Supplemental Figures and Table

Subject#	Age	Status	Sleep Onset	Sleep Offset	Sleep Duration (h)
100471	84	C	22:30	3:00	4.5
100784	79	C	22:30	6:00	7.5
100290	55	C	21:00	2:30	5.5
100475	49	C	0:42	5:30	4.8
100487	51	C	23:08	5:15	6.1
100913	57	C	NA	NA	5.5
100727	29	C	23:30	5:30	6.0
100481	43	NC	0:00	8:00	8.0
100469	50	NC	22:00	5:45	7.8
100889	39	NC	23:30	6:15	6.8
100730	28	NC	10:30	7:00	8.5
100729	26	NC	10:30	7:00	8.5
FNSS *n* = 7					5.7 ± 0.4
Control *n* = 5					7.9 ± 0.3

Table S1. Sleep schedule comparison for human subjects, Related to Figure 1

Ages were at the time of data collection. C = mutation carrier, NC = non-mutation carrier. Sleep offset is local standard clock time of average final morning awakening, and sleep onset is evening time of first falling asleep during extended vacations based on structured interviews. Two-tailed Student's *t*-test, P=0.0016. Values are mean ± SEM.

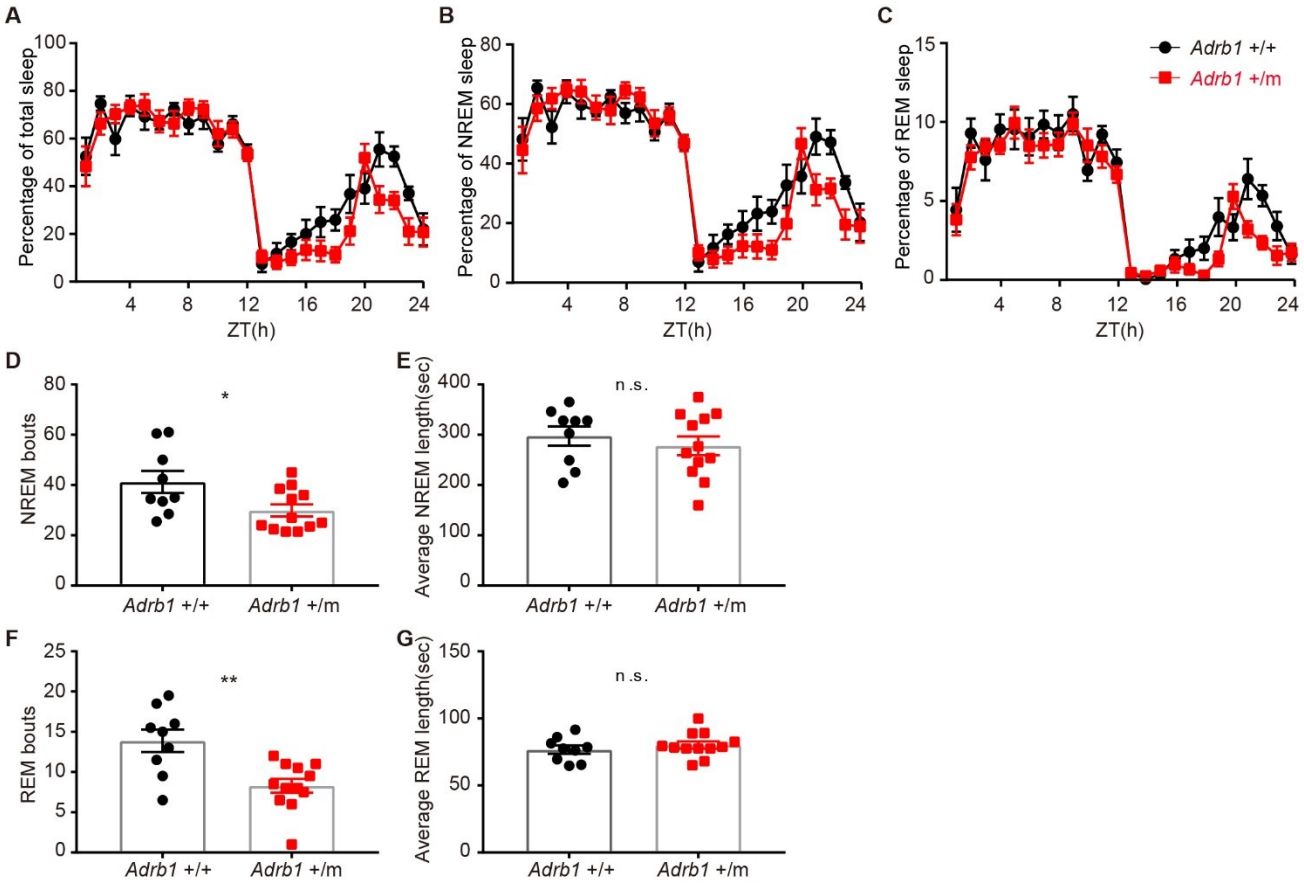

Figure S1. Additional sleep analysis in the *Adrb1-A187V* mouse model, Related to Figure 3

(A-C)Percentage of total sleep (A), NREM sleep(B) and REM sleep (C) were plotted hourly in 24 hours for *Adrb1* +/+ (N=9) and +/m (N=13) mice.

(D-E) NREM sleep bouts and average NREM sleep episode length during the dark phase were calculated for *Adrb1* +/+ (N=9) and +/m (N=13) mice.

(F-G) REM sleep bouts and average REM sleep episode length during the dark phase were calculated for *Adrb1* +/+ (N=9) and +/m (N=13) mice.

* P<0.05, **P<0.01, n.s.=not significant. Two-tailed Student's *t*-test.

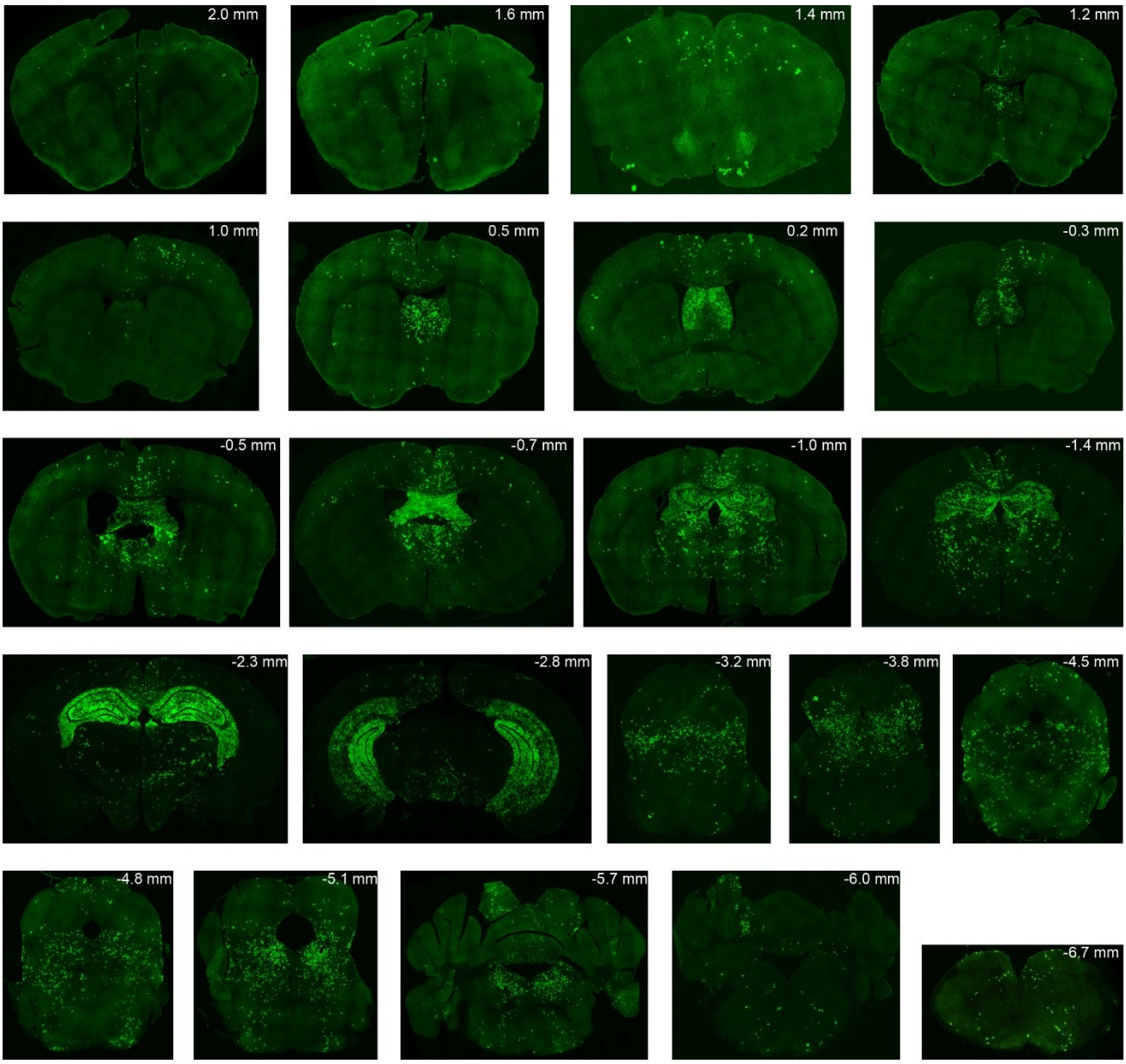

Figure S2. Brain-wide map of *ADRB1-Cre* reporter mice, Related to Figure 4

Representative coronal sections from *ADRB1-Cre*;loxP-flanked-ChR2–eYFP mice (N=2) show the ADRB1 positive cells throughout the brain. Distance relative to Bregma is displayed in the upper right corner of each figure.

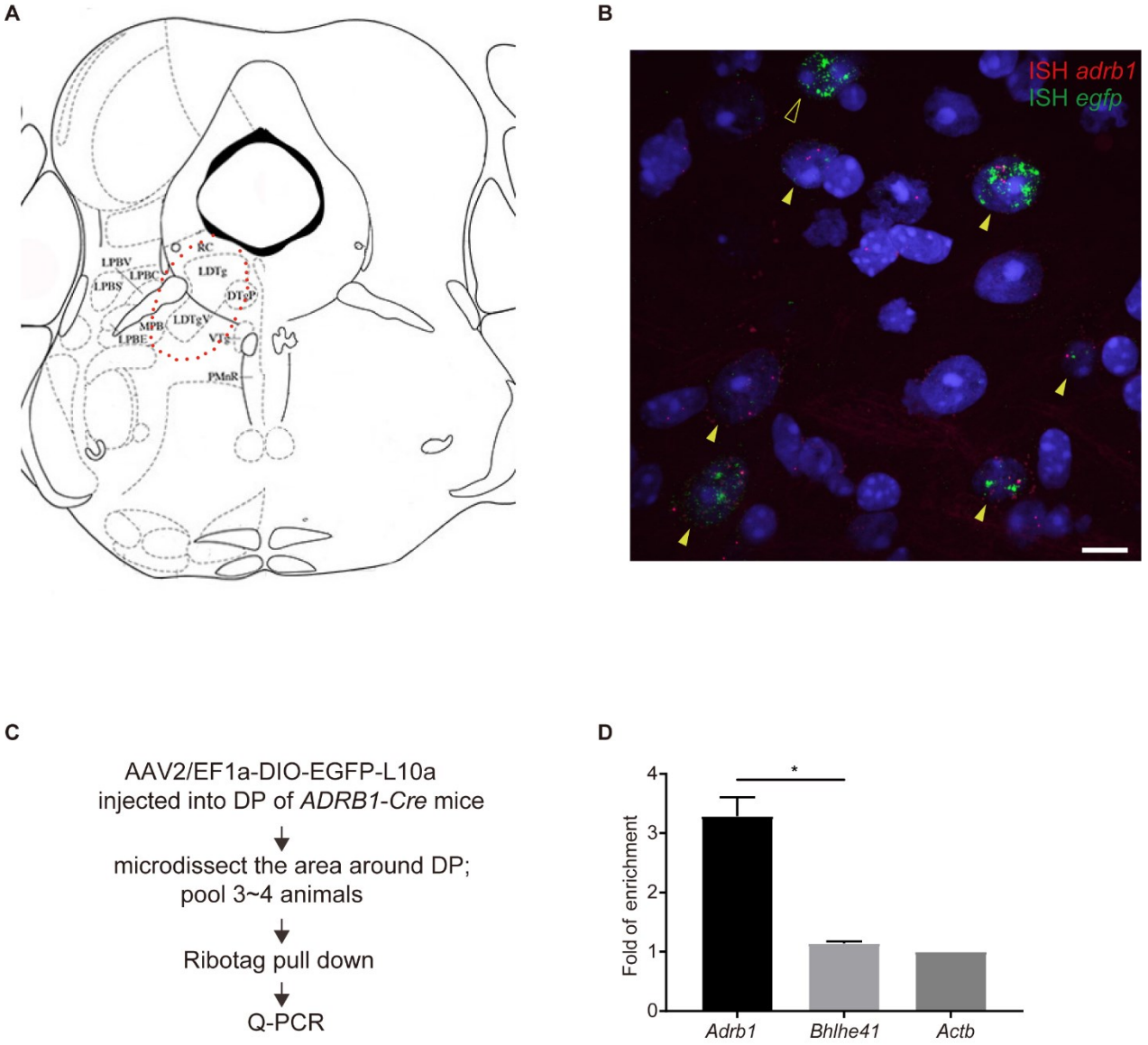

Figure S3. Characterization of *ADRB1*$^+$ cells in DP, Related to Figure 4

(A) Schematic of anatomy for dorsal pons (DP) area.

(B) Representative ISH image for endogenous *Adrb1* mRNA and *gfp* mRNA in the virus (AAV2/EF1a-DIO-EGFP-L10a) infused *ADRB1-Cre* mice (N=3). Yellow arrowheads indicate *Adrb1*$^+$*gfp*$^+$cells. Open arrowhead indicates *Adrb1*$^-$*gfp*$^+$ cells. In total, ~79.5% (163/205) *gfp*$^+$ cells are also *Adrb1*$^+$. Scale bar, 10 μm.

(C) Schematic of TRAP experiment workflow.

(D) q-RTPCR analysis of *Adrb1* transcripts expressed in DP after immunoprecipitation of polysomes from virus (AAV2/EF1a-DIO-EGFP- L10a) infused *ADRB1-Cre* mice. * P<0.05, two-tailed Student's *t*-test. Error bars represent ± SEM of four independent experiments.

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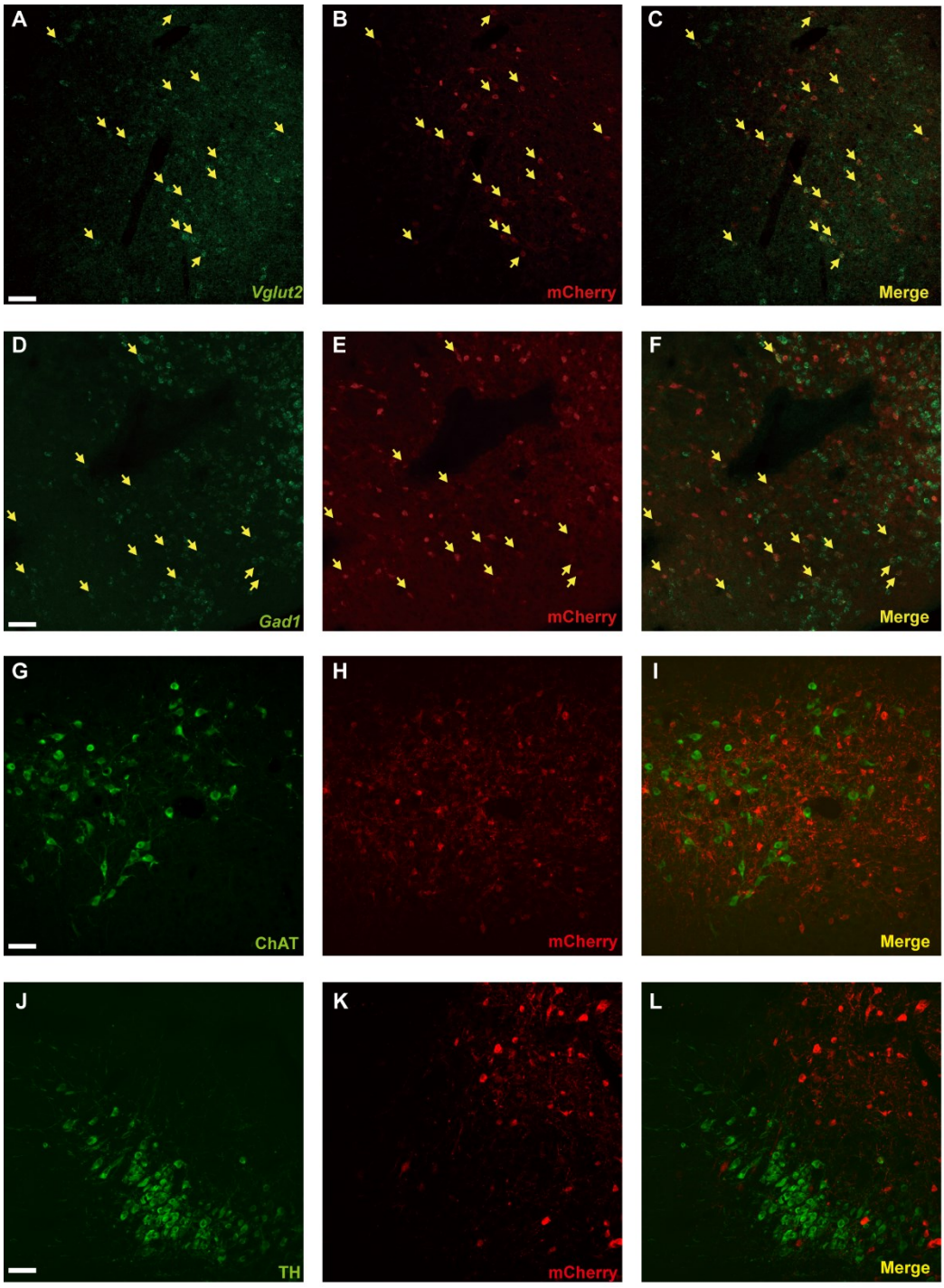


Figure S4. Expression of neurotransmitters in DP *ADRB1*$^{+}$cells, Related to Figure 4

(A-C) Co-localization of *Vglut2* mRNA (A, green) with mCherry (*ADRB1*) (B, magenta). Merged image is shown in C.

(D-F) Co-localization of *Gad1* mRNA (D, green) with mCherry (*ADRB1*) (E, magenta). Merged image is shown in F.

(G-I) Co-staining of anti-ChAT (G, green) with anti-Cherry (*ADRB1*) (H, magenta). Merged image is shown in I.

(J-L) Co-staining of anti-TH (J, green) with anti-Cherry (*ADRB1*) (K, magenta). Merged image is shown in L. Arrowheads indicate cells in which co-expression was detected. Scale bars, 30 μm. N=3 mice.

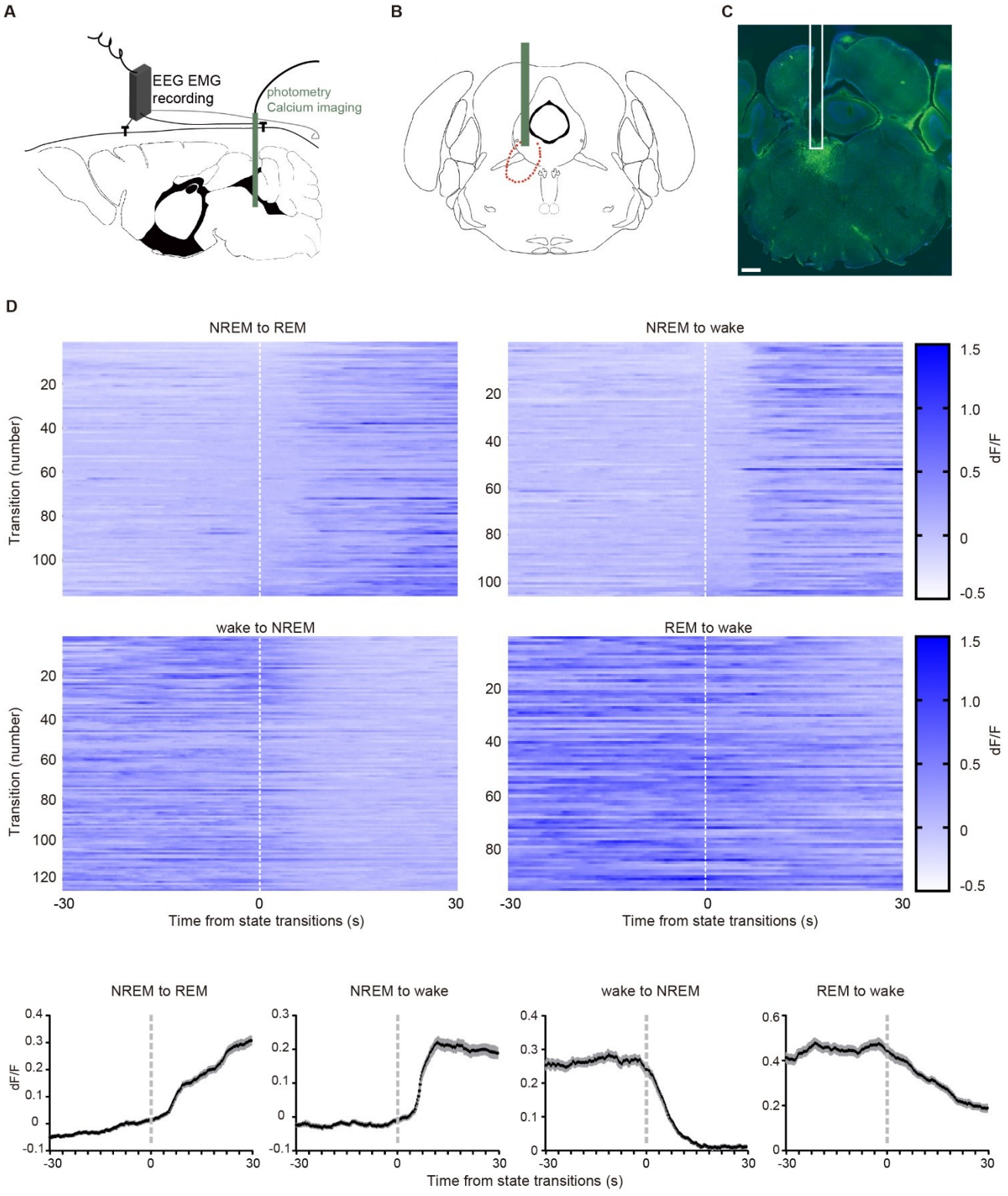


Figure S5. Photometry setup and population activity of DP *ADRB1*$^{+}$ neurons during the state transitions, Related to Figure 4

(A) Schematic of simultaneous fiber photometry/EEG/EMG set up for recording the activity of DP *ADRB1*$^{+}$ neurons across the different sleep/wake states.

(B and C) Schematic (B) and representative slice (C) showing viral expression and the placement of the fiber tip

above the DP.

(D) Fluorescence aligned to arousal state transitions. Top, individual transitions with color-coded fluorescence intensity (NREM-REM, n=116; NREM-wake, n=105; wake-NREM, n=125; REM-wake, n=95; N=6 mice). Bottom, average response from all the transitions. Shaded areas represent SEM.

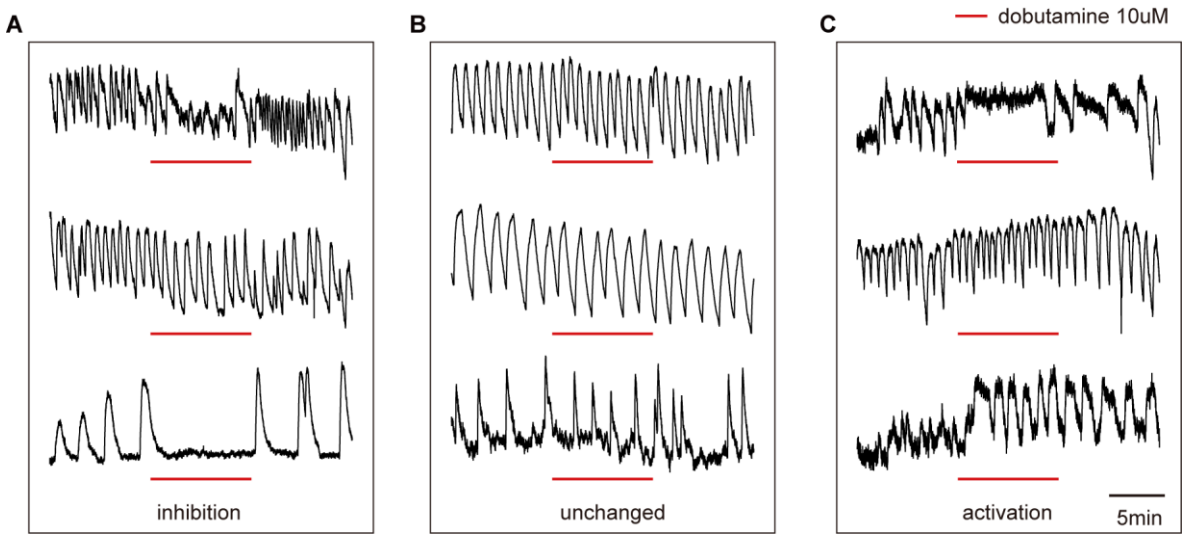
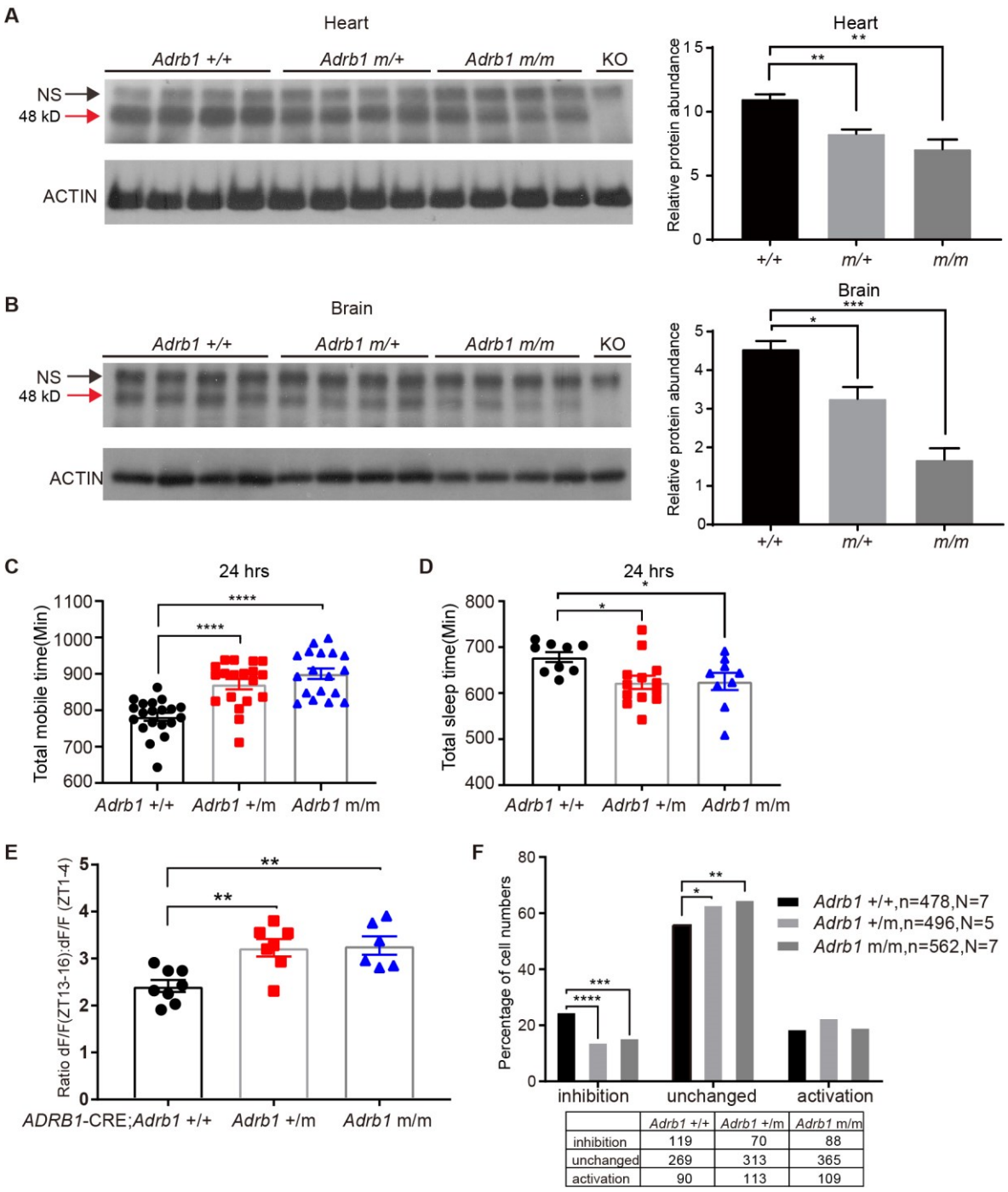


Figure S6. Categories of DP *ADRB1*$^{+}$ cells with differential responses to dobutamine treatment, Related to Figure 6

(A-C) Representative GCaMP fluorescence traces from cells that were inhibited (A), unaffected (B) and activated(C) by dobutamine treatment.



	Adrb1 +/+	*Adrb1* +/m	*Adrb1* m/m
inhibition	119	70	88
unchanged	269	313	365
activation	90	113	109

Figure S7. Analysis of *Adrb1-A187V* homozygous mouse model, Related to Figures 2C, 2D, 3A, 3D, 6B and 6D

(A and B) Western blotting results of endogenous β_1AR protein from the heart (A) and brain (B) lysates of *Adrb1*+/+, *Adrb1*+/m, and *Adrb1*m/m animals. N=4 mice per group. NS, non-specific band. Quantified results are shown on the right. Related to Figures 2C and 2D

(C) Total mobile time by ANY-maze within 24 hours were calculated in *Adrb1* +/+ (N=20), +/m (N=19) and m/m (N=18) mice. Related to Figure 3A

(D) Total sleep time by EEG/EMG within 24 hours were calculated in *Adrb1* +/+ (N=9), +/m (N=13) and m/m (N=9) mice. Related to Figure 3D

(E) Quantified ratio for photometry fluorescence at ZT1-4 and ZT13-16 from *ADRB1-Cre*; *Adrb1*+/+ (N=8), *ADRB1-Cre*; *Adrb1*+/m (N=7) and *ADRB1-Cre*; *Adrb1*m/m (N=6) mice. Related to Figure 6B

(F) Percentage of *ADRB1*$^{+}$cells that respond differentially to dobutamine treatment in *Adrb1*+/+ (N=7), *Adrb1*+/m (N=5) and *Adrb1* m/m (N=7) brain slices. The bottom table shows the original cell numbers in different categories. Related to Figure 6D

* P<0.05, **P<0.01, *** P<0.001, **** P<0.0001. Two-tailed Student's *t*-test for (A)-(E). Error bars represent ± SEM. Chi-square test for (F).