## Supplemental Methods

### Slice electrophysiology of leptin effects on AIC pyramidal neurons

To study the effects of leptin on insula pyramidal neurons, we initially studied experimentally-naïve, chow-fed female Wistar rats (*n*=6 rats) to determine an effective leptin concentration and A-P range of AIC to target. These rats received stereotactic infusion of AAV5-CaMKIIa-eYFP to facilitate identification of CaMKIIa-expressing pyramidal neurons ($7.4x10^{12}$ vg/ml dissolved in 350 mM NaCl + 5% Sorbitol in 1x PBS) (UNC Chapel Hill Viral Vector Core) into the AIC (AP +2.8, ML +/- 4.8 from bregma; from dura DV -4.6 mm; with a 10-min post-injection wait before injector removal). Following at least 2 weeks for surgical recovery and transduction, slices were prepared as described above, and rat leptin (60 nM) (R&D Biosystems, Minneapolis, Minn., USA) was bath applied in aCSF for 7 min to investigate its functional effect on insular cortex neurons with and without Blockers. To explore effects of acute energy state, subsets of females were sacrificed in fed vs. 24-hr fasted states (*n*=3 rats/group). To investigate whether effects of leptin were uniform across the A-P range of the AIC, responses of cells (*n*=19) were compared from 0 to +3.5 mm from bregma.

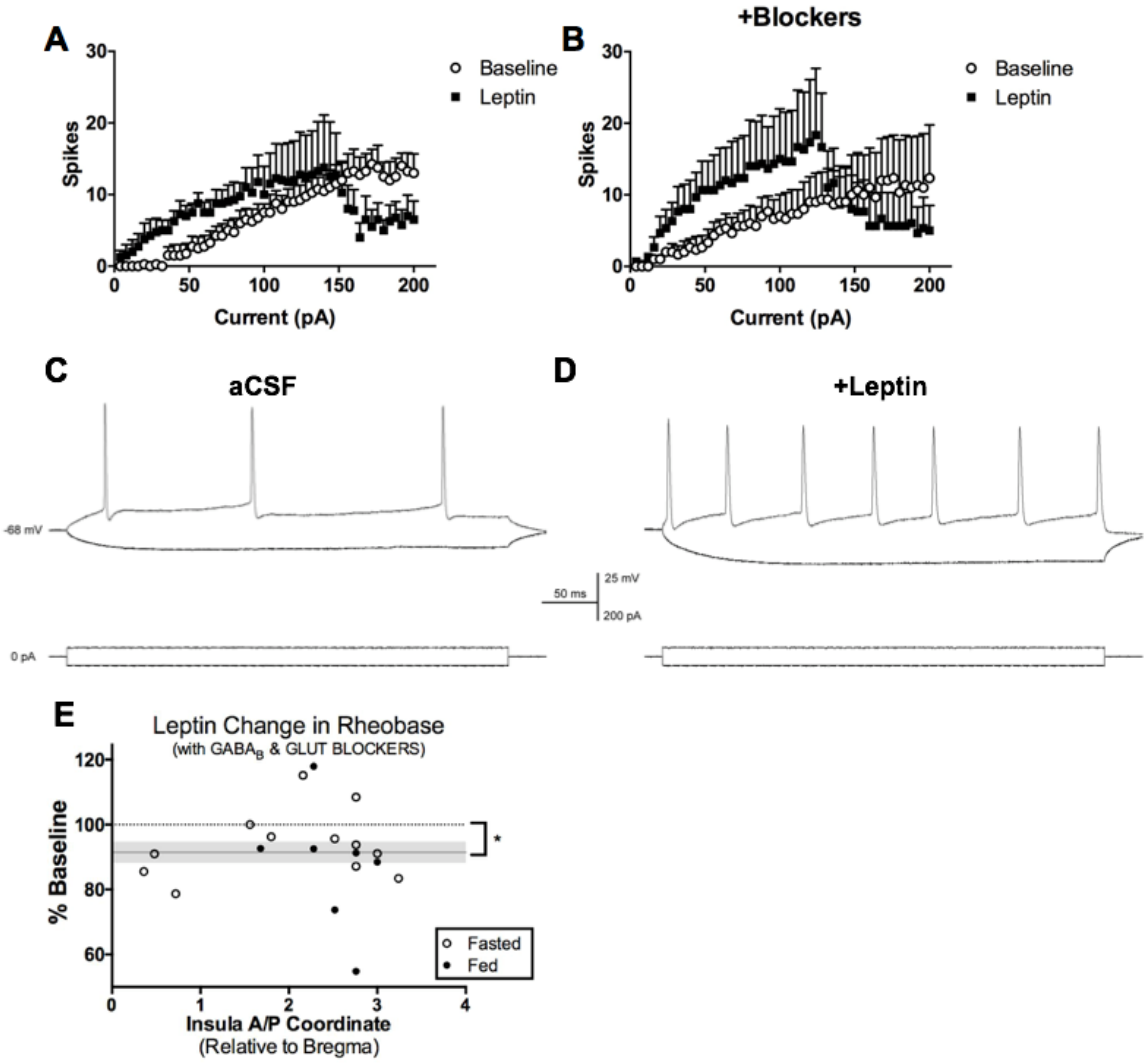
## Supplemental Results

### Functional effects of leptin in the insular cortex

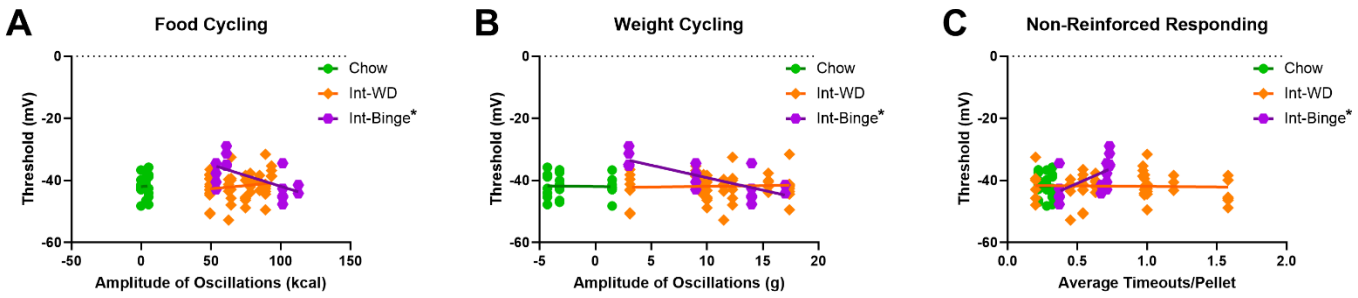
In whole-cell current clamp recordings from layer V pyramidal AIC neurons of diet-schedule naïve rats, bath application of leptin in aCSF increased AP spiking frequency (Treatment X Injected Current interaction $p<0.0001$, *n*=4 cells/condition) (Fig S1A). Despite low activity in pyramidal neurons, leptin was sufficient to evoke this effect when $GABA_A$ mediated inhibitory neurotransmission was isolated by applying Blockers (Treatment X Current interaction $p<0.017$,

*n*=3 cells/condition) (Fig S1B). Representative traces showing these effects are illustrated for an AIC neuron at baseline (aCSF) and with leptin applied (Fig S1C&D, respectively). Further, leptin reduced mean rheobase from baseline in the presence of Blockers ($t(18)=2.63$, $p<0.02$) (Fig. S1E). This action was seen regardless of fed vs. fasted energy state or A/P position within the agranular insula.

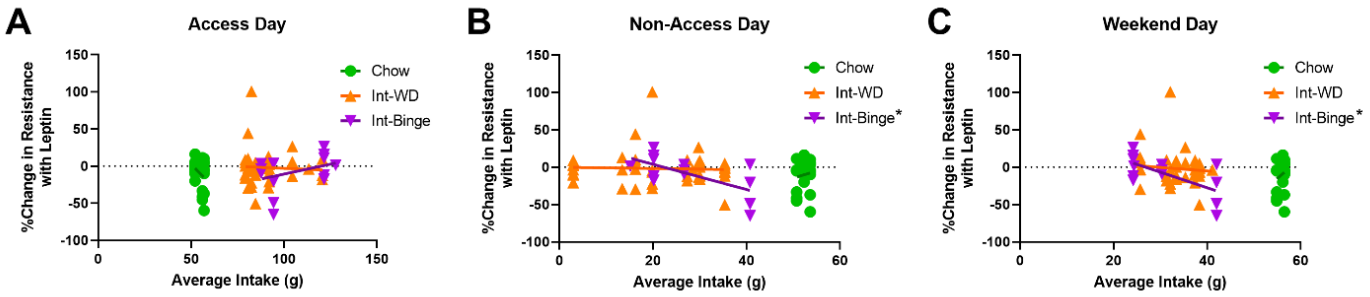
**Supplemental Figure 1: Effect of leptin in pyramidal neurons in the insular cortex.** A) In females, application of leptin (60 nM) increases spiking frequency of AAV-CamKII-GFP-labeled pyramidal neurons, B) in what may be a $GABA_A$-dependent manner. Data shown are *M* + SEM. This effect is depicted for a representative trace, where C) shows spiking in aCSF alone and D) is following the superfusion of leptin. E) In females, regardless of energy state, and A/P position within the anterior insula, leptin reduced mean rheobase (the minimum current required to generate an action potential) from baseline. $M \pm SEM$ denoted by shading. *$p$<0.05.



**Supplemental Figure 2: AIC pyramidal neuron action potential threshold correlates with feeding behaviors in rats that exhibited binge intake during intermittent access to palatable foods.** Graphs show correlation between AIC pyramidal neuron action potential threshold and A) the amplitude of cyclic oscillations in food intake, B) amplitude of cyclic oscillations in weight, and C) persistent responding during non-reinforced timeout periods, for chow fed rats and intermittent access rats that were sacrificed 24 hours after their last access session (Int-WD) or immediately after their last access session (Int-Binge). There were significant correlations of threshold to cyclic amplitudes in food intake ($r$= -0.61), weight ($r$= -0.42), and to non-reinforced persistent responding ($r$= 0.58) for the Int-Binge group, such that rats with a higher amplitude of cyclic oscillations and more persistent responding had more depolarized action potential thresholds. *$p$<0.05.



**Supplemental Figure 3: Leptin has less effect on AIC pyramidal neuron resistance in rats with binge intake during intermittent access to palatable foods.** Graphs show correlation between average intake, on A) intermittent access days, B) non-access days, and C) weekends, and the magnitude of the leptin effect on neuron resistance, for chow fed rats and intermittent access rats that were sacrificed 24 hours after their last access session (Int-WD) or immediately after their last access session (Int-Binge). There were significant correlations of the effect of leptin on neuron resistance to average intake on non-access days ($r$= -0.65) and weekends ($r$= -0.66) for the Int-Binge group, such that rats with a higher binge intake on access days and less consumption on other days had AIC neurons that were less sensitive to leptin. *$p$<0.05.



**Supplemental Table 1**

Electrophysiology measures of AIC neurons from CHOW and PREF diet rats under Baseline conditions and in the presence of Blockers and Blockers+Leptin.

| **Neuronal Measure** | **Baseline** | | **Blockers** | | **Blockers+Leptin** | |
|---|---|---|---|---|---|---|
| | CHOW | PREF | CHOW | PREF | CHOW | PREF |
| Resistance (MΩ) | 236.6 ±20.5 | 188.6 ±14.3 | 238.7 ±20.5 | 207.5 ±17.5 | 219.0 ±22.9 | 204.6 ±20.4 |
| Rheobase (pA) | 88.7 ±9.5 | 91.5 ±7.0 | 89.8 ±9.8 | 109.6 ±13.4 | 95.4 ±12.2 | 109.6 ±13.4 |
| AP Threshold (mV) | -41.9 ±0.8 | -43.1±0.5 | -40.9 ±0.9 | -40.5 ±0.6 | -40.1 ±1.2 | -38.9 ±1.2 |
| AP Amplitude (mV) | ***87.0 ±2.0** | ***92.0 ±1.3** | 84.2 ±2.5 | 87.7 ±1.8 | 80.3 ±3.2 | 87.2 ±1.3 |

Data are presented as *mean* ±*SEM*. **p* < 0.05, CHOW vs PREF.