

## SUPPLEMENTARY DATA

### Materials and Methods:

#### Genetic association studies: single-nucleotide polymorphism (SNP) selection, clinical data collection, and genotyping platform

We searched the GWAS Central database (<http://www.gwascentral.org>) for associations of phenotypes near/within the *Slc2a10* (GLUT10) gene. Search criteria included a *p*-value threshold of 0.05 and 10 kb-flanking regions. SNPs within the region were identified from meta-analysis of a glucose and insulin-related traits study [1], a 2-h glucose challenge study [2], an adult body mass index study in a British population [3], and a T2D study [4] as listed in S1 Table.

#### Measurement of food intake and metabolic rate

Mice were housed individually for measurement of food intake using Tecniplast® Metabolic Cage (TECNIPLAST S.p.A, Via I Maggio, Italy), or their metabolic rate was measured using the CLAMS-home cage (CLAMS-HC) system (Columbus Instruments, Columbus, OH, USA) in the Taiwan Mouse Clinic at Academia Sinica. The first readings were taken after a 48-h acclimation period. Heat production, RER, oxygen consumption rate (VO<sub>2</sub>), and carbon dioxide production (VCO<sub>2</sub>) rates were determined. VO<sub>2</sub>, VCO<sub>2</sub>, and heat were measured every 17 min during a 76-h period at the indicated temperature and were normalized to body weight.

#### Mice

*Glut10*<sup>G128E</sup> mice were backcrossed for more than 10 generations from a C3H background onto the C57BL/6J background, which is widely used in metabolic disease research [5, 6]. *Glut10*<sup>G128E</sup> mice on a C57BL/6J background had similar phenotypes to those previously observed in *Glut10*<sup>G128E</sup> mice on the C3H background. *Glut10*<sup>G128E</sup> mice and their wild-type littermates (WT) control mice were produced from heterozygous breeding pairs. Animals were weaned at 3 weeks of age onto CD and genotyped at 4 weeks of age. *Glut10*<sup>G128E</sup> and WT mice were randomly assigned to CD or HFD at 5 weeks of age. Male mice were used in this study. No data were excluded in the analysis. Mice were weighed weekly. For fasting blood glucose measurement, GTT, and ITT studies, blood samples were collected from mice at indicated time points using a 30-gauge needle to puncture the tip of the tail. At the conclusion of CD and HFD-feeding, animals were killed by CO<sub>2</sub> asphyxiation. Blood was collected from cardiac puncture for blood chemistry and adipokines assays, and tissues were dissected immediately, weighed, and snap frozen in liquid nitrogen (for RT-PCR or western blot assays) or fixed (for histological analysis and immunohistochemistry studies).

#### Fluorescent microbead perfusion for vasculature studies

The functional architecture of the adipose vasculature in mice was examined by intravenous injection of fluorescent microbeads as described [7]. In brief, mice were injected with 150 µl of fluorescent microbeads in PBS (red fluorescence, 0.1 µm in diameter microbeads, Invitrogen) in their tail vein, anaesthetized 5-10 min later, and then perfused through the heart with 6 ml of 1:10 PBS dilution of fluorescent microbeads. Tissues were then dissected, embedded, sectioned, and analyzed of blood vessel density using fluorescence microscopy

#### Isolation of stromal vascular fraction (SVF) cells

SVF cells were freshly isolated from eWATs of WT and *GLUT10*<sup>G128E</sup> male mice (at age 6–8 weeks) as previously described [8].

#### OCR measurements

OCR was measured using the Seahorse XF24 extracellular flux analyzer as previously described [9].

#### ROS measurements

Intracellular and mitochondrial ROS were measured using 2',7'-dichlorodihydrofluorescein

diacetate (DCFH-DA; Sigma-Aldrich, St. Louis, MO, USA) and MitoSox Red (Thermo Fisher, Miami, FL, USA), respectively.

### Primer sequences used for PCR:

| Gene               | Forward primer (5'- 3')  | Reverse Primer (5'- 3')   |
|--------------------|--------------------------|---------------------------|
| <b>RT-qPCR</b>     |                          |                           |
| <i>36B4</i>        | TCCAGGCTTTGGGCATCA       | CTTTATCAGCTGCACATCACTCAGA |
| <i>β-actin</i>     | CCAACCGTGAAAAGATGACC     | ACCAGAGGCATACAGGGGACA     |
| <i>Gapdh</i>       | ATGACCACAGTCCATGCCATC    | GAGCTTCCCCTTCAGCTCTG      |
| <i>Emr1</i>        | TGTAACCGGATGGCAAAC       | CTGTACCCACATGGCTGATG      |
| <i>Tnfa</i>        | CACAGAAAGCATGATCCGCGACGT | CGGCAGAGAGGAGGTTGACTTTCT  |
| <i>Tgf β</i>       | CTCCCGTGGCTTCTAGTGC      | GCCTTAGTTTGGACAGGATCTG    |
| <i>Il-6</i>        | TCCAGTTGCCTTCTTGGGAC     | GTACTCCAGAAGACCAGAGG      |
| <i>Adiponectin</i> | CCTGGAGAGAAGGGAGAGAAA    | GTTCCCAATGTACCCATTCG      |
| <i>Leptin</i>      | CCGTGGCTTTGGTCCATCTG     | AGGCAAGCTGGTGAGGATCTG     |
| <i>Pgc1α</i>       | CCCTGCCATTGTTAAGACC      | TGCTGCTGTTCTGTTTTTC       |
| <i>Pgc1β</i>       | TGCGGAGACACAGATGAAGA     | GGCTTGTATGGAGGTGTGGT      |
| <i>Ppar</i>        | GCCCTTTGGTGACTTTATGG     | CAGCTGGCGACATACAGTAC      |
| <i>C/Ebpa</i>      | AGCTTACAACAGGCCAGGTTT    | CGGCTGGCCGACATACAGTAC     |
| <i>Dlk1/Pref1</i>  | GACCCACCCTGTGACCCC       | CAGGCAGCTCGTGCACCCC       |
| <i>Fabp4/aP2</i>   | CCCTGCGTGATCAATGGT       | CACAGAAGTTGCCTGAGAAGC     |
| <i>Slc2a1</i>      | TCAACACGGCCTTCACTG       | CACGATGCTCAGATAGGACAT     |
| <i>Slc2a2</i>      | TCTTCACGGCTGTCTCTGTG     | AATCATCCCCTTAGGAACA       |
| <i>Slc2a3</i>      | ATGGCAGTGGCTGGTTGTTC     | TCCTGAGCTGAAGAGAATGTC C   |
| <i>Slc2a4</i>      | GGTTGTCTTGACCCCTCCAG     | TTCGGGTTTAGCACCCCTCC      |
| <i>Slc2a6</i>      | TTGGTGCTGTGAGGCT         | TGGCACAAACTGGACGTA        |
| <i>Slc2a8</i>      | GCTCCTCATGTGAGAGATCTT C  | GGCTGTGATTTGTTCCAGAGTC    |
| <i>Slc2a9</i>      | TGCTTCTCGTCTTCGCCACAATA  | CTCTTGGCAAATGCCTGGCTG ATT |
| <i>Slc2a10</i>     | CCTGGCTTTCATCTACCTACTTGT | ATGGTACTGAATACCGATGCGC    |
| <i>Slc2a12</i>     | TGAGTTTGTCTACACGCTCCTT   | GGCAATTTCCGCAATGTACA      |
| <i>Slc23a1</i>     | CAGCAGGGACTTCCACCA       | CCACACAGGTGAAGATGGTA      |
| <i>Slc23a2</i>     | CAGCAGGGACTTCCACCA       | CATCTGTGCGTGCATAGTAGC     |
| <b>ChIP-PCR</b>    |                          |                           |
| <i>Pparg</i>       | CCTGAATTTACCCGAGCTGA     | TTATCTCGGAGGCGGTAAGA      |
| <i>Cebpa</i>       | ACTGGCGCCTTCGATCCGAGA    | AGCTTCGGGTGCGGAATGGC      |

### Antibodies used for western blotting and immunohistochemistry

#### Antibodies

| Secondary antibody               | Anti   | Cat. #     | Company       | WB       | IHC   |
|----------------------------------|--------|------------|---------------|----------|-------|
| Anti-Mouse IgG (Fc Specific)     | mouse  | A0168      | Sigma-Aldrich | 1:5,000  |       |
| Anti-Rabbit IgG (whole molecule) | rabbit | A0545      | Sigma-Aldrich | 1:10,000 | 1:100 |
| Primary antibody                 | Source | Cat        | Company       |          |       |
| Beta-actin                       | rabbit | GTX109639  | Genetex       | 1:10,000 |       |
| DLK1                             | rabbit | 10636-1-AP | Proteintech   | 1:500    |       |
| FABP4                            | rabbit | 12802-1-AP | Proteintech   | 1:7,000  |       |
| C/EBPα                           | rabbit | sc-61      | Santa Cruz    | 1:400    |       |
| PPARγ                            | rabbit | sc-7196    | Santa Cruz    | 1:800    |       |
| Collagen I                       | rabbit | ab34710    | Abcam,        | 1:1,000  |       |

|            |        |         |           |         |      |
|------------|--------|---------|-----------|---------|------|
| CD68       | rabbit | A13286  | Abclonal, | 1:50    | 1:50 |
| Collagen I | rabbit | ab34710 | Abcam,    | 1:1,000 |      |

**A position map is shown for the Proteome Profiler Mouse Adipokine Array kit from R&D Systems.**

|   |               |         |         |         |         |         |          |        |        |          |        |             |    |    |    |    |    |    |    |    |    |    |    |    |
|---|---------------|---------|---------|---------|---------|---------|----------|--------|--------|----------|--------|-------------|----|----|----|----|----|----|----|----|----|----|----|----|
|   | 1             | 2       | 3       | 4       | 5       | 6       | 7        | 8      | 9      | 10       | 11     | 12          | 13 | 14 | 15 | 16 | 17 | 18 | 19 | 20 | 21 | 22 | 23 | 24 |
| A | PC            |         |         |         |         |         |          |        |        |          |        |             |    |    |    |    |    |    |    |    |    |    | PC |    |
| B | Adiponectin   | AgRP    | ANGPTL3 | CRP     | CD26    | ESM-1   | Fetuin A | FGF-1  | FGF-21 | HGF      | ICAM-1 | IGF-I       |    |    |    |    |    |    |    |    |    |    |    |    |
| C | IGF-II        | IGFBP-1 | IGFBP-2 | IGFBP-3 | IGFBP-5 | IGFBP-6 | IL-6     | IL-10  | IL-11  | Leptin   | LIF    | Lipocalin-2 |    |    |    |    |    |    |    |    |    |    |    |    |
| D | MCP-1         | CSF-1   | OSM     | PTX 2   | PTX 3   | Pref-1  | RAGE     | RANTES | RBP4   | Resistin | PAI-1  | TIMP-1      |    |    |    |    |    |    |    |    |    |    |    |    |
| E | TNF- $\alpha$ | VEGF-A  |         |         |         |         |          |        |        |          |        |             |    |    |    |    |    |    |    |    |    |    |    |    |
| F | PC            |         |         |         |         |         |          |        |        |          |        |             |    |    |    |    |    |    |    |    |    |    | NC |    |

The adipokine signals with observable changes in Fig. 3I are shaded grey in the table.

## References

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