Supporting Information

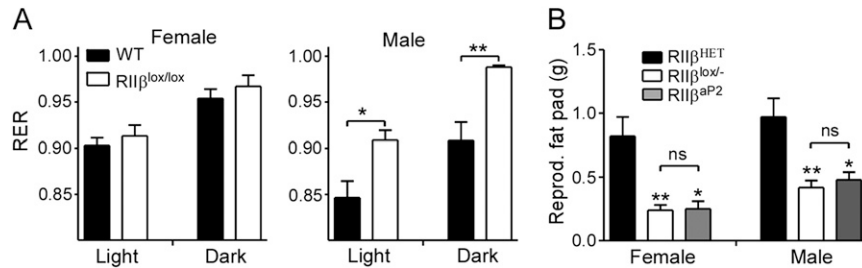## Zheng et al. 10.1073/pnas.1219542110

Fig. S1. Respiratory exchange ratio and additional fat-pad data on aP2-Cre/RIIβ$^{lox/-}$ mice. (*A*) Respiratory exchange ratio (RER) of WT and RIIβ$^{lox/lox}$ mice during light and dark phases was measured over 2 d and averaged. For both sexes, $n = 8$ for each genotype; values represent mean ± SEM. **$P < 0.01$. (*B*) Weight of reproductive fat pads in heterozygote (HET) ($n = 15$ for male and 16 for female), RIIβ$^{lox/-}$ ($n = 14$ for male and 13 for female), and RIIβ^{aP2} ($n = 9$ for male and 8 for female) mice. Values represent mean ± SEM ***$P < 0.001$ compared with HET.
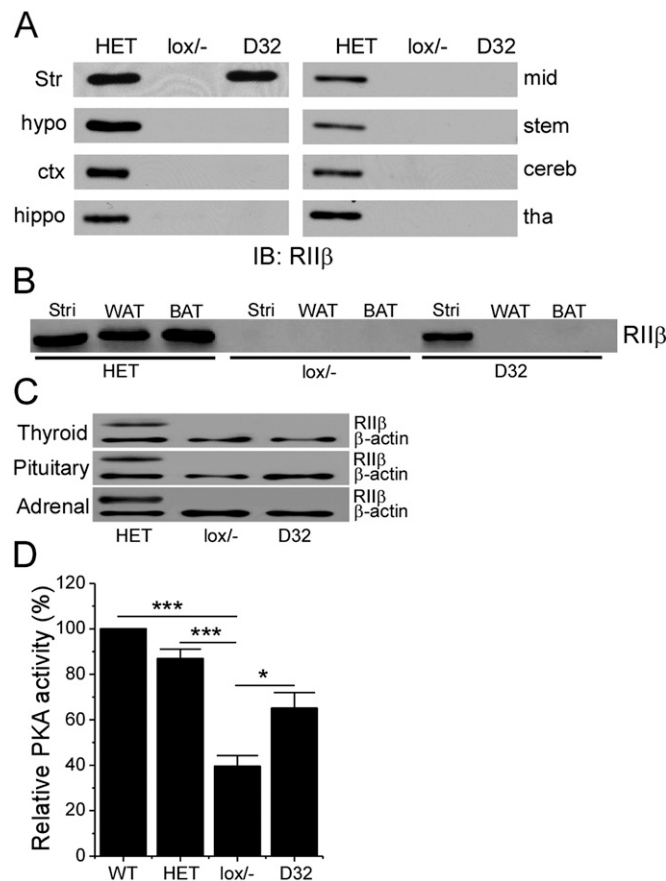
Fig. S2. Specificity of D32-Cre–induced RIIβ expression and the effect on protein kinase A (PKA) activity in the striatum. (*A–C*) Immunoblots of RIIβ in different brain regions and periphery tissues, including brown adipose tissue (BAT), white adipose tissue (WAT), pituitary, thyroid, and adrenal glands of HET, RIIβ$^{lox/-}$, and RIIβ^{D32} mice. (*D*) Total PKA activity (in presence of 5 μM cAMP) of striatal extracts from WT, HET, RIIβ$^{lox/-}$, and RIIβ^{D32} mice. *$P < 0.05$, ***$P < 0.001$.

Fig. S3. Specificity of D1R-Cre– and D2R-Cre–induced RIIβ expression. (*A–C*) Immunoblots for RIIβ in different brain regions (*A*) and periphery tissues, including pituitary (Pit), thyroid (Thy), and adrenal (Adr) (*B*), and BAT and WAT (*C*) of HET, RIIβ$^{lox/−}$, RIIβ^{D1R}, and RIIβ^{D2R} mice. (*D*) Immunostaining of RIIβ and dynorphin in the dorsal striatum of RIIβ^{D2R} mice. Cells with double staining are indicated by arrowheads.

Fig. S4. Effects of selective RIIβ expression in paraventricular hypothalamus (PVH), ventromedial hypothalamus (VMH), agouti-related peptide (AgRP)/neuropeptide Y (NPY), or proopiomelanocortin (PMOC) neurons on locomotion and adiposity. (*A*) Immunostaining for RIIβ in single-minded 1 (Sim1)-Cre/RIIβ$^{lox/−}$, steroidogenic factor 1 (Sf1)-Cre/RIIβ$^{lox/−}$, Agrp-Cre/RIIβ$^{lox/−}$, and Pomc-Cre/RIIβ$^{lox/−}$ mice. 3V, third ventricle; Arc, arcuate; ME, median eminence. (*B*) Locomotor activity and (*C*) reproductive fat-pad weight of Sim1-Cre/RIIβ$^{lox/−}$, Sf1-Cre/RIIβ$^{lox/−}$, Agrp-Cre/RIIβ$^{lox/−}$, and Pomc-Cre/RIIβ$^{lox/−}$ mice compared with HET and RIIβ$^{lox/−}$ controls at 12–16 wk of age (*n* = 6–14 for each group). Error bars are shown as SEM. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

Fig. S5. (*A*) Immunostaining of RIIβ in posterior striatum and hypothalamus of Vgat-ires-Cre/RIIβ[lox/−] (RIIβ[Vgat]) mice. CTX, cortex; Hippo, hippocampus, Tha, thalamus; VMH, ventromedial hypothalamus. (*B*) Western blots showed that RIIβ was not expressed in BAT and WAT of RIIβ[Vgat] mice. Increased level of RIα protein was observed in both RIIβ[lox/−] and RIIβ[Vgat] mice. (*C*) Major fat pads, including reproductive, inguinal, and retroperitoneal fat pads of 12-wk-old male HET, RIIβ[lox/−], and RIIβ[Vgat] mice (*n* = 6 for each group). Data are expressed as mean ± SEM. **$P < 0.01$, ***$P < 0.001$, unpaired *t* test compared with HET control or as indicated.