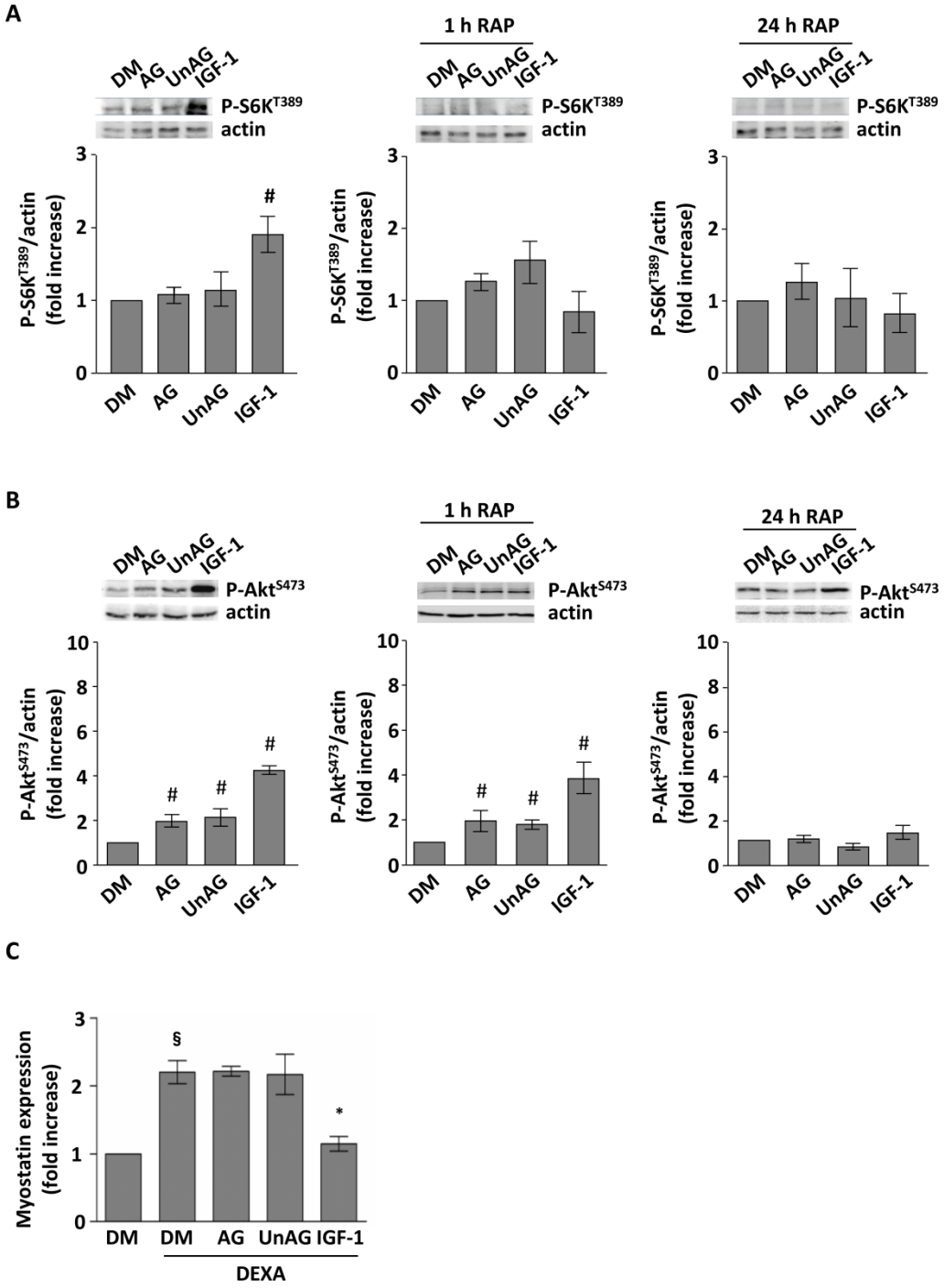

Figure S1.

Long rapamycin treatment impairs both mTORC1 and mTORC2 activities in C2C12 myotubes. (**A**-**B**) Phosphorylation of S6K^{T389} and Akt^{S473} detected by western blotting upon treatment for 20 min with 1 μM AG or UnAG. Upper panels: representative blots; lower panels: quantifications of 3 independent experiments. Rapamycin (RAP) 20 ng/ml was used for 1 h or 24 h. After short rapamycin treatment (1 h), mTORC1-mediated phosphorylation of S6K^{T389} is completely abolished (**A**), while the activity of mTORC2-mediated phosphorylation of Akt^{S473} is spared (**B**). On the other hand, upon 24 h of rapamycin treatment, Akt^{S473} phosphorylation is also abrogated, indicating that long rapamycin treatments affect mTORC2 activity as well as mTORC1 in C2C12 myotubes. #$P < 0.05$ versus control cells in DM.

(**C**) AG and UnAG do not affect dexamethasone (DEXA)-induced myostatin expression, measured by real-time RT-PCR. C2C12 myotubes were treated in DM for 24 h with 1 μM DEXA in the presence or absence of 10 nM AG or UnAG, or 10 ng/ml IGF-1 and processed for myostatin expression analysis. §$P < 0.01$ versus control cells in DM; *$P < 0.01$ versus DEXA-treated cells.

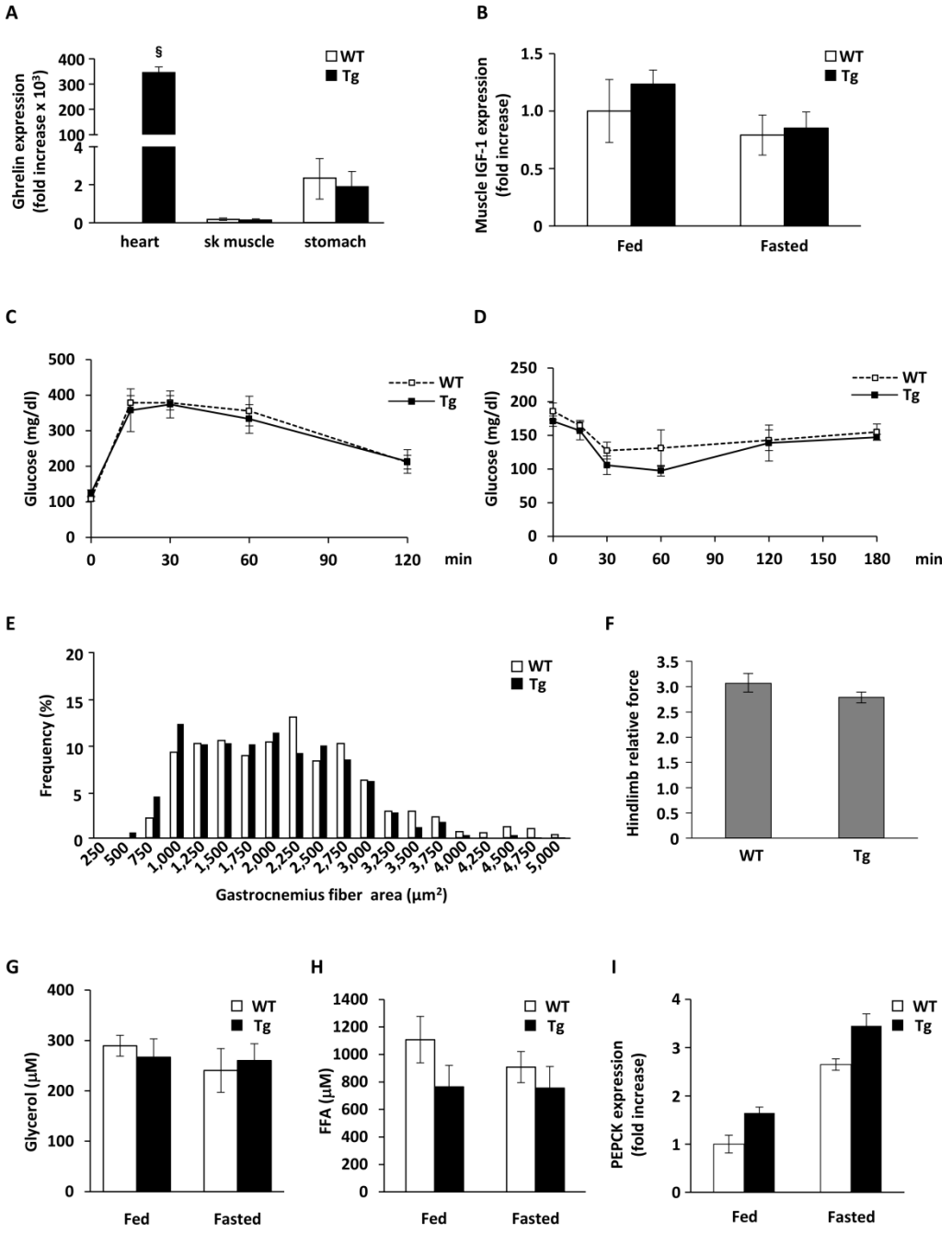

Figure S2.

(**A**) Ghrelin expression is specifically induced in the hearts of *Myh6/Ghrl* mice without any leaky expression in skeletal muscle. The expression in the stomach, the main source of AG and UnAG, is not altered in *Myh6/Ghrl* animals. §$P < 0.01$ versus WT mice. (**B**) IGF-1 expression in skeletal muscle is not altered in *Myh6/Ghrl* mice, either fed or starved animals (fed WT and Tg, n = 5; starved WT, n = 5; staved Tg, n = 6). (**C** and **D**) *Myh6/Ghrl* mice do not feature significant differences in glucose uptake (**C**) and insulin resistance (**D**) compared to WT littermates. For the glucose tolerance test, mice were injected after 16 h of fasting (WT and Tg, n = 5). (**E** and **F**) *Myh6/Ghrl* mice do not feature significant differences in muscle-fiber distribution and force compared to WT littermates. (**E**) Gastrocnemii were removed from fed animals and mean fiber CSA and distribution were analyzed (WT and Tg, n = 3). Mean gastrocnemius CSA of fed animals ± SEM (μm²) WT: 2,165.65 ± 290.85; Tg: 1,871.15 ± 100.86. (**F**) The weight that WT and Tg animals managed to hold up before losing grip was measured through a Grip Meter device and normalized to the weight of animals (WT and Tg, n = 5). (**G-I**) *Myh6/Ghrl* mice do not feature significant differences in plasmatic glycerol (**G**) and FFA (**H**), nor in liver PEPCK expression (**I**) compared to WT animals, in either fed or starved animals (n = 5 for each group).

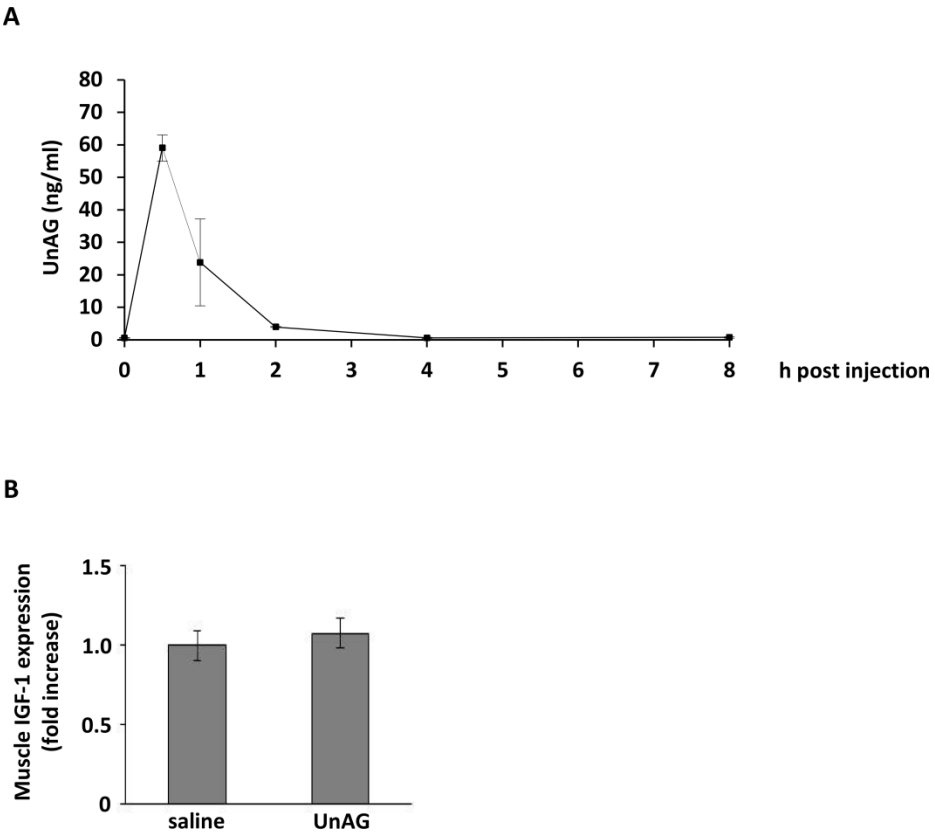

Figure S3.
(**A**) UnAG plasma concentration upon UnAG treatment. WT mice (n = 2) were injected s.c. with 100 µg/kg UnAG and, at the indicated time points, blood samples were collected by retro-orbital puncture and processed for EIA determination of plasmatic UnAG concentration. Each sample was loaded in triplicates and mean values ± SD are represented. (**B**) IGF-1 expression in skeletal muscle is not altered after UnAG injection compared to saline treatment (n = 5 for each group).