Supplementary Materials for

WTAP maintains macrophage homeostasis to attenuate HFD-induced obesity by promoting IDH1-mediated α-ketoglutarate production

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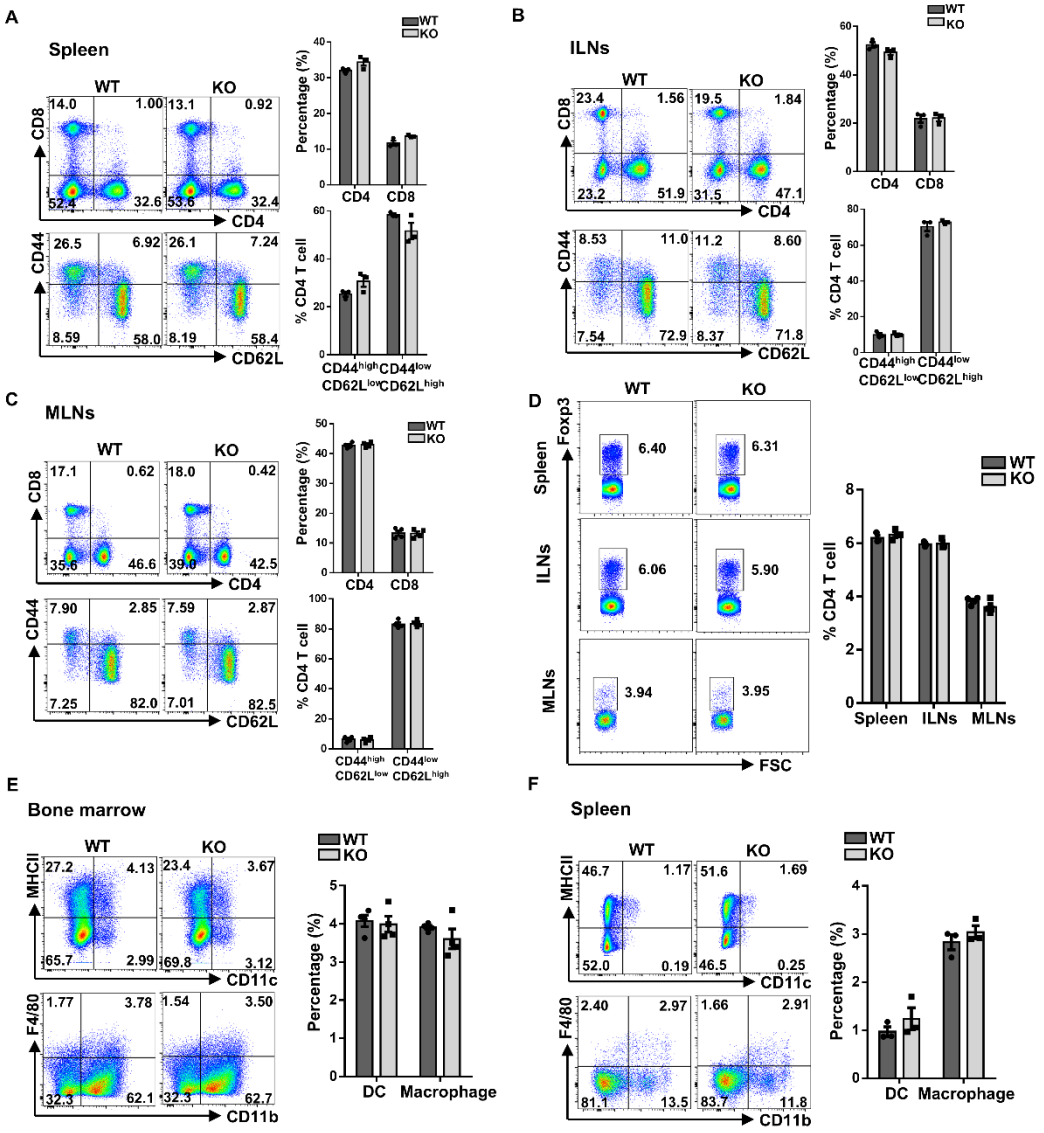

Fig. S1. Depletion of *Wtap* has no effects on either T cell activation or myeloid cell development and maturation. A-C Representative FACS proportion of $CD4^+$, $CD8^+$, $CD4^+$ effector memory ($CD44^{high}CD62L^{low}$) and naive ($CD44^{low}CD62L^{high}$) T cells in the spleen (n = 3), inguinal lymph nodes (ILN) (n = 3) and mesenteric lymph nodes (MLN) (n = 4) from WT and KO mice. **D** Representative FACS proportion of $Foxp3^+$ Tregs gated from $CD4^+$ T cells in spleen, ILN and MLN. **E-F** Representative FACS proportion and percentage of $CD11c^+MHCII^+$ DCs and $F4/80^+CD11b^+$ macrophages from bone marrow (E) and spleen (F). Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test. All results presented no significant differences.

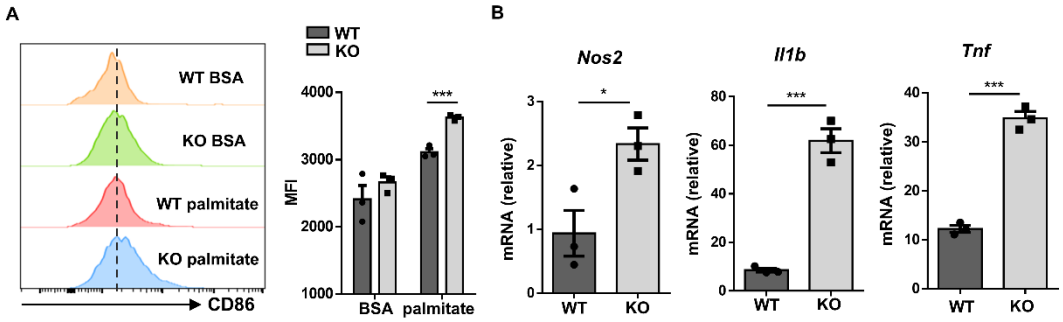

Fig. S2. *Wtap* deletion promotes macrophage activation in response to palmitate stimulation. A-B Expression of CD86 and quantified MFI in F4/80^{+} macrophages under treatment of BSA or palmitate. **C** RT-qPCR analysis of *Nos2, Il1b and Tnf* in WT and KO BMDMs with addition of palmitate. Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test. $^{*}P < 0.05$; $^{***}P < 0.001$.

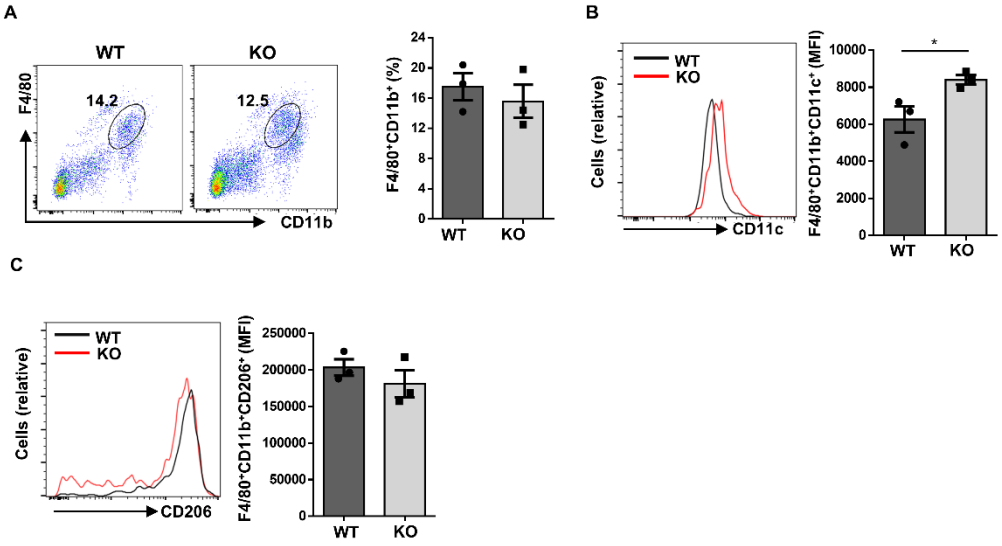

Fig. S3. The effects of *Wtap* deficiency on ATMs under ND. A Representative frequency plots of total macrophages in epWAT from WT and KO mice (n = 3) **B-C** Expression of CD11c and CD206 in F4/80^{+}CD11b^{+} macrophages from WT and KO mice. Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test. $^{*}P < 0.05$.

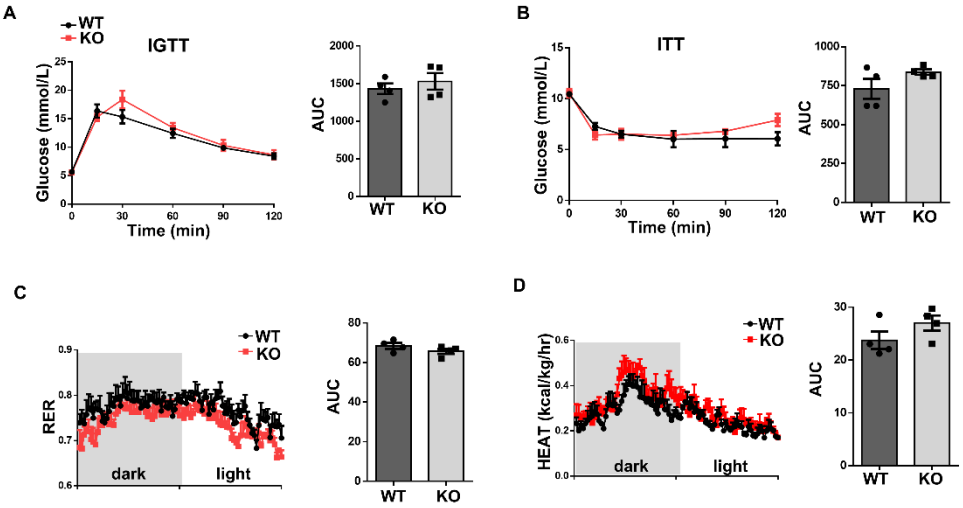

Fig. S4. KO mice present comparable glucose tolerance, insulin sensitivity, RER and heat production with WT mice fed with ND. A-B The results of intraperitoneal glucose tolerance test (IGTT) (I) and insulin resistance test (ITT), and the analysis of areas under curves. **C-D** The monitoring of real-time RER and heat production of ND mice for 24 h, and the areas under the curves were quantitatively analyzed. Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test. All results presented no significant differences.

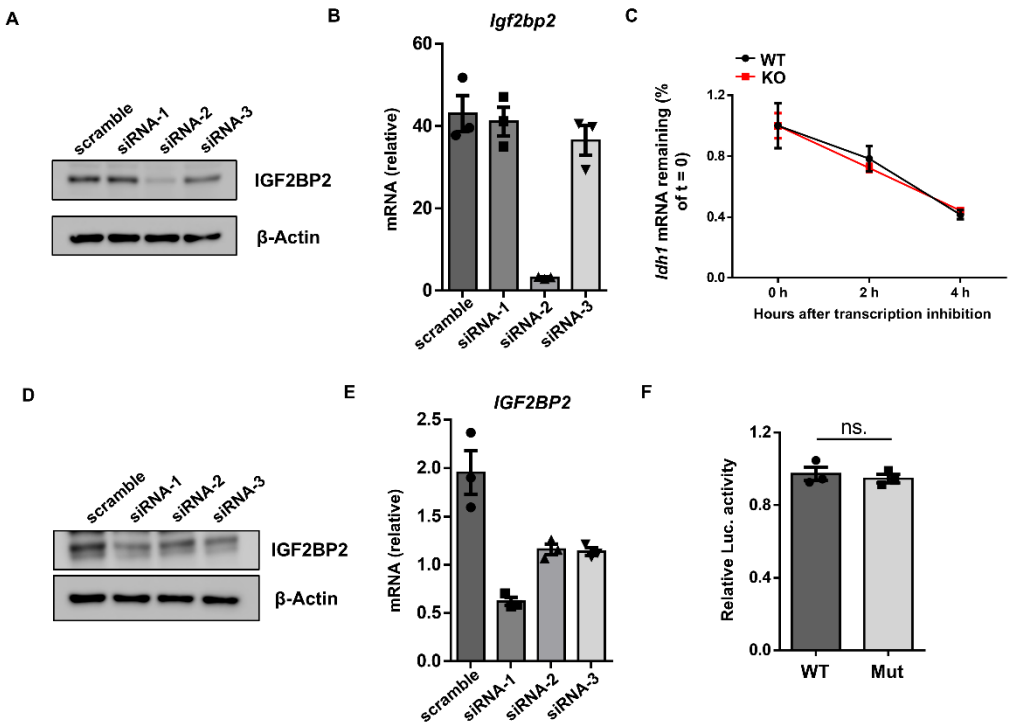

Fig. S5. IGF2BP2 modulates *Idh1* mRNA stability and translation. **A-B** Knockdown efficiency of IGF2BP2-specific siRNAs in BMDMs at the protein (A) and mRNA (B) levels **C** RT-qPCR analysis of *Idh1* mRNA stability in BMDMs treated with 5 μg/mL Actinomycin D for 0, 2, and 4 hours following IGF2BP2 knockdown. **D-E** Knockdown efficiency of IGF2BP2-specific siRNAs in HEK293T cells at the protein (D) and mRNA (E) levels **F** Relative luciferase activities of HEK293T cells co-transfected with WT or mutant plasmid and IGF2BP2 siRNA. Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test. ns. no significance.

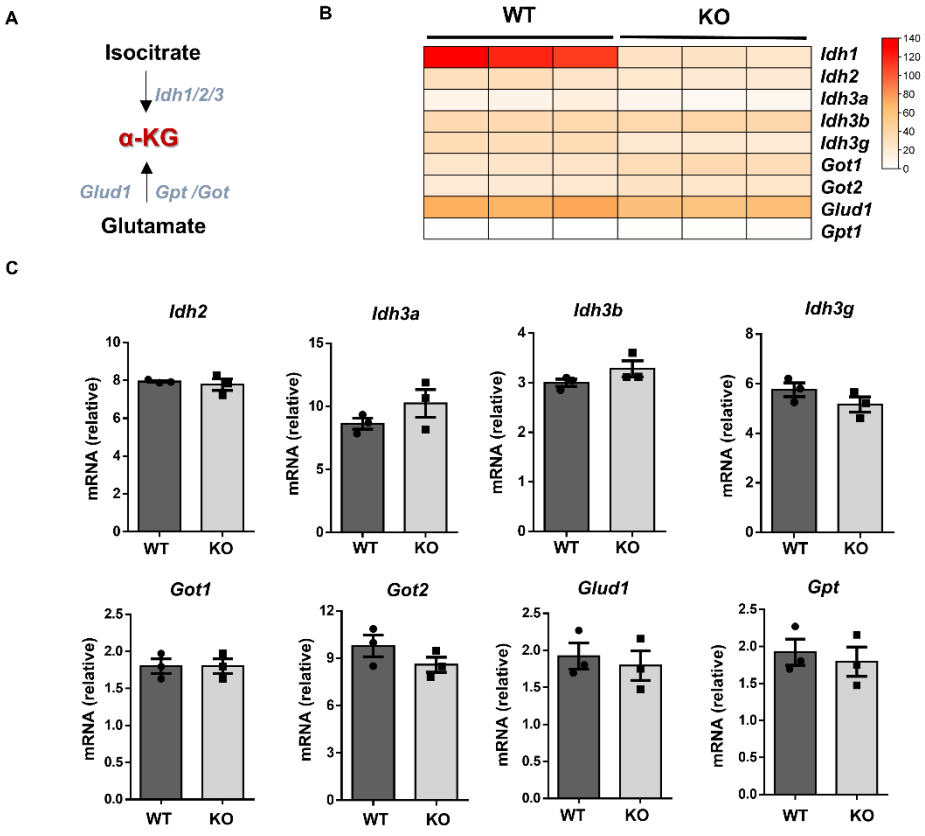

Fig. S6. Expression profile of the genes critical for α-KG production. A Alpha-KG productions are mainly from isocitrate and glutamate. **B** Heatmap of the vital enzyme genes related to α-KG production. **C** RT-qPCR analysis of abovementioned genes. Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test. All results presented no significant differences.

Table S1. Clinical characteristics of subjects

	Non-obese	Obese
Number	12	46
Male/ female (n)	2/10	15/31
Age (years)	32.17 ± 2.007	33.65 ± 1.173
BMI (kg/m^2)	27.75 ± 0.9062	38.25 ± 0.9697
Fasting Bood Glucose (mmol/L)	4.576 ± 0.1298	6.290 ± 0.3673
Insulin (uU/ml)	16.29 ± 2.795	35.50 ± 4.017
c-peptide (nmol/L)	1.810 ± 0.3674	1.533 ± 0.1034
HbA1c (%)	5.790 ± 0.4661	5.950 ± 0.1423
Triglyceride (mmol/l)	1.184 ± 0.1512	1.588 ± 0.1078
Total cholesterol (mmol/l)	4.781 ± 0.2800	5.125 ± 0.1727
LDL (mmol/L)	2.686 ± 0.1375	3.256 ± 0.1488
HDL (mmol/L)	1.198 ± 0.08451	1.193 ± 0.04704

Data were exhibited as mean ± SEM and analyzed by unpaired Student's *t* test.

Table S2. Primer sequences for RT-qPCR

Gene	Forward (5'-3')	Reverse (5'-3')
Nos2	GTTCTCAGCCCAACAATACAAGA	GTGGACGGGTCGATGTCAC
Il1b	TACGGACCCCAAAAGATGA	TGCTGCTGCGAGATTTGAAG
Tnf	ACTGAACTTCGGGGTGATCG	GGCTACAGGCTTGTCACTCG
Arg1	CTCCAAGCCAAAGTCCTTAGAG	AGGAGCTGTCATTAGGGACATC
Ym1	CAGGTCTGGCAATTCTTCTGAA	GTCTTGCTCATGTGTGTAAGTGA
Retnla	CTGGGTTCTCCACCTCTTCA	TGCTGGGATGACTGCTACTG
β-Actin	AGAGGGAAATCGTGCGTGAC	CAATAGTGATGACCTGGCCGT
Ucp1	AGGCTTCCAGTACCATTAGGT	CTGAGTGAGGCAAAGCTGATTT
Idh1	ATGCAAGGAGATGAAATGACACG	GCATCACGATTCTCTATGCCTAA
Cox5a	GGAAGACCCTAATCTAGTCCCG	GTTGGGGCATCGCTGACTC
Cox7a	GCTCTGGTCCGGTCTTTTAGC	GTACTGGGAGGTCATTGTCGG
Cox8b	TGTGGGGATCTCAGCCATAGT	AGTGGGCTAAGACCCATCCTG
Wtap	TAGACCCAGCGATCAACTTGT	CCTGTTTGGCTATCAGGCGTA
Igf2bp1	CGGCAACCTCAACGAGAGT	CGGCAACCTCAACGAGAGT

Igf2bp2	GTCCTACTCAAGTCCGGCTAC	CATATTCAGCCAACAGCCCAT
Igf2bp3	CCTGGTGAAGACGGGCTAC	TCAACTTCCATCGGTTTCCCA
***WTAP* (human)**	CTTCCCAAGAAGGTTCGATTGA	TCAGACTCTCTTAGGCCAGTTAC
***IDH1* (human)**	TGTGGTAGAGATGCAAGGAGA	TTGGTGACTTGGTCGTTGGTG
***IGF2BP2* (human)**	AGTGGAATTGCATGGGAAAATCA	CAACGGCGGTTTCTGTGTC
Idh2	GGAGAAGCCGGTAGTGGAGAT	GGTCTGGTCACGGTTTGGAA
Idh3a	TGGGTGTCCAAGGTCTCTC	CTCCCACTGAATAGGTGCTTTG
Idh3b	TGGAGAGGTCTCGGAACATCT	AGCCTTGAACACTTCCTTGAC
Idh3g	GGTGCTGCAAAGGCAATGC	TATGCCGCCCACCATACTTAG
Got1	GCGCCTCCATCAGTCTTTG	ATTCATCTGTGCGGTACGCTC
Got2	GGACCTCCAGATCCCATCCT	GGTTTTCCGTTATCATCCCGGTA
Glud1	CCCAACTTCTTCAAGATGGTGG	GGTTTTCCGTTATCATCCCGGTA
Gpt	TCCAGGCTTCAAGGAATGGAC	CAAGGCACGTTGCACGATG