Drd1-Cre;ROSA26^{td⊤}



Figure S1. *Drd1a* **Driven tdTomato Expression is Present Throughout the SCN. Related to Figure 1.** Five representative coronal sections outlining tdTomato red fluorescent protein expression within the SCN. Scale bar, 100 µm.



Figure S2. Drd1-KO Mice Have a Normal Response to Ligh Induced Phase-shifts or Masking. Related to Figure 3. (A) Representative double-plotted actograms of a 15 minute light pulse at CT 14 and (B) CT 22 (yellow dots indicate time of light pulses in DD). The dark blue line represents an extended regression line derived by activity onsets prior to CNO; the red line follows actual onset of activity after CNO. (C) Quantification of phase shift in response to CT 14 and CT 22 light pulses; p > 0.05, Student's two-tailed t-test; n = 6-9/group. ns: not significant. Data reported as mean \pm SEM. (D) Representative single-plotted actograms comparing the reponses to a 3 hour light pulse from ZT 14 – ZT 17 between wild-type and Drd1-KO mice. White background indicates the time of light exposure. (E) % of total wheel running activity during ZT 14 – ZT 17 on the day prior (light off) and during the 3 hour light exposure (light on).



Figure S3. Drd1 Expression Within the SCN Modulates the Rate of Photoentrainment in Response to a 6-hour Delay in the LD Cycle. Related to Figures 3 and 5. (A) Group analysis of activity offsets comparing wild-type and Drd1-KO mice; F (1,20) = 5.20 ; p = 0.0337, repeated-measures two-way ANOVA with Bonferroni *post hoc* comparison; n = 12/group. * p < 0.05, ** p < 0.01, *** p < 0.001. (B) Frequency of mice per number of days required to entrain wheel running activity to the new light cycle. (C) Rate of entrainment calculated by the slope of activity offsets; repeated-measures two-way ANOVA with Bonferroni *post hoc* comparison, *** p < 0.001. (D) Group analysis of activity offsets comparing Drd1-KO-ChR2 and Drd1-KO-Rescue mice; F (1, 12) = 23.33; p = 0.0004, repeated-measures two-way ANOVA with Bonferroni *post hoc* comparison; n = 8/group. * p < 0.05, *** p < 0.001. (E) Frequency of mice per number of days required to entrain wheel running activity to the new light cycle; repeated-measures two-way ANOVA with Bonferroni *post hoc* comparison; n = 8/group. * p < 0.05, *** p < 0.001. (E) Frequency of mice per number of days required to entrain wheel running activity to the new light cycle. (F) Rate of entrainment calculated by the slope of activity offsets; repeated-measures two-way ANOVA with Bonferroni *post hoc* comparison; n = 8/group. * p < 0.05, *** p < 0.001. (E) Frequency of mice per number of days required to entrain wheel running activity to the new light cycle. (F) Rate of entrainment calculated by the slope of activity offsets; repeated-measures two-way ANOVA with Bonferroni *post hoc* comparison, *** p < 0.001. Data are represented as mean \pm SEM.



Figure S4. SCN or NAc-targeted Retrograde Tracing. Related to Figure 6. (A) Fluorescent immunohistochemistry with TH (green) and retrobeads (red) in DA neuron populations after retrobead delievery to the SCN. No colocalization was observed in the olfactory bulb (OB), zona incerta (ZI), substantia nigra pars compacta (SNc) or retrorubral field (RRF). Scale bar, 50 µm. (B) Coronal diagram indicating retrobead target sites to the nucleus accumbens (NAc). Red dots specify positive NAc targeting. (C) Pseudo-colored images of retrobeads (beads; red) and DAPI (blue) confirming targeting of the NAc. Scale bar, 100 µm. (D) Fluorescent immunohistochemistry of retrobeads (red) colocalized within TH (green) positive neurons in the VTA. Scale bar, 50 µm.



Figure S5. VTA-DA Neuron Activation Mediated c-Fos Expression and Behavioral Responses. Related to Figure 7. (A) Double-fluorescent immunohistochemistry with mCherry (top) and c-Fos (middle) antibody labelling within the VTA, 120 minutes after CNO (1 mg/kg) injection to DAT-Cre-mCherry (left) or DAT-Cre-hM3Dq (right) mice. Pseudo-colored images (mCherry: red and c-Fos: green) are merged at the bottom panel. IPN: interpeduncular nucleus. Scale bar, 100 μ m. (B) High magnification images of fluorescent immunohistochemistry with mCherry, c-Fos, and DAPI labelling within the VTA, 120 minutes after CNO (1 mg/kg) injection to DAT-Cre-hM3Dq mice. Pseudo-colored images (mCherry: red, c-Fos: green, and DAPI: blue) are merged at the bottom panel. Scale bar, 50 μ m. (C). Representative double-plotted actograms of a 6-hour light cycle advance followed by immediate release to DD comparing DAT-Cre-hM3Dq mice treated with an i.p. injection of saline (blue dot) or CNO (green dot; 1mg/kg). Red lines outline ZT 12 prior to LD shift. (D) Group analysis of day 1 and day 2 cumulative phase shift; F (1, 18) = 0.1291; p = 0.7235, repeated-measures two-way ANOVA; n = 10/group. Data are represented as mean \pm SEM.

Reference Figure	Genotype	Free-Running Period (h)	Phase Angle of Entrainment (h)	% of Light Activity
Figure 2	Drd1-Cre-mCherry	23.63 ± 0.44	-0.08 ± 0.5	3.72 ± 1.42
	Drd1-Cre-hM3Dq	23.57 ± 0.06	-0.03 ± 0.02	1.96 ± 0.44
Figure 3	Wild-type	23.61 ± 0.04	$\textbf{-0.07} \pm 0.02$	3.25 ± 0.64
	Drd1-KO	23.57 ± 0.04	$\textbf{-0.08} \pm 0.02$	3.86 ± 0.57
Figure 5	Drd1-KO-mCherry	23.52 ± 0.12	$\textbf{-0.04} \pm 0.02$	3.34 ± 0.85
	Drd1-KO-Rescue	23.54 ± 0.06	0.05 ± 0.04	6.94 ± 2.22
	Drd1-KO-ChR2	23.52 ± 0.04	-0.06 ± 0.02	3.60 ± 1.08
Figure 7	DAT-Cre-mCherry	23.56 ± 0.05	-0.05 ± 0.04	6.76 ± 2.22
	DAT-Cre-hM3Dq	23.53 ± 0.04	-0.04 ± 0.04	4.40 ± 1.58

Table S1. Phenotypic Characteristics of Experimental Mice. Related to Figures 2, 3, 5 and 7. Viral treatment had no effect on free running period in constant darkness, the phase angle of entrainment, or % of total wheel running activity during the light period in a LD cycle. Figure 2; Drd1-Cre-mCherry vs Drd1-Cre-hM3Dq: Free-running period: p = 0.44, n = 7-9/group; Phase angle of entrainment: p = 0.40, n = 7-9/group; % of light activity: p = 0.27, n = 7-9/group. Student's two-tailed t-test. Figure 3; Wild-type vs Drd1-KO: Free-running period: p = 0.46, n = 16/group; Phase angle of entrainment: p = 0.65, n = 24/group; % of light activity: p = 0.22, n = 26/group. Student's two-tailed t-test. Figure 5; Drd1-KO-ChR2 vs Drd1-KO-mCherry vs Drd1-KO-Rescue: Free-running period: F(2, 19) = 0.03667, p = 0.9641, n = 6-8/group; Phase angle of entrainment: F(2, 30) = 0.07922, p = 0.9240, n = 8-13/group; % of light activity: F(2, 27) = 1.976, p = 0.1582, n = 8-12/group; One-way ANOVA. Figure 7; DAT-Cre-mCherry vs DAT-Cre-MM3Dq: Free-running period: p = 0.77, n = 5-7/group. Phase angle of entrainment: p = 0.88, n = 8-9/group. % of light activity: p = 0.40, 8-9/group. Student's two-tailed t-test. Data are represented as mean \pm SEM.