

## SUPPLEMENTAL INFORMATION

### FIGURE LEGENDS

#### Figure S1. Generation of Ghrl-DTR Mice (related to Experimental Procedures)

(A) Schematic of cross between Ghrl-Cre and iDTR alleles to generate active diphtheria toxin receptor (DTR) molecules on ghrelin cells. (B) Chow-fed control and Ghrl-DTR (male, 8-week old) were injected with 8-10 ng/g body weight of DTX. Mice were sacrificed 3 days after injection. Blood was drawn from the interior vena cava. Plasma ghrelin and des-acyl ghrelin levels were measured as described in Experimental Procedures. Stomach, duodenum, liver and kidney were harvested, mRNA was isolated from these tissues, and quantitative RT-PCR analysis of Cre, DTR, and ghrelin were performed on mRNAs from the indicated tissue. The number above each bar represents the mean  $C_t$  value from 3-4 mice. *Hbegf*, DTR gene; *Neo*, neomycin resistance gene; pA, polyadenylation signal; STOP, transcriptional stop cassette; SA, splice acceptor; FRT, flip recombinase site; loxP, cre recombinase site.

#### Figure S2. Circadian Variation of Ghrelin and des-Acyl Ghrelin in Wild Type Mice (related to Figure 1)

Chow-fed wild type mice (male, 8 weeks old; C57BL/6J and DBA mixed background) were bled from the tail vein every 4 hr for one 24-hr period. Plasma ghrelin and des-acyl ghrelin were measured by ELISA. Bars represent mean $\pm$ SEM of values from 6 mice.

#### Figure S3. Plasma Ghrelin Levels in Mice Two Days after Injection of DTX (related to Figure 6)

Chow-fed control and Ghrl-DTR littermates (male, 8-12 weeks old; same animals used in Figure 6) were housed individually and injected with DTX on day 0. On day 2, blood was drawn from

the tail vein, and plasma ghrelin and des-acyl ghrelin were measured to verify ablation in Ghrl-DTR mice. Each value represents mean $\pm$ SEM of 20 mice. Each group of 20 was randomly divided into four dosing groups (n=5 mice per group) and then used for the study in Figure 6.

**Figure S4. Plasma GH Levels in Mice 10 min after Injection of Ghrelin (related to Figure 6)**

Two weeks after the experiment in Figure 6, the same chow-fed control and Ghrl-DTR littermates were injected subcutaneously at 2 p.m. with the indicated doses of ghrelin. Ten minutes after injection, mice were anesthetized with isoflurane and blood was drawn by retro-orbital bleed, after which levels of GH were measured by ELISA. Each value represents mean $\pm$ SEM of 5 mice.

**Figure S1**

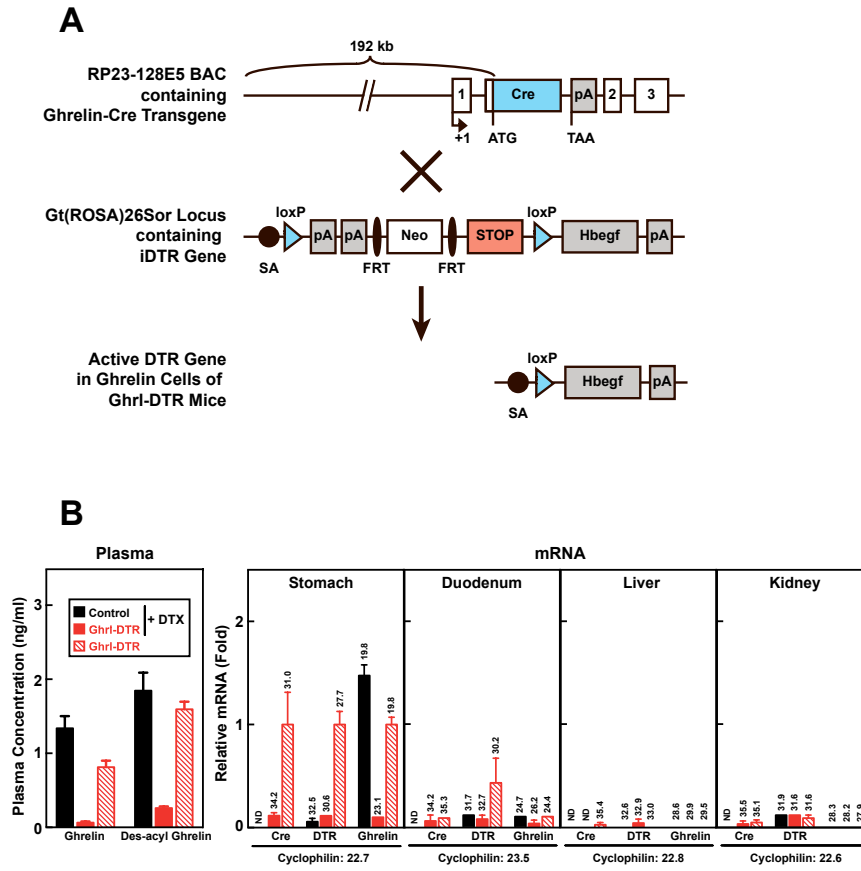


Figure S2

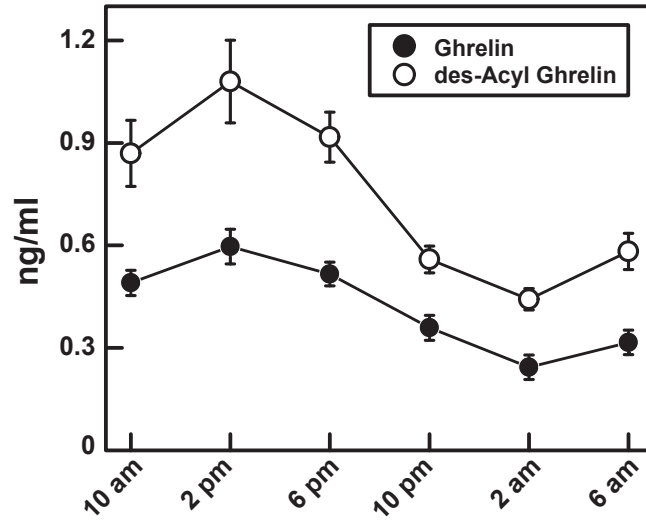


Figure S3

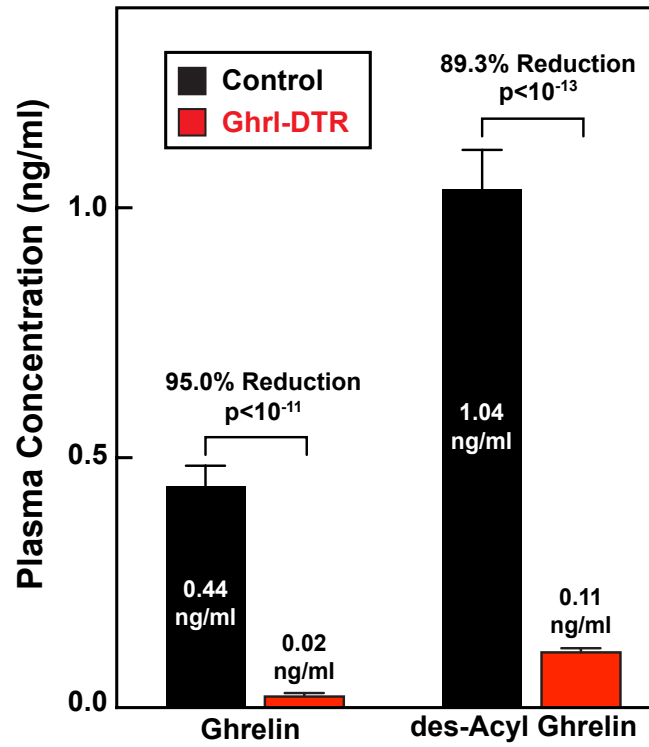


Figure S4

