGGTTGGGTGGATACC
GP130	CGGACAGCTTGAACAGAATGT	ACCATCCCACTCACACCTCA
GAPDH	ACAACTTTGGTATCGTGGAAGG	GCCATCACGCCACAGTTTC
Gene name (human)	siRNA sequences (5’-3’)	
YTHDF1	GCUCCAUUAAGUACUCCAU	
METTL3	AGGAGCCAGCCAAGAAATCAA	