**Supplementary Table 1.** Summary table of the effects of NN1213 and sCT compared to vehicle on body weight again and food intake. → no change, ↓ decrease.

| Group                        | Body weight Gain | Food Intake |
|------------------------------|------------------|-------------|
| WT + NN1213                  | ↓↓               | ↓ →         |
| RAMP1/3 KO + NN1213          | →                | ↓           |
| RAMP3 KO + NN1213            | →                | →           |
| RAMP1 KO + NN1213            | ↓↓               | →           |
| WT + sCT                     | →                | →           |
| RAMP1/3 KO + sCT             | ↓↓               | →           |
| RAMP3 KO + sCT               | ↓↓               | →           |
| RAMP1 KO + sCT               | →                | →           |
Supplementary Figure 1. Efficacy of NN1213 compared to that of sCT in cells transfected with (A) mouse CTRb (mCTR), (B) mouse CTRb and mouse RAMP1 (mAMY1), (C) mouse CTRb and mouse RAMP2 (mAMY2) and (D) mouse CTRb and mouse RAMP3 (mAMY3). All data are expressed as mean ± SEM, N=6/group.
Supplementary Figure 2. Obesity induction period in WT and RAMP1/3, RAMP3 and RAMP1KO mice fed with 45% HFD ad libitum for 20 weeks. A-C: Weekly body weight (g) in WT, RAMP1/3KO (A), RAMP3KO (B), RAMP1KO (C) mice over 20 weeks on 45% HFD. D-F: Weekly body weight gain (g) WT, RAMP1/3KO (D), RAMP3KO (E), RAMP1KO (F) mice over 20 weeks on 45% HFD. G-H: Weekly cumulative food intake (kcal) WT, RAMP1/3KO (G), RAMP3KO (H), RAMP1KO (I) mice over 20 weeks on 45% HFD. J-K: Non-fasting blood glucose level (mmol/l) of WT, RAMP1/3KO (J), RAMP3KO (K), RAMP1KO (L) mice maintained on 45% HFD for 20 weeks measured on a bi-weekly base. All data are expressed as mean ± SEM, N=24/group. Statistics: two-way ANOVA with repeated measured followed by Sidak post-hoc comparison. Symbols denote significant differences between the two genotypes; *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001.
Supplementary Figure 3. CTR and RAMP1, RAMP2 or RAMP3 are colocalized within the same cell in the NTS and AP of WT HFD-fed neurons in mice. Representative NTS 20X, NTS 63X, AP 20X and 63X images of in situ hybridization of CTR (Calcr1A; green), RAMP1 (white), RAMP2 (red) or RAMP3 (cyan) in WT (A-D), RAMP1KO (E-H), RAMP3KO (I-L) and RAMP1/3 KO (M-P) mice. The box indicates the NTS and AP 63X magnification presented in B, F, J, L and D, H, L, P. The empty arrows indicate no colocalization with CTR and the filled arrows indicate RAMP-CTR colocalization. The overlay of RAMP1 and RAMP2 generates a white-pink signal.
Fluorescent in situ hybridization of the AP/NTS regions

Fresh frozen brains from WT, RAMP1 KO, RAMP3 KO and RAMP1/3 KO were cut in 14 µm section onto superfrost plus slides (Life Technologies). The mRNA signal of interest was detected using the RNAscope® Multiplex Fluorescent Reagent Kit v2 Assay (Cat No. 323100, Advanced Cell Diagnostics) and 4-plex ancilliary kit (Cat No. 323120). Specific probes targeting mouse CTR, RAMP1, RAMP2 or RAMP3 (Probe-Mm-Calcr, Cat No. 494071-C1; Probe-Mm-RAMP1, Cat No. 532684-C4; Probe-Mm-RAMP2, Cat No. 451811-C2, and Probe-Mm-RAMP3, Cat No. 497131-C3, Advanced Cell Diagnostics) were used according to the manufacturer’s instructions. Probes were detected using Opal reagents (Akoya Biosciences; CTR:1:500 and RAMPs 1:1500 dilution). The slides were counterstained with DAPI and coverslipped using hardset fluorescent mounting medium before being scanned using a confocal microscope (Zeiss SP8 confocal system (CTR: HD1 FITC laser 20%, RAMP1: HD1 cy3 laser 5%, RAMP2: HD1 Texas Red laser 5%, RAMP3: HD1 cy5 laser 20%, DAPI: HD2 405 laser 10%, Z-stack 10 µm, and step size 0.5 µm). The AP and NTS were scanned at 20X and 63X.