

Supplemental Information

Estradiol Regulates Energy Balance

by Ameliorating Hypothalamic

Ceramide-Induced ER Stress

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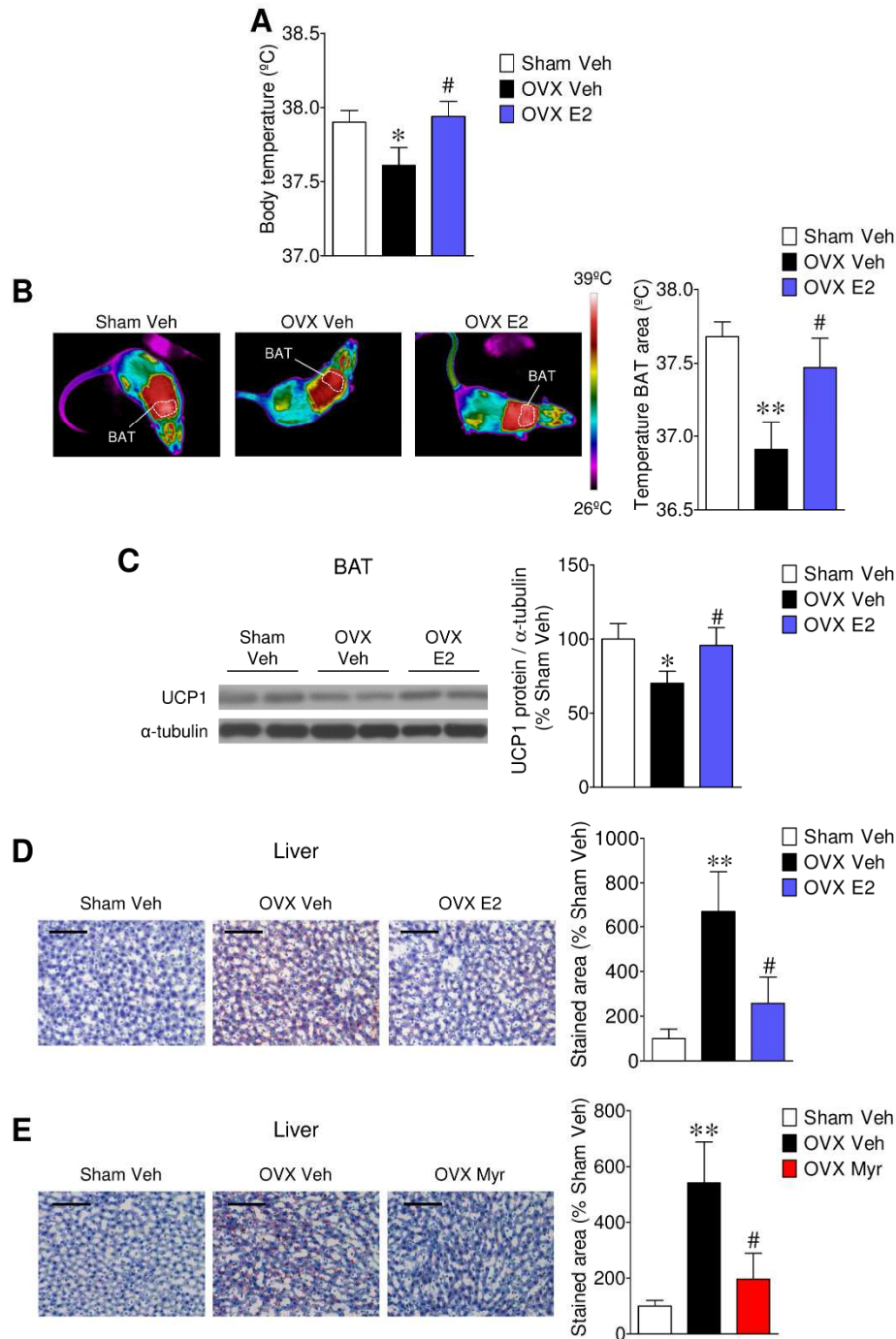


FIGURE S1 related to Figure 1. Effect of central E2 and myriocin on BAT thermogenesis and liver steatosis in OVX rats

(A) Body temperature, (B) representative infrared thermal images (left panel) and temperature of BAT area (right panel) and (C) representative western blot autoradiographic images (left panel) and protein levels of UCP1 in the BAT (right panel) and (D) representative Oil Red O images (left panel; 20X; scale bar: 100 μ m), and staining analysis (right panel) of Sham rats or OVX rats ICV treated with vehicle or E2 (n=7-9 animals per group). (E) Representative Oil Red O images (left panel; 20X; scale bar: 100 μ m) and staining analysis (right panel) of Sham rats or OVX rats ICV treated with vehicle or myriocin (n=7-10 animals per group). All data are expressed as mean $\pm$ SEM. * and ** P<0.05 and 0.01 vs. Sham vehicle; # P<0.05 vs. OVX vehicle. For the western blot analyses, representative images for all proteins are shown; in the case of the loading controls a representative gel is displayed for clarity, although each band of each protein was always corrected by its own internal control band (α -tubulin). The bands for each picture come always from the same gel, although they have been spliced for clarity.

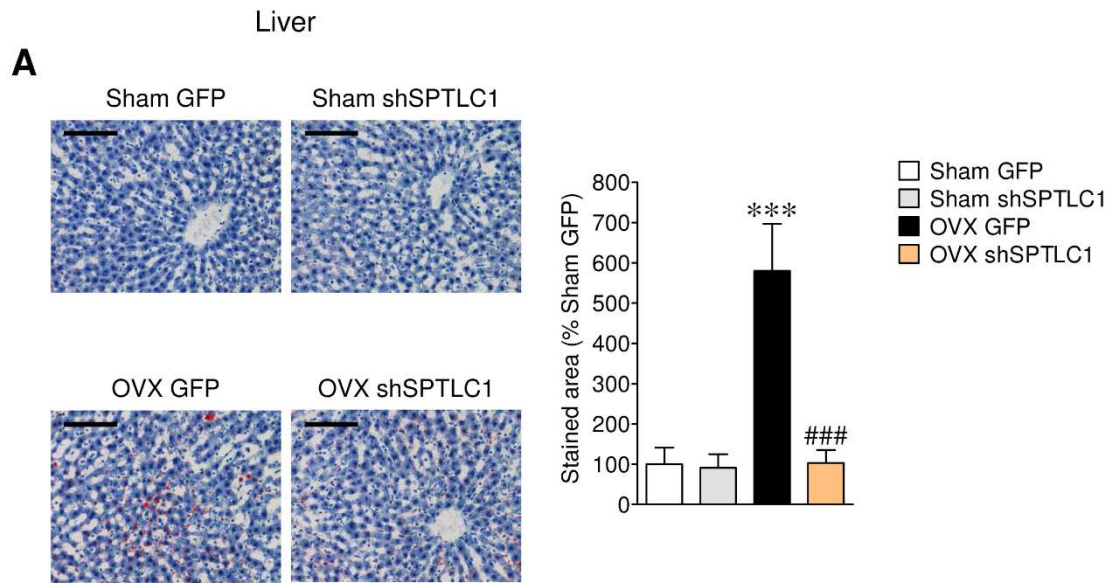


FIGURE S2 related to Figures 3 and 4. Effect of SPTLC1 downregulation in the VMH of OVX rats on liver steatosis

(A) Representative Oil Red O images (left panel; 20X; scale bar: 100 μ m) and staining analysis (right panel) of Sham or OVX rats stereotactically treated in the VMH with adenoviruses encoding GFP or shSPTLC1 (n=7-8 animals per group). All data are expressed as mean $\pm$ SEM. *** P<0.001 vs. Sham GFP; ### P<0.001 vs. OVX GFP.

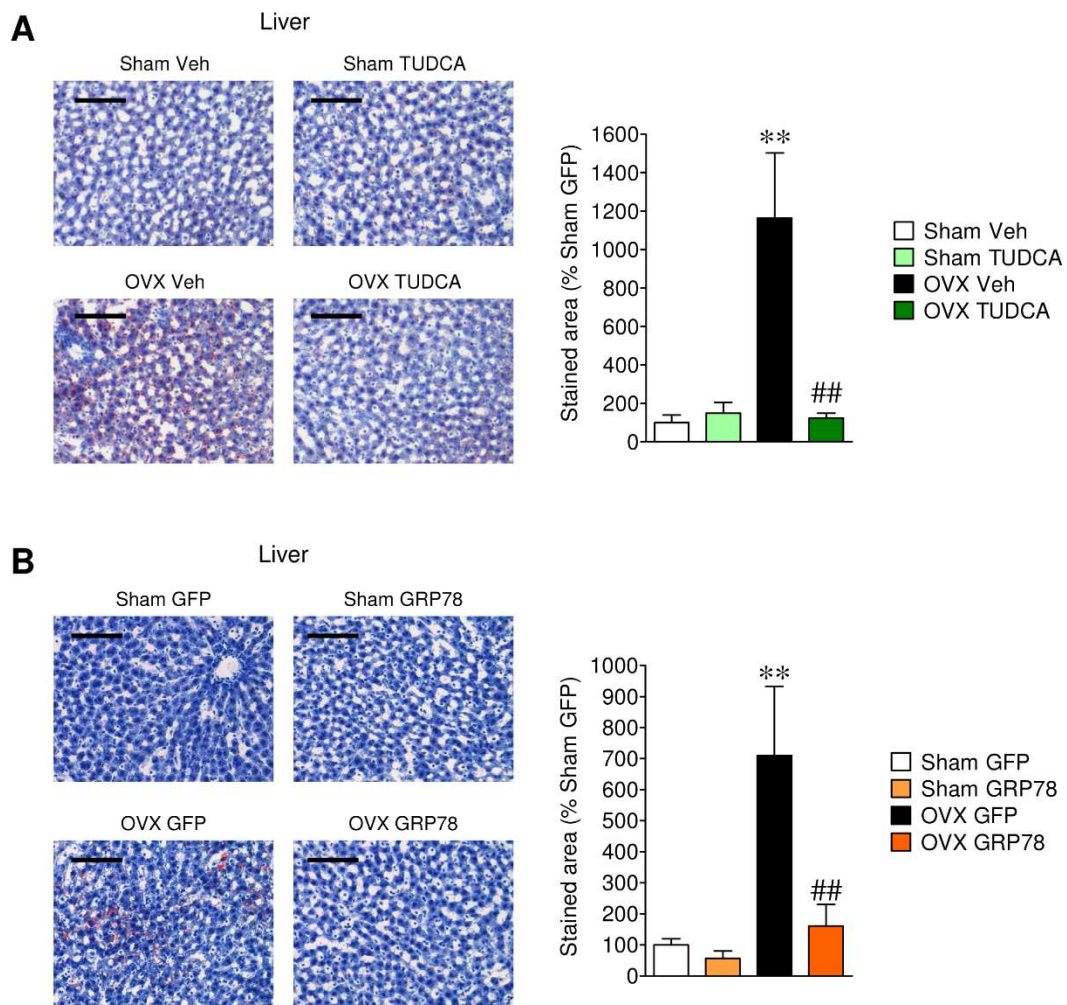


FIGURE S3 related to Figures 5 and 6. Effect of central TUDCA or GRP78 overexpression in the VMH of OVX rats on liver steatosis

(A) Representative Oil Red O images (left panel; 20X; scale bar: 100 μ m) and staining analysis (right panel) of Sham or OVX rats ICV treated with vehicle or TUDCA (n=8 animals per group). (B) Representative Oil Red O images (left panel; 20X; scale bar: 100 μ m), staining analysis (right panel) of Sham or OVX rats stereotactically treated in the VMH with adenoviruses encoding GFP or GRP78 (n=7-9 animals per group). All data are expressed as mean $\pm$ SEM. ** P<0.01 vs. Sham vehicle or Sham GFP. ## P<0.01 vs. OVX vehicle or OVX GFP.