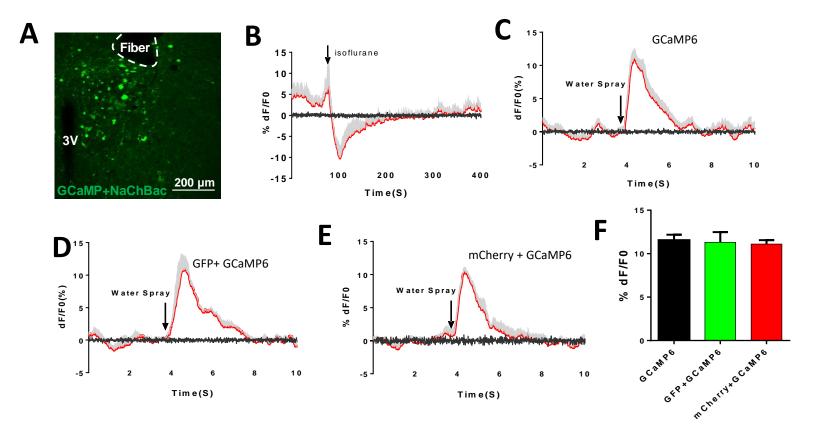
## Appendix Data

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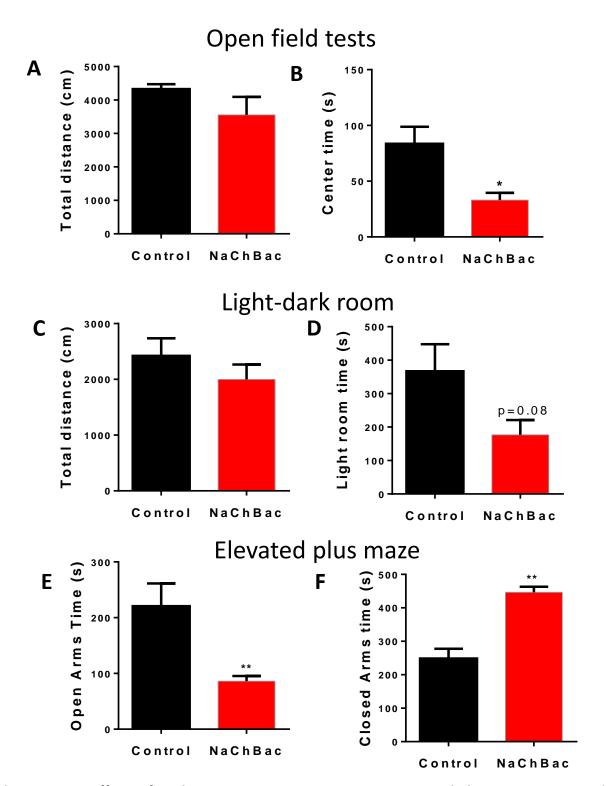
Page 2: Appendix Figure S1:Effects of isoflurane and coexpression of GFP or mCherry on GCaMP6 recording;

Page 3: Appendix Figure S2:Effects of NachBac expression in PVH CRH neurons on behaviors.

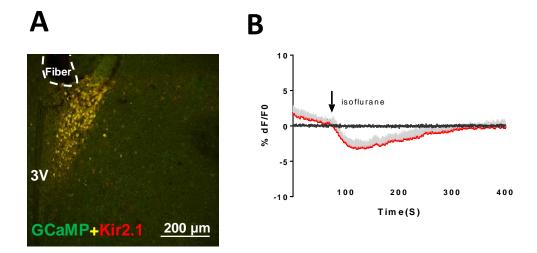
Page 4: Appendix Figure S3:Effects of isoflurane on PVH CRH neurons expressing Kir2.1 expression.



Appendix Figure S1. Effects of isoflurane and coexpression of GFP or mCherry on GCaMP6 recording. A) A presentative picture showing expressing of NachBac and GCaMP6 in CRH neurons of PVH with fiber implantation trace. B) Traces showing NachBac-expressing CRH neurons response to inhalation of isoflurane. (C-F) Fiber photometry recording on PVH CRH neuron response to water spray with expression of GCaMP6m alone (C), GCaMP6m+GFP (D), GCaMP6m+mCherry (E) and comparison between the 3 group (F). n=4 each. Data presented as mean+/-SEM, 1-way ANOVA tests.



**Appendix Figure S2. Effects of NachBac expression in PVH CRH neurons on behaviors.** CRH-Cre male mice (7-8 weeks old, n=6-7) were injected with control AAV-Flex-GFP or NachBac vectors to bilateral PVH, and behavioral tests were conducted 4 weeks later for open field tests (A-B), light-dark room tests (C-D) and elevated plus maze (E and F). \*p<0.05 and \*\*p<0.01, unpaired Student's t tests.



Appendix Figure S3. Effects of isoflurane on PVH CRH neurons expressing Kir2.1 expression. A) A representative image showing expression of GCaMP6m and Kir2.1 in PVH CRH neurons with optic fiber implantation trace and B) traces showing Kir2.1-expressing CRH neurons response to inhalation of isoflurane (E).