# Effect of hypothalamic leptin resistance on energy metabolism in skeletal muscle

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# Leptin

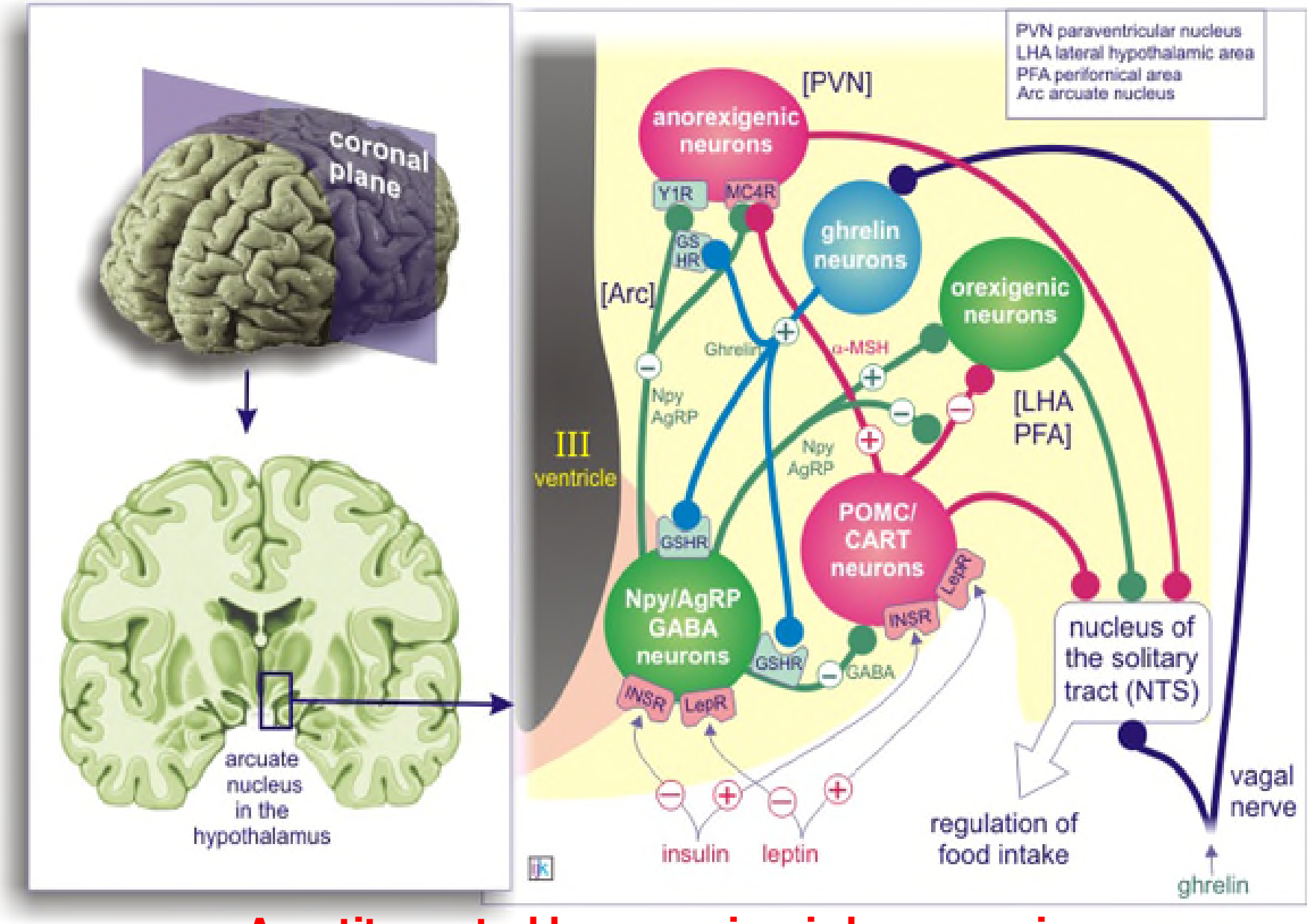
**Zhang Y, Proenca R, Maffei M, Barone M, Leopold L, Friedman JM.** Positional cloning of the mouse obese gene and its human homologue. *Nature 372: 425–432, 1994.*

**Halaas JL, Gajiwala KS, Maffei M, Cohen SL, Chait BT, Rabinowitz D, Lallone RL, Burley SK, Friedman JM.** Weight-reducing effects of the plasma protein encoded by the obese gene. *Science 269: 543–546, 1995.*

Leptin is a suitable gateway for peripheral molecules to access brain tissue

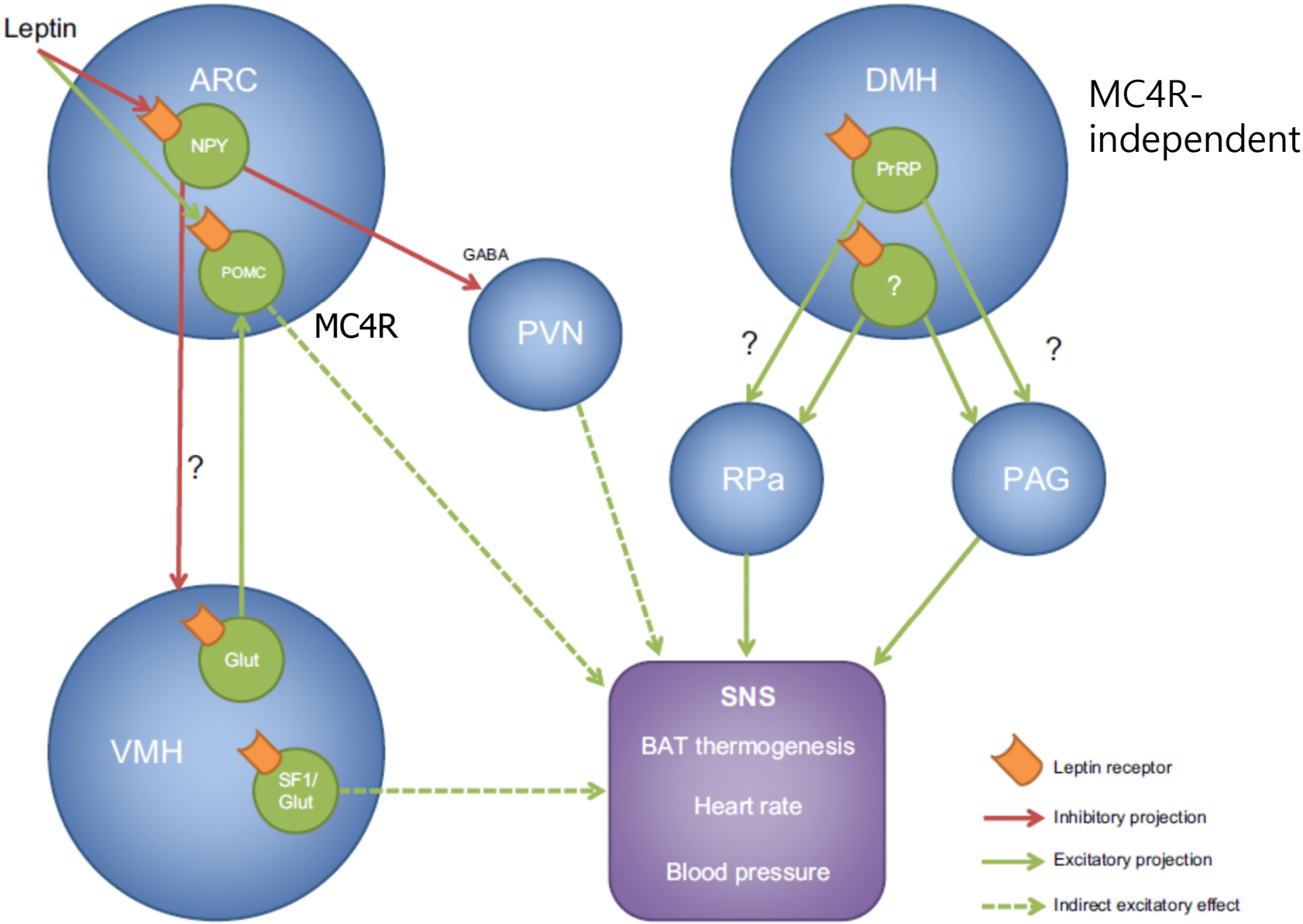
Arcuate nucleus (ARC) at the border of third ventricle
Neuropeptide Y (NPY) and Proopiomelanocortin (POMC) neurons
→ related with food intake and glucose metabolism

# Food intake regulation



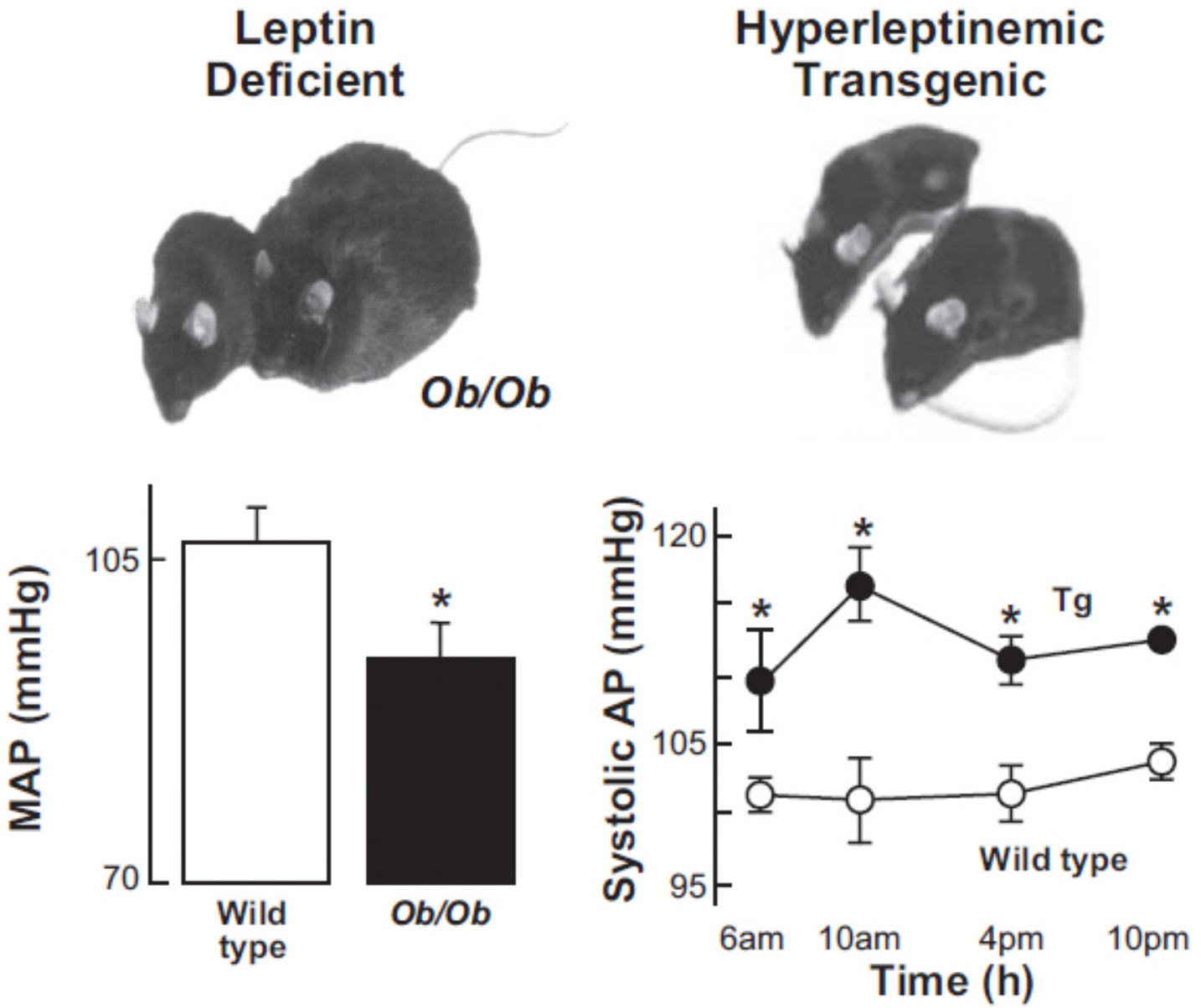

**Appetite control by anorexigenic hormones is mediated by melanocortin 4 receptors (MC4R)**

# Energy expenditure and cardiovascular regulation




MC4R-independent

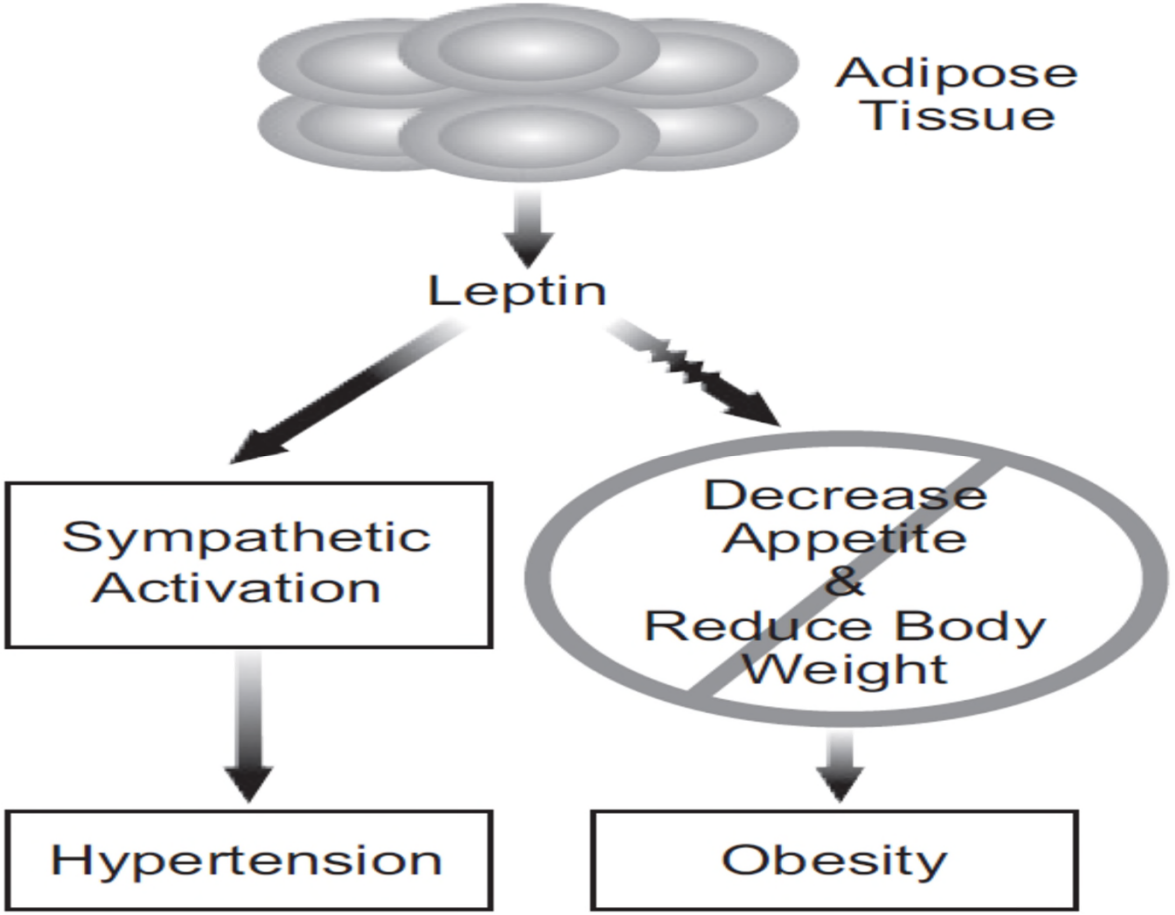
*Pandit R et al, 2017, AJP*




*Mark AL 2013, AJP*

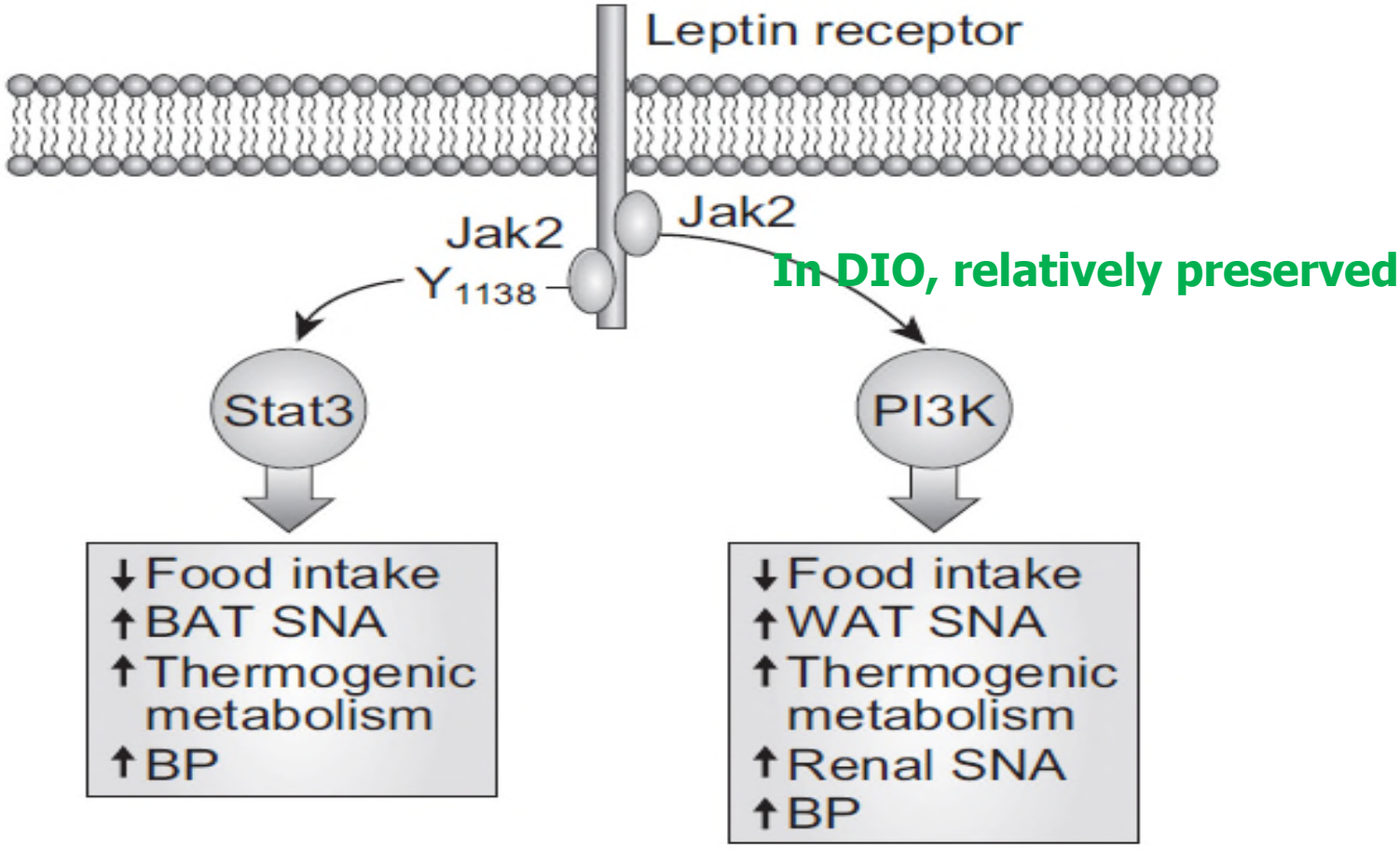
# Selective leptin resistance

Obesity is associated with high blood pressure and hyperleptinemia, implying that leptin resistance does not influence the leptin-mediated hypothalamic activation of renal sympathetic nervous activity



# Possible cause of selective leptin resistance

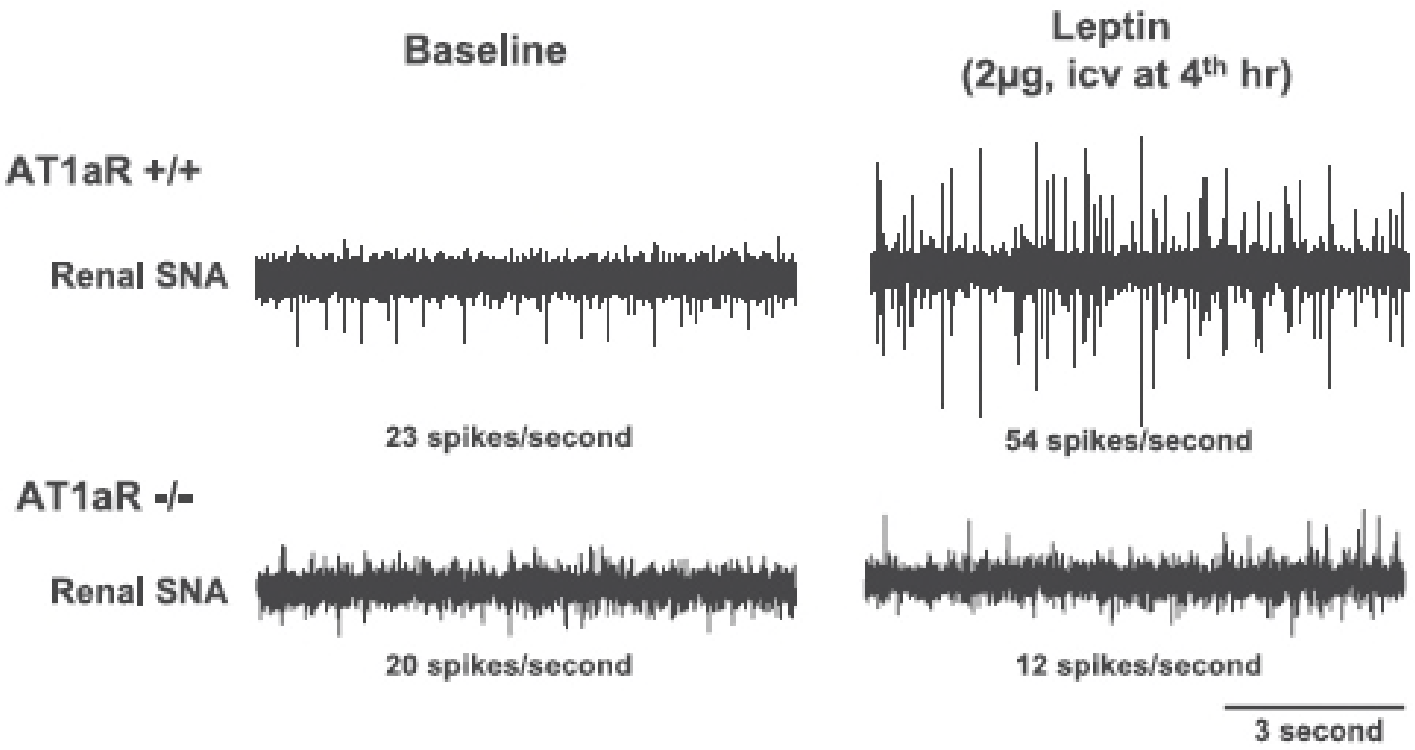
Pathway-specific leptin action: STAT3, PI3K, and ERK/MAPK

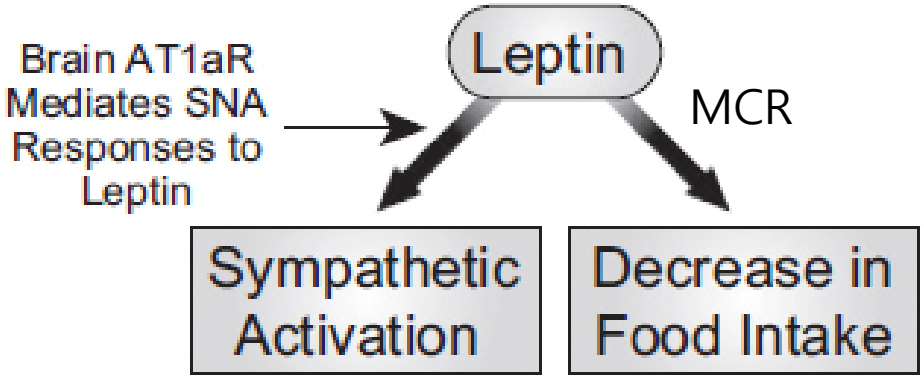



Very intriguing: SNS activation by leptin is also divided

*Mark AL, 2013, AJP*

# Receptor-specific leptin action



# Physiological role of leptin and SNA in BAT and skeletal muscle

## In BAT

- Leptin was shown to increase glucose utilization through ObRb and the JAK/STAT pathway.

- Leptin increases the expression of some metabolic enzymes and PPAR activity in preadipocytes and BAT

- UCP-1 gene expression in BAT was induced by leptin, but through sympathetic innervation, β3

## In Skeletal muscle

- Leptin directly exerts its action, which is like the action of insulin, by facilitating glycogen synthesis, glucose uptake, and promoting cell survival

- Leptin deficiency (e.g., *ob/ob* mice) caused decreased muscle mass and increased expression of the muscle-wasting protein myostatin, which was recovered by administration of exogenous leptin.

-SNS mainly via β2 receptors, increases muscle mass by activating the Akt/mTOR pathway

## Epac2a deficiency as a leptin resistance model

Epac2a knock-out mice exhibited obesity-prone nature
and resistance to leptin for the regulation of food intake

However,
The distribution of Epac2a in the distinct nuclei of the hypothalamus and its role in the leptin-mediated hypothalamic regulation of SNA has not been fully clarified.

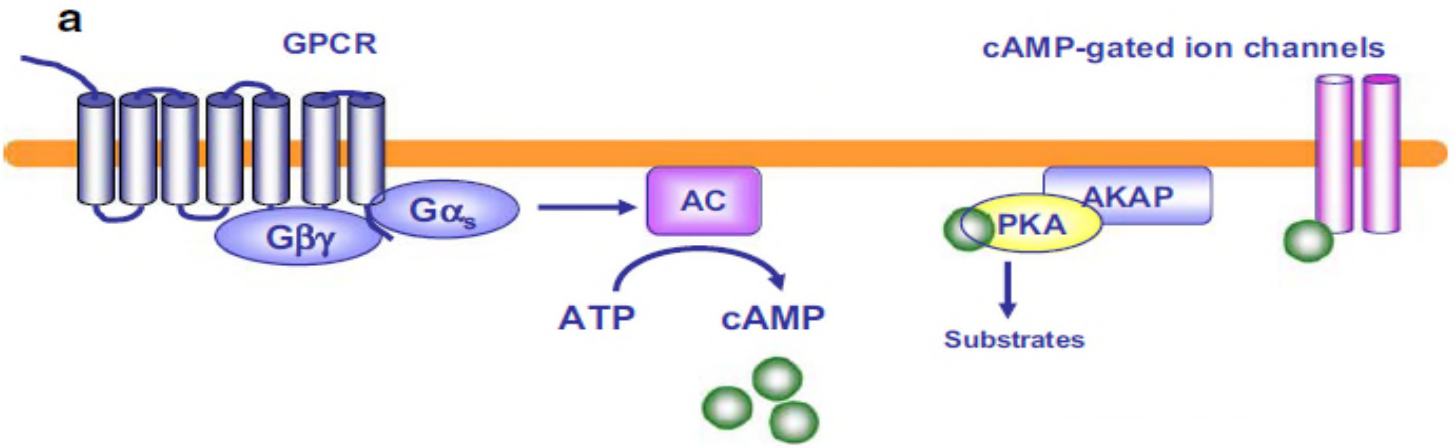
## Purpose of this research

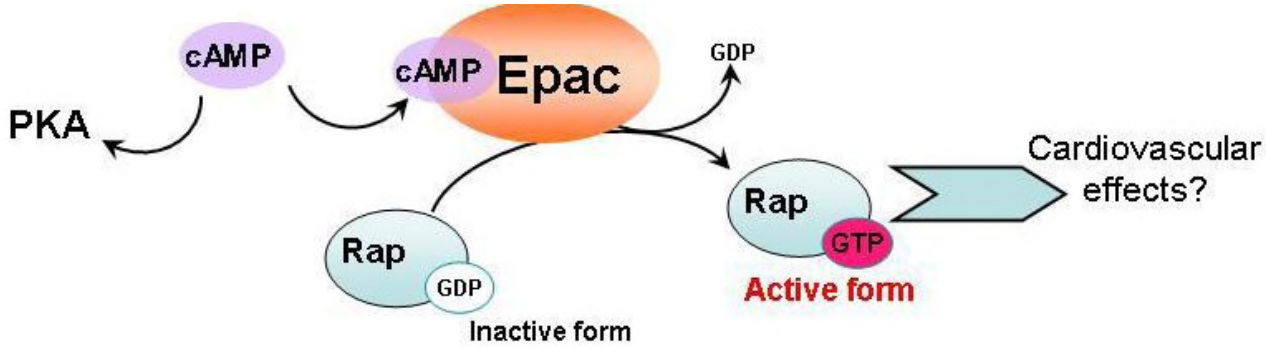
to evaluate the possible changes in SNA in Epac2a-KO mice, by observing the response of peripheral tissues to dexamethasone-induced muscle atrophy and cold stress.

We chose 7 weeks of age of mice
After that, adipose tissue mass
and food intake increase can be detected

Selection of pure hyperleptinemic state before significant changes in food intake, body weight, blood glucose, insulin resistance, adipokines, and etc is to rule out their possible influence on BAT and skeletal muscle

# Epac (exchange protein directly activated by cAMP) as a cAMP target

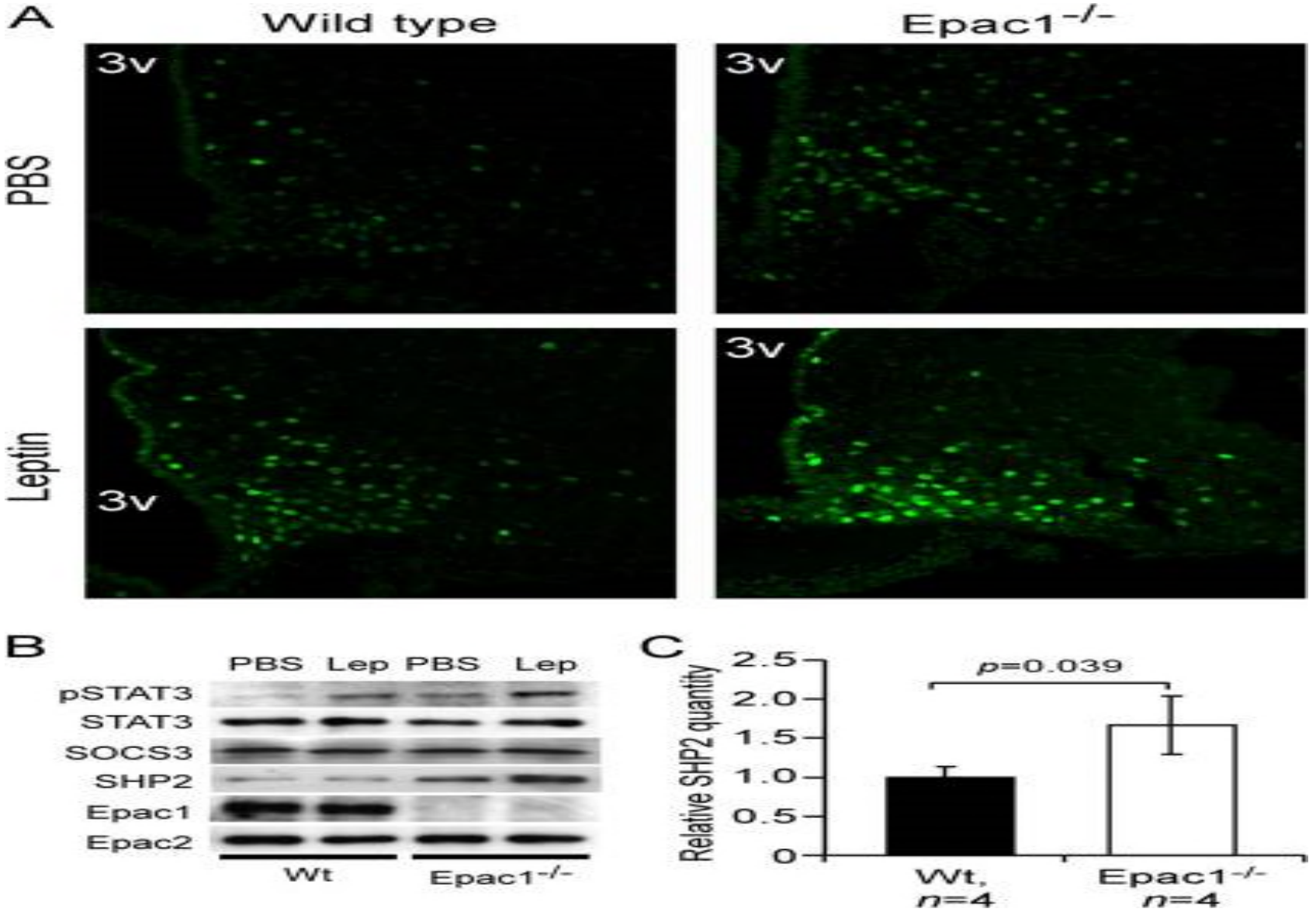


Epac1: Ubiquitous

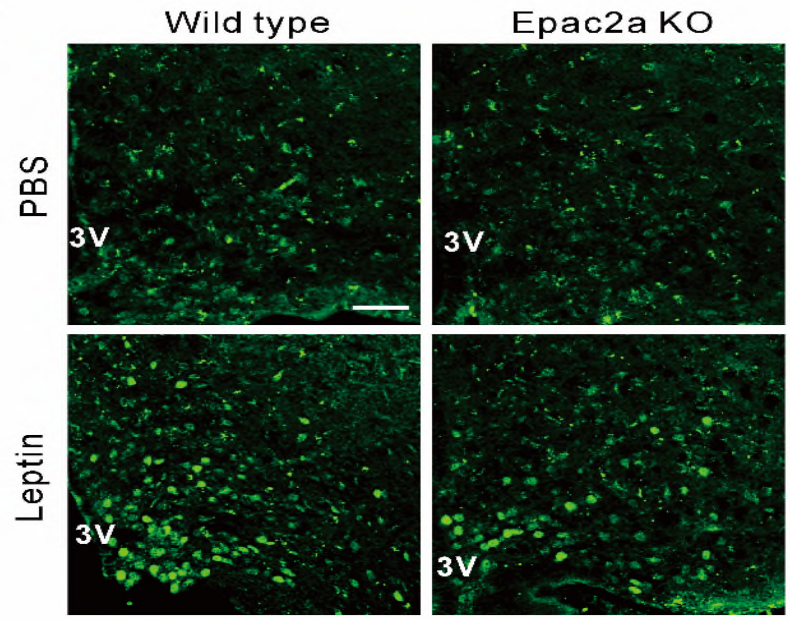
Epac2:
Brain/Heart/β-cell type (Epac2a)
Adrenal type (Epac2b)
Liver type (Epac2c)

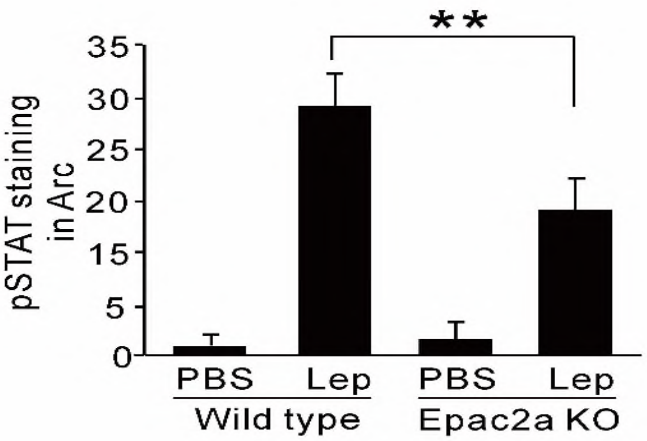
# Epac1 deficiency increases hypothalamic leptin sensitivity

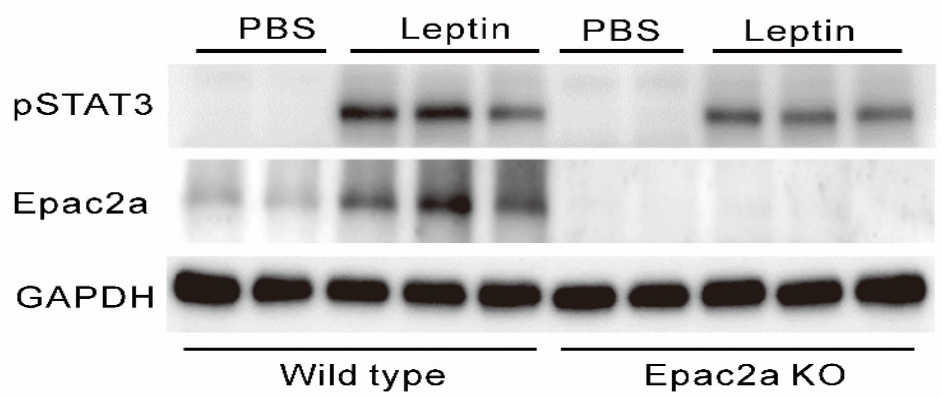


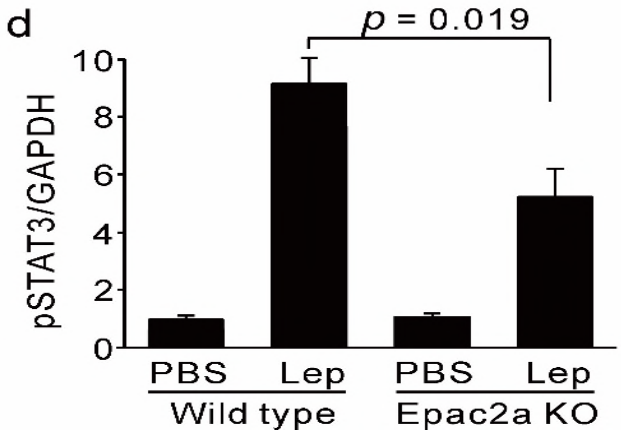

*Yan j, MCB, 2012*

# Epac2a deficiency decreases hypothalamic leptin sensitivity

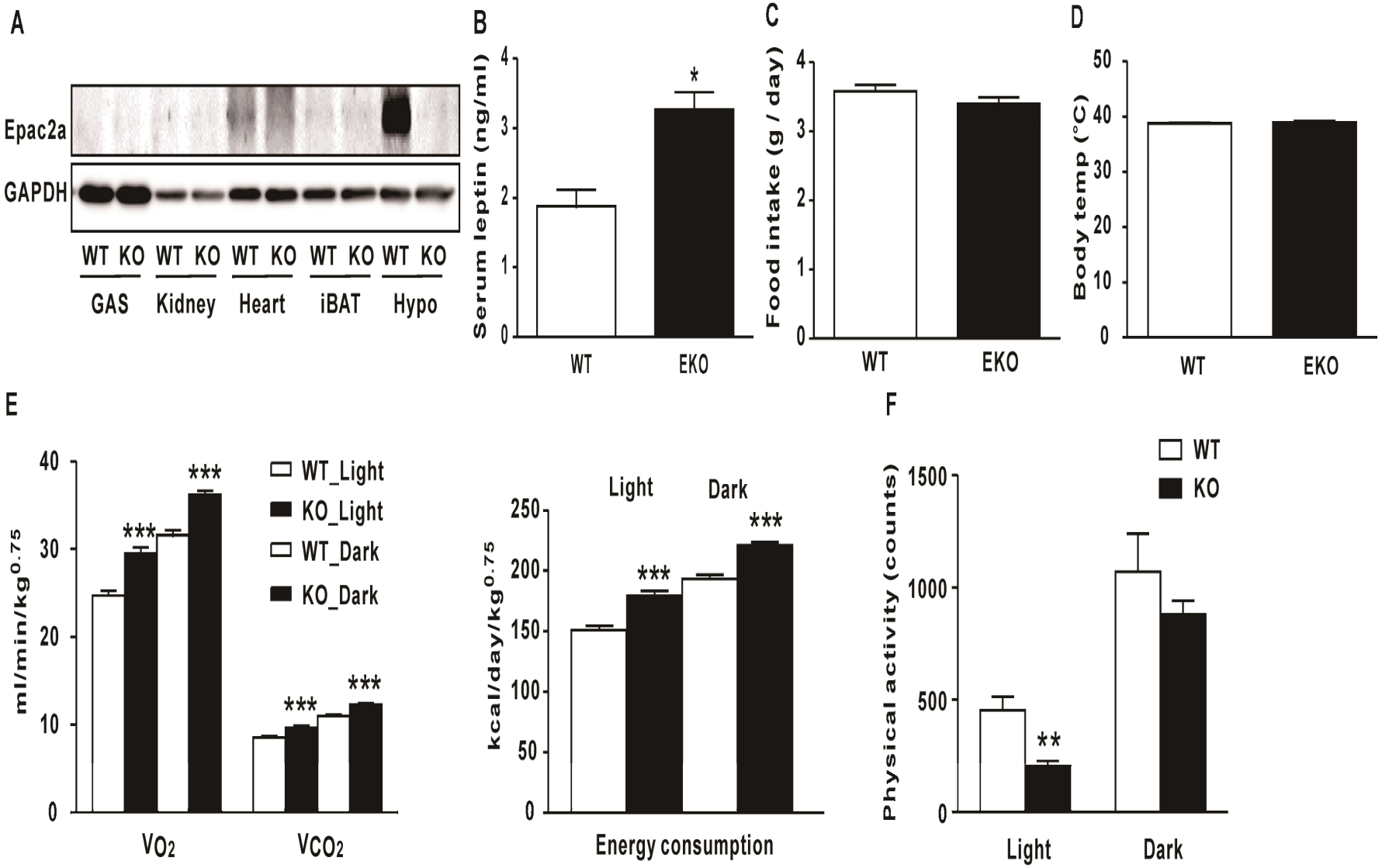


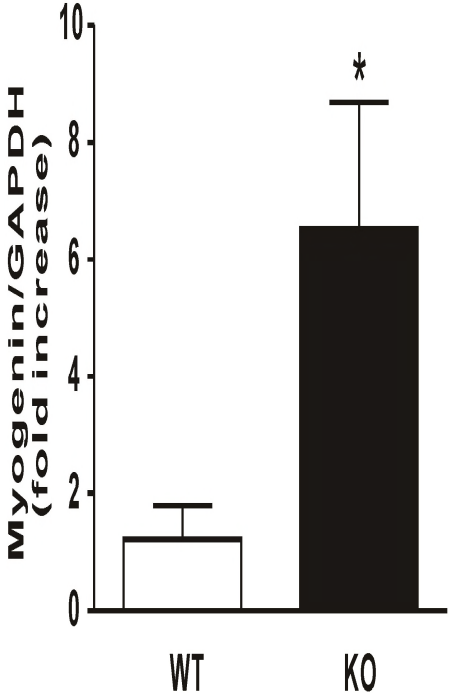



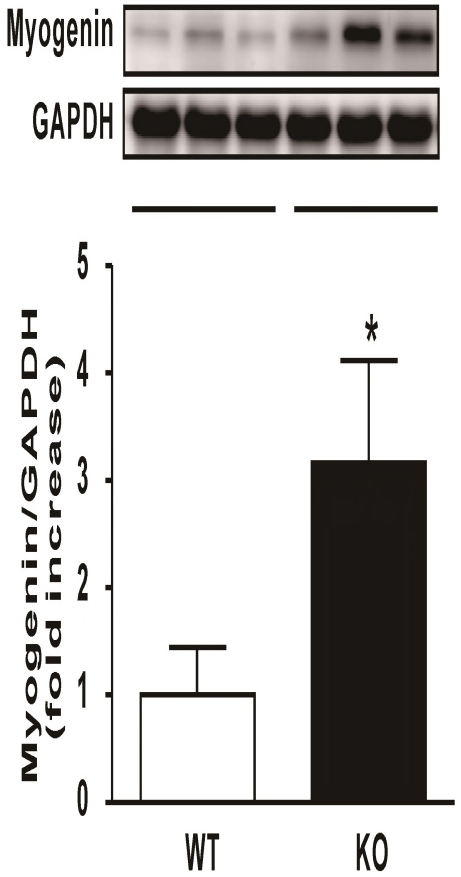



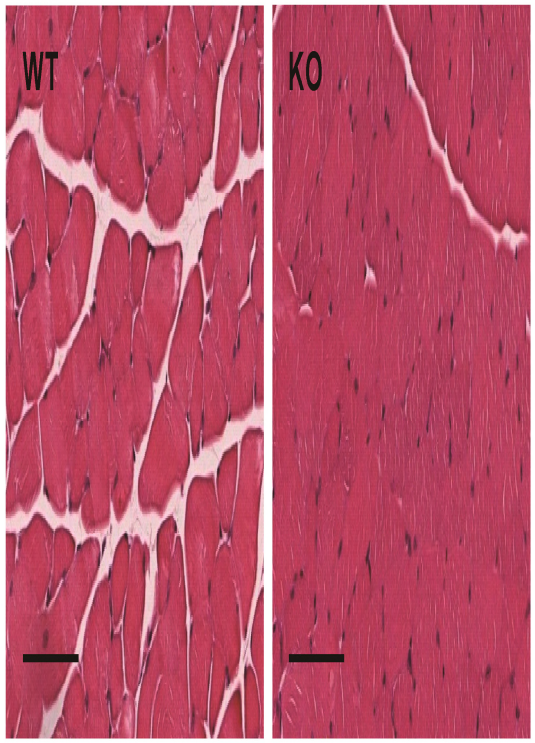
# Epac2a KO mice exhibit greater EE and lower physical activity



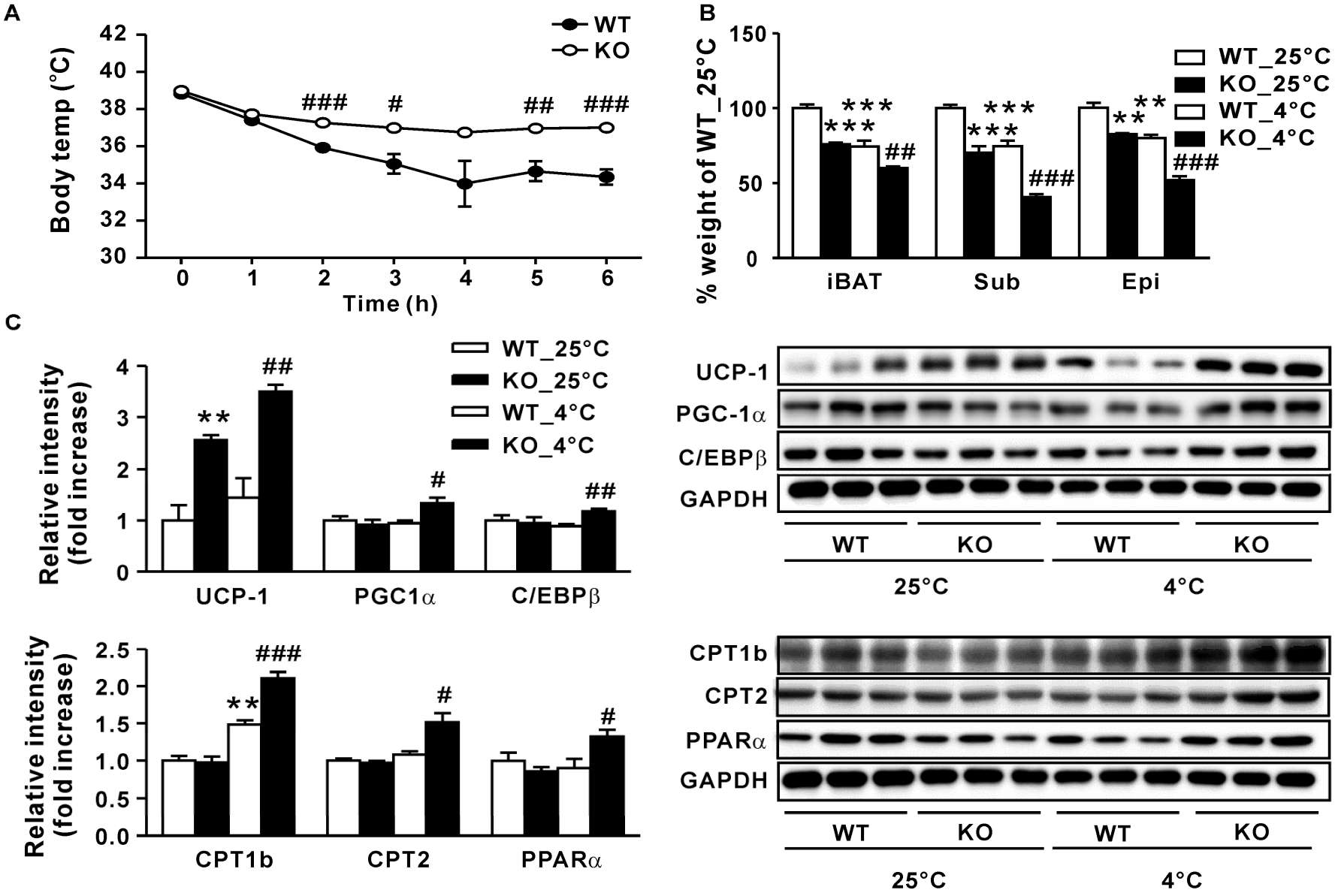
# Epac2a KO mice exhibit greater myogenin and muscle density



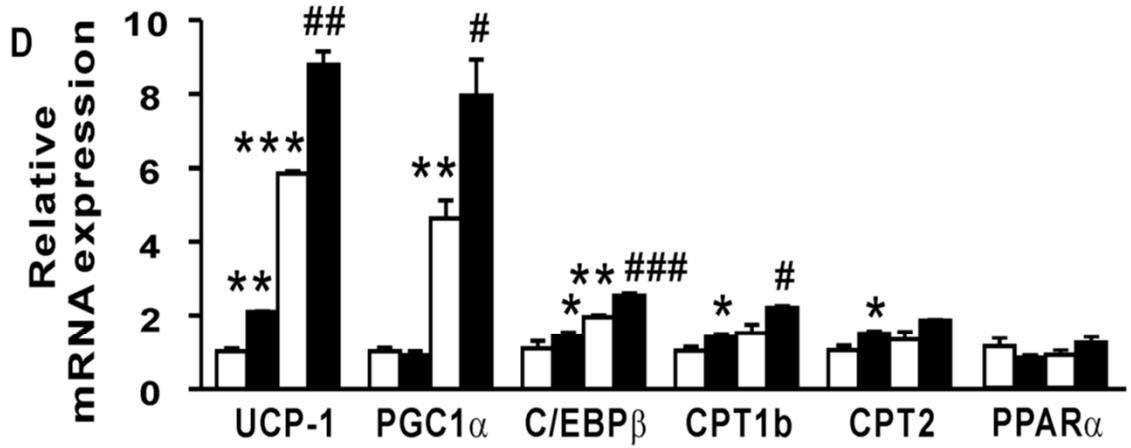



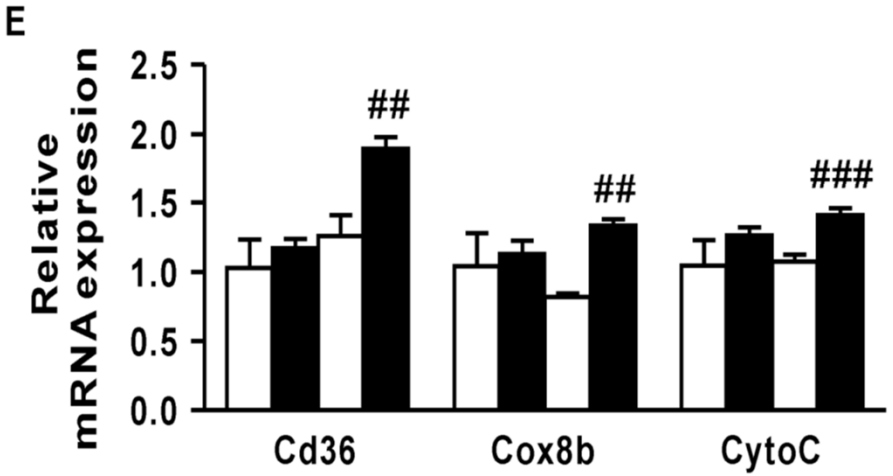
# Epac2a KO mice exhibit greater tolerance to cold stress



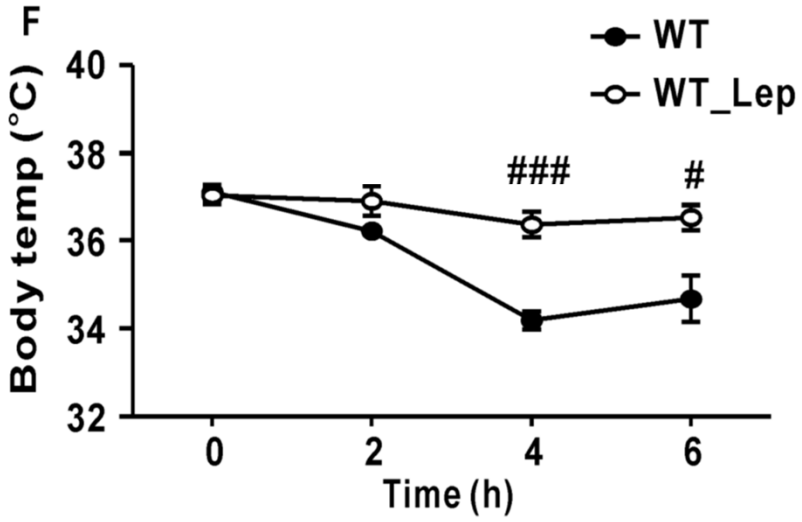
# Exogenous leptin increases tolerance to cold stress

Thermogenic factors

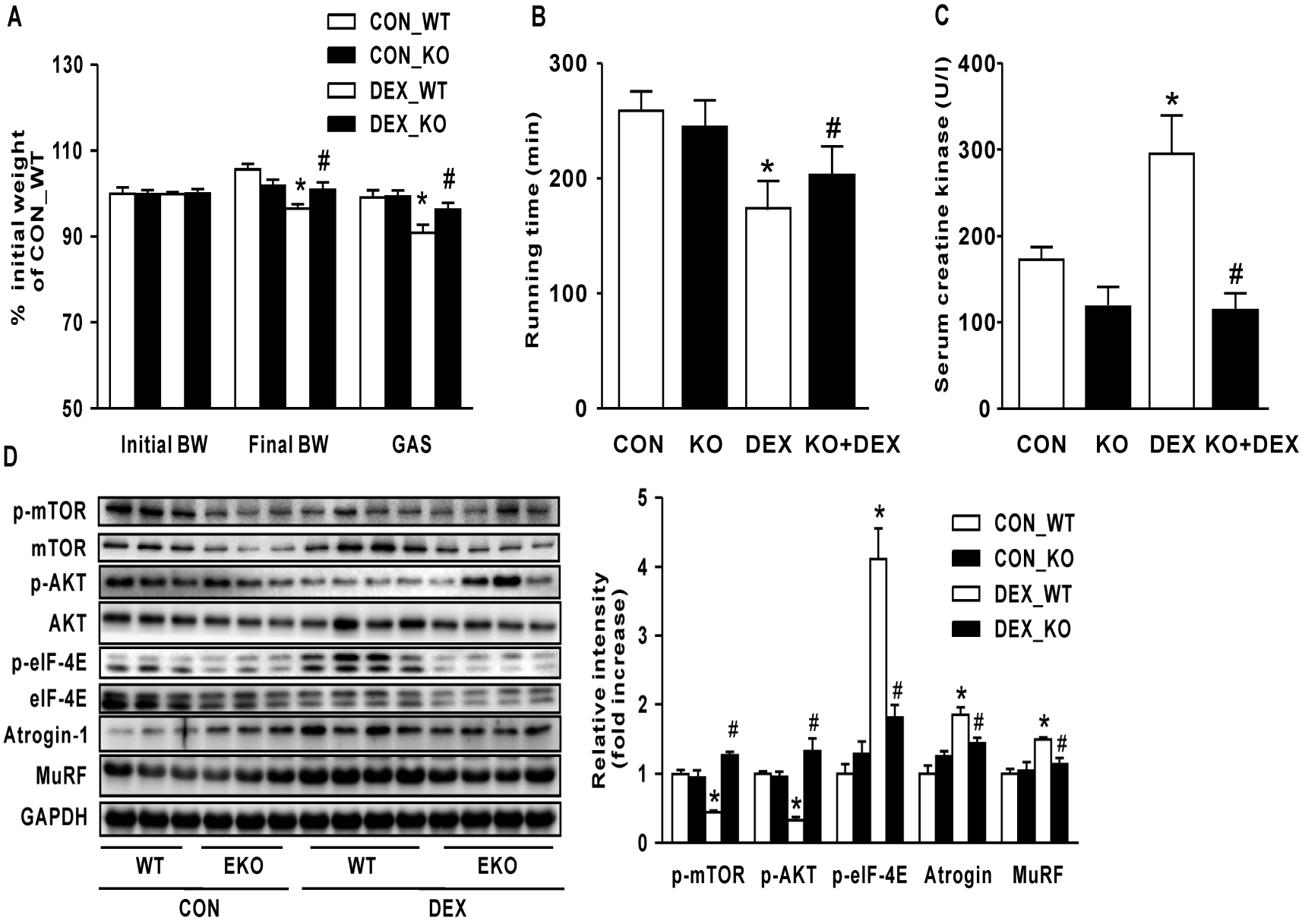


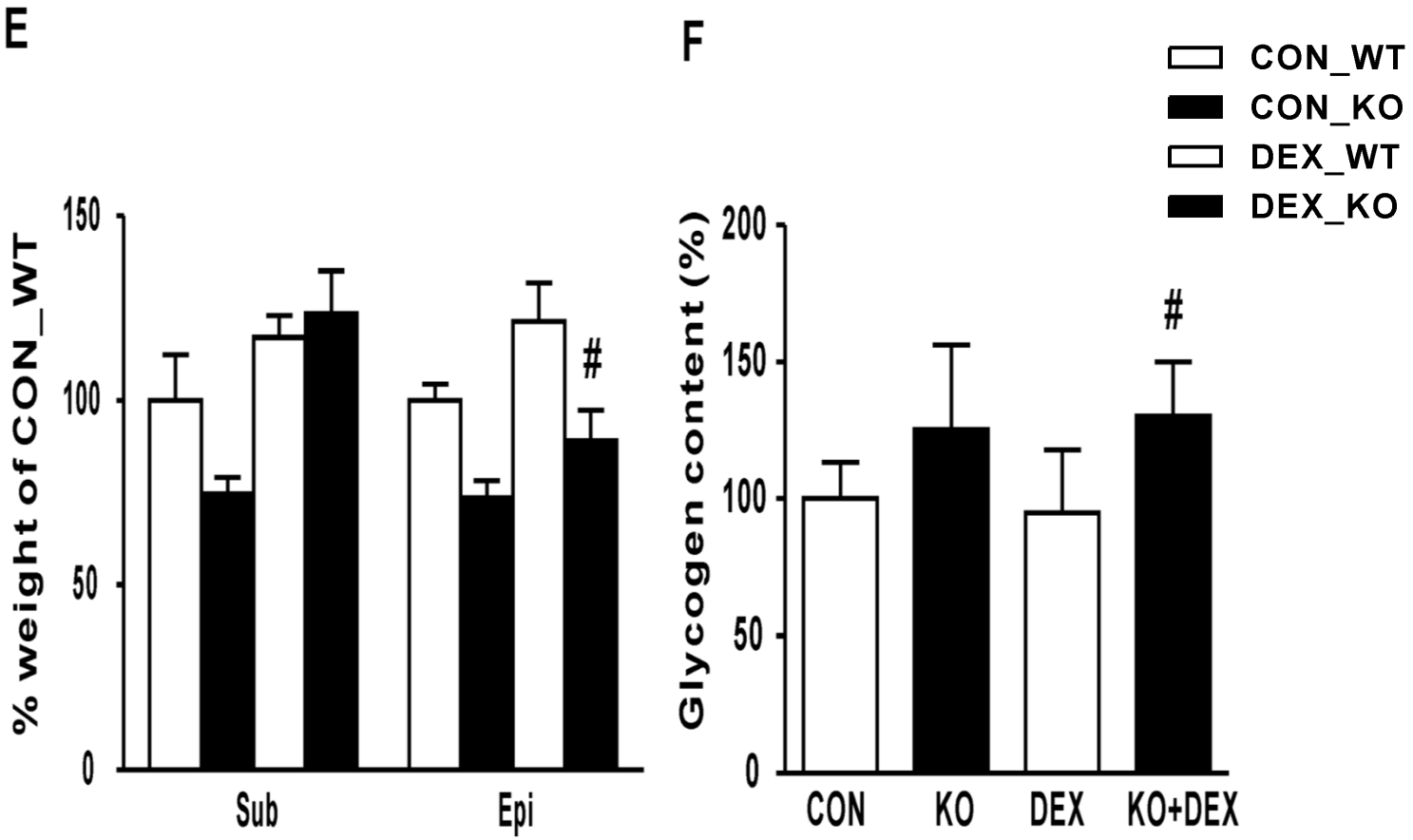

Fatty acid uptake and
mitochondrial oxidative metabolism



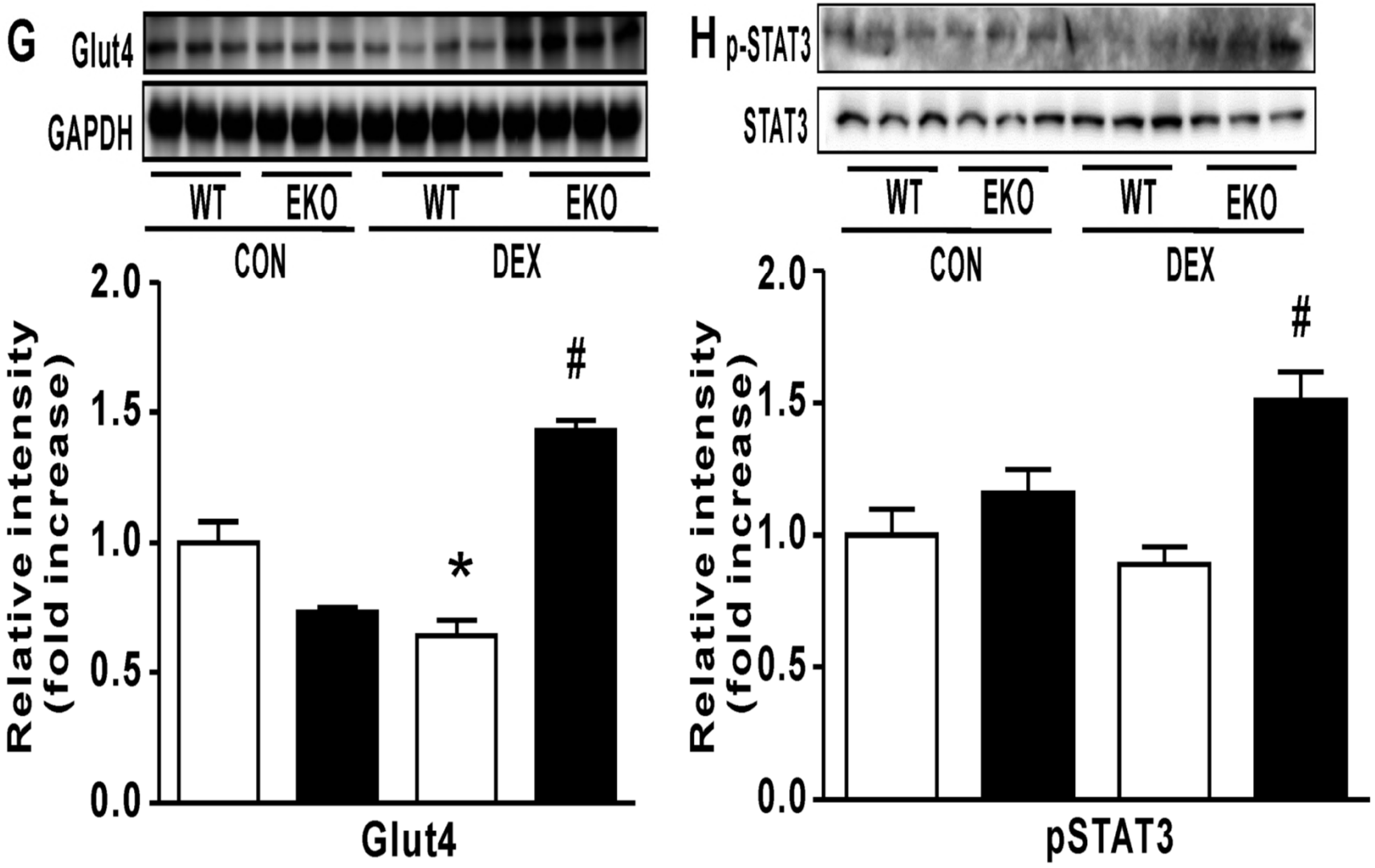
# Epac2a KO mice exhibit greater tolerance to dexametasone



# Fat mass and glycogen content



# Glut4 and pSTAT3



# pPKA levels in BAT and skeletal mucle

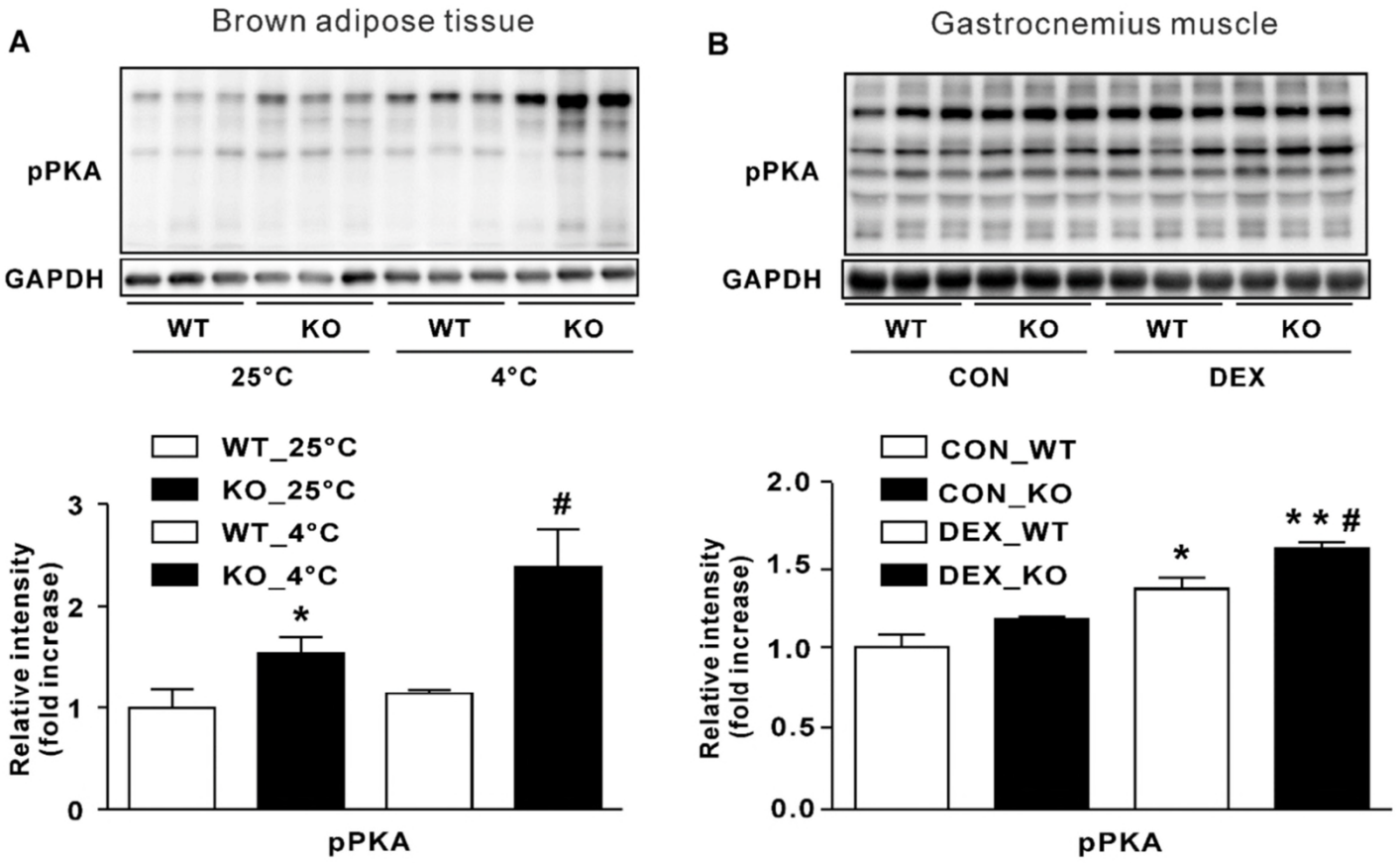



Fig. S1. (A) pPKA protein expression in iBAT before and after cold exposure. (B) pPKA protein expression in gastrocnemius muscle before and after 7-day dexamethasone treatment.
*p<0.05, **p<0.01 compared to WT; #p<0.05 compared to WT treated with cold or dexamethasone stress. For details, see the main text.

# TG levels in BAT before and after cold stress

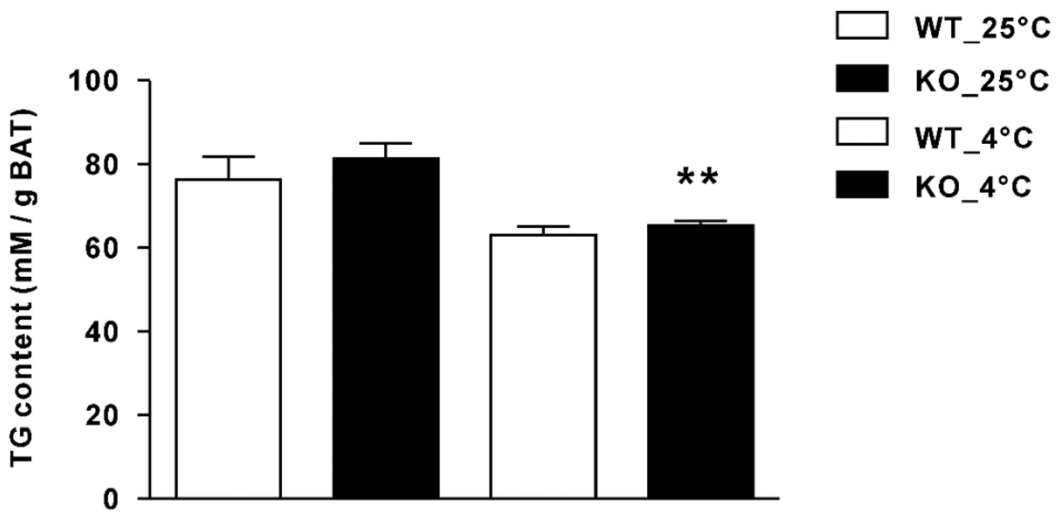
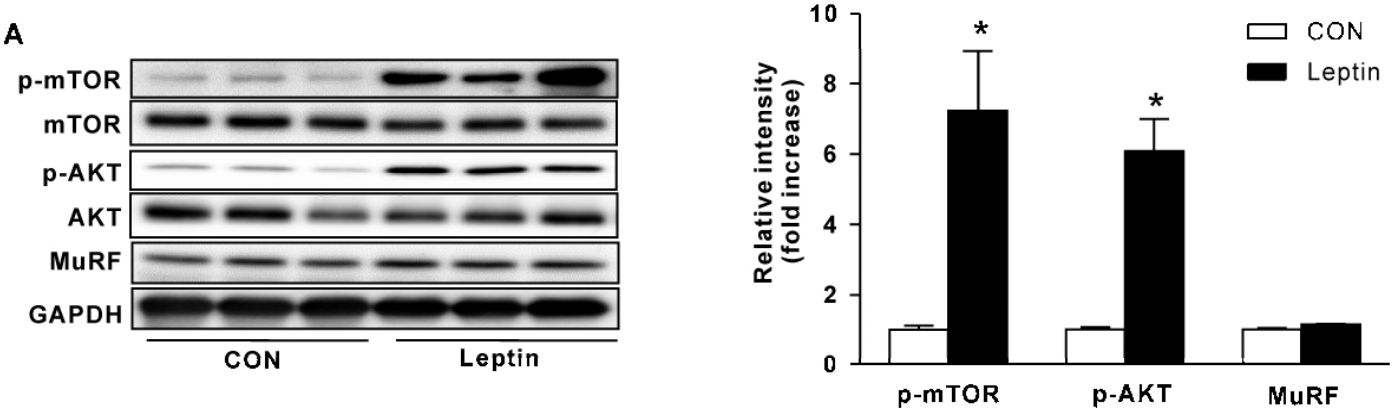
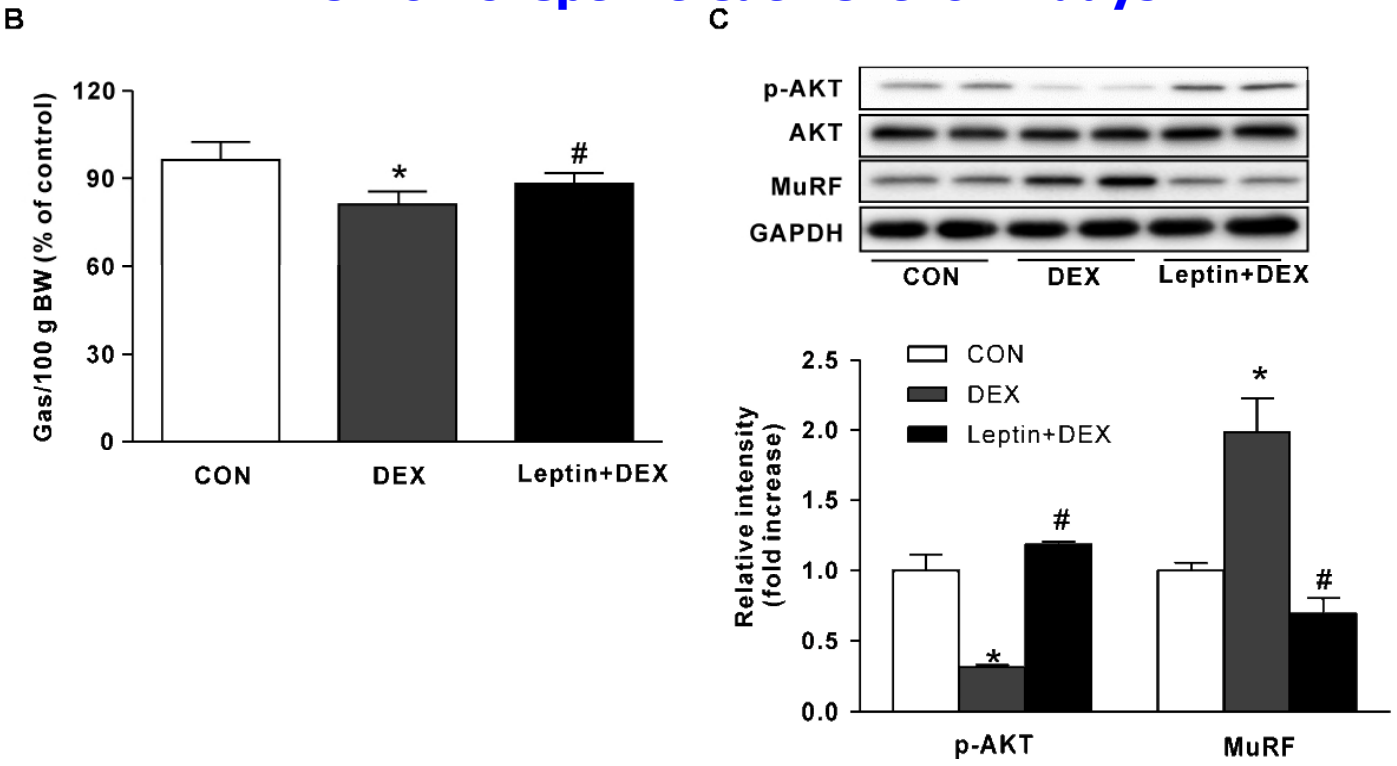



Fig. 2S. Triglyceride content (TG) of iBAT before and after cold stress.
**p<0.01 compared to KO at ambient temperature. For details, see the main text.

**WT mice were administered with leptin (i.p.). After 45 min, an *ex vivo* study of the gastrocnemius muscle was conducted.**




**Chronic leptin treatment for 7 days**



# In summary

- We confirmed that Epac2a expression was not detected in gastrocnemius muscle and BAT in WT mice

- Leptin-mediated activation of SNA from the hypothalamus, projecting to BAT and skeletal muscle, may not be impaired at least in Epac2a deficiency-type leptin resistance

- Skeletal muscle and BAT in Epac2a-KO mice have normal leptin and adrenergic signaling through their own respective plasmalemmal receptors.

## In conclusion,

Epac2a in the hypothalamus only affects leptin-mediated signaling for appetite control, but not SNA regulation

If function of BAT and skeletal muscle tends to be impaired in obesity, other factors,
such as insulin resistance, glucose intolerance, unexpected adipokines, and lower pAMPK, etc. might be the cause.

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*Thank you for your attention!*