Supplemental Tables and Figures

Supplemental Table 1. Functional Activity (EC50 in cAMP Assay) of PYY and GIP peptides at their respective mouse receptors.

Supplemental Table 2. Summary of the mean pharmacokinetic parameters following a single subcutaneous (SC) dose of LAGIPRA or LAPYY to male CD-1 mice.

Supplemental Figure 1: Analog structures. Structure long-acting GIPR agonist (A, LAGIPRA), long-acting PYY analog (B, LAPYY) and short-acting GIPR agonist (C, GIPRA). Compound 4 in Osterbaard et al., WO 2016/198682 and Sequence ID Numbers 354 and 123 in Taiji et al., WO 2018/181864.

Supplemental Figure 2. *In vivo* characterization of GIPR and Y2R agonists in mice. Wild-type (WT) and Lean wild-type (C57BL/6J, A and E) and germline, whole-body GIPR mice ($Gipr^{-/-}$, B and F) dosed subcutaneously with vehicle, a long (A, 100 nmol/kg or 300 nmol/kg) or short-acting (E, 3 nmol/kg or 30 nmol/kg) glucose-dependent insulinotropic polypeptide receptor agonist 15-min (short-acting analogs) or 16-hours (long-acting agents) prior to an ipGTT, n=5 per group. Twenty-four-hour body weight and food intake in lean WT mice dose with a long-acting PYY analog (C and D). Data are presented as mean ± SEM. P<0.05* compared to vehicle. Statistical analyses performed included one-way ANOVA, followed by Dunnett's multiple comparisons test where appropriate.

Supplemental Figure 3: GIPR agonism attenuates PYY-induced aversion in mice. Saccharin preference ratio, 24-hour body weight and food intake in lean C57/Bl6 mice dosed subcutaneously (SC) with a long-acting peptide tyrosine-tyrosine analog (LAPYY) alone (A, C and E) or in combination (B, E and F) with a long-acting glucose-dependent

insulinotropic polypeptide receptor agonist (LAGIPRA), n=6 per group. Values are presented as mean ± SEM. *P<0.05 vs vehicle. $^{\$}$P<0.05 vs PYY only group. Statistical analyses performed included student unpaired t-test or one-way ANOVA, followed by Dunnett's multiple comparisons test where appropriate.

Supplemental Figure 4. GIPR agonism induced cFos expression in Npy2r+ neurons in the area postrema. Mice were subcutaneously injected with vehicle (A), LAPYY (B, 100 nmol/kg), GIPRA (C, 10nmol/kg) or combination (D, GIPRA (10 nmol/kg) + LAPYY 100 nmol/kg). Mouse brains were collected 45 minutes post injection. N=6 per group. RNA triple in situ was conducted on area postrema using probes against *cFos* (green), *Gipr* (purple) and *Npy2r* (white). Arrows in panels C and D points to Y2R+ cells expressing cFos. (E) Quantification of Y2R+ neurons that also express cFos. Scale bars: 20um. Values are presented as mean ± SEM. Statistical analyses used one-way ANOVA followed by Dunnett's multiple comparison test *p<0.05, ****p<0.0001.

Supplemental Figure 5. cFos cell count in other brain regions. Values are presented as mean ± SEM. Statistical analyses performed included one-way *ANOVA* followed by Dunnett's multiple comparison test. A) DMX: dorsal motor complex. B) ARC: arcuate nucleus of the hypothalamus. C) DHM: dorsal medial hypothalamus. D) VMH: ventral medial hypothalamus. E) PVH: paraventricular hypothalamus. F) ACB: nucleus accumbens. G) VTA: ventral tegmental area.

Supplemental Figure 6. cFos expression in PBN_D neurons induced by PYY. Mice were subcutaneously injected with vehicle, LAPYY (100nmol/kg), GIPRA (10nmol/kg) or combination (GIPRA (10 nmol/kg) + LAPYY (100 nmol/kg)). Mouse brains were collected

120 minutes post injection. Immunohistochemistry was conducted on parabrachial nucleus using antibodies against cFos (green), CGRP (purple) and FoxP2 (red). Note that many cFos+ cells in PYY injected mice are present in dorsal PBN_D and do not express FoxP2 or CGRP (arrowheads). Scale bars: 50um.