

SUPPLEMENTARY DATA

**Supplementary Table 1.** *Western blot analysis for diet-induced weight gain and loss in rats experiment.*

About 250 mg of rat rWAT were homogenized in three volumes of RIPA buffer (50mM Tris-HCl, pH 7.4, 150mM NaCl, 0.25% deoxycholic acid, 1% NP-40, 1 mM EDTA), supplemented with protease and phosphatase inhibitors. Lysates were cleared by centrifugation at 14000g for 20 min. The protein concentration was determined by BCA Protein Assay Kit (Thermo Scientific). Forty micrograms of protein from three-four samples per group were mixed with 4x sample buffer (8% SDS, 40% glycerol, 250mM TrisHCl pH=6.8, 0.008% bromophenol blue and 20% beta-mercaptoethanol) and boiled for 5 min at 100°C. Electrophoresis was performed on 1.5mm thick 4% stacking/ 7% resolving polyacrylamide gel for 2.5h at 100V in Mini-Protean 3 cell (Bio-Rad). The proteins were transferred to a 0.45µm nitrocellulose membrane by semi-dry transfer at constant 3mA/cm<sup>2</sup> for 80min. The membrane was blocked for 1h with Odyssey blocking buffer (Li-cor) at room temperature and then incubated with rabbit Rb antibody (C-15) (Santa Cruz, SC 50) 1:500 overnight at 4°C, washed and incubated with IRDye 800CW Goat anti-Rabbit secondary antibody (Li-cor) 1:10000 for 40min. After scanning by Odyssey scanner, the membrane was stripped for 6min in NewBlot™ Nitrocellulose 1X Stripping Buffer (Li-cor), washed and incubated with rabbit Phospho-Rb (Ser780) Antibody (Cell Signalling, 9307) 1:1000 for 1h at RT, washed and again incubated with the secondary antibody and scanned. Densitometry analysis of the bands was performed using the Odyssey V3.0 software.

<i>Gene</i>	<b>Assay number or primer sequence</b>
<b>Human (Homo sapiens)</b>	
Cyclophilin A, PPIA	4333763
Retinoblastoma, RB1	Hs01078066_m1
Peroxisome proliferator-activated receptor gamma, PPAR $\gamma$	Hs00234592_m1
Solute carrier family member 4, SLC2A4 or GLUT4	Hs00168966_m1
Insulin receptor substrate 1, IRS1	Hs00178563_m1
Fatty acid synthase, FASN	Hs00188012_m1
Leptin, LEP	Hs00174877_m1
Acetyl-CoA carboxylase, ACC	Hs00167385_m1
<b>Mouse (Mus musculus)</b>	
Peroxisome proliferator-activated receptor gamma, <i>Ppar<math>\gamma</math></i>	Mm01184322_m1
<i>Adiponectin, Adipoq</i>	Mm00456425_m1
Fatty acid synthase, <i>Fasn</i>	Mm00662319_m1
<i>Prdm16</i>	Mm00712556_m1
<i>Ucp1</i>	Mm01244861_m1
<b>Rat (Rattus norvegicus)</b>	
Eif4e	Forward 5'-GCAGCAGAGACGAAAGTGACC-3' // Reverse 5'-TGACAACAGCTCCACACACG-3'
Eif2s1	Forward 5'-ATTCCAGAGGACTGCCTGGGTC-3' // Reverse 5'-ACAGCTTGTGGGGTCAAACGC-3'
Retinoblastoma, Rb1	Forward 5'-TGCCTCCACCAGGCCTCCTAC-3' // Reverse 5'-ACCTCCAGGAATCCGCAAGGGT-3'

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**Supplementary Figure 1.** Effects of transient Rb1 (41% reduction for 36 h) knockdown in fully differentiated 3T3-L1 adipocytes using siPORT method on the expression of the denoted genes. Treatments: SiC, control, non-targeted siRNA; siRb1, Rb1-targeted siRNA.

