Arcuate nucleus glucokinase regulates glucose intake

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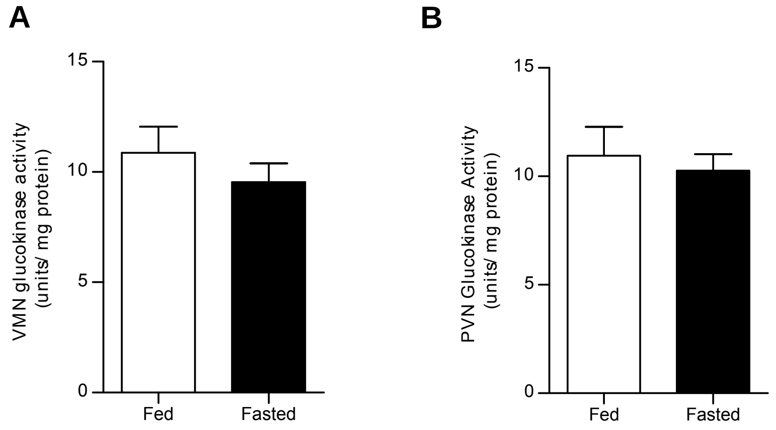
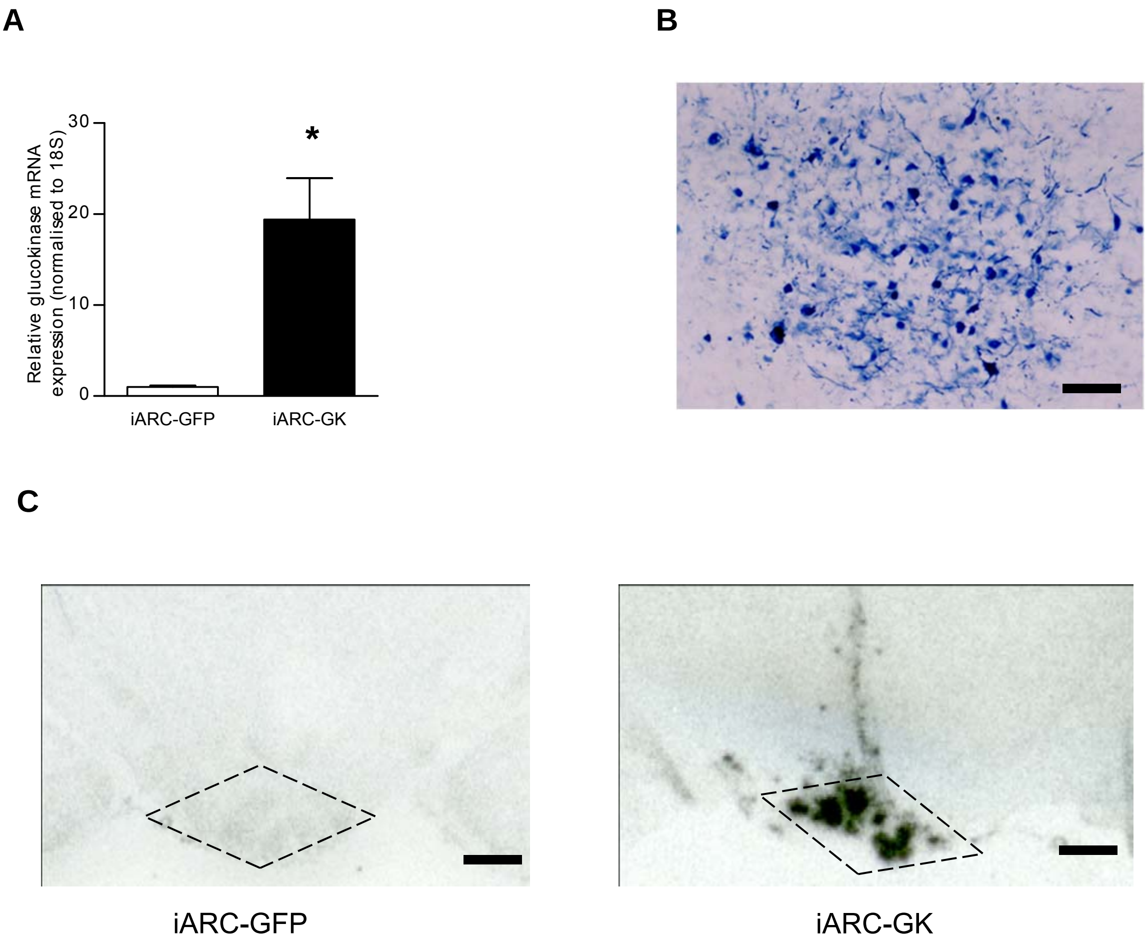
Supplementary Figure 1



Supplementary Figure 1. Effect of nutritional state on VMN and PVN glucokinase activity **(A)** Glucokinase activity in homogenate supernatants from the VMN of chow-fed and 24 hour fasted rats (n=10) and **(B)** PVN of chow-fed and 24 hour fasted rats (n=8-9).

Data presented as mean ± s.e.m.

Supplementary Figure 2

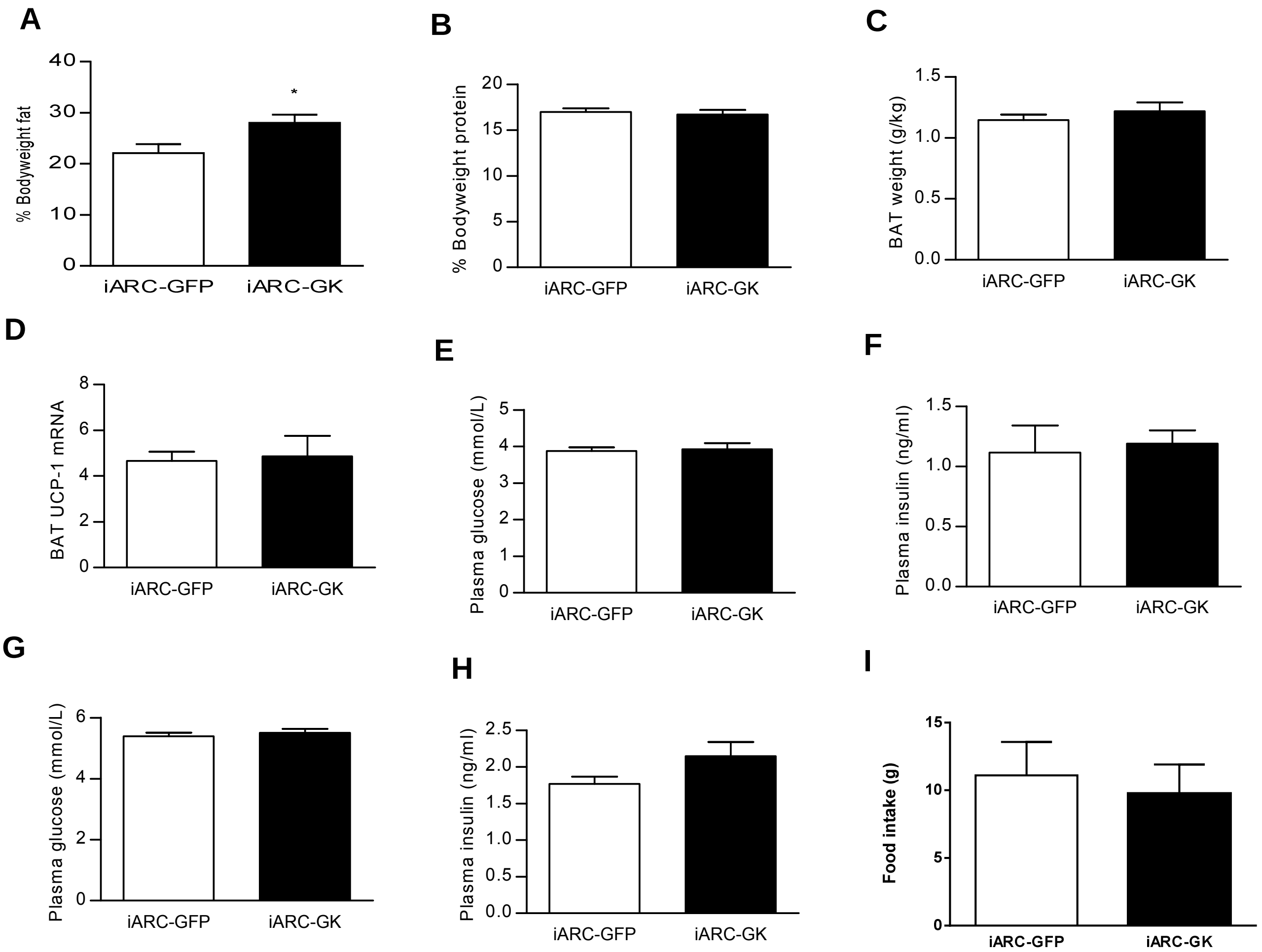


Supplementary Figure 2. Hypothalamic glucokinase mRNA expression following stereotactic injection of rAAV-GK into the arcuate nucleus of male Wistar rats

- **(A)** Relative hypothalamic glucokinase mRNA expression in iARC-GFP and iARC-GK rats (n=9-12).
- **(B)** Immuncytochemical detection of GFP in the arcuate nucleus in iARC-GFP rat. Scale bar = 20μm
- **(C)** In-situ hybridization of hypothalamic glucokinase mRNA in chow-fed iARC-GFP and iARC-GK rats (darkfield photomicrograph of 35S-silvergrains). Dashed area indicates approximate location of the ARC. Scale bar = 1mm.

Data presented as mean ± s.e.m. **P<0.01 versus control.

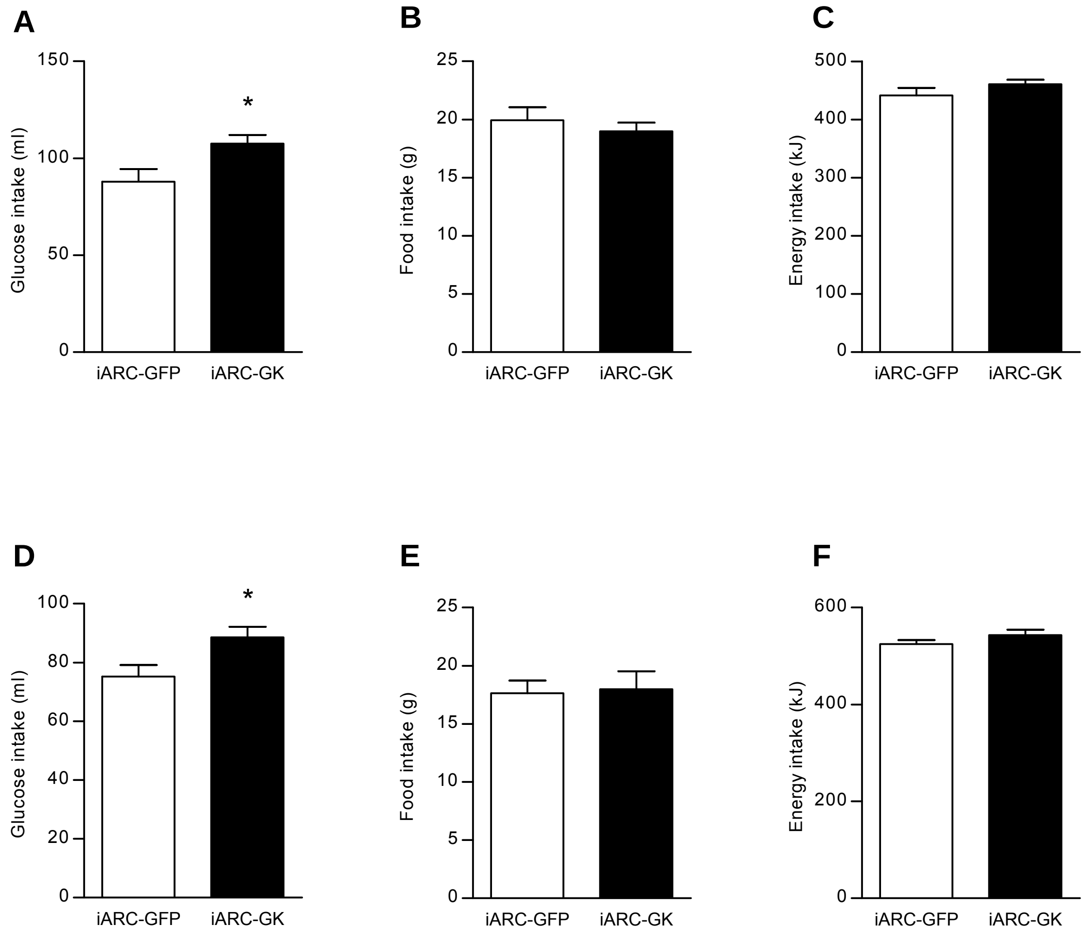
Supplementary Figure 3



Supplementary Figure 3. Effect of increased arcuate nucleus glucokinase on body composition, BAT, glucose homeostasis and fasting induced food intake.

(A) Percentage body fat measured using body composition analysis in iARC-GFP and iARC-GK rats on normal chow diet for 33 days after recovery from surgery (n=12-15); **(B)** Percentage body protein measured using body composition analysis in iARC-GFP and iARC-GK rats on normal chow diet for 33 days after recovery from surgery (n=12-15). ; **(C)** BAT weight corrected to bodyweight (n=10-13); **(D)** UCP-1 BAT mRNA expression normalised to 28S ribosomal RNA in iARC-GFP and iARC-GK rats on normal chow diet for 33 days after recovery from surgery (n=6-11). fasting plasma insulin, **(E)** fasting plasma glucose fed plasma glucose and **(F)** fasting plasma insulin **(G)** fed plasma glucose; and **(H)** fed plasma insulin in iARC-GFP and iARC-GK rats on normal chow diet (n=9-13). **(I)** 2 hour food intake following 48 hour fast(n=12-15). Data presented as mean ± s.e.m.

Supplementary Figure 4



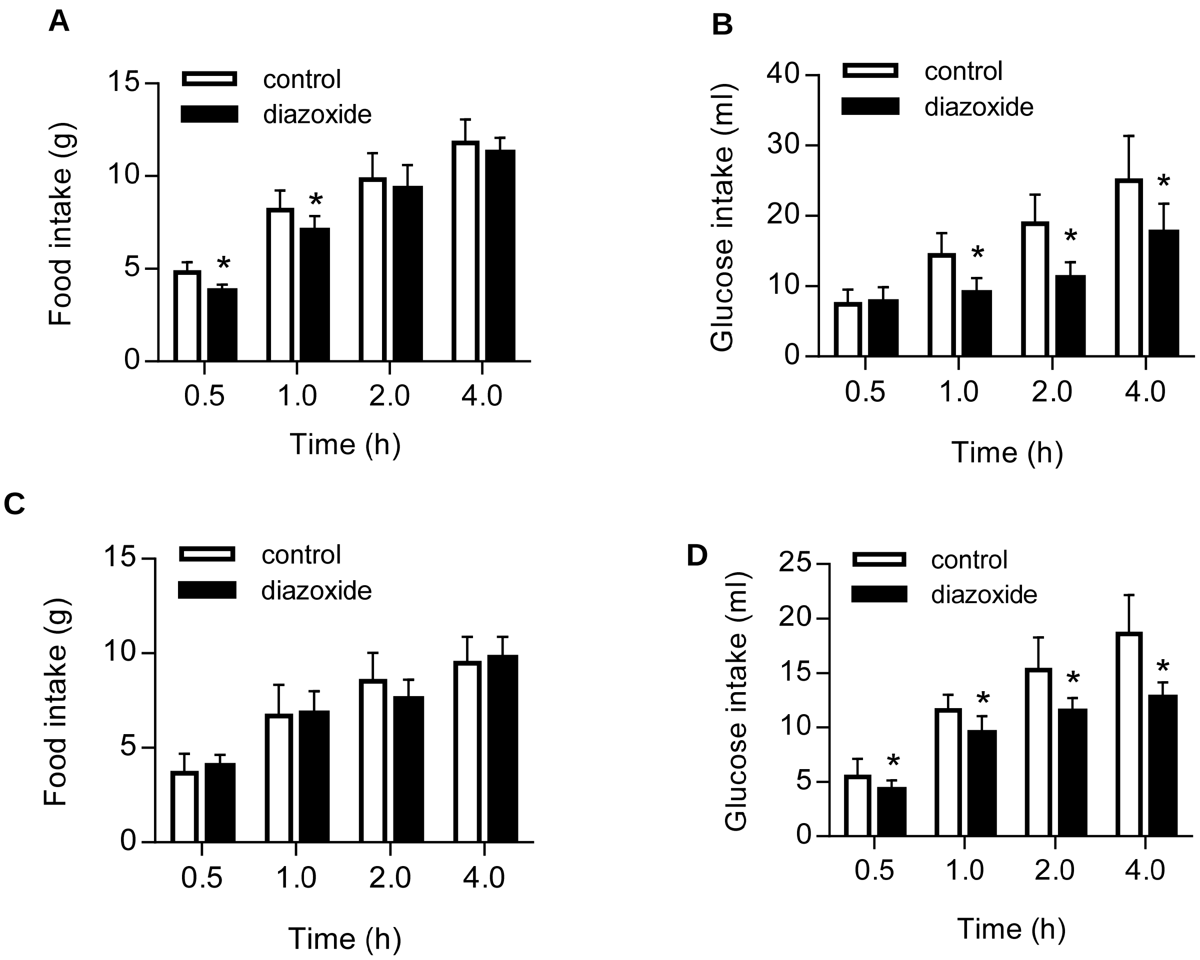
Supplementary Figure 4. Effect of increased arcuate nucleus glucokinase activity on food intake and glucose appetite with ad libitum access to normal chow diet and 10% glucose solutions

(A) Glucose, **(B)** food and **(C)** energy intake in iARC-GFP (filled circles) or iARC-GK (open squares) rats after 24 hours during a 24-hour feeding study with ad libitum 10% glucose and normal chow intake (n=7).

(D) Glucose, **(E)** food and **(F)** energy intake in iARC-GFP (filled circles) or iARC-GK (open squares) rats after 24 hours during a 24-hour feeding study with ad libitum 20% glucose and normal chow intake (n=8).

Data presented as mean ± s.e.m. *P<0.05 versus corresponding control values.

Supplementary figure 5



Supplementary Figure 5. Effect of intra arcuate injection of diazoxide on food intake and glucose appetite with ad libitum access to either normal chow diet or 2% glucose solutions or both chow and glucose

A Food intake after injection of 1 nmol diazoxide or vehicle (control) in rats (n=9), when only chow is available

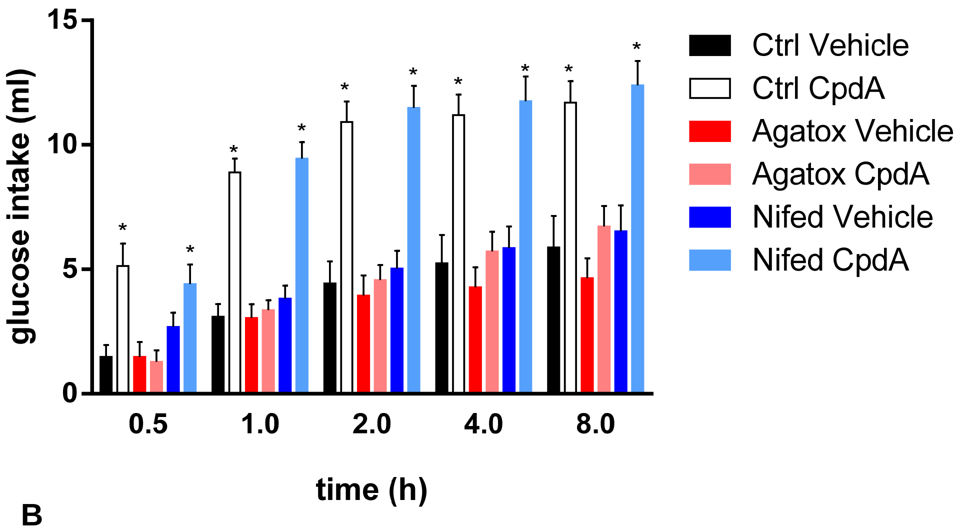
B 2% glucose intake after injection of 1 nmol diazoxide or vehicle (control) in rats (n=9), when only glucose is available

C Food intake after injection of 1 nmol diazoxide or vehicle (control) in rats (n=9), when both chow and glucose available

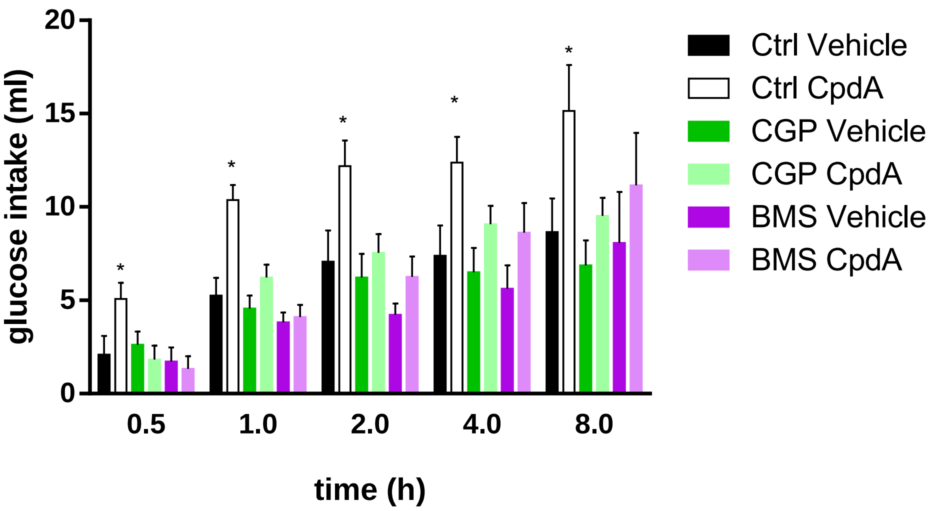
D 2% glucose intake after injection of 1 nmol diazoxide or vehicle (control) in rats (n=9), when both chow and glucose available

Supplementary figure 6

A



B



Supplementary figure 6: Effect of calcium channel blockers and NPY receptor antagonists on compound A induced glucose intake.

(A) 2% w/v glucose solution intake after intra-arcuate injection of nifedipine, ω-agatoxin IVA or vehicle and subsequent injection of CpdA or control, in rats time was measured from the end of the second injection (n=15).

(B) 2% w/v glucose solution intake after intra-peritoneal injection of BMS-193885, CGP-71683 or vehicle and subsequent intra-arcuate injection of CpdA or control, in rats time was measured from the end of the second injection (n=14) Data presented as mean ± s.e.m. *P<0.00001 vs corresponding vehicle injected group