P-0395

Role of SENP2 in the regulation of browning of white adipose tissue

Ji Seon Lee[1], Weiping Han[5], Young-Bum Kim[4], Sung Soo Chung[1], Kyong Soo Park [2,3]*

1. Biomedical Research Institute, Seoul National University Hospital, Seoul, Korea 2. Department of Molecular Medicine and Biopharmaceutical Sciences, Graduate School of Convergence Science and Technology, and College of Medicine or College of Pharmacy, Seoul National University, Seoul, Korea 3. Department of Internal Medicine, Seoul National University College of Medicine, Seoul, Korea 4. Division of Endocrinology, Diabetes and Metabolism, Beth Israel Deaconess Medical Center and Harvard Medical School, Boston, Massachusetts, USA 5. Laboratory of Metabolic Medicine, Singapore Bioimaging Consortium, Agency for Science, Technology and Research (A*STAR), Singapore, Singapore

Abstract SUMO-specific proteases 2 (SENP2) regulates lipid metabolism and also plays a critical role in adipogenesis of 3T3L1. Browning of white adipose tissue has protective effects against diet-induced obesity and insulin resistance, therefore molecular mechanism related to browning process is an important question. To investigate the role of SENP2 in adipocytes in detail, we generated adipocyte-specific SENP2 knockout mice (SENP2-aKO) using adiponectin-cre mice. The weights of adipose tissue of SENP2-aKO mice were lower than those of WT mice. SC-WAT of SENP2-aKO mice had numerous multilocular adipocytes, and increased thermogenic response to cold exposure. The mRNA levels of brown adipocyte-specific genes, such as Ucp1 and Cidea, were much higher in the SC-WAT of SENP2-aKO mice compared to WT mice, indicating browning of SC-WAT in SENP2-aKO mice. Consistently, brown adipocyte-specific genes were significantly increased in the SENP2-aKO SVF-derived adipocytes, while expression of Hoxc10, a key negative regulator of browning, was suppressed. When Hoxc10 was overexpressed, SENP2 KO-induced brown adipocyte-specific gene expression was disappeared. Using siRNA-mediated knock-down, transient transfection and reporter assays, we demonstrated that sumoylated form of C/EBPβ efficiently suppresses transcription of Hoxc10, and SENP2 maintains high level of Hoxc10 during differentiation of white adipocytes through desumoylation of C/EBPβ.

Aim of study The aim of this study is to investigate function of SENP2 in adipocytes by using SENP2-aKO mice.

Materials and Methods SENP2-aKO were generated using adiponectin-cre mice. Mice were fed with a high fat diet (HFD) for 12 weeks and GTTs were performed to examine the effects of SENP2 KO on HFD-induced obesity and insulin resistance. Also, metabolic activities of the mice were measured by using CLAMS. For in vitro study, stromal vascular fractions (SVF) were isolated from subcutaneous fat (SC-WAT) of WT mice and SENP2-aKO mice, followed by induction of adipocyte differentiation.

Results

Fig 1. SENP2-aKO mice are protected from HFD-induced obesity and insulin resistance and increase energy expenditure (A) Body weights (B) Glucose tolerance test (C) O_2 consumption rate (VO_2)

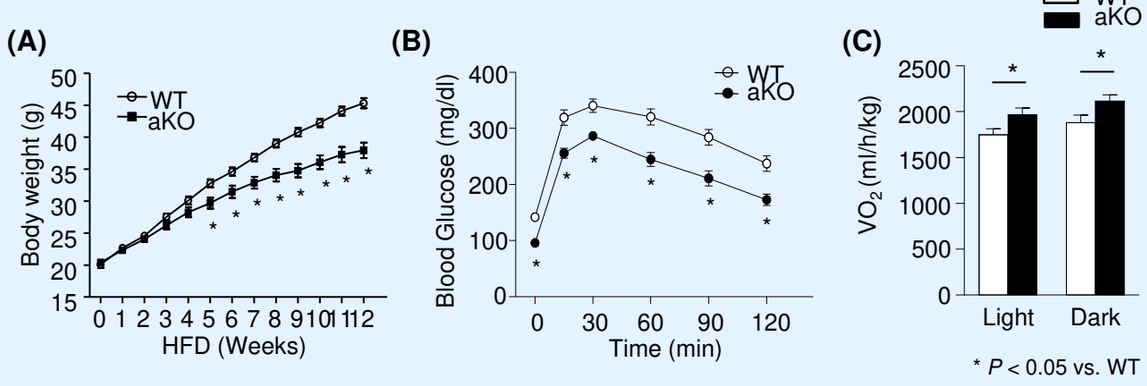


Fig 2. Enhanced thermogenesis and induced browning of iWAT in SENP2-aKO mice (A) Rectal temperature after exposure to 4 °C. (B) IHC staining of UCP1 in iWAT (C) Results of qPCR using mRNAs from iWAT

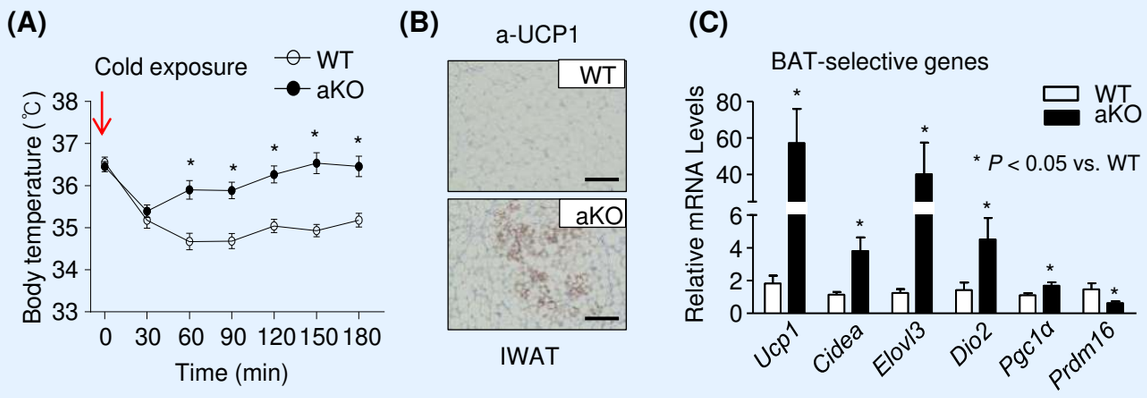


Fig 3. Reduction of Hoxc10, a key negative regulator of browning, is necessary for the browning of scWAT in SENP2-aKO (A) Gene expression in SVF-derived adipocytes. (B) Hoxc10 was overexpressed using AAV-Hoxc10 (C) Western blot (D) and mRNA expression in SVF-derived adipocytes after Hoxc10 overexpression

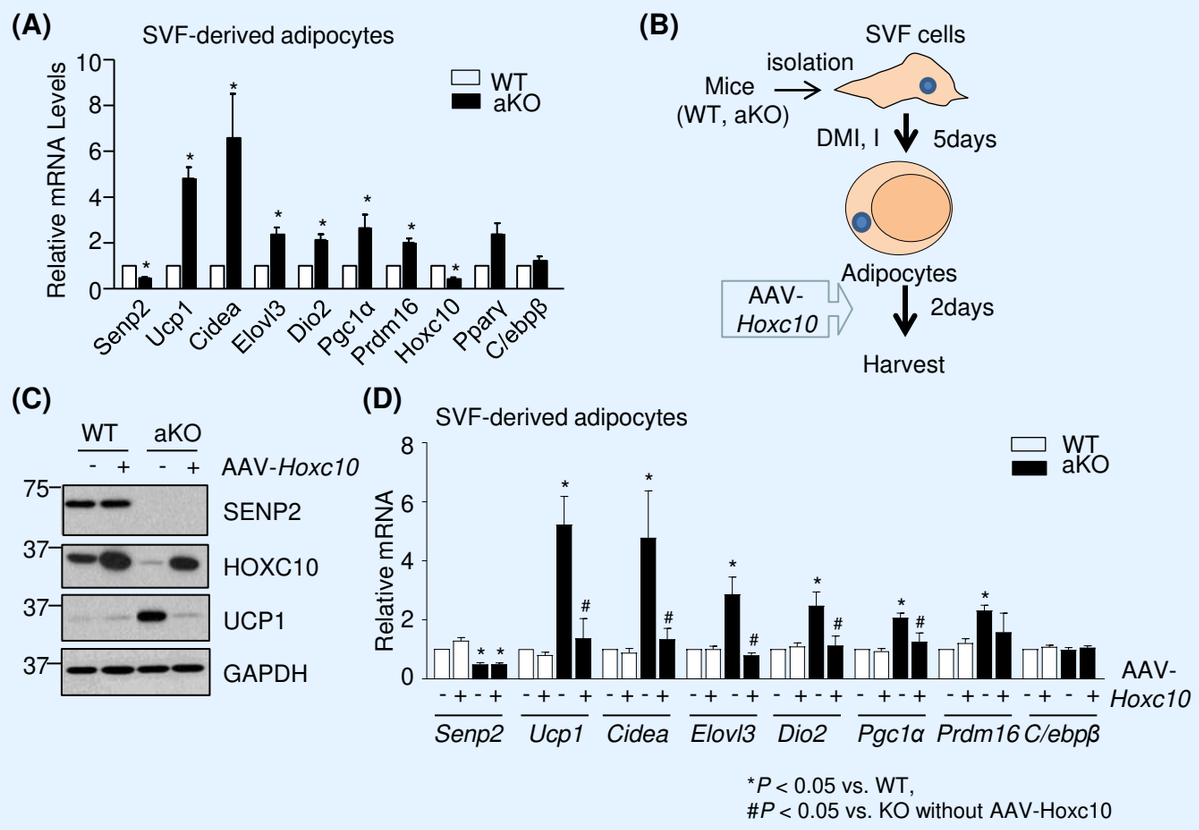


Fig 4. C/EBPβ mediates Hoxc10 reduction by SENP2 depletion SVF-derived adipocytes were transfected with siRNAs (100 nM) of each transcription factor. (A) q-PCR and (B) immunoblot analysis were performed.

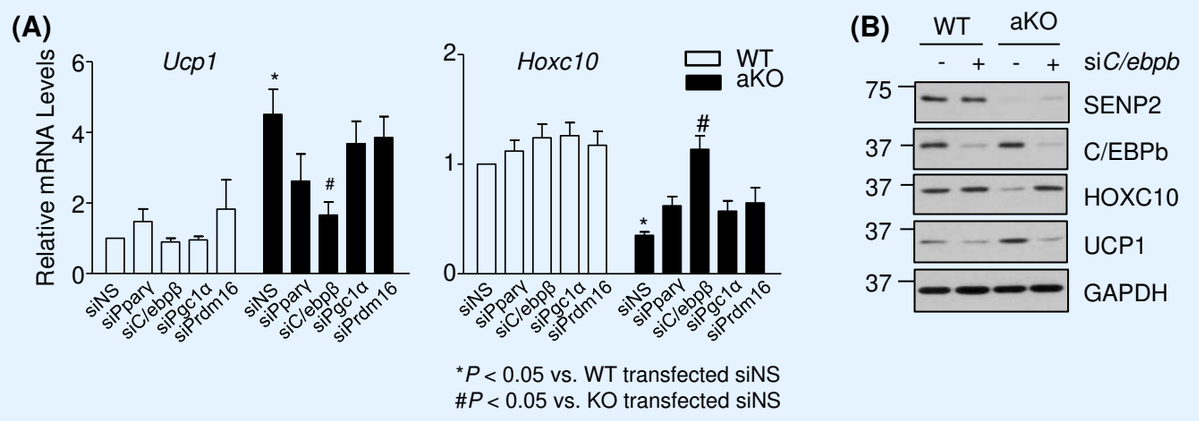
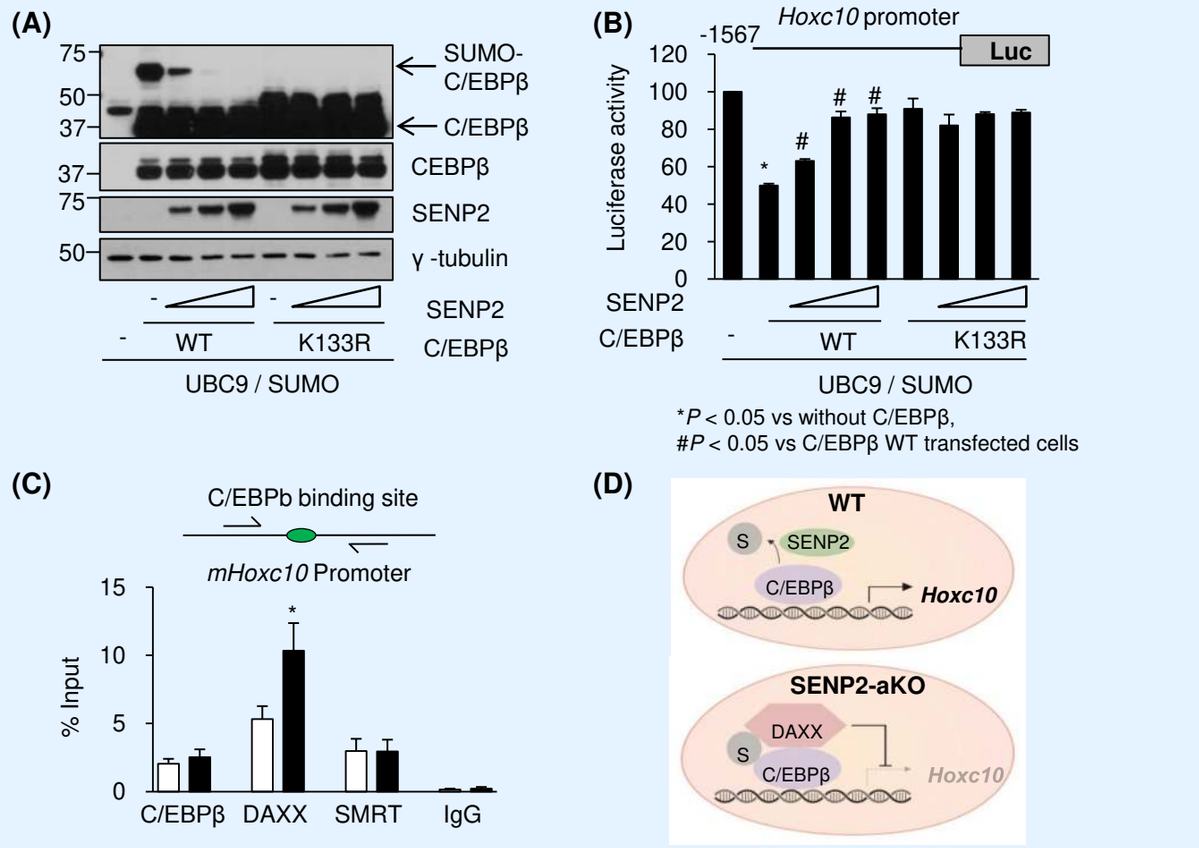


Fig 6. Sumoylated form of C/EBPβ efficiently suppresses Hoxc10 transcription (A) Immunoblot analysis (B) Luciferase activities (C) ChIP- qPCR (D) A proposed model of transcriptional regulation of Hoxc10 by C/EBPβ sumoylation



Conclusion Adipocyte-specific loss of SENP2 promotes browning in white adipose tissue through suppression of Hoxc10 transcription, which results in enhanced energy expenditure and alleviation of HFD-induced obesity and insulin resistance.