Supplementary Materials for

An adiponectin-frontothalamic pathway mediates the rapid antidepressant effects of acute exercise

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This PDF file includes:

Figs. S1 to S5
Table S1

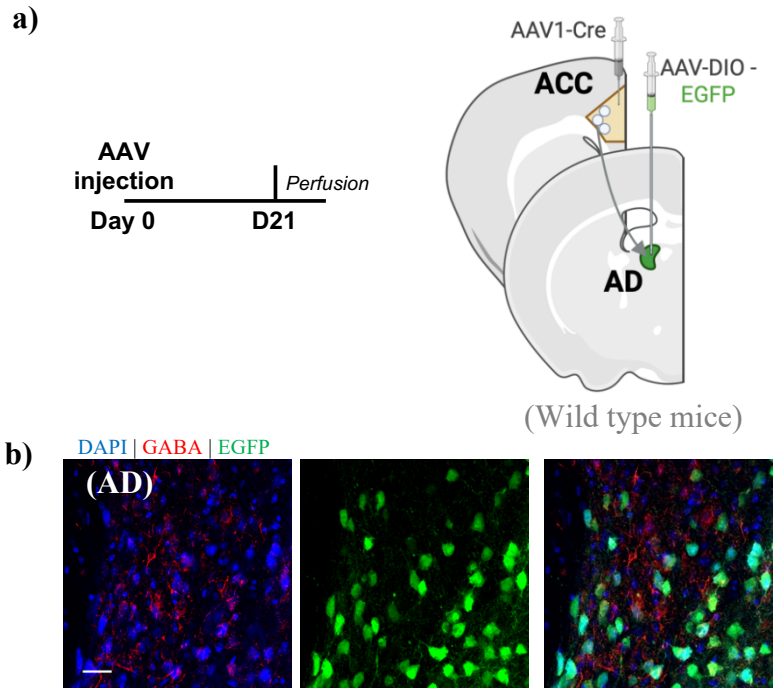


Fig. S1. ACC projection to the AD is not a GABAergic circuit. (**a**) Experimental timeline (left) and injection site (right). The anterograde AAV1 was unilaterally injected in the ACC, while a Cre-dependent AAV driving the expression of EGFP was injected ipsilaterally in the AD. Animals were sacrificed 21 days later for immunohistochemical analysis. (**b**) Co-expression of EGFP with GABA is not seen in the AD (scale bars: 30 μm).

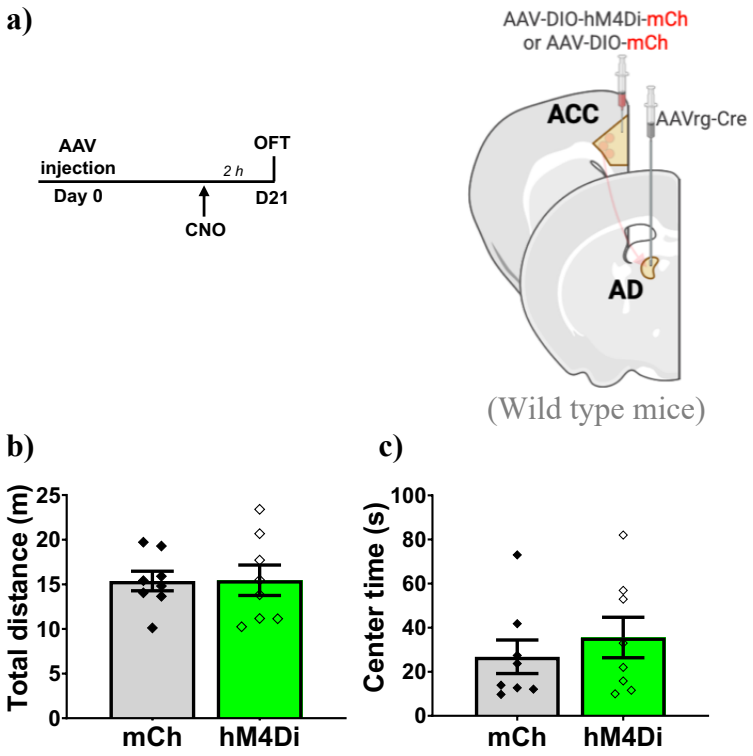


Fig. S2. Inhibition of the ACC-AD pathway does not affect locomotor activity. (**a**) Mice were injected with AAVrg-Cre in the AD, and AAV-DIO-hM4Di-mCh or AAV-DIO-mCh in the ACC. Animals were treated with CNO (1 mg/kg, i.p.) 2 h before behavioral assessment in the OFT. (**b**) Total distance traveled and (**c**) time spent in the center of the OFT 2 h after CNO treatment (n = 8/group). Data presented as mean ± SEM, unpaired t-test.

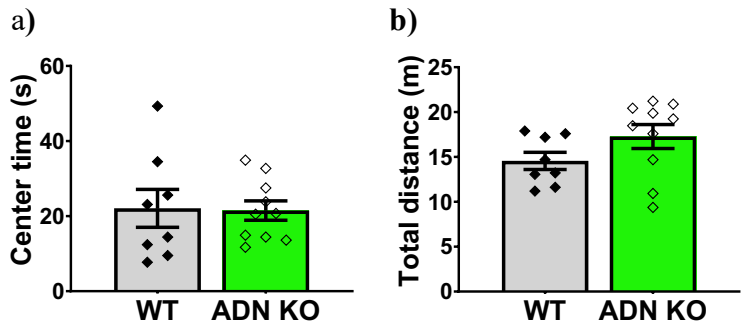


Fig. S3. Adiponectin deficiency does not affect anxiety-like behavior or spontaneous locomotor activity. Adiponectin knockout mice (ADN KO) were subjected to the OFT. (**a**) Adiponectin deficiency did not affect the time spent in the center or (**b**) the total distance traveled in the OFT compared to wild-type (WT) mice (n = 8-10/group). Data presented as mean ± SEM; unpaired t-test.

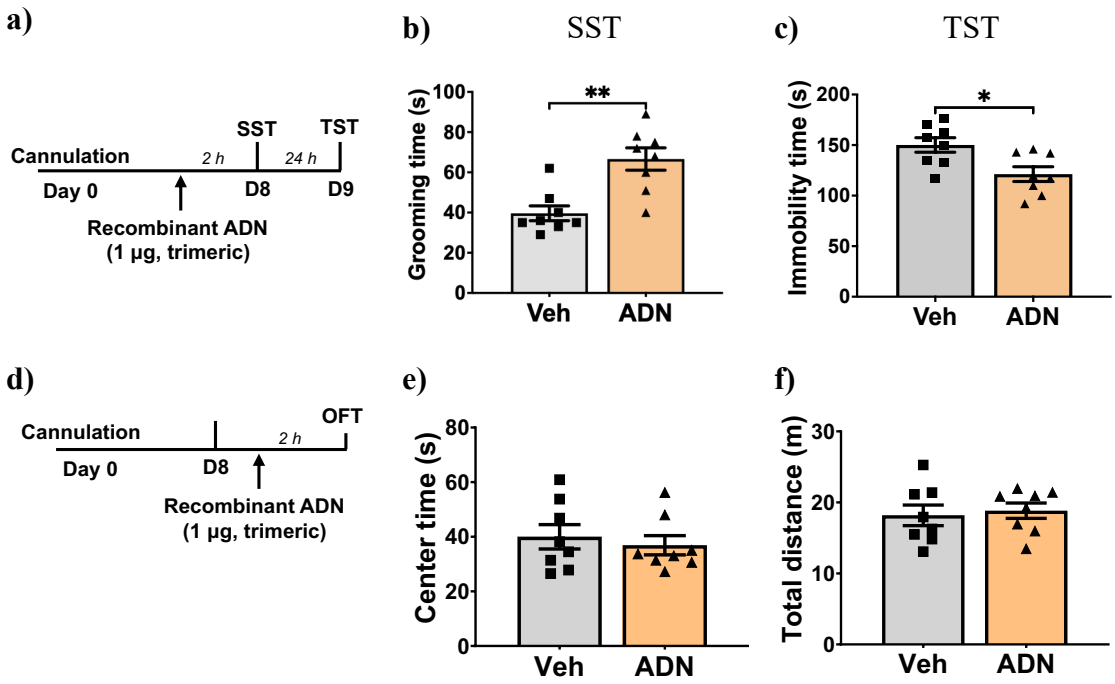


Fig. S4. Single intracerebroventricular infusion with recombinant adiponectin mimics the rapid antidepressant effects of acute exercise. (**a**) On the eighth day after surgery, cannulated mice received a single intracerebroventricular infusion of recombinant adiponectin (1 µg, trimeric) or vehicle, followed by behavioral assessment. (**b**) Expression of grooming time in the SST and (**c**) immobility time in the TST after infusion (n = 8/group). (**d**) Another cohort of cannulated mice received a single intracerebroventricular infusion of recombinant adiponectin (same dosage and isoform) or vehicle 2 hours before OFT. **(e)** Time spent in the center and (**f**) total distance traveled in the OFT (n = 8/group). Data presented as mean ± SEM; unpaired t-test, * $p < 0.05$, ** $p < 0.01$.

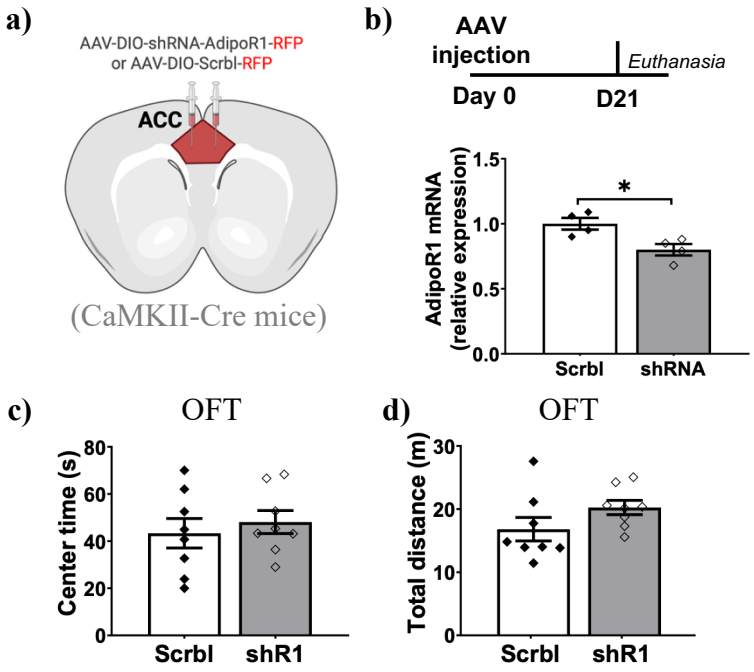


Fig. S5. Knockdown of AdipoR1 in ACC glutamatergic neurons. (**a**) AAVrg-Cre was injected in the AD of CaMKII-Cre mice, while AAV-DIO-shRNA-AdipoR1-RFP or AAV-DIO-shRNA-Scrbl-RFP was injected in the ACC to knock down the AdipoR1 in glutamatergic neurons. (**b**) Significant reduction in AdipoR1 mRNA expression in the ACC (n = 4/group). (**c**) In a separate cohort of mice, anxiety-like behavior and (**d**) spontaneous locomotor activity were assessed in the OFT 21 days after AdipoR1 knockdown in the ACC (n = 8/group). Data presented as mean ± SEM; unpaired t-test, * $p < 0.05$.

	Participants (n = 40)
Age (*M, SD*)	27.03 (5.25)
Sex (*%*)	
Male	15 (37.50)
Female	25 (62.50)
Physical activity level (*%*)*	
Low	8 (20.00)
Moderate	19 (47.50)
High	13 (32.50)
BMI (*M, SD*)	21.26 (2.25)

***Based on the International Physical Activity Questionnaire (IPAQ)

Table S1. Participants' demographic characteristics.