

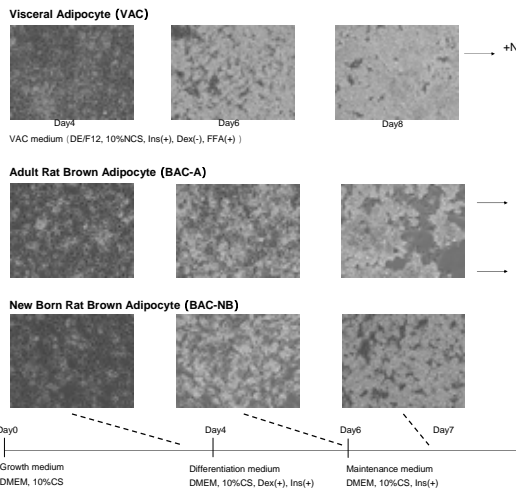
Conversion of white adipocyte into brown adipocyte within visceral adipocyte culture system, and concerning adult rat brown adipocyte.

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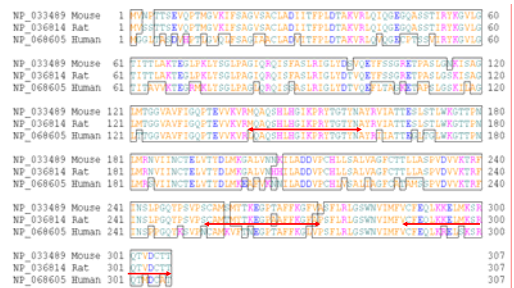
Mitochondrial uncoupling protein 1 (UCP1) is a key molecule for metabolic thermogenesis in brown adipose tissue (BAT). Generally, visceral adipose tissue is recognized as a white adipose tissue (WAT).⁽¹⁾

Recently, several researchers have shown that conversion of WAT into BAT by feeding seaweed lipids or the cold exposure in vivo.^(2,3) Here, we demonstrate that white adipocytes can be converted into brown adipocytes (BAC) by the addition of norepinephrine in vitro. Norepinephrine (1×10^{-6} M) increases the both UCP1 gene and protein expression in visceral adipocyte (VAC) culture at 6 hours stimulation. In addition, we show that the differences of between brown adipocytes isolated from adult rat and new born rat.⁽⁴⁾

Primary cultured adipocyte



UCP1 Amino Acid Sequence



- 1) NP_036814_rUCP-1_#141
CMQAQSHLHGKPRYGTYN
C: Additional Cys for conjugation
- 2) NP_036814_rUCP-1_#254
CAMTMYTKEGPAAFKFGFA
- 3) NP_036814_rUCP-1_#288
CFEQLKMLKSRQTVDCTT

All peptides were synthesized by F-moc method.

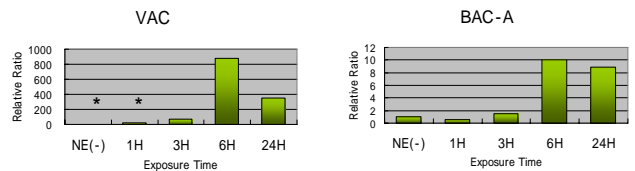
Peptide-KLH conjugation as antigen (Ag) were prepared using MBS method.

One Leghorn Hen was immunized for each Ag. 0.1mg of Ag was injected with FCA (1st injection) and FIA (3 or 5 boosts) at 2 weeks interval.

Conversion of white adipocyte into brown

Time dependency of UCP1 gene expression with NE(1uM) stimulation. (day7)

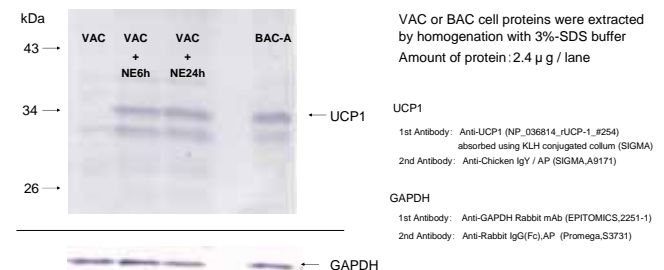
UCP1 gene expression (Real time PCR)



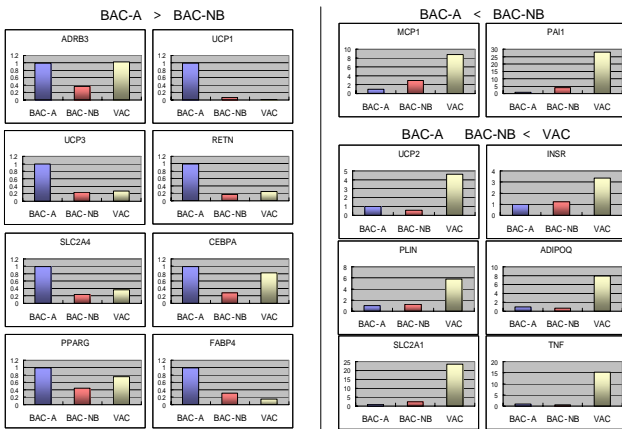
* : CP Value was lower than linear range

Reference gene: GAPDH

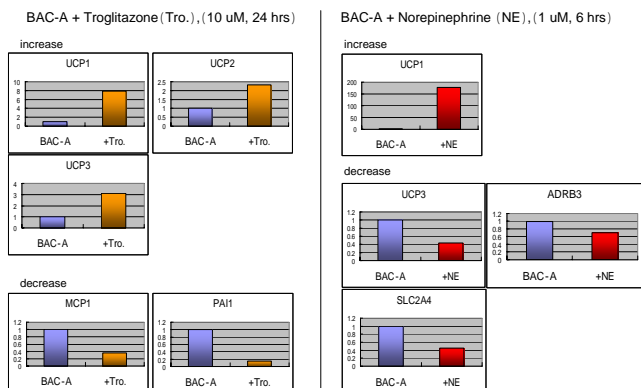
UCP1 Protein expression (Western blot)



Gene expression comparison between BAC-adult, BAC-new born and VAC. (day7)



Adipocyte marker gene expression changes by Troglitazone and Norepinephrine. (day7)



Materials

BAC-A : Rat Brown Adipocyte Culture kit (Adult) (Primary Cell, BAT10)
BAC-NB : Rat Brown Adipocyte Culture kit (New Born) (Primary Cell, BAT01)
VAC : Visceral Adipocyte Culture kit (Primary Cell, VAC01)
Primers : Real Time PCR Primer Set (For Rat Adipocyte) : (Primary Cell, PCRR1)

References

1. Newly developed primary culture of rat visceral adipocytes and their in vitro characteristics. Shimizu K, Sakai M, Ando M, Chiji H, Kawada T, Mineo H, Taira T. Cell Biol Int. 2006 Apr;30(4):381-8. Epub 2006 Mar 10.
2. The emergence of cold-induced brown adipocytes in mouse white fat depots is determined predominantly by white to brown adipocyte transdifferentiation. Barbatelli G, Murano I, Maden L, Hao Q, Jimenez M, Kristiansen K, Giacchino JP, De Matteis R, Cinti S. Am J Physiol Endocrinol Metab. 2010 Jun;298(6):E1244-53. Epub 2010 Mar 30.
3. Fucoxanthin from edible seaweed, Ulvaria pinnatifida, shows antiobesity effect through UCP1 expression in white adipose tissues. Maeda H, Hosokawa M, Sashima T, Funayama K, Miyashita K. Biochem Biophys Res Commun. 2005 Jul 1;332(2):392-7.
4. Effect of adenosine deaminase, N6-phenylisopropyladenosine and hypothyroidism on the responsiveness of rat brown adipocytes to noradrenaline. Woodward JA, Saggerson ED. Biochem. J. (1986) 238 (395-403)