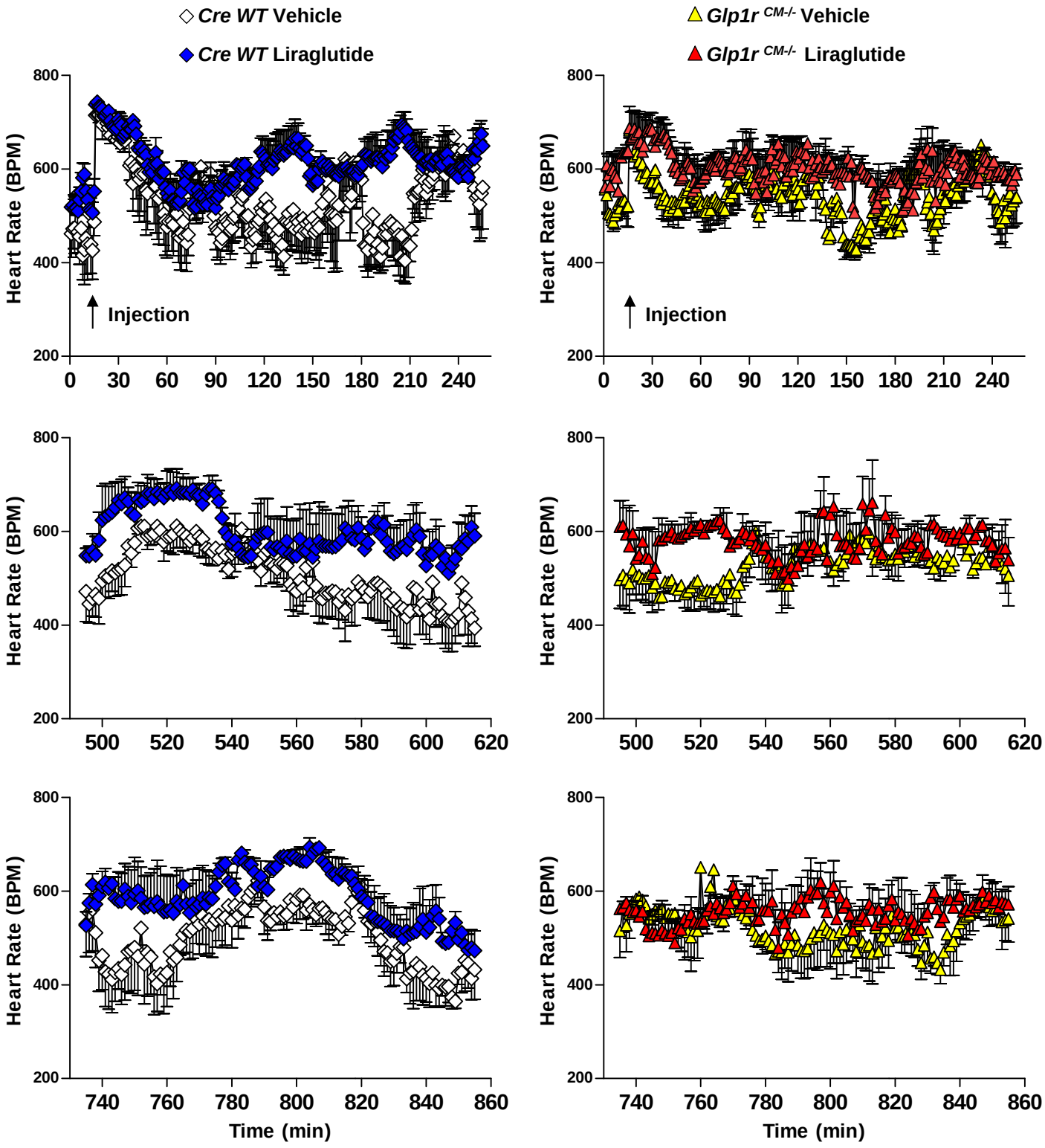
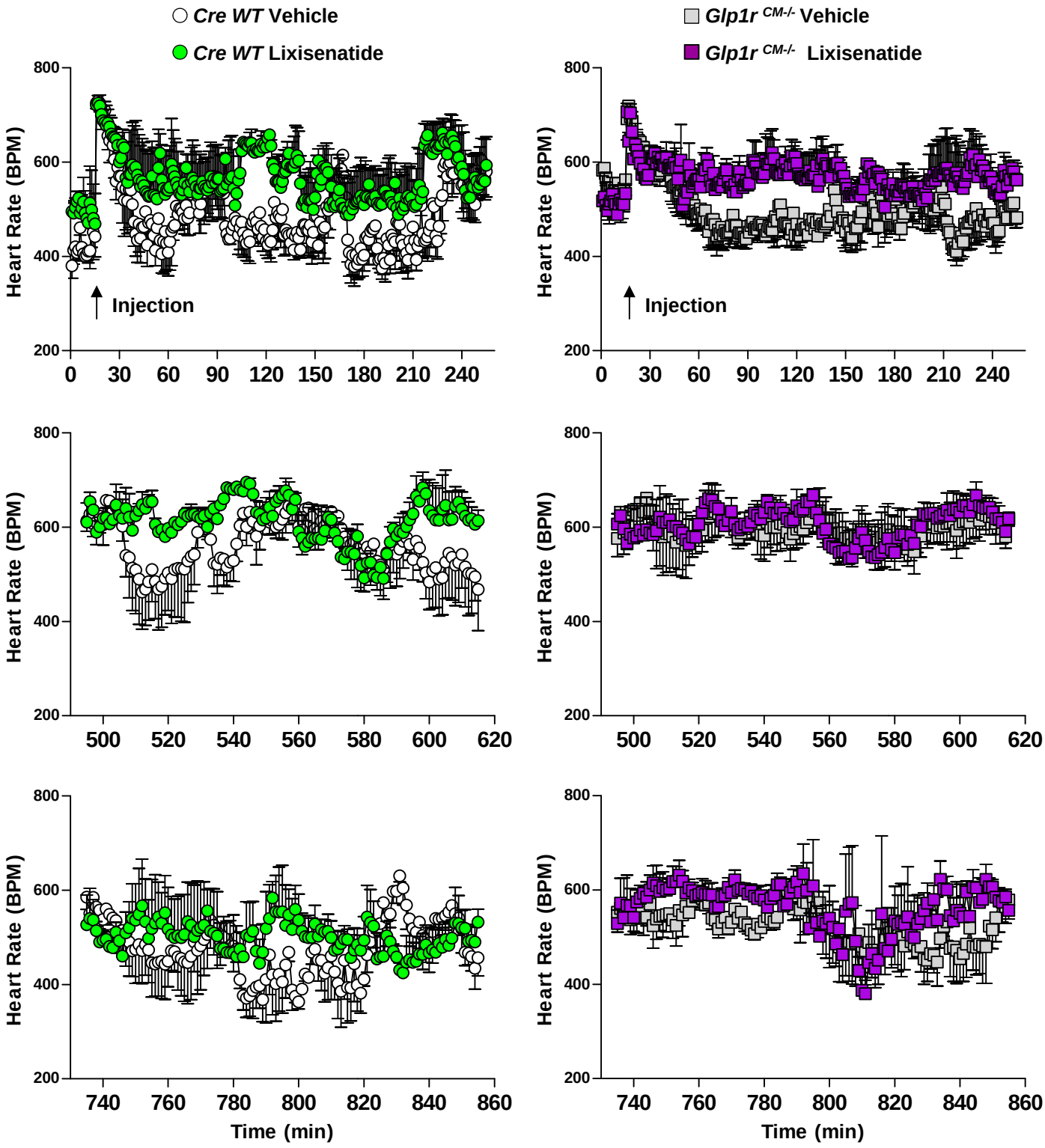
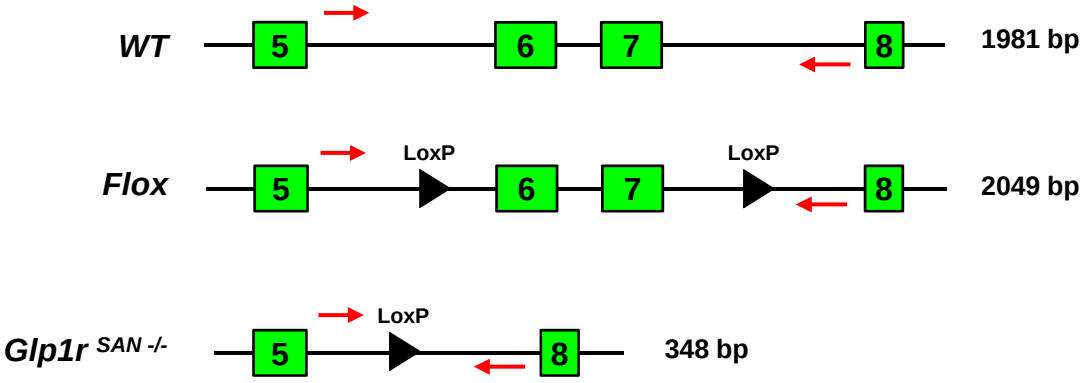

Cre WT Vehicle
Cre WT Liraglutide
Glp1r CM-/- Vehicle
Glp1r CM-/- Liraglutide
Heart Rate (BPM)
Injection
Time (min)

Sup. Figure 1

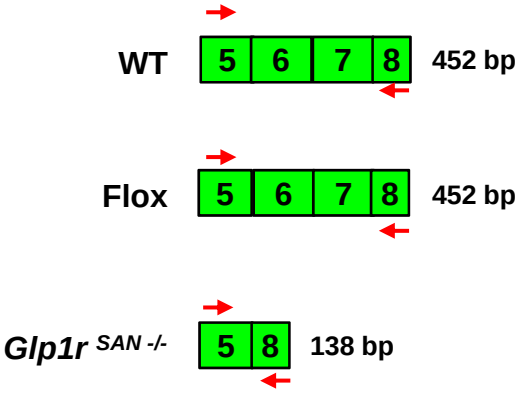

Cre WT Vehicle
Cre WT Lixisenatide
Glp1r CM-/- Vehicle
Glp1r CM-/- Lixisenatide
Heart Rate (BPM)
Injection
Time (min)

Sup. Figure 2

Glp1r Genomic DNA:

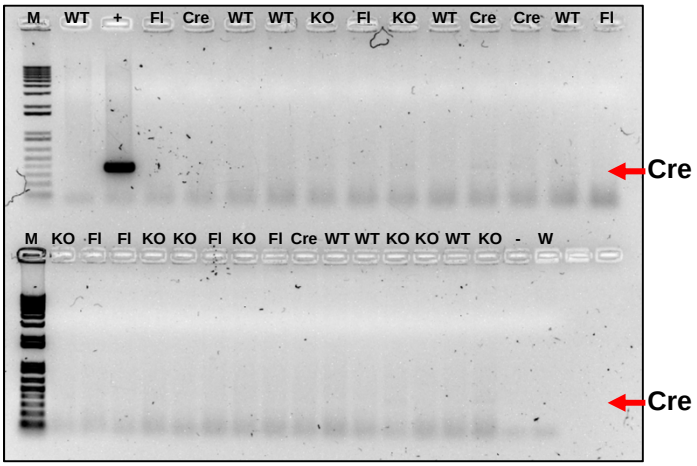


Glp1r cDNA:

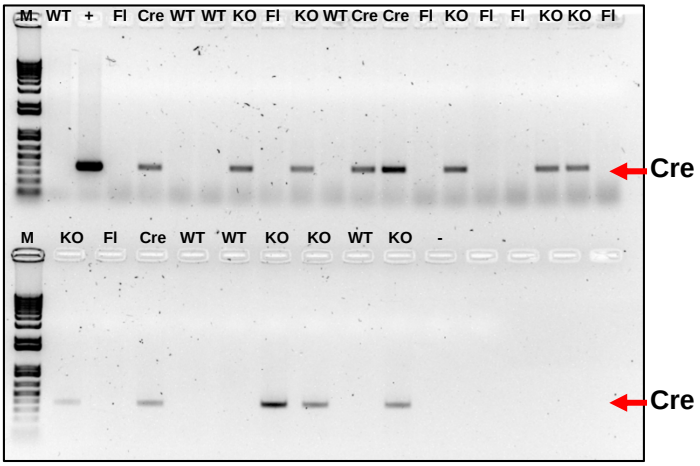


Sup. Figure 3

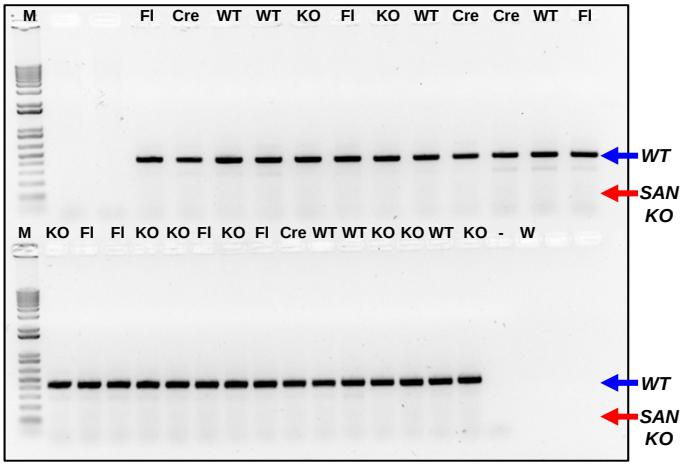
Cre mRNA Expression Left Atrium



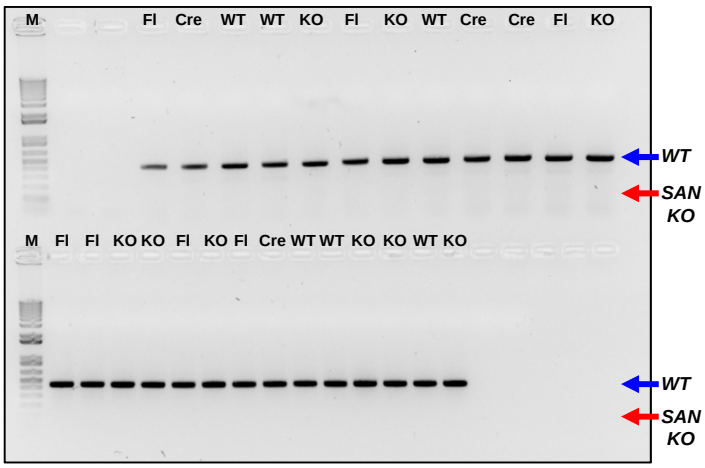
Cre mRNA Expression Right Atrium



Glp1r Exon 5-8 mRNA Expression Left Atrium



Glp1r Exon 5-8 mRNA Expression Right Atrium

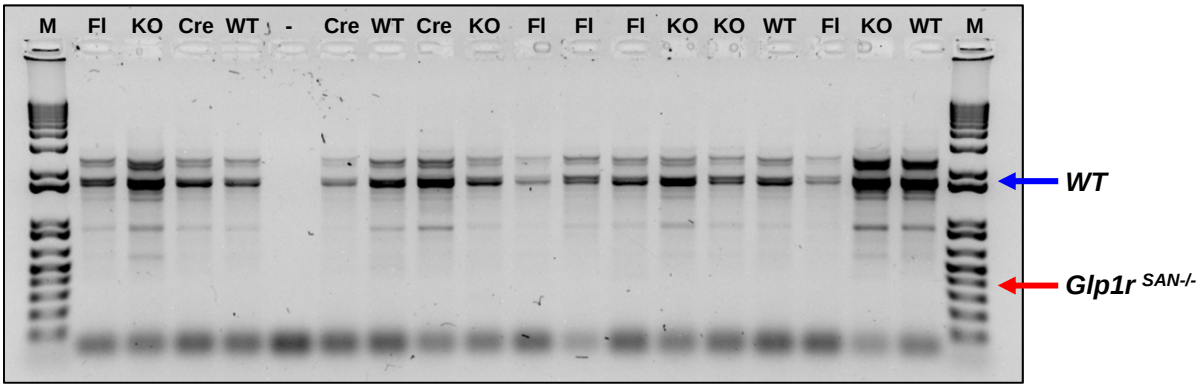


***WT* = 452 bp**

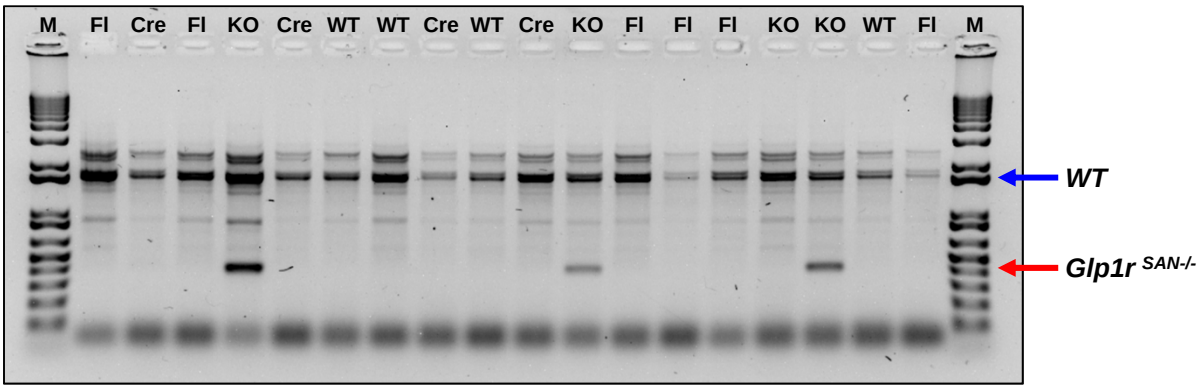
***Glp1r* $^{SAN-/-}$ *(SAN KO)* = 138 bp**

Sup. Figure 4

Left Atrium Genomic DNA



Right Atrium Genomic DNA

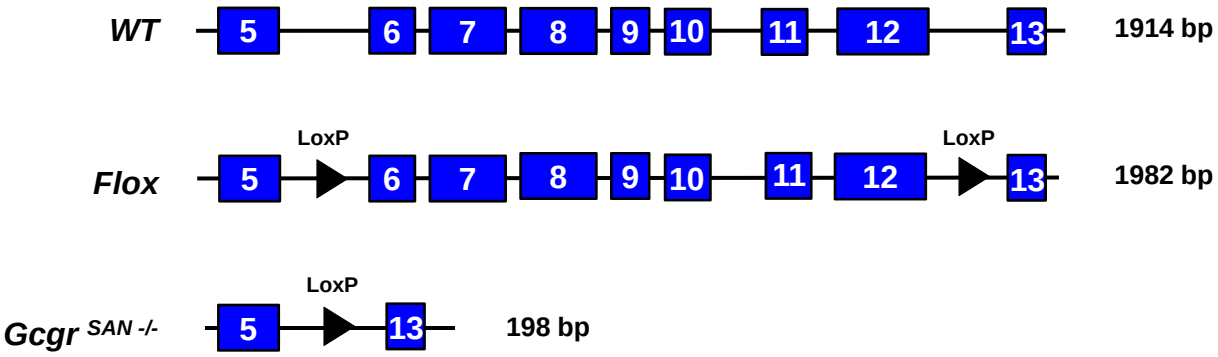


***WT* = 1981 bp**

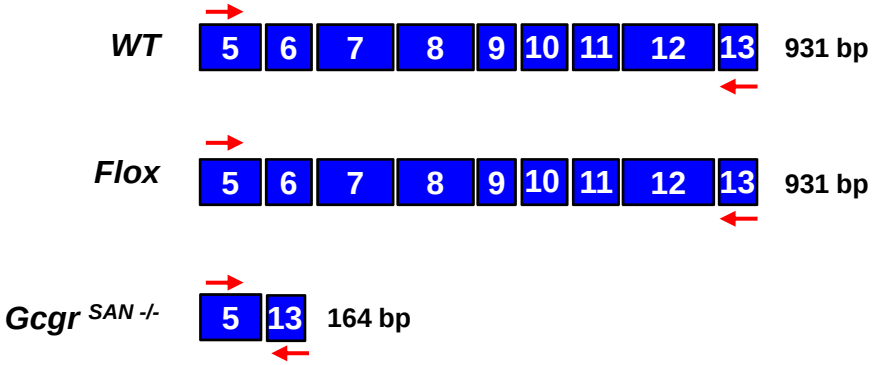
***Glp1r* $^{SAN-/-}$ = 348 bp**

Sup. Figure 5

Gcgr Genomic DNA

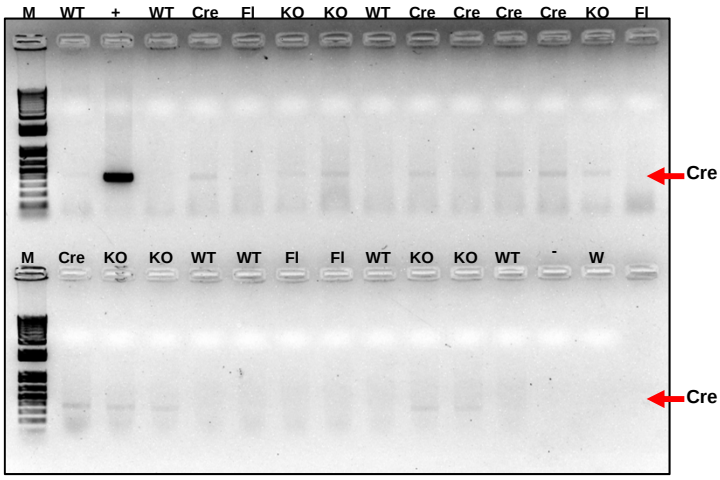


Gcgr cDNA

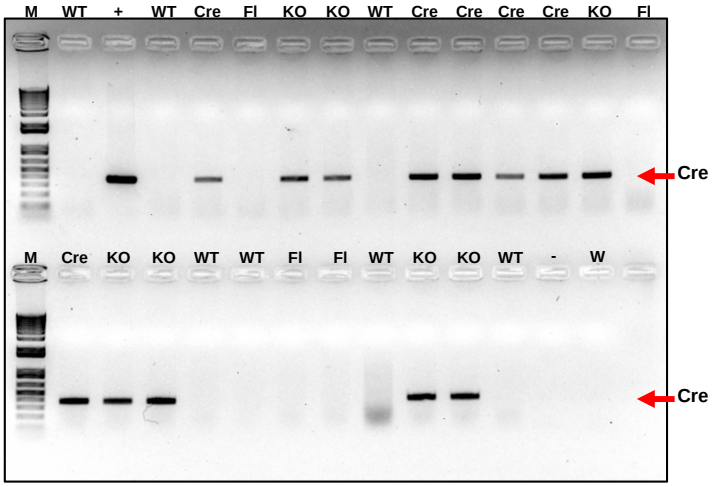


Sup. Figure 6

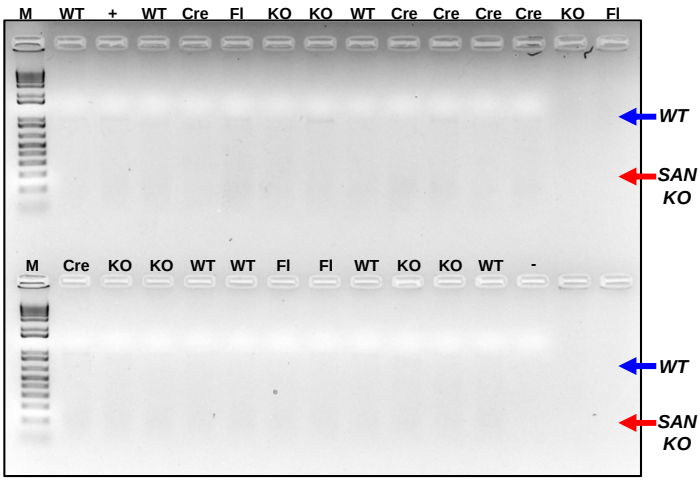
Cre mRNA Expression Left Atrium



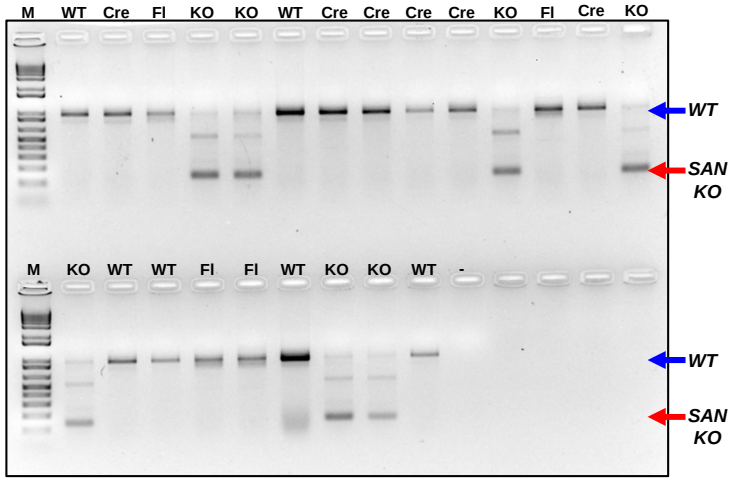
Cre mRNA Expression Right Atrium



Gcgr Exon 5-13 mRNA Expression Left Atrium



Gcgr Exon 5-13 mRNA Expression Right Atrium



***WT* = 931 bp**

***Gcgr* $^{SAN-/-}$ *(SAN KO)* = 164 bp**

Sup. Figure 7

Supplementary Figure 1. Acute administration of liraglutide is associated with prolonged increases in heart rate in mice. The graphs show minute-minute heart rate recordings of the data in Figures 2A and B. All injections were administered 15 min after the start of data collection. Values are mean ± SE; n=3 mice/group.

Supplementary Figure 2. Acute administration of lixisenatide is associated with short-term increases in heart rate in mice. The graphs show minute-minute heart rate recordings following ip injection of vehicle or 10 μg/kg lixisenatide in *αMHC-Cre* (*Cre WT*) and *Glp1r*$^{CM-/-}$ mice. All injections were administered 15 min after the start of data collection. Values are mean ± SE; n=3 mice/group.

Supplementary Figure 3. Schematic of *Glp1r* genomic DNA, cDNA, and expected PCR product sizes from wild-type (*WT*), *Fl/Fl Glp1r* (Flox), and sinoatrial node-specific *Glp1r* knockout mice (*Glp1r* $^{SAN-\backslash-}$). Arrows show positions of PCR primers.

Supplementary Figure 4. Agarose gels of PCR products generated by amplification of cDNA samples from the left atrium (left panels) and right atrium (right panels) from wild-type (*WT*), *Fl/Fl Glp1r* (Fl), *Hcn4-Cre* (Cre) and sinoatrial node-specific *Glp1r* knockout (KO) mice. Upper panels are Cre PCR products (red arrow) and show that Cre is only expressed in the right atrium as would be expected for a sinoatrial node-specific Cre driver. Lower panels are PCR products from primers designed to amplify exons 5 to 8 of the Glp1r. In both the left and right atrium, only the full-length (452 bp) wild-type exon 5 to 8 product (blue arrow, WT) is amplified, suggesting that, although the Glp1r is expressed in the mouse atria, it is not expressed in HCN4-positive sinoatrial node cells. The predicted position of a truncated (138 bp) *Glp1r* $^{SAN-/-}$ exon 5 to 8 product is also shown (red arrow, SAN KO). M, molecular weight marker; +, positive control; -, negative control; w, water.

Supplementary Figure 5. Agarose gels of PCR products generated by amplification of *Glp1r* genomic DNA sequences flanking exons 6 to 7 of the *Glp1r* (see Supp. Fig. 3). Genomic DNA was isolated from the left and right atria of wild-type (*WT*), *Fl/Fl Glp1r* (Fl), *Hcn4-Cre* (Cre) and sinoatrial node-specific *Glp1r* knockout (KO) mice. A full-length (1981 bp) product is amplified in all atrial genomic DNA samples (blue arrow, WT). Whereas a 348 bp truncated product, where exons 6 and 7 have been excised, is generated only from right atria genomic DNA from sinoatrial node-specific *Glp1r* knockout mice (red arrow, *Glp1r* $^{SAN-/-}$), but not control mice. This data confirms that Cre recombinase is able to truncate the *Glp1r* genomic DNA sequence flanked by loxP sites in the right atrium and supports the notion that the *Glp1r* is expressed in the mouse atria, but is not expressed in HCN4-positive sinoatrial node cells. M, molecular weight marker.

Supplementary Figure 6. Schematic of *Gcgr* genomic DNA, cDNA, and expected PCR product sizes from wild-type (*WT*), *Fl/Fl Gcgr* (Flox), and sinoatrial node-specific *Gcgr* knockout (*Gcgr* $^{SAN\ -/-}$) mice. Arrows show positions of PCR primers.

Supplementary Figure 7. Agarose gels of PCR products generated by amplification of cDNA samples from the left atrium (left panels) and right atrium (right panels) from wild-type (*WT*), *Fl/Fl Gcgr* (Fl), *HCN4-Cre* (Cre) and sinoatrial node-specific *Gcgr* knockout (KO) mice. Upper panels are Cre PCR products (red arrows) and show that Cre is highly expressed in the right atrium as would be expected for a sinoatrial node-specific Cre driver. Lower panels are PCR products from primers designed to amplify exons 5 to 13 of the *Gcgr*. The *Gcgr* is not expressed in the left atrium. In the right atrium, the full-length (931 bp) exon 5 to 13 PCR product (blue arrow, WT) is amplified in all mice. Whereas a 164 bp truncated product, where exons 6 to 12 have been deleted, is amplified only in the sinoatrial node-specific *Gcgr* knockout mouse (red arrow, SAN KO). M, molecular weight marker; +, positive control; -, negative control; w, water.