## **Supplementary Materials**

## **Figure description**

**Figure S1. Representative Western blots showing UCP3 antibody specificity.** 25 µg of total protein from skeletal muscles (SkM) of UCP3 knockout mice (KO) and their wild type controls (wt) were loaded per lane. Recombinant UCP3 (5 ng) was used as a positive control (A). Silver staining visualizes recombinant proteins mUCP1, mUCP2, mUCP3, mUCP4-eGFP and mUCP5 (150 ng from each) loaded as a cross-reactivity control (B). GAPDH was used as a loading control for SkM. Eight mice per group were tested.

Figure S2. Representative Western blots and quantification showing UCP2 expression in UCP1<sup>-/-</sup> and UCP3<sup>-/-</sup> mice. No compensatory expression of UCP2 in BAT (A), spleen and thymus (B) of UCP1<sup>-/-</sup> mice and in skeletal muscles, heart, BAT (C), thymus and spleen (D) of UCP3<sup>-/-</sup> mice. The relative amount is the intensity ratio between the standard (recombinant mUCP2, 5 ng) and the sample. 25  $\mu$ g of total protein were loaded per lane. Data are presented as a mean values from three mice  $\pm$  SD.

Figure S3. Representative Western blots and quantification showing UCP1 and UCP3 expression in UCP2<sup>-/-</sup> mice. (A) Lack of compensatory UCP1 expression in BAT of UCP2<sup>-/-</sup> and wt mice. (B) Lack of significant UCP3 up-regulation in BAT, skeletal muscles, heart. Data are presented as mean values from four mice  $\pm$  SD. 25 µg of total protein were loaded per lane. The relative amount was calculated as an intensity ratio between the protein standard (recombinant mUCP1 or mUCP3) and the intensity of samples from wt or KO mice.



## Figure S1. Hilse et al. (2015)



Figure S2. Hilse et al. (2015)



Α

1.2

1.0

0.8

0.6

0.4

0.2

0.0

UCP1 amount, r.u.







SkM

• ко

⊐ wt

Т

