Figure S1

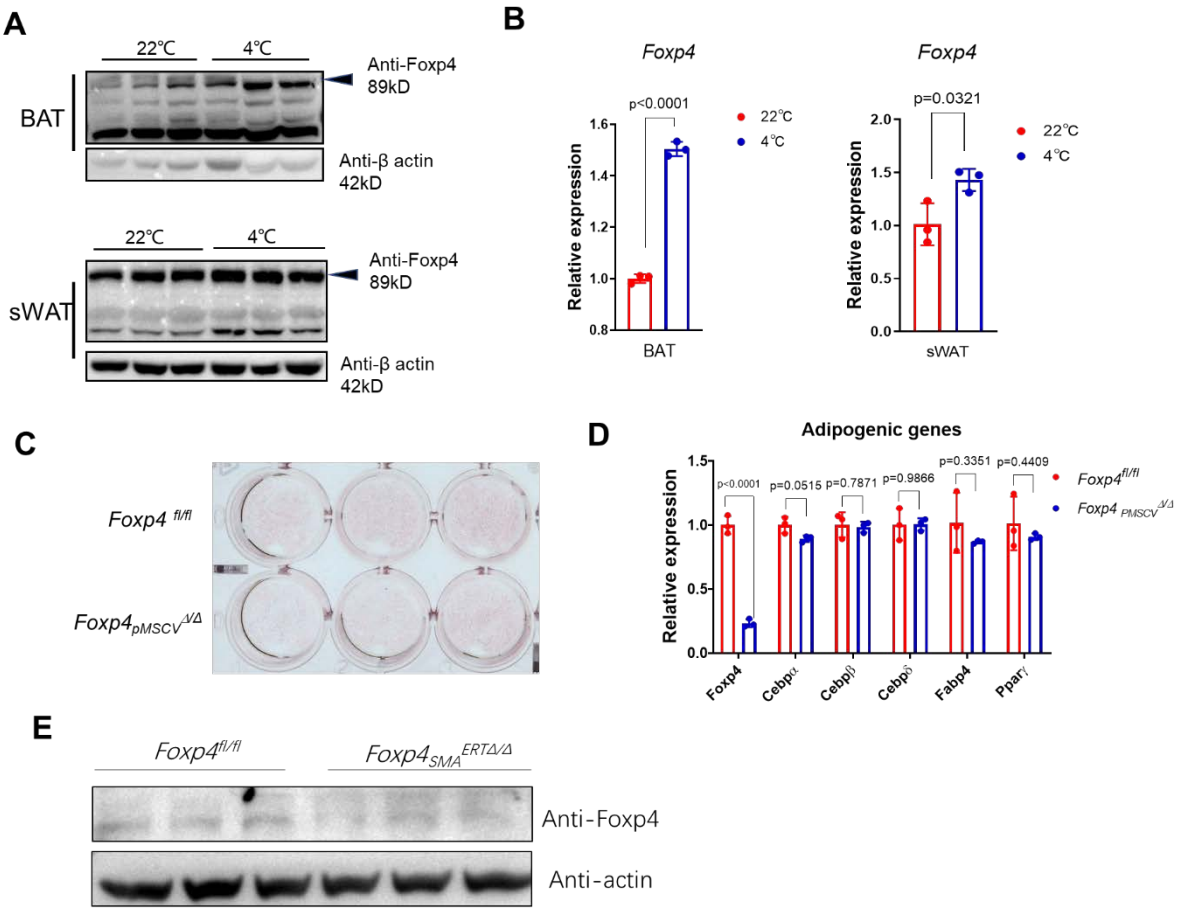

Fig. S1. Expression of *Foxp4* in adipose tissues at cold exposure, by *pMSCV-Cre* or *SMA-CreER*.

(A) Western blot for FOXP4 protein in BAT and sWAT of mice under room temperature (22℃) and one-week cold exposure (4℃). Three independent experiments were replicated.

(B) mRNA levels of *Foxp4* expression in BAT and sWAT from mice of (A). n, 3.

(C) Oil Red O staining for 8-day white adipocyte differentiation from *pMSCV-Cre*-transfected SVF of sWAT from $Foxp4^{fl/fl}$ mice.

(D) mRNA levels of adipogeniesis markers in cells of (C). n, 3.

(E) FOXP4 protein expression in SVF cells from $Foxp4_{Sma}^{ErtΔ/Δ}$ mice. Three independent experiments were replicated.

Figure S2

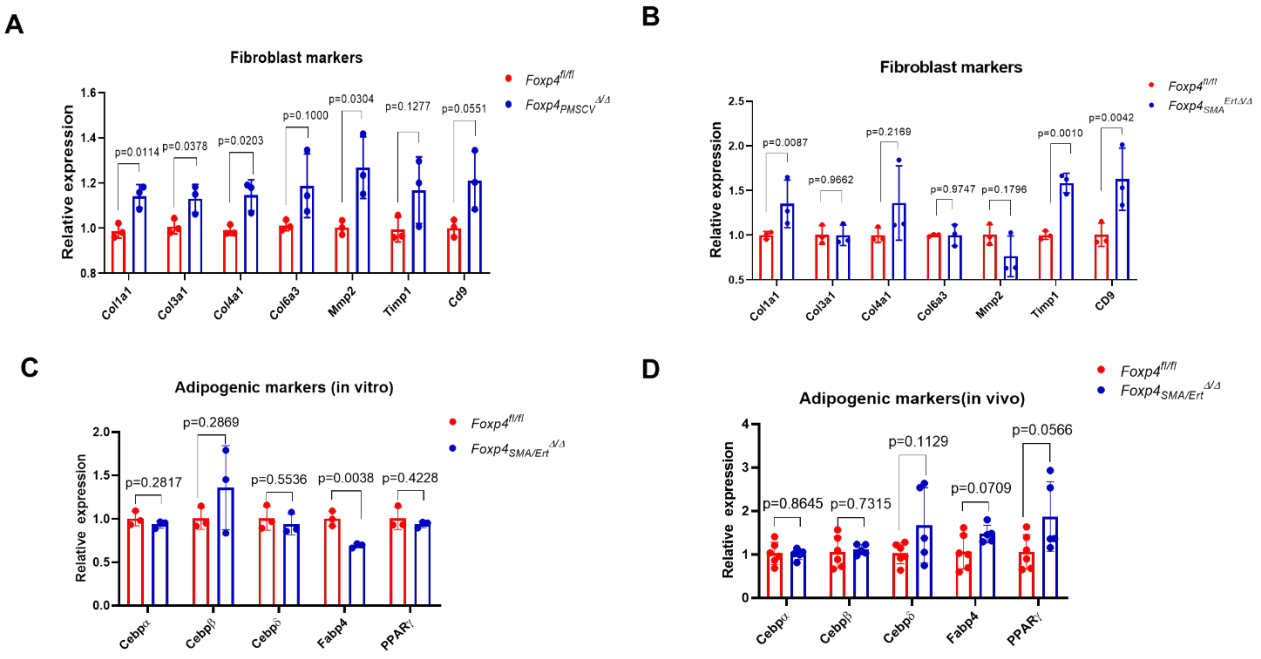

Fig. S2. *In vitro* beige adipocyte differentiation with *Foxp4* deficiency by *pMSCV-Cre*.

(A) mRNA levels of fibroblast cell marker genes in beige differentiation from SVF with *Foxp4* inactivation induced by *pMSCV-Cre*.

(B) mRNA levels of fibroblast cell marker genes in beige differentiation from SVF of $Foxp4_{Sma}^{Ert\Delta/\Delta}$ mice with *Foxp4* inactivation induced by 4-OH tamoxifen in cultures.

(C) mRNA levels of general adipogenic markers in beige differentiation from SVF of $Foxp4_{Sma}^{Ert\Delta/\Delta}$ mice with *Foxp4* inactivation induced by tamoxifen injection.

(D) mRNA levels of general adipogenic markers in beige differentiation from SVF of $Foxp4_{Sma}^{Ert\Delta/\Delta}$ mice with *Foxp4* inactivation induced by 4-OH tamoxifen in cultures.

Figure S3

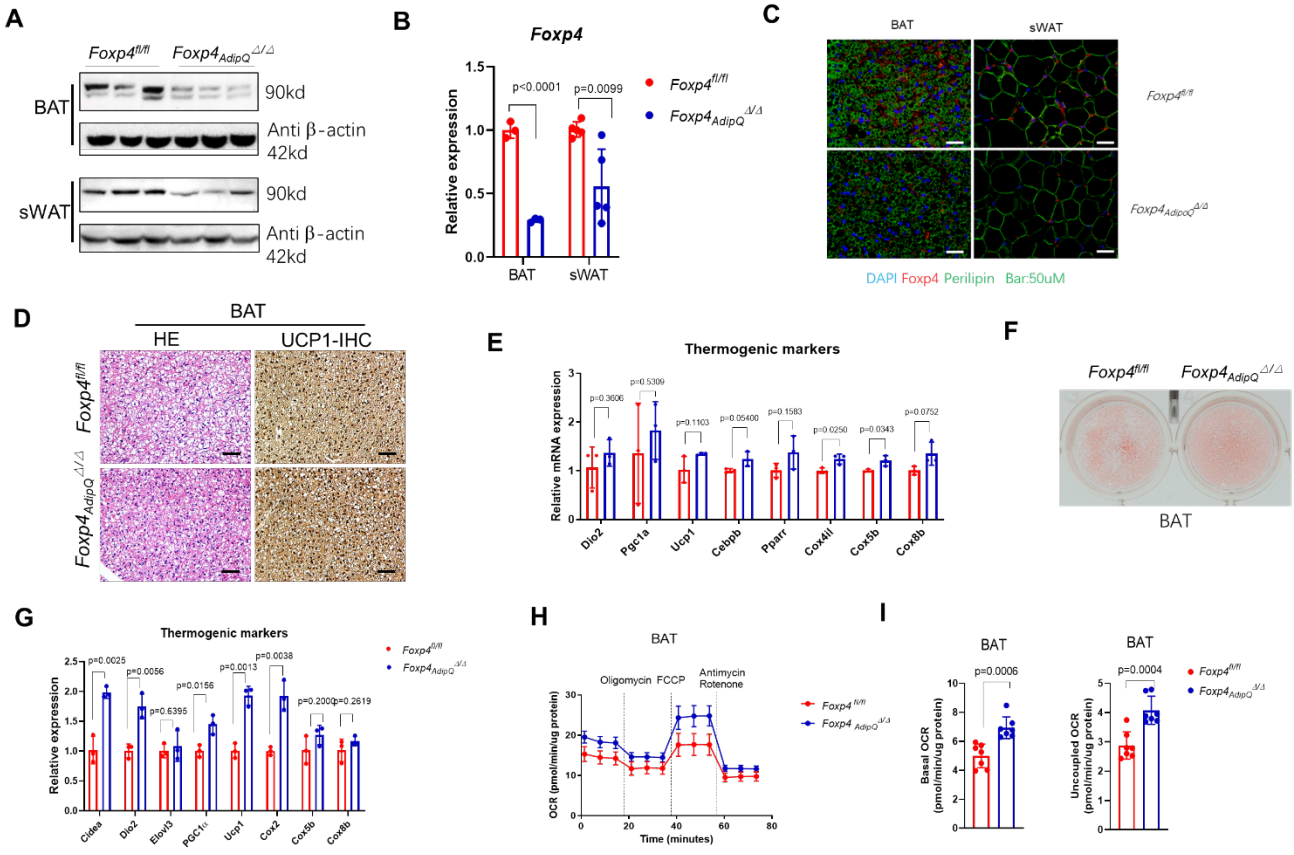

Fig. S3. Thermogenesis in BAT of *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice.

(A) Western blot for FOXP4 protein in BAT and sWAT of *Foxp4*$^{fl/fl}$ and *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice at age of 2 months old. Three independent experiments were replicated.

(B) Assessment of *Foxp4* mRNA expression in BAT and sWAT from mice by qPCR. n, 3~5.

(C) Immunofluorescence examination for FOXP4 on sections from BAT and sWAT of 2-month-old *Foxp4*$^{fl/fl}$ and *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice.

(D) H&E and immunohistochemical staining (IHC) for UCP1 on BAT sections from *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice. Three independent experiments were replicated.

(E) mRNA levels of thermogenic and mitochondrial markers in BAT.

(F) Oil Red O staining 8 day post brown adipocyte differentiation from BAT-SVF of *Foxp4*$^{fl/fl}$ and *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice at age of 8 weeks. Three independent experiments were replicated.

(G) mRNA levels of thermogenic markers for brown adipocytes in (F). n, 3.

(H) Oxygen consumption rate (OCR) was measured for brown adipocytes from (F).

Uncoupled respiration was recorded after oligomycin inhibition of ATP synthesis, and maximal respiration following stimulation with carbonyl cyanide 4-(trifluoromethoxy) phenylhydrazone (FCCP). n,7.

(I) Quantitative analysis of basal and uncoupled OCR in (H). n, 7.

Figure S4

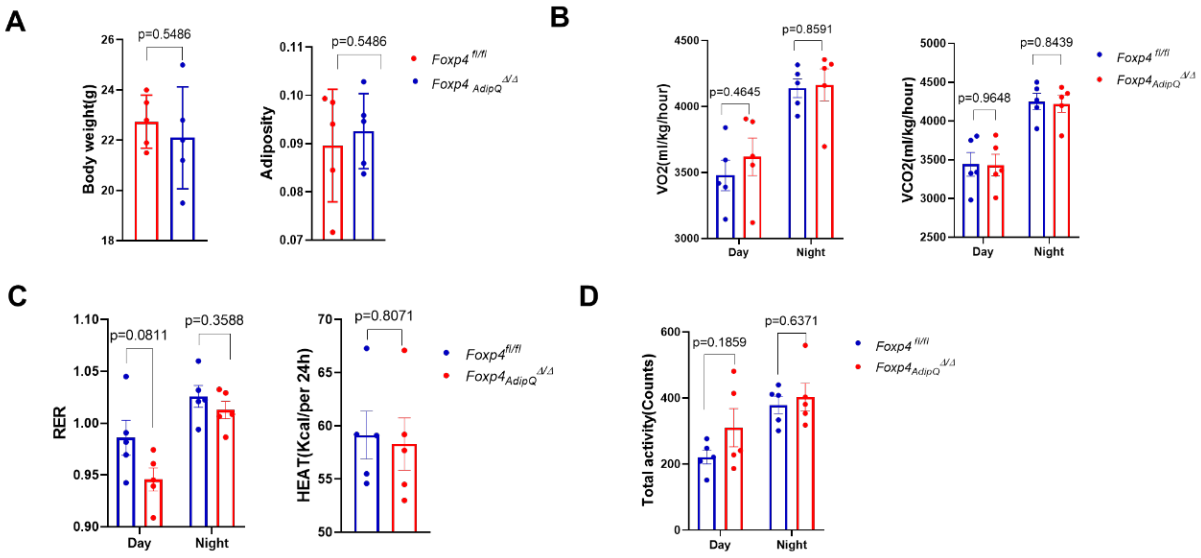

Fig. S4. Metabolic analysis for *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice.

(A) Body weight and relative adiposity of *Foxp4*$^{fl/fl}$ and *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice during day and night in metabolic cages at age of 3 months old. n, 5.

(B) Quantification of O_2 and CO_2 consumption of mice under room temperature. n, 5.

(C) RER and heat production of mice. n, 5.

(D) Total activity of mice. n, 5.

Figure S5

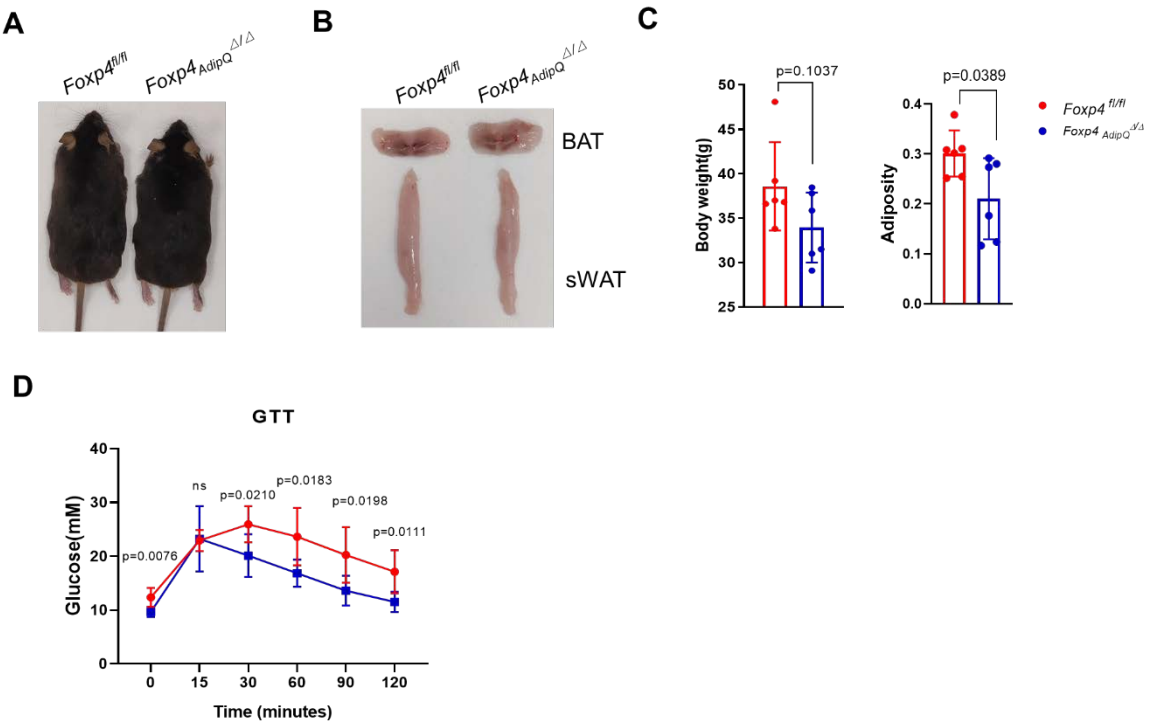

Fig. S5. *Foxp4* deficiency protects mice from HFD-fed obesity.

(A) Dorsal view of representative *Foxp4*$^{fl/fl}$ and *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice of after 8-week feeding with HFD at age of 2 months.

(B) Representative fat depot of BAT and sWAT from mice of (A).

(C) Body weight and relative adiposity of HFD-fed mice of (A). n, 6.

(D) GTT of HFD-fed mice. n, 6.

Figure S6

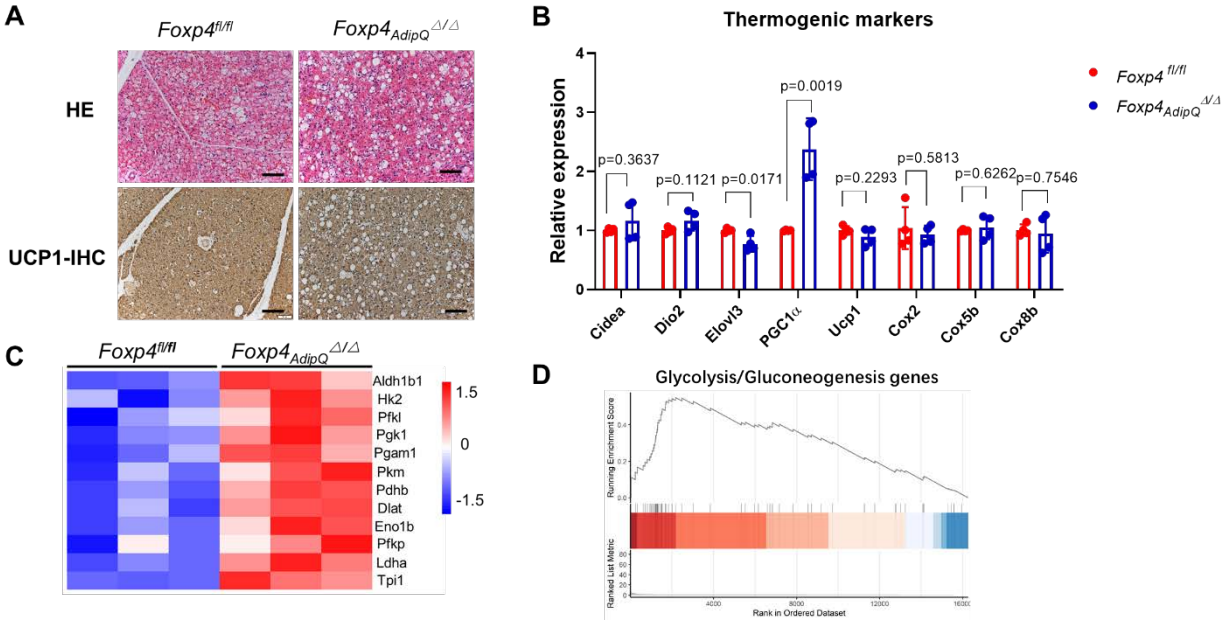

Fig. S6. BAT thermogenesis in *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice upon cold exposure.

(A) H&E and immunohistochemistry (IHC) staining for UCP1 protein on BAT sections from *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice after one-week cold exposure at 4℃. Three independent experiments were replicated.

(B) Thermogenesis in BAT of (A) assessed by qPCR with selective markers (*Cidea*, *Dio2*, *Elovl3*, *PGC1α*, *Ucp1*, *Cox2*, *Cox5b*, *Cox8b*). n, 3.

(C) Heatmap depicting the mRNA levels of glycolytic genes in beige adipocytes from sWAT in *Foxp4*$_{AdipQ}^{\Delta/\Delta}$ mice after one-week cold exposure at 4℃.

(D) Gene set enrichment analysis of glycolytic marker gene expressions in sWAT of (C).

Figure S7

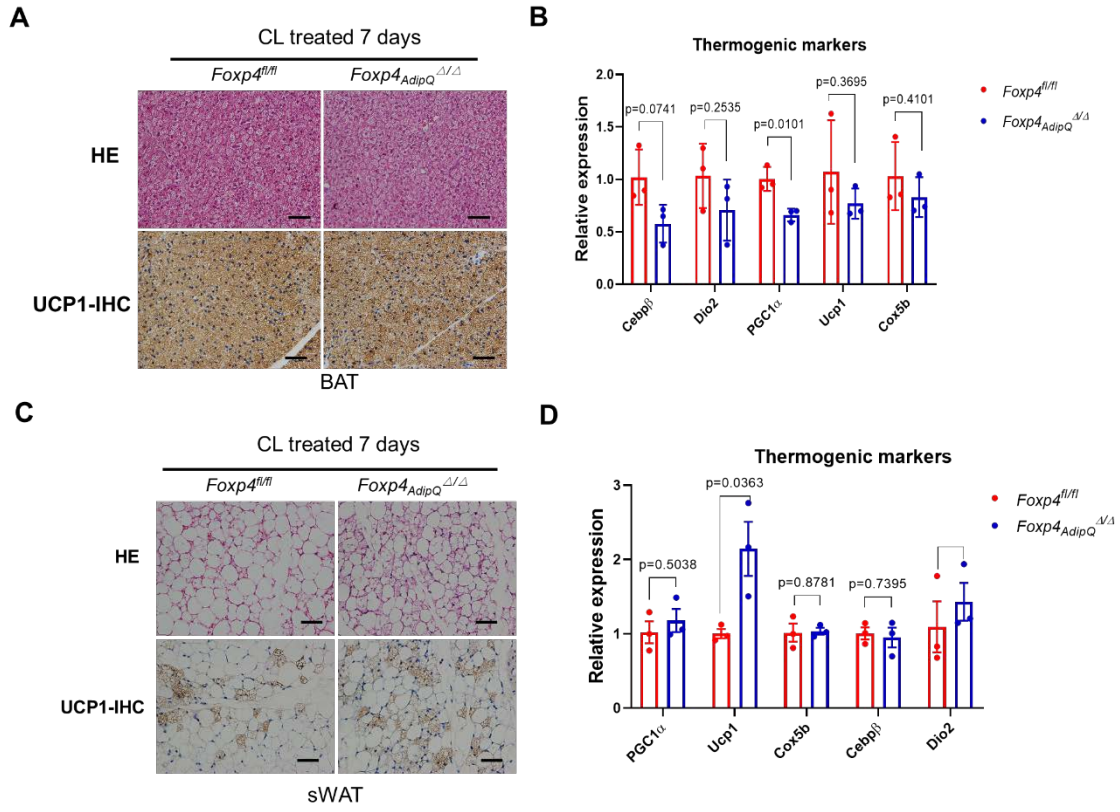

Fig. S7. Thermogenic activation in CL-316,243-stimulated *Foxp4*$^{AdipQ\Delta/\Delta}$ mice.

(A, C) H&E and IHC staining for UCP1 on BAT (A) and sWAT (C) sections from mice stimulated with CL-316,243 for 7 days. Three independent experiments were replicated.

(B, D) qPCR analysis for thermogenic gene expressions in BAT (B) and sWAT (D) from CL-316,243-stimulated mice. n, 3.

Figure S8

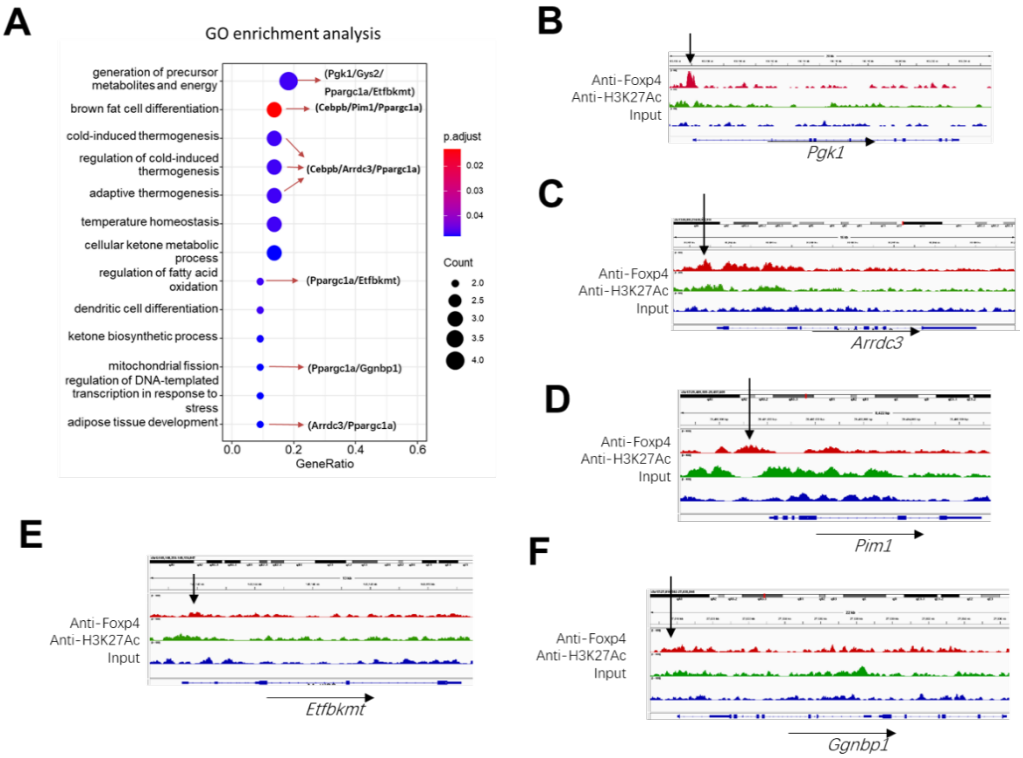

Fig. S8. Crossover examinations of RNA-seq and ChIP-seq data to identify putative FOXP4 targeting genes.

(A) GO enrichment analysis for several putative FOXP4-targeting genes.

(B-F) ChIP-seq profile showed the FOXP4 binding sites (black arrows) within *Pgk1*, *Arrdc3*, *Pim1*, *Etfbkmt*, *Ggnbp1* gene regions.

Table S1. Primers for qPCR and genotyping

primer for qPCR		
Foxp4	F	GTGTCTGTGGCCATGATGTC
	R	TCTTTGGGCTGCTGTTTTCC
Adrb3	F	GGCCCTCTCTAGTTCCCAG
	R	TAGCCATCAAACCTGTTGAGC
Ucp1	F	ACTGCCACACCTCCAGTCATT
	R	CTTTGCCTCACTCAGGATTGG
PGC1α	F	AGCCGTGACCACTGACAACGAG
	R	GCTGCATGGTTCTGAGTGCTAAG
Cebpα	F	TGGACAAGAACAGCAACGAG
	R	TCACTGGTCAACTCCAGCAC
Cebpβ	F	ACGACTTCCTCTCCGACCTCT
	R	CGAGGCTCACGTAACCGTAGT
Dio2	F	CAGTGTGGTGCACGTCTCCAATC
	R	TGAACCAAAGTTGACCACCAG
Prdm16	F	CCACCAGCGAGGACTTCAC
	R	GGAGGACTCTCGTAGCTCGAA
Cox2	F	GCAAGCATAAGACTGGACCAAA
	R	TTGTTGGCATCTGTGTAAGAGAATC
Cox4il	F	ACCAAGCGAATGCTGGACAT
	R	GGCGGAGAAGCCCTGAA
β-actin	F	AGAGGGAAATCGTGCGTGACA
	R	CACTGTGTTGGCATAGAGGTC
Elovl3	F	TCCGCGTTCTCATGTAGGTCT
	R	GGACCTGATGCAACCCTATGA
Cox5b	F	GCTGCATCTGTGAAGAGGACAAC
	R	CAGCTTGTAATGGGTTCCACAGT
Cox8b	F	TGTGGGGATCTCAGCCATAGT
	R	AGTGGGCTAAGACCCATCCTG
PPARα	F	GCGTACGGCAATGGCTTTAT
	R	GAACGGCTTCCTCAGGTTCTT
PPARγ	F	GGAAAGACAACGGACAAATCAC
	R	TACGGATCGAAACTGGCAC
Cox7a1	F	CAGCGTCATGGTCAGTCTGT
	R	AGAAAACCGTGTGGCAGAGA
Cidea	F	TGCTCTTCTGTATCGCCCAGT
	R	GCCGTGTTAAGGAATCTGCTG

Rgs2	F	GAGAAAATGAAGCGGACACTCT
	R	GCAGCCAGCCCATATTTACTG
CD137	F	CGTGCAGAACTCCTGTGATAAC
	R	GTCCACCTATGCTGGAGAAGG
Them26	F	ACCCTGTCATCCCACAGAG
	R	TGTTTGGTGGAGTCCTAAGGTC
Tbx1	F	GGCAGGCAGACGAATGTTC
	R	TTGTCATCTACGGGCACAAAG
Cd40	F	TTGTTGACAGCGGTCCATCTA
	R	CCATCGTGGAGGTACTGTTTG
Ear2	F	CCTGTAACCCCAGAACTCCA
	R	CAGATGAGCAAAGGTGCAAA
Klhl13	F	AGAATTGGTTGCTGCAATACTCC
	R	AAGGCACAGTTTCAAGTGCTG
Slc27a1	F	CTGGGACTTCCGTGGACCT
	R	TCTTGCAGACGATACGCAGAA
primer for genotyping		
Foxp4-Floxed	F	TGGAGGGACTGGGATTAGAAC
	R	ACGGGAGGCTGAACAACAC
Cre	F	TTTCCCGCAGAACCTGAAGA
	R	GGTGCTAACCAGCGTTTTCGT
***Pdgfrα*-Luc**		
Pdgfrα-promoter	F	CAGAGGGCAGGCATTTGGTAGT
	R	GCTTACTGGGACGAACACCA