Supplemental Figure 1. 7 days ICV leptin treatment did not change plasma leptin levels and STAT-3 phosphorylation in gastrocnemius muscles in 10% fat diet fed and 60% fat diet fed mice

A, Plasma leptin levels as determined by ELISA assay. B. Representative immunoblots and densitometric analysis of phosphorylated and total STAT-3

Values are mean ± S.E. of 9 for each group

Supplemental Figure 2. Acute ICV leptin injection induces an increased phosphorylation of STAT3, decreased phosphorylation of AMPK and ACC in the hypothalamus of C57/BL mice.

Representative immunoblots and densitometric analysis of A, STAT3 phosphorylation, B, AMPK phosphorylation and C, ACC phosphorylation in hypothalamus of C57/BL mice. Mice on normal chow diet were given ICV leptin injection (5ug). Hypothalamic tissues were collected 3 hrs after injection.

Supplemental Figure 3. Effect of 7 days of ICV leptin treatment on hypothalamic expression of FOXO1

Representative immunoblots and densitometric analysis of FOXO1 in the hypothalamus

Values are mean ± S.E. of 6 for each group. * p<0.05 when compared to respective control
Supplemental Figure 1

(a) Plasma Leptin (pg/mL)

(b) Western Blot Analysis

- **P-STAT3**
- **STAT3**
- **Actin**
Supplemental Figure 3

**Western Blot Analysis**

- **FOXO1/Actin**
  - 10% fat diet
  - 60% fat diet
  - Vehicle
  - Leptin

- **Actin**
  - Vehicle
  - Leptin

**Graphical Representation**

- Comparison of FOXO1/Actin expression levels between 10% and 60% fat diets under Vehicle and Leptin conditions.

- Statistical significance indicated by asterisks (*) on the graph.